REVIEW

by Assoc. Prof. Dr. Heliana Irzhi Dundarova
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of the dissertation titled: "Wintering Patterns of Cave-Dwelling Bat Species in Bulgaria in the
Context of Global Climate Change" submitted in fulfilment of the requirements for the
educational and scientific degree "Doctor" Field of study: 4.3 Biological Sciences,

Specialty: Ecology and Ecosystem Conservation Author: Niya Lyubenova Toshkova Scientific Advisor: Prof. Dr. Vasil Valkov Popov (IBER-BAS)

1. General Description of the Submitted Materials

This review is prepared in accordance with Article 9 of the Regulations for the Acquisition of Academic Degrees and Holding Academic Positions at IBER-BAS, adopted on 22.07.2014 and last amended on 25.11.2022, and pursuant to Order No. 29/04.04.2025 of the Director of IBER-BAS, related to the decisions of the Scientific Council (Protocol No. 40/28.03.2025) and the Doctoral Committee held on 23.04.2025.

Niya Lyubenova Toshkova is a full-time doctoral student in the Department of Ecosystem Research, Ecological Risk, and Conservation Biology, IBER-BAS. The dissertation is titled "Wintering Patterns of Cave-Dwelling Bat Species in Bulgaria in the Context of Global Climate Change" in the field of study 4.3 Biological Sciences, specialty "Ecology and Ecosystem Conservation".

The set of materials and documents presented in digital format complies with the requirements of the BRAADB Act, the Regulations for its implementation and the Regulations for the conditions and procedure for the acquisition of scientific degrees and academic positions at the IBER-BAS. The candidate has included three published scientific articles—two in a Q2-ranked journal with impact factor 1.3 (Biodiversity Data Journal), and one in a Q3-ranked journal without impact factor (Historia Naturalis Bulgarica). These publications meet the national criteria for doctoral qualifications.

2. Brief Biographical Data of the Doctoral Candidate

Niya Lyubenova Toshkova (10.03.1992) holds two master's degrees from the Faculty of Biology, Sofia University "St. Kliment Ohridski"—in Molecular Biology and Mycology. During her PhD studies, she specialized in bat ecophysiology and participated in Erasmus+ programs at Sorbonne University, Réunion University, and the University of Porto. She is the author of over 20 scientific publications (Q1–Q4), including studies on temperature sensitivity of antibodies, the relationship between climate change and bats, and caves as reservoirs for zoonotic diseases. Toshkova has led projects for bat monitoring (under the Ministry of

Environment and Water), participated in the Earth Hologenome Initiative, co-authored the ClimBats database, and is active in science communication and education, including the development of educational games and expeditions.

3. Relevance of the Topic and Appropriateness of the Aims and Tasks

The dissertation addresses a specific and ecologically significant issue - the wintering behavior of cave-dwelling bats in Bulgaria under conditions of climate change. The topic is scientifically grounded and timely, intersecting zoology, ecology, and climate science. Cave-dwelling bats serve as suitable bioindicators for environmental monitoring. The choice of study sites - Parnitsite, Devetashka Cave, Ivanova Voda, and Balabanova Dupka - is well justified, as these are important subterranean habitats of conservation value.

The main goal - to identify and analyze wintering traits of bats in the context of climate variability - is clearly formulated and achievable through four interconnected research tasks: winter activity patterns, feeding behavior, health status before and after hibernation, and optimization of monitoring methods. These tasks are logically structured and cover essential dimensions of the subject. The working hypotheses regarding species-specific responses, elevation effects, temperature correlations, and physiological adaptations are well substantiated.

Toshkova demonstrates awareness of the current state of research both nationally and internationally. The theoretical framework is well defined, based on a selection of relevant literature that supports the methodology and interpretation of results.

4. Familiarity with the Research Problem

The candidate demonstrates strong knowledge of current scientific literature, with carefully selected and well-discussed references. The reviewed publications align precisely with the research issues under discussion, reflecting a solid theoretical foundation - likely a result of her international academic experience.

