REVIEW

Position on the Dissertation of Ilka Tsvetanova Tsvetkova-Ivanova on "Studies of the Mechanisms of Cell Death and the Role of the Effector Gasdermin D in the Induction of the NLRP3 Inflammasome: Importance for Male Fertility," for the Award of the Doctorate in Immunology 01.06.23 by Academician Dr. Bogdan Petrunov

Biologist Tsvetkova's dissertation work is devoted to the important medical and social issue of male fertility. It presents detailed data on specific immunological impacts and control mechanisms on Sertoli cells, exploring potential pathways for their cell death involving numerous biologically active factors. This experimental laboratory work is conducted at a high theoretical and practical level under the guidance of Prof. Hayrabedyan, showcasing the candidate's excellent preparation for serious scientific study.

The dissertation, comprising 136 standard pages, is structured conventionally with sections on literature review, materials and methodologies, results and discussion, conclusions, contributions, and a literature review. The literature review is exceptionally well-prepared, reflecting Tsvetkova's awareness and includes modern insights on Sertoli cells, caspases, gasdermin, and types of cell death, based on 213 sources, predominantly from the last ten years.

The dissertation's topic is detailed in its title, and it sets out seven tasks clearly outlining the study's logic and information necessary to achieve the goal. The study investigates the complex nature of cell death involving certain caspases and the activation of the inflammasome signaling pathway. It is a theoretical experimental study that unveils new aspects of the intimate mechanisms of Sertoli cell death and their significant role in sperm development and activity. The use of a wide range of materials, including established commercial kits, modern laboratory methods (such as cell culture, flow cytometry, PCR, ELISA, inverse microscopy), and consistent statistical result processing, is commendable. The dissertation is well-illustrated and written in a scientifically sound language, further evidencing the candidate's solid theoretical training. The competent discussion and corresponding conclusions are well-aligned with the results obtained.

The main contributions of the dissertation include:

1. The establishment of the fact that activation of the caspase-1-pyroptotic inflammasome signaling pathway leads to less destructive programmed cell death.
2. The discovery of the molecule/receptor CD300a in Sertoli cells and its regulatory activity, which has a two-way action with respect to caspase-1 and caspase-3, showing that CD300a is crucial for the expression of gasdermin, which in turn affects caspases differently — inhibiting caspase-1 and activating caspase-3.
3. The identification of a mechanism of reverse regulation that protects Sertoli cells from gasdermin D in terms of caspase-1 and caspase-3, a major factor in their programmed cell death.

These results contribute significantly to understanding the complex mechanisms of Sertoli cell death, thereby enhancing knowledge of male fertility disorders.

Regarding the dissertation, the candidate has two publications in impact factor journals and has participated in international scientific meetings, making her research known globally. She also contributes to a scientific project related to her dissertation work, funded by the Ministry of Education and Science.

The autoreferat of the dissertation is excellently prepared, providing a comprehensive overview of the experimental work, results, discussions, and conclusions.

Mrs. Ilka Tsvetkova-Ivanova, born in 1994, graduated in 2017 from Sofia University with a Bachelor's in Molecular Biology and a Master's in Developmental Biology in 2019. Since 2019, after a competitive exam, she has been an assistant in the Laboratory of Reproduction and OMIC Technologies at IBIR.

In conclusion, the dissertation by Ilka Tsvetkova fully meets the standards for a rigorous laboratory study conducted at a modern experimental level on a topic of both theoretical and practical importance related to male fertility. The results are persuasive, and their analysis reflects the candidate's solid theoretical and practical training, forming a foundation for her future development as a researcher-immunologist. Thus, I highly endorse the dissertation and vote "YES" for awarding the title of "Doctor" in "Immunology" to Mrs. Ilka Tsvetanova Tsvetkova-Ivanova.

Academician Dr. Bogdan Petrunov