References

1. Fu, Y., Dou, Q., Smalla, K., Wang, Y., Johnson, T. A., Brandt, K. K., Mei, Z., Liao, M., Hashsham, S. A., Schäffer, A., Smidt, H., Zhang, T., Li, H., Stedtfeld, R., Sheng, H., Chai, B., Virta, M., Jiang, X., Wang, F., Zhu, Y.-G., & Tiedje., J. M. (2023). Gut microbiota research nexus: One Health relationship between human, animal, and environmental resistomes. *mLife,* 2, 350–364. https://doi.org/10.1002/mlf2.12101
2. Lu, S., Na, K., Li, Y., Zhang, L., Fang, Y., & Guo, X. (2022). Bacillus-derived probiotics: metabolites and mechanisms involved in bacteria-host interactions. *Crit Rev Food Sci Nutr, 6*, 1–14. https://doi.org/10.1080/10408398. 2022.2118659
3. Safronova L. A., Pylypiuk Y. V., Skorochod I. O., Polova Z. M. (2024). Probiotics and their potential for the prevention and treatment of infections. *Microbiological journal.* 6, 74–91. https://doi.org/10.15407/microbiolj86.06.074

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MONITORING OF BACTERIAL DISEASE OUTBREAKS IN RAINBOW TROUT (*ONCORHYNCHUS MYKISS*) TO IMPROVE PREVENTIVE STRATEGIES FOR THE NATIONAL AQUACULTURE

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Abstract. During monitoring studies of trout farms 3 strains of *A. salmonicida* (A.salmUA1/2025RTV, A.salmUA1/2026WFV and A.salmUA2/2026BTCh), 2 strains of *F. psychrophila* (F.psyUA1/2025RTV and F.psyUA1/2026RTCh) and one strain of *Y. ruckeri* bacteria (Y.ruckUA1/2026RTZ) were isolated from rainbow trout, whitefish and brook trout. Using PCR, fragments of species-specific genes and the 16S rRNA gene of the isolated bacteria were amplified for further genetic characterization. Development of inactivated autogenous vaccines for oral administration based on isolated bacterial strains will help to decrease antibiotic use at the local level and prevent economic losses of trout farms in Ukraine.
Keywords: bacterial diseases, aquaculture, PCR, 16SrDNA, vaccine, rainbow trout.

Today, aquaculture is the most dynamic and fastest growing sector of the animal agriculture and is crucial for food security in many countries. Like other livestock sectors, aquaculture requires adequate veterinary support and control of infectious diseases (Buchatsky et al., 2020). Vaccination of fish with locally developed autogenous vaccines can provide a local and cost-effective means of reducing mortality in aquaculture facilities, as well as contributing to the rational use of antibiotics. A comprehensive scientific and methodological approach to assessing the possibilities of introducing autogenous vaccination in national aquaculture will allow using a number of key advantages of immunobiotechnological tools in terms of efficiency and flexibility of animal welfare, transboundary biosecurity, and the economy of local farms (Rud et al., 2022).

Pathogenic bacteria from the genera *Aeromonas*, *Flavobacterium* and *Yersinia* cause morbidity and high mortality in salmon farming, which encourages producers to constantly use antimicrobial drugs for therapeutic and prophylactic purposes. The lack of immunoprophylactic agents for aquaculture on the market and the constant growth of production volumes require comprehensive solutions to improve the epizootic situation in fish farms. Monitoring studies of trout farms and assessment of the epizootic situation are very important for the biosecurity of the industry and make it possible to respond in a timely manner to the outbreaks (Vinh et al., 2026). However, for the prevention and control of infectious diseases in aquaculture, it is important to introduce immunoprophylactic agents that can solve the global problem of antimicrobial resistance at the local level. Therefore, the goals of our research were to monitor the main bacterial diseases of rainbow trout in national aquaculture and select strains of pathogenic microorganisms for the development of inactivated autogenous vaccines.

In our monitoring studies, we focused our efforts on the identification of three main pathogenic bacteria: namely the causative agent of bacterial cold water diseases (BCWD) and rainbow trout fry syndrome (RTFS) bacteria *Flavobacterium psychrophila*; the causative agent of furunculosis *Aeromonas salmonicida*; and the causative agent of enteric red mouth disease or "yersiniosis" – *Yersinia ruckeri*. For this, we used selective media ECA, FLPA, BHIA, TSA+CBB and SWA. Gram stain and PCR methods were used to identify the strains. We also applied the diffusion disk method to determine susceptibility to the main antibiotics allowed in aquaculture.