5. Methodology

The dissertation employs a multidisciplinary approach, integrating ecological methods (acoustic monitoring), taxonomic methods (microscopy), molecular techniques (metabarcoding), and physiological assays (oxidative stress markers). The methodology is well described and corresponds directly to the research objectives, with only minor remarks indicated later in this review.

6. Structure and Evaluation of the Dissertation

The dissertation comprises 148 pages, following the structured format adopted by IBER-BAS, consisting of published and accepted scientific articles. It includes: Dedication (1 page), Table of Contents (1 page), Abbreviations (1 page), Introduction (2 pages), Objectives and Tasks (4 pages), Literature Review, Materials and Methods, Results and Discussion (118 pages), three published articles, one unpublished manuscript, one protocol (preprint on protocols.io), and one national monitoring protocol (uploaded to Figshare). The Conclusion (15 pages), Acknowledgments (2 pages), References (2 pages), and a Declaration of Authenticity (1 page) complete the thesis.

7. Contributions and Scientific/Practical Significance

- 1. Fundamental Scientific Contributions:
- 2. First study in Bulgaria examining winter activity in four major bat hibernation sites of European importance.
- 3. First documentation of bat flight activity at -8° C, among the coldest such records globally.
- 4. Established link between external temperature dynamics and acoustic activity patterns.
- 5. Recorded diurnal activity of wintering bats under favorable temperature conditions.
- 6. Historical and current climate data reveal decreasing sub-zero days and increasing days over 5°C—suggesting extended winter activity.
- 7. First identification in Bulgaria and the Balkans of Psorergatoides kerivoulae mites on *Myotis blythii*, with histopathological analysis of wing lesions.
- 8. First evidence in bats of pre- and post-hibernation differences in glutathione and 8-OHdG levels—markers of oxidative stress adaptation. First integrated study of winter diet using guano morphology, metabarcoding, and insect activity monitoring.

Scientific-Applied Contributions:

- 1. All raw data is publicly accessible, promoting transparency and further research use.
- 2. Acoustic data are uploaded to the ChiroVox database for broader comparative use.
- 3. Developed a new methodology for assessing Wing Damage Index in bats using ImageJ.
- 4. Provided recommendations for updating the national bat monitoring methodology.

8. Evaluation of Publications

The two core publications are in Q2-ranked peer-reviewed journals, with Toshkova as first author - demonstrating her leadership in the conception, execution, and writing of the studies. These works clearly reflect the quality and significance of the research findings.

9. Individual Contribution of the Candidate

The dissertation is the independent work of the candidate, encompassing complex research design, analysis, and interpretation. Toshkova has demonstrated diligence, consistency, and innovation, and is now a fully developed researcher in the field of ecology.

10. Thesis Summary

The abstract reflects the methodological, theoretical, and applied contributions of the dissertation, complying with the relevant academic standards and institutional requirements.

11. Critical Remarks and Recommendations

1. Figure 1 (Objectives and Tasks chapter): Avoid mixing Cyrillic and Latin in the same diagram; consider labeling species with numbers and explaining them in the legend.

2. Task Formulations: Tasks currently include methodological elements, which should be described in the Materials and Methods section instead.

3. Description of Publications: Clarify the number of published articles and those under review or in press.

4. Protocols.io Submission: The name of IBER-BAS should be included as a participating institution in all online protocol materials.

Reasoned conclusion

The dissertation is a scientifically sound and well-structured study with both theoretical and applied value in the field of bat ecology under climate change. The candidate has demonstrated the necessary competencies to conduct independent scientific research. Based on this conclusion, I propose to the Honourable Scientific Jury to award the degree of Doctor of Education and Science to Niya Lyubenova Toshkova in the professional field 4.3 "Biological Sciences" and the scientific speciality: "Ecology and Conservation of Ecosystems."

Date: 11.06.2025 Reviewer:

Assoc. Prof. Dr. H. Dundarova