Institute: National Research Institute of Animal Production

Title: Analysis of myogenesis process to improve production efficiency and product quality for in vitro produced artificial meat.

Name of potential supervisor: dr hab. Katarzyna Piórkowska, prof. IZ

Background information:

The advancement of cell culture techniques and knowledge make cultured meat approaches more real. Based on the cellular ability to differentiate and self-renew, three-dimensional structures are often constructed and composed from cells of animal muscle tissue origin.

The advantages of the artificially grown meat support approach to the industrial use of laboratory techniques. Artificial meat is associated with less environmental pollution, including lower use of raw materials and available land; and lowered waste production. In addition, synthetic meat is related to improving the quality of the product itself in terms of sensory (e.g. enrichment with omega-3 acids), medicinal and technological features. Artificial meat is safer because it is not associated with the zoonotic disease. And it is also available to vegans, including supplementary role to non-meat products.

As part of the proposed research, appropriate culture techniques will be developed: different cell phenotype composition, the composition of the culture medium, time and parameters of incubation, the suitable carrier for cells in vitro culture, which can compete with traditional meat production techniques. Nevertheless, the production of "artificial meat" requires multi-disciplinary research in cell biology, myogenesis processes and pre-industrial biotechnology, which the proposed dissertation intends to combine.

The main question to be addressed in the project:

The study aims to verify the techniques of myogenic cells culture and their influence on cell differentiation and the formation of muscle fibres *in vitro* conditions.

The proposed approach will answer the questions about the *in vitro* myogenesis process, its changes in gene expression in a strictly defined pool of input cells. The possibility of controlling these processes might reduce costs, help obtain a higher quality product, safer for the consumer. Furthermore, the heterogeneous cell mixture is assumed to supplement the nutritional and signalling needs of the cultured cells to reduce production costs.

The analysis of reliable markers of myogenesis processes allows for the easy manipulation of cell composition and the description of the proper culture conditions. Also, based on the expanded RNA expression profile of cells, the final product - "artificial meat" - will be assessed in terms of quality features, including chemical composition and selected sensory and technological features. Moreover, the research approach will include analysing the influence of the expression profile on the differentiating pseudo-tissue, identifying individual genes and signalling pathways that can potentially have a beneficial effect on forming "artificial meat" in the *in vitro* process.

Information on the methods/description of work:

The planned analyses will be carried out at the Department of Animal Molecular Biology of the National Research Institute of Animal Production or at cooperating research centres. The starting point for the cultivation and differentiation of cell co-cultures will be well-defined primary myogenic cell lines of the *Bos taurus* species banked in the ZBMZ IZ-PIB collection or obtained from slaughter material. The total size of the collection will include five individuals, gradually included in the research procedures. For cultivation on large-area flasks on various cell adhesion carriers, co-cultures of myogenic and adipogenic cells and cells with high secretory abilities will be carried out [incl. fibroblasts]. The benchmark for these procedures will be high-throughput spinner-flask suspension cultures, where there will be an attempt to integrate those two approaches in the "artificial meat" culture in subsequent steps.

Quality control will analyse changes in the expression of selected genes in the *in vitro* production process, where available protein markers will be controlled. Depending on the type of protein and research needs, it will be examined using classic proteomic methods such as western-blot, immunofluorescence techniques (including ELISA), and flow cytometer.

Additional information (e.g., special requirements from the candidate):

The potential candidate should have extensive knowledge of molecular genetics and/or cell biology and biochemistry and/or bio-material sciences. Basic molecular techniques, including RNA isolation and PCR techniques, should be done with ease. A high level of written and spoken English is a requirement.

Experience in cell cultures techniques is an asset, as well as experience with bio-compatible materials.

Place/name of potential collaborator: -

References:

Sissel Beate Rønning (Editor), 2019; „**Myogenesis, Methods and Protocols**." Methods in Molecular Biology, Humana Press, ISBN 978-1-4939-8896-9 ISBN 978-1-4939-8897-6 (eBook) https://doi.org/10.1007/978-1-4939-8897-6.

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