Identification of *A. salmonicida, F. psychrophila* and *Y. ruckeri* bacteria was carried out in rainbow trout (*Oncorhyncyus mykiss*) and American char (*Salvelinus fontinalis*) with characteristic external clinical signs of diseases. To isolate *A. salmonicida* strains, trout or char with boils (furuncles) on the side or in the tail part of the body were selected. *F. psychrophila* bacteria were isolated from rainbow trout fry from ulcers formed in the form of a "saddle" behind the dorsal fin and from internal organs. In the case of *Y. ruckeri* bacteria, characteristic external and internal hemorrhages, exophthalmos and redness of the oral cavity in rainbow trout were recorded. For molecular diagnostics of bacteria, the target genes 16S rRNA, VapA and gyrB for *A. salmonicida*, the ompA gene for *Y. ruckeri* and the 16S rRNA gene for *F. psychrophila* were used.

As a result of our research conducted in 2025–2026, 3 strains of *A. salmonicida* bacteria (A.salmUA1/2025RTV, A.salmUA1/2026WFV and A.salmUA2/2026BTCh) were isolated from rainbow trout, whitefish (*Coregonus sp*.) and brook trout, respectively, two strains of *F. psychrophila* bacteria (F.psyUA1/2025RTV and F.psyUA1/2026RTCh) were isolated from rainbow trout and one strain of *Y. ruckeri* bacteria (Y.ruckUA1/2026RTZ) was isolated from rainbow trout. Using PCR, fragments of species-specific genes and the 16S rRNA gene of the isolated bacteria were amplified for further genetic characterization.

The isolated bacteria *A. salmonicida, F. psychrophila* and *Y. ruckeri* were characterized by a significant level of antibiotic resistance, which indicates an incorrect approach to the treatment of infectious diseases by national producers, who often do not turn to specialized laboratories, but select a treatment method empirically. Most often, the studied bacteria were resistant to oxytetracycline, trimethoprim and sulfamethoxazole. Since the rational use of antibiotics is a global public health issue, in which attention is focused on the supply of food products within the framework of the "One Health" concept, the creation of inactivated autogenous vaccines for oral administration based on isolated bacterial strains will help to solve the global problem of antimicrobial resistance at the local level and prevent economic losses of trout farms in Ukraine.

References

1. Buchatskyi L.P., Rud Yu.P., Matvienko N.M. Viral infections of sturgeons and salmon. Kyiv: DIA, 2020. 240 p. ISBN 978–617–7785–10–0.
2. Rud Yu.P., Zaloilo O.V., Zaloilo I.A., Buchatskyi L.P. Antiviral vaccines for fish (review) / Ribogospod. nauka Ukr. 2022. No. 3(61). P. 71—104. https://doi.org/10.15407/fsu2022.03.071
3. Vinh, N. T., Dong, H. T., Turner, J. K., Senapin, S., & Turner, W. A. (2026). Back to basics: locally produced vaccines offer a practical alternative to antibiotics for prevention of streptococcosis in farmed tilapia (*Oreochromis* spp.). Frontiers in Veterinary Science, 13. https://doi.org/10.3389/fvets.2026.1740757

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DIAGNOSTIC CAPABILITIES OF MALDI-TOF MS IN THE IDENTIFICATION OF RARE BACTERIAL ISOLATES

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Abstract. MALDI-TOF mass spectrometry has transformed clinical microbiology by enabling rapid, highly accurate identification of microorganisms based on unique protein spectral profiles, overcoming the limitations of conventional biochemical panels when applied to atypical strains. To characterize rare bacterial isolates identified by MALDI-TOF MS and assess their clinical significance. Methods: Clinical specimens (blood, sputum, wound discharge, urine, respiratory material) collected in 2025/2026 at the Center for Microbiological Research, Danylo Halytsky Lviv National Medical University, were analyzed using a Bruker MALDI-TOF mass spectrometer with reference database comparison. Results: Six rare pathogens were identified – *Elizabethkingia meningoseptica*, *Chryseobacterium indologenes*, *Ochrobactrum anthropi*, *Myroides odoratimimus*, *Sphingomonas paucimobilis*, and *Kocuria kristinae* – many of which are difficult to distinguish from more common species by conventional culture-based methods, risking misclassification and diagnostic delay.
Keywords: MALDI-TOF, rare pathogens, microorganism identification, clinical microbiology.

Introduction. The introduction of matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) into clinical microbiology practice has opened fundamentally new possibilities for rapid and highly accurate identification of microorganisms. Unlike conventional biochemical panels, which often prove uninformative or misleading when applied to strains of pathogens with atypical properties, MALDI-TOF enables identification of microorganisms based on a unique protein spectral profile considerably faster than conventional methods.

Aim. To characterize rare isolates recovered from clinical material and identified by MALDI-TOF mass spectrometry, and to assess their clinical significance.

Materials and methods. The study was conducted at the Center for Microbiological Research of Danylo Halytsky Lviv National Medical University using a MALDI-TOF mass spectrometer (Bruker, Germany). Strain identification was performed by comparing the acquired spectral profiles with a reference database. The material comprised clinical specimens (sputum, blood, wound discharge, urine, and respiratory material) collected during 2025/2026.