EPIZONE short – term mission scientific report Aleksandra Makuch

Host Institute: Agence Nationale de Sécurité Sanitaire (ANSES), Ploufragan, France Home Institute: National Veterinary Research Institute (NVRI), Puławy, Poland Period of research stay: 03.09.2024 – 02.10.2024

Purpose of the visit

The purpose of the short – term mission was to improve the management of the strategy for control, prevention and laboratory diagnosis of animal botulism cases in Poland. The realization of this purpose is made possible by getting acquainted with the principles of botulinum cluster gene detection, taking into account the specificity of samples and cultures subjected to analysis using molecular biology methods. It was extremely important for the task to familiarize oneself with methods of detecting *C. botulinum* group III, distinguishing between mosaic toxotypes, which are increasingly noted as a contributing factor to outbreaks of botulism in animals. The results of the visit to Agence Nationale de Sécurité Sanitaire in France will allow the development of a strategy for botulism management and handling of monitoring samples and samples from animal botulism cases, as well as the establishment of a reference laboratory in Poland and participation in ILC and PT testing in the future.

Work carried out during the visit

Botulism is a rare neuroparalytic disease in humans as well as wild and farmed animals. The disease is caused by exposure to botulinum neurotoxins (BoNTs) - the most potent naturally occurring toxins in the environment, produced by anaerobic spore-forming bacteria belonging to the *Clostridium botulinum* species and other bacteria of the *Clostridium* genus capable of producing botulinum toxins. These microorganisms pose a significant problem, especially among animals raised on farms, due to their potential to cause serious economic losses resulting from the high animal mortality observed during botulism outbreaks. In Poland, botulinum toxin poisoning of animals remains an underestimated but increasingly common problem. Over the past few years, a significant increase in the animal botulism outbreaks has been observed in our country, affecting mainly poultry farms and waterfowl. In the last year, economic losses caused by botulinum toxin poisoning are estimated at tens of thousands of animals. The

mandatory reporting of botulism cases prevents the collection of adequate data suggesting potential sources and the detailed number of affected animals, which is the main reason for the underestimation of the scale of the disease.

The present short-term mission was aimed at gaining experience from world-renowned specialists, necessary to develop at NVRI a strategy for the management and monitoring of cases of botulism in animals. During my fellowship at ANSES, I became familiar with the methods of collecting field samples and the principles of culturing anaerobic microflora from biological material from suspected cases of botulism. A particularly important step was the discussion of the most effective microbial substrates for obtaining the best possible expression of genes that determine the production of botulinum neurotoxins. During my fellowship I became familiar with methods of detecting C. botulinum group III, including the differentiation of mosaic types of toxins, which are increasingly noted as a contributor to botulism outbreaks in animals. I also had the pleasure of giving a presentation titled "Introduction to botulism management strategy elaboration at NVRI in Puławy". During the presentation, I discussed the structure of my home institute and the scope of research on botulism cases in Poland. The final stage of the fellowship was a hands-on review with laboratory staff at ANSES of a preliminary strategy for botulism management and handling of monitoring samples and samples from animal botulism cases, taking into account species specificity and biosafety elements, as well as sample transport. This strategy will be developed and implemented at NVRI to standardize procedures for handling samples from animal botulism outbreaks. I express confidence that my fellowship will allow me to use the skills I have acquired in gaining international collaborations and in my future career.

> Aleksandra Makuch Puławy, 23.10.2024

