

## ANNOTATION

**dissertation work of Tulepova Gulmira Kairbekovna  
on the topic “Development of a nano - platform for a diagnostic drug  
brucellosis in cattle” submitted for the degree of Doctor of Philosophy (PhD)  
in the educational program 6D120100 – «Veterinary medicine»**

**Relevance of the research topic.** One of the directions for the development of a safe diagnostic kit and brucella vaccine is the production of plant-based brucella antigens. Each cell of the plant is a capsule with the target substance, surrounded by a strong cell wall that protects it from the enzymatic systems of the body. After eating such a plant in its raw form, the animal's body forms an active acquired immunity against the disease. The use of plant systems is a promising direction in the development of protein production technology. Plant systems provide the possibility of both stable expression of a foreign gene due to the transformation of nuclear DNA and fast, highly efficient production of the desired protein due to extra-chromosomal amplicons.

The obtained brucellosis antigens further serve as the basis for the production of a recombinant vaccine against brucellosis in cattle. Plant systems for the production of recombinant vaccines are an attractive alternative compared to other eukaryotic systems due to their low cost, high productivity and safety, since they do not contain viruses pathogenic to humans and animals.

An animal with brucellosis produces antibodies against a field strain of the brucellosis pathogen. Vaccinated animals also produce antibodies against the Brucella vaccine strain. However, diagnostic preparations existing on the world market are not capable of differentially detecting antibodies developed to the vaccine strain from antibodies developed to the field brucellosis strain. As a result, vaccinated animals are diagnosed as sick and the animal is slaughtered according to veterinary requirements.

Since the existing diagnostic kits do not have specificity and are not able to detect antibodies to all strains of brucellosis, and do not allow differential diagnosis of vaccinated animals from patients, a high risk of total infection of cattle is created. As a result, early diagnosis is vital to quarantine to prevent further spread.

**This dissertation study aimed** to perfect the methods of creating a platform by developing brucellosis antigens in plants for a diagnostic kit against cattle brucellosis.

**Research objectives:**

1. Isolation of DNA genome from Brucella vaccine strain B.abortus rb19.
2. Determination of the nucleotide sequence of surface antigens of brucellosis according to the gene bank.
3. Isolation and preparation of surface Brucella antibodies Omp25 and Omp16 for cloning into bacterial cells.
4. Vectoring of plant virus products of modified capsid proteins.

5. Insertion of the surface brucellosis antigens Omp25 and Omp16 into the VLP platform.

**Materials and research methods.** Dissertation work used methods for isolating DNA genome; PCR – amplification; molecular cloning of DNA fragments; alloyage and sequencing; obtaining and purifying protein expression; isolation of DNA plasmid from bacterial cells; nucleotide sequence of surface antigens of brucellosis; purification cloning and subcloning of brucellosis surface antigens; cloning of viral vectors for expression in plants; metal chelate affinity chromatography method; fluorescence polarization analysis; methods for obtaining VLPs/antigenic peptides of virus particles in plants.

**The main provisions for defense.** Preparation of a VLP platform by combining the gene of grapevine virus A and surface antigens of brucellosis (Omp25 and Omp16).

For the first time, based on a special method, a nano – platform has been created that provides rapid and accurate screening by detecting antibodies against the surface antigens of all three brucella vaccine strains rb19, rb51 and rb82. A nano – platform was created by expressing the gene encoding surface antigens in plants.

As a result of the research work, a 1 patent of Republic of Kazakhstan was obtained for one useful model “Method for obtaining brucellosis antigen for diagnostics and prevention of brucellosis in farm animals”. “National Institute of Intellectual Property” RSE №35533. 25.02.2022.

**Description of the main results of the study.** Judging from the introduction to the dissertation, there is sufficient approbation of research results in form of 8 scientific publications, including 3 in the proceedings to international scientific conferences, 3 of those published in publications, by the Recommendation Committee of the Ministry of Education and Science of the Republic of Kazakhstan, 1 in a journal, included into Scopus database «Advancements in Life Sciences» – International Quarterly Journal of Life Sciences. ISSN 2310-5380. Volume 11. Issue 2-May 2024-Pakistan (44%) and 1 patent of RK.

**Justification of the novelty and importance of the results obtained.** Date obtained in the course of scientific and industrial work “Kazakh national agrarian research university specialty of 6D120100 – “Veterinary medicine”, recommendations were made on effective measures to introduce into the educational process of doctoral students on the subject of “Immunogenetics during infectious diseases of animals and to create vaccines against brucellosis of cattle”.

**Relevance to scientific development directions or state programs.** Scientific research work №101 within the framework of the Grant Funding Program for Scientific Research in 2018-2020, subject: “Development of the ViroN-Brucella diagnostic kit for treatment of brucellosis of cattle in the territory of the Republic of Kazakhstan” 217-implemented according to the budget program project.

**Description of the doctoral student’s contribution to the preparation of each publication.** The dissertation is a completed scientific research work carried out by the author personally and meets the requirements for dissertations for

obtaining the PhD degree of the Ministry of Science and Higher Education of the Republic of Kazakhstan. The publication of the research results in the dissertation work under the author's name, photo materials and conclusions, which determine the accuracy and validity of the results of the work, are proven by the author's certificate of the received innovation patent, which confirms that the experimental work was carried out independently.

**Scope and structure of the dissertation.** The written dissertation consists of 75 pages, including 15 pages of appendixes, containing 11 figures and 6 tables. The list of references includes 119 references. This dissertation represents a large body of work and supplies a wide-ranging survey of recent methods and tools.