REFEREE REPORT

to the PhD thesis entitled "Changes in the antioxidative status of developing brain under acrylamide influence with the use of in ovo model"

PhD student: Marta Batoryna

Reviewer: prof. Ing. Adriana Kolesárová, PhD., Department of Animal Physiology, Faculty of Biotechnology and Food Sciences, Slovak University of Agriculture in Nitra

The developing fetus may be exposed to a range of environmental toxins that have longterm consequences for neurodevelopment. Some of the harmful substances are blocked by natural biological barriers, such as the placental and blood-brain barrier. Nevertheless, there is a group of toxins which can cross these barriers and influence the embryo. One of the toxins is acrylamide (ACR). Potential risk of exposure to ACR stems from the prevalence of this toxin in carbohydrate-rich processed food and tobacco smoke, as well as occupational exposure. Therefore, in this study various structures of the brain were examined for the influence of ACR on antioxidative status. The aim of this study was to estimate the impact of ACR on the oxidative status and cytoarchitecture of the developing embryo brain with the use of in ovo model. The experiment was conducted with chicken embryos.

General background of the thesis, written on 12 pages, is focused on Chemical properties of acrylamide, Exposure risk to ACR, Metabolism of ACR, Mechanisms of ACR neurotoxicity, Brain development, Histological differentiation of brain, Brain development and apoptosis, Activity of antioxidant enzymes of the chicken embryo, and Antioxidant system in different brain regions. Rich literature review provides current state of the studied topic at the national and international level and suggests that the PhD student is well oriented in this area. More than 94 literature sources of foreign and domestic authors including literature sources showen in 3 articles (Journal of Environmental Science and Health and The Journal of Microbiology, Biotechnology and Food Sciences) were used in the thesis.

I'd like to highlight the strong aspect of this work - the choice of appropriate contemporary methods to analyze chicken responses to ACR exposure. In the experiments using these methods, the PhD student, Marta Batoryna obtained a large body of interesting results, which prove that studied ACR can modulate certain functions of brain cells. All of the observed effects suggest that ACR has a serious influence on the developing brain through several different mechanisms.

The other strong aspect of the thesis is that the results of her study are published in international journals Journal of Environmental Science and Health and The Journal of Microbiology, Biotechnology and Food Sciences, two of them are Current Contents journals and one in databases Web of Science ansd Scopus.

To the thesis I have following comments for discussion, objections and questions:

- 1) Which examined brain structures are highly vulnerable?
- 2) Please explain the apoptosis signaling pathways with caspase-3 involvement and describe the mechanism of action of ACR in brain cells.
- 3) Do you expect the influence of ACR on brain structures of other animal species, including humans?

The doctoral thesis of Marta Batoryna presents an interesting and actual research in the field of nerve toxicology. Structure and scope of the work conforms to the standards of this kind of manuscripts. In this work rich experimental material was collected and analyzed using appropriate methodology and modern equipment. The study brings a large volume of results that expand knowledge in this field of research. These aspects are the strengths of the present thesis.

However, the attached publications of Marta Batoryna evidences about her potential ability to process, analyze and publish obtained scientific results.

In case of successful defence, I propose to award Marta Batoryna academic degree "philosophiae doctor" (PhD).

Nitra, 26.06.2019

prof. Ing. Adriana Kolesárová, PhD.