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Porto, January 29th 2021

Informal PhD thesis review

PhD candidate: Lobov Arseniy Andreevich

Thesis Title: Gamete interaction proteins as factors of reproductive isolation of cryptic species of the genus *Littorina* Férussac, 1822

University: Saint-Petersburg State University

To whom it may concern:

The thesis addresses a very relevant topic in evolutionary biology: the mechanisms underlying speciation in the genus *Littorina*, namely the role of gametic incompatibility and postcopulatory prezygotic reproductive isolation. Some species of this genus, in particular *L. saxatilis*, have been widely studied in terms of adaptation and ecological speciation, but little is known about the contribution of gametic incompatibility to diversification within the genus. By attempting to fill this gap, the candidate also aimed to give a general contribution to understand these processes in organisms with internal fertilization, allowing the comparison to better-characterized systems with external fertilization (e.g. mollusks and echinoderms).

The goals of the thesis are clear and precise. The candidate implemented very different techniques and seems to have gain a vast array of skills on different disciplines that will be important for his future research career. Among the various achievements, I would like to highlight the identification of a particular interesting gamete recognition protein candidate (LOSP) that he suggested to be involved in gamete incompatibilities between some *Littorina* species. The author was also able to characterize where and when this parasperm protein was expressed. Finally, the phylogenetic and divergence analyses of the LOSP gene across several species led the candidate suggest that it has been evolving under the influence of selection. Although alternative hypotheses probably exist to explain the phylogenetic/divergence patterns obtained, I think the results are intriguing and open doors for future studies testing his and others evolutionary hypothesis for the role of LOSP (and other gamete recognition proteins) in reproductive isolation across different *Littorina* species.

Based on all the exposed, I think that the candidate achieved the scientific maturity and research experience necessary to defend his PhD thesis with success.

Yours Sincerely,



Rui Faria