

PUBLIC LECTURE EVALUATION

Masaryk University

Faculty	Faculty of Science
Procedure field	Molecular Biology and Genetics
Applicant	Mgr. Petr Králík, Ph.D.
Lecture date	September, 26, 2025
Lecture topic	Proč potřebujeme detekovat tolik mikroorganismů současně? (Why we need to detect so many microorganisms simultaneously?)
Persons present (number)	32
Designated evaluators (board members)	prof. RNDr. Jiří Doškař, CSc. on site doc. Mgr. Monika Vítězová, Ph.D. on site prof. RNDr. Aleš Knoll, Ph.D., on line prof. MVDr. Alois Čížek, CSc. on line

In the opening part of the lecture, Dr. Králík addressed three major topics relevant to the application of PCR-based techniques in microbial detection and quantification. First, he discussed the limitations and challenges of data interpretation when qPCR is applied for quantifying microorganisms in food matrices and in animals, particularly in relation to decision-making processes. Second, he introduced the methods for viability detection of microorganisms using qPCR, highlighting how they can distinguish live from dead cells. Finally, he focused on multiplex detection of dozens of pathogens in food matrices and clinical samples. For each of these areas, Dr. Králík briefly outlined the background, summarized the current state-of-the-art, and emphasized how the topics are interconnected within microbial diagnostics in food and feed matrices, in animals, and in clinical samples. He also provided a concise introduction to paratuberculosis infection and its causal agent, *Mycobacterium avium* subsp. paratuberculosis (MAP). In this context, Dr. Králík presented an overview of existing knowledge on paratuberculosis in animals, described the main routes of transmission, and outlined the debated relationship between MAP and Crohn's disease.

In the second part of the lecture, Dr. Králík concentrated on multiplex DNA detection of pathogens by various technologies, with particular emphasis on their strengths and weaknesses. He then moved on to describe multiplex methods based on ligation assays and the visualization of ligation products using suspension arrays. He traced the historical development of multiplex ligation approaches, explained the use of different ligation strategies, and described the conditions required for hybridization of amplified ligation products onto microspheres in suspension arrays. After summarizing the methodological principles of ligation assays, Dr. Králík illustrated their practical application in multiplex pathogen detection and provided concrete case examples that demonstrated their utility and robustness.

In the final part of his presentation, Dr. Králík discussed the future prospects of multiplex technologies, drawing on current, though as yet unpublished, research results. He suggested several potential directions for further development and highlighted their possible impact on the future of microbial diagnostics.

At the conclusion of the lecture, Dr. Králík responded to selected questions raised by the opponents in their assessments of his habilitation thesis, further clarifying key aspects of his research and practical applications.

In the following discussion Dr. Králík answered the questions of the board members and the attendees:

1. Will the MOL-PCR method work even if multiple pathogen species are present in one sample? Bude metoda MOL-PCR funkční i v případě přítomnosti více druhů patogenů v jednom vzorku?)
2. When validating the method, did you first test individual pathogens and then their mixtures? (Při validaci metody jste testovali nejdříve jednotlivé patogeny a pak jejich směsi?)
3. What standards are used for method validation? (Jaké standardy se využívají pro validaci metod?)
4. How do you address strain variability within a species during detection? (Jak řešíte variabilitu kmenů v rámci druhu při detekci?)
5. Do you perform the validation process repeatedly when changing the chemical used? (Provádíte validační proces opakovaně při změně používané chemikálie?)

Conclusion

The lecture delivered by Petr Králík entitled “Proč potřebujeme detekovat tolik mikroorganismů současně? (Why we need to detect so many microorganisms simultaneously?)” and delivered as part of the habilitation procedure, **demonstrated** sufficient scholarly qualifications and pedagogical capabilities expected of applicants participating in a habilitation procedure in the field of Molecular Biology and Genetics.

The lecture took place on-site at 10.00 a.m. – 11.30 a.m. The above-mentioned members of the board attended the lecture and provided its evaluation. All designated evaluators are familiar with the text of the evaluation and agree with it.

Date: September, 26, 2025

Jiří Doškař

Monika Vítězová

Aleš Knoll

Alois Čížek