Review of the doctoral thesis "Exploring the marine biodiversity with environmental DNA" submitted to the Institute of Oceanology of the Polish Academy of Sciences (IO PAN) by M. Sc. Ngoc Loi Nguyen.

Reviewer: Prof. Dr. Thorsten Stoeck, Kaiserslautern, Germany

Declaration

I hereby declare that I have no personal or professional relation to the applicant and that I have no Conflict of Interest to conduct this review.

Review

Marine sedimentary archives hold extensive marine biodiversity, including benthic and pelagic organisms. Environmental DNA (eDNA) preserved in these sediments offers insights into both current and past marine ecosystems. Recent high-throughput sequencing technologies have enhanced eDNA studies, especially for microbial biomes, although challenges and biases remain. The thesis of Ngoc Loi Nguyen investigates the application of eDNA in understanding marine biodiversity by focusing on three main research areas:

First, it explored eukaryotic biodiversity in the water column and surface sediments, revealing that plankton DNA in sediments differs significantly from that in water samples. Planktonic DNA, while abundant in sediments, does not accurately represent water column diversity, highlighting potential biases in historical biodiversity reconstruction.

Second, it examined the diversity of benthic foraminifera, showing that eDNA metabarcoding reveals a much higher phylogenetic diversity than traditional microscopy-based (morphological) methods. Results obtained from samples that were collected in the Svalbard Archipelago identified numerous new foraminiferal lineages and potential ecological indicators of water mass characteristics, enhancing our understanding of Arctic marine environments. A second study conducted in this context identified unique deep-sea foraminiferal lineages, suggesting limited migration between shallow and deep-sea habitats.

Third, the thesis reviews advances in marine sediment ancient (seda) DNA research, emphasizing its potential in paleoceanographic studies. A systematic review of 55 studies outlines the taphonomic processes, key issues, and applications of sedaDNA, predicting its routine use in future research to understand biodiversity changes over geological timescales and anthropogenic impacts.

Overall, this PhD thesis underscores the significant potential and limitations of eDNA metabarcoding in marine biodiversity studies, contributing valuable insights into the diversity of deep-sea and polar foraminifera and the biases in eDNA taphonomy. The findings demonstrate the importance of eDNA metabarcoding in advancing marine biodiversity surveys over time and space.

These studies were summarized in four manuscripts, three of which are already published in high-ranking scientific journals. The fourth manuscript (taxonomic and

abundance biases affect the record of marine eukaryotic plankton) is currently under review with the journal Molecular Ecology Resources".

The thesis is well written, its structure is clear and self-explaining. Before the candidate presents the four papers in the thesis, Ngoc Loi Nguyen presents a synthesis consisting of a summary of all four main chapters. The purpose of the research program conducted is convincingly motivated; all methods are explained in a way that they are reproducible, and Ngoc Loi Nguyen uses the available state-of-the-art tools and technologies to produce and analyze the data; the results are presented adequately in high-quality figures, tables and descriptions; and the discussion(s) are very solid, convincing and sufficiently critical where needed.

The obtained results are of high importance and advanced our knowledge in the specific fields of research. This is obviously not only my opinion, but also the one of several anonymous reviewers who gave their green light to publish hitherto three of the four manuscripts resulting from this thesis thus far in high-ranking scientific journals. More manuscripts are currently in preparation. Based on my decades-long experience in reviewing PhD theses, I feel confident to say that the quality of this thesis most certainly belong to the best 10% in its field and the production of four high-quality manuscripts is an extraordinary achievement. And all of this under consideration that the duration of this thesis was only three years and the research subject as well as the methodologies were everything but trivial. Very impressive!

Without doubts, Ngoc Loi Nguyen is an excellent scientist who earns to be awarded the degree of doctor.

Prof. Dr. Thorsten Stoeck, Kaiserslautern May 23rd 2024