

nizing, chest cavity lavage — in a usual microtest tube 1.5 ml. The same samples and one more probe of parenchymatous bodies in a test tube were taken from fresh carcasses for homogenizing with addition 2%-formalin in 1000 µl volume for detection of *Yersinia pestis* capsule antigen (F1). Spinal or bone marrow from birds-of-prey food debris, mummified carcasses, bones were taken in two test tubes for homogenizing (one tube with formalin).

In MLMD ectoparasite taxonomic identification was performed. After the necessary sample preparation the agent express-diagnostics was conducted in all received probes with the subsequent bacteriological examination only the positive samples. 100-µl liquid phase samples from agonizing, dead and birds' pecked animals, ectoparasites found out on them, bone remains were used for ICH-tests (FBUN GNTS PMB, Obolensk). All samples were examined by real-time PCR on a Rotor Gene Q instrument (Qiagen, Germany) and RNGA-RNAt. Specific fragments of *Y. pestis* DNA were amplified from all 39 ICH-positive samples at early cycles (since the 5th) and *Y. pestis* cultures were isolated. In total 60 positive responses were received in PCR including 50 replies that were confirmed by F1 detection in RNGA-RNAt, 47 *Y. pestis* subsp. *pestis* cultures were isolated.

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CLONING OF THE YERSINIA PESTIS TRANSALDOLASE GENE

A.L. Trukhachev, M.G. Meloyan, I.E. Arsenyeva, S.A. Lebedeva

Rostov-on-Don Plague Control Research Institute, Rostov-on-Don, Russia

One of the antigenic complexes of the plague pathogen is fraction V (FV) (Bozhko et al., 2006). Investigation of the components of FV using two-dimensional protein electrophoresis, Western blott and mass-spectrometric analysis allowed determining that the composition of FV includes transaldolase, which has immunochemical activity with the monoclonal antibodies against FV (Arsenieva et al., 2017; Trukhachev et al., 2017). Transaldolase is an enzyme of the pentose phosphate cycle. The characteristics of the *Y. pestis* transaldolase and the *Y. pseudotuberculosis* transaldolase are similar. Perhaps, transaldolase of pathogenic *Yersinia* is the "moonlighting protein" (González-Rodríguez et al., 2012, He Y. et al., 2015).

The aim of the study was the cloning of the transaldolase gene of *Y. pestis* (*talB*) into a high-copy vector. After amplification of DNA with talF- and talR-primers, the 1059-bp PCR fragment including the *talB* gene of *Y. pestis* was cloned into the corresponding sites of pGEM-T using the set of reagents pGEM[®]-T Easy Vector Systems (Promega), resulting in pGEM-T-tal. Transformation of *E. coli* strains was performed using standard protocols (Sambrook J., 2001). The recombinant clones was selected on LB agar containing 100 µg/ml ampicillin, 0.5 mM IPTG and 80 µg/ml X-Gal. The few selected recombinant clones were detected insert of about 1000 nucleotide pairs in the plasmid vector with help the method extraction plasmid DNA (Kado et al., 1981). Analysis of the recombinant DNA of these strains using talF-, talR- and pT7-primers in PCR showed that there was an embedding of a fragment containing a *talB* under the control of the T7 promoter. The resulting recombinant *E. coli* pGEM-T-tal strain which carried the plasmid containing the *Y. pestis talB* gene reacted with the horse hyper immune antiplague serum and serum from rabbits

immunized against FV in the gel precipitation reaction. The control strain containing only the vector plasmid pGEM-T didn't react with the serum. The *Y. pestis EV* strain served as a positive control.

Thus, recombinant strain of *E. coli* pGEM-T-tal containing gene of immunologically active *Y. pestis* transaldolase in plasmid was obtained. A tool for further study of immunogenic and protective properties of one of the components of FV *Y. pestis* was created.

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THE PRESENT EPIDEMIOLOGICAL CHARACTERISTICS OF YERSINIOSIS IN THE RUSSIAN FEDERATION

E.A. Voskresenskaya¹, G.I. Kokorina¹, E.B. Ezhlova², Yu.V. Demina², N.D. Pakschina², O.N. Skudareva²

¹St. Petersburg Pasteur Institute, St. Petersburg, Russia;

²Federal Service for Surveillance on Consumer Rights Protection and Human Wellbeing, Moscow, Russia

The objective of this study was to carry out a retrospective analysis of pseudotuberculosis and intestinal yersiniosis surveillance data in the Russian Federation during the period 2013–2015.

Federal statistical observation data and federal subjects of Russia data were analyzed.

Presently the intestinal yersiniosis prevails in the etiological structure of yersiniosis, with proportion of 60%. A statistically significant decreasing trend in incidence is observed, the annual average incidence of pseudotuberculosis and intestinal yersiniosis per 100 000 population is 0.82±0.05 and 1.90±0.11 correspondingly.

The intensity of the epidemic process of yersiniosis varies greatly across different regions of the country. The registration of pseudotuberculosis is noted in approximately 49% of the entities of the Russian Federation, intestinal yersiniosis is registered more evenly — in 77% of the entities. The maximum incidence of yersiniosis is noted in a number of the entities of the North-West Federal District, the Siberian Federal District, the Far Eastern Federal District, where morbidity rates exceeded the federal average rate by 2–15 times.

The proportion of outbreak morbidity of pseudotuberculosis decreases, sporadic cases prevail. For the time period 2013–2015 10 outbreaks with a total of 110 diseased persons were reported. The incidence of intestinal yersiniosis is sporadic.

In the age structure of patients with pseudotuberculosis children predominate (65%) mainly in the age group 3–6 years (32%). In 2015 incidence in this age group was 5.2 per 100 000 persons, this is 17 times higher compared with adults and 2 times higher compared with children in the age group 1–2, 7–14 years. The ratio of children and adults with intestinal yersiniosis is practically 1:1 — 45 and 55%. The maximum incidence is noted among children in the age group 1–2, 3–6 and 7–14 years (3–32%) — 3.5, 3.4, 2.9 per 100 000 persons respectively. The incidence among adults was lowest (0.8 per 100 000 persons).

During this epidemiological study it was shown what the pathogens are principally transmitted to humans through fresh vegetables (11–61%) and fruits (3–32%). Thus meat and meat products, milk and dairy products are often not investigated as the possible sources of intestinal yersiniosis infection. The diagnosis is confirmed mainly by the serological methods — 49–91% of cases, by the PCR — only 1–15%.