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Doctoral dissertation review of Maima Matin, MSc entitled „Nutraceutical Intervention with Ginger (Zingiber officinale) Modulates Age-Related Liver Changes in Mice"

The author of the doctoral dissertation is Maima Matin, who completed her master's degree in biotechnology at the North South University, Dhaka, Bangladesh, and previously completed her bachelor's degree in pharmacy at East West University, Dhaka, Bangladesh. The candidate has not previously applied for a doctoral degree, and the work submitted for review was not the basis for applying for a doctoral degree. The candidate's place of work and professional experience are unknown from the submitted documentation. However, the candidate has attended three scientific conferences (in Poland and Bulgaria), actively participated in research projects, and completed a course for people working with laboratory animals, organized by the Polish Society for Laboratory Animal Sciences (PolLASA). She completed two short-term research internships: Visited Ludwig Boltzmann Institute for Digital Health and Patient Safety at the Medical University of Vienna to learn computational methods and data analysis approaches and visiting abroad in Sofia, Bulgaria at the Department of Biochemical Pharmacology and Drug Design, Institute of Molecular Biology "Roumen Tsanev", Bulgarian Academy of Sciences (IMB-BAS). The candidate's scientific activity was recorded in 35 papers published in foreign journals with a total impressive Impact Factor of 172.31 and a Hirsch index of 9.

Maima Matin Maima Matin's doctoral dissertation was performed at the Institute of Genetics and Animal Biotechnology of the Polish Academy of Sciences in Jastrzębiec, Poland, under research supervisors associate professors Atanas G. Atanasov, PhD and Artur Jóźwik, PhD. The submitted doctoral dissertation **entitled „Nutraceutical Intervention with Ginger (Zingiber officinale) Modulates Age-Related Liver Changes in Mice"** was prepared as a series of three publications: one review, providing theoretical background for the research described in two subsequent original papers. All papers were published in peer-reviewed journals with Impact Factor.

The work was prepared in a typical layout of experimental format, consisting of a title page, statements of supervisors and the author, acknowledgments and a table of contents, which precisely

indicates the subsequent elements of the substantive part of the work. In this part, the author provided the following: a list of abbreviations, a list of publications constituting the doctoral dissertation, an abstract in Polish and English, an introduction in which the author presents the current knowledge on the biological importance of ginger, then presents the research aim and hypotheses as well as specific objectives, the methods, a description of the results together with a graphical presentation, a discussion, a summary, bibliography, appendices and statements of co-authors about the contribution to individual publications.

The work is written in a very accessible style, with a thorough and comprehensive introduction to emphasize the importance and potential of ginger for health, cell physiology, and liver metabolism, while also highlighting areas that remain unexplored. The methods and materials are well-described including technical details. The methods used were very well selected, ensuring complete verification of the hypotheses. The author presents the results in a simple and logical manner, easily interpreted through the graphs, which are thoroughly described, including statistical tools. The discussion is conducted at a very high scientific level, in relation to the latest scientific achievements of other authors. The numerous references (not numbered, about several dozen items) were carefully selected and indicate a thorough understanding of the research topic and the current knowledge in the field of research.

The research is comprehensive and exhaustive, and the reader feels saturated with the topic within the scope of the research. However, in my opinion, some elements or sections could have been presented in a different way, sometimes important information is missing, which is often unnecessarily sought in the author's publications. The interesting research problem addressed raises further questions, which I will soon ask the author myself. Below I present my comments, to which I would like to ask the author of the work to respond:

1. The introduction describes the mechanisms occurring in the liver, mixing examples from humans and mice - are they similar? Similarly, the section “Relevance of Nutritional Interventions in Age-Related Liver Dysfunction” but the diet of humans is different from that of mice. I understand that the importance of supplementing the human diet with ginger is great, but in the context of this work, it could only be a briefly described thread highlighting the potential of translating this research to humans.
2. It would be better to number the hypotheses to make it easier to determine whether the hypothesis was confirmed, e.g. 1, 2 or 3. The work lacks answers to the hypotheses posed
3. The method description states that the study was performed in three replicates. Were these replicates technical or biological? How many mice were used in total? What was the rationale behind the selection of a given mouse strain or dosage? This information is all described in publications, but in the dissertation, this information is also very important and should be included instead of, for example, technical issues related to standard analysis.

4. The study only indicates the positive effects of ginger, but have you analyzed the effect of ginger therapy on other systems, especially the digestive tract, because it is known that it can damage the digestive tract mucosa and cause allergic reactions? Have you examined the gastric mucosa of the tested mice in any way?
5. The given amount of ginger (at two doses: 0.6% and 1.8% ginger by weight) in the diet referred to what? The daily weight of the food or the weight of the mouse? What were your considerations when determining the ginger dosage? Comparing the dose proportionally, for a human weight e.g. 60 kg, it would be between 360 g and 1 kg of ginger per day. To compare this to the realities of human nutrition, this level is impossible to achieve. However, we know that the amount of food a mouse consumes relative to its weight is not proportional to the food consumed by humans. If we consider the amount of food consumed by humans daily, 0.6-1.8% may represent about 6 or 18 g/1 kg of food. However, a mouse consumes about 10-15% of its body weight, which is about nine times more food than a human. I suspect such analyses were considered, but I'd like the author to comment on whether the amount of daily food consumed by mice was taken into account, compared to the amount consumed by humans, which is much smaller. On the other hand, metabolism, diet, and digestion are also different between these species. Was this even relevant, given the clear subject of the work? However, the narrative alludes to the human species.
6. Can the results of studies on mice be applied to the impact on humans - however, we have a different species, different daily food intake, different diet, different flow of substances through the body, different metabolism e.t.c.. Can you say that the same impact will be observed in humans when using the same proportion of ginger daily?
7. The work is complete, very interesting, and provides many important results that can be used for further research, especially in humans. However, the author lacks an explanation of what constitutes novelty in the work, which results are innovative, or which make a new contribution to ginger research.

Considering the enormous amount of conducted research and the obtained results, the above-mentioned comments and shortcomings do not diminish the value of the doctoral dissertation submitted for review, which I rate very highly. I extend my appreciation to the candidate, **Maima Matin** and her supervisors, professors Atanas G. Atanasov and Artur Jóźwik. The presented dissertation clearly proved the practical and development potential of the obtained research results and the significant impact on the economic and social environment by promoting the functional food. Maima Matin's doctoral dissertation presents the candidate's general theoretical knowledge in the field of agricultural sciences, in the discipline of animal husbandry and fisheries, as well as the ability to independently conduct scientific work

I hereby certify that the submitted doctoral dissertation represents a significant contribution to current knowledge of ginger and its biological impact on the organisms and, at the same time, an original solution to a scientific problem. **Maima Matin**'s doctoral dissertation, entitled "**Nutraceutical Intervention with Ginger (Zingiber officinale) Modulates Age-Related Liver Changes in Mice"** meets the requirements specified in Article 187 of the Act of 20 July 2018, the Law on Higher Education and Science. Therefore, I wholeheartedly request that the Scientific Council of the Institute of Genetics and Animal Biotechnology of the Polish Academy of Sciences in Jastrzębiec, Poland admit **Maima Matin** to the next stages of the doctoral process.

dr hab. Wioletta Adamus-Białek, prof. UJK

Signed by / Podpisano przez:
Wioletta Adamus-Białek
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