REVIEW

From Assoc. Prof. Dr. Diana Yordanova Zasheva

For the dissertation on

"Influence of Specific MicroRNAs in Tumor Pathogenesis by Altering Autophagy Processes and Innate Immune Signaling"

by PhD student Radostina Petkova Tsvetankova,

Laboratory for Reproductive OMIC Technologies, Institute of Biology and Immunology of Reproduction "Acad. Kiril Bratanov" at the Bulgarian Academy of Sciences,

for acquiring the educational and scientific degree 'Philosophy Doctor'

in Professional Field 4.3. Biological Sciences, with a specialization in Immunology,

Code: 01.06.23.

This opinion is prepared in accordance with the requirements of the Law on the Development of the Academic Staff of the Republic of Bulgaria and the Rules for its Application at the Institute of Biology and Immunology of Reproduction "Acad. Kiril Bratanov" at the Bulgarian Academy of Sciences.

Topicality of the Scientific Problem

The dissertation addresses the mechanisms of autophagy and innate immune signaling under the influence of microRNAs in prostate cancer pathogenesis. Prostate cancer, being the fifth leading cause of death among men, highlights the social significance of this research. The exploration of changes in signaling pathways that direct cells toward various autophagy mechanisms is particularly relevant. The disease is characterized by increased levels of oxidative stress, inflammatory processes, and cell-structural remodeling, crucial for initiating various autophagy forms. Moreover, identifying new therapeutic targets and reliable markers for diagnosis and understanding the impact of microRNAs on signaling pathways is vital for developing personalized therapies, underlining the study's exceptional relevance and importance.

Argumentation of the Research Done in the Dissertation

The PhD student presents a well-grounded 43-page literature review detailing the changes occurring in prostate cancer, the regulatory mechanisms and alterations associated with microRNAs in prostate carcinogenesis, and the interplay between autophagy and apoptosis processes. Special attention is given to NF-κB-mediated pro-inflammatory signaling in

carcinoma pathogenesis, particularly prostate carcinoma. This comprehensive review logically sets the stage for elucidating the influence of microRNA-141 and its role in autophagy processes and pro-inflammatory signaling alteration through MARK1 gene inhibition, aiming to identify new diagnostic and therapeutic targets for prostate cancer.

Purpose & Objectives

The literature review leads to a precise and clear definition of the dissertation's purpose and the subsequent study tasks.

Materials and Methods

Described over 13 pages, this section is divided into materials and methods. It details all necessary materials, chemicals, and kits used in the research. The methods section is thorough and reproducible, employing state-of-the-art techniques such as transfection with RNAs, real-time PCR analysis, methylation analysis, flow cytometry, nanopore sequencing, fluorescence microscopy, and determination of alkaline phosphatase activities, alongside modern statistical analysis methods.

Results

The results detailed over 45 pages and divided into 12 sections, present 32 figures, 2 annexes, 3 tables, and one diagram of exceptional quality. The study found that microRNA-141 mimic and inhibitor and RNA inhibitor of the MAPK1 gene influence the expression of ATG16L and LC3 in two prostate carcinoma cell lines. The results elucidate various molecular mechanisms, paving the way for new therapeutic strategies.

Discussion

Spanning 20 pages with two diagrams of gene regulatory networks, the discussion compares the results with recent literature. It concludes with potential new therapeutic methods for treating prostate carcinoma, particularly in the PC3 cell line, after restoring miRNA-141 expression.

Nine conclusions are drawn, aligning with the obtained results and leading to two original contributions. The study validates miRNA-141's role in regulating macroautophagy and mitophagy, offering new therapeutic avenues for metastatic tumors characteristic of the PC3 cell line. Combining miRNA-141 restoration with selective MAPk1 gene inhibition may provide a personalized strategy for treating androgen-resistant prostate cancer.

Question for the PhD Student: What rationale led to choosing miRNA-141 for exploring its association with the studied processes?

Citation and Support:

The dissertation cites 359 recent literary sources, supported by 4 projects from the Laboratory for Reproductive OMICs. The findings have been published in two international journals with impact factors and presented at five scientific events.

Conclusion:

The dissertation offers significant insights into prostate cancer's molecular mechanisms, contributing to new therapeutic approaches. The PhD student has demonstrated proficiency in modern methodologies and analytical skills. Therefore, I strongly recommend the esteemed members of the scientific jury to vote in favor of awarding Radostina Petkova Tsvetankova the educational and scientific degree of "Philosophy Doctor".

17.01.2024 Assoc. Prof. Dr. Diana Zasheva