REVIEW

Of the member of the dissertation council for the dissertation of Asya Genrikovna Davidian on the topic: FUNCTIONING OF THE NUCLEOLUS ORGANIZER REGION IN THE GROWING OOCYTES OF SAUROPSIDA, submitted for the degree of Candidate of Biological sciences in a Scientific specialty 1.5.23. Developmental biology, embryology

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I have read thoroughly the thesis from Ms Davidian and find it well written in English and an easy read to follow through all the information and thought processes. This I commend the candidate upon. There are at least five published peer-reviewed articles in well-known reputable journals that have come from Miss Davidian's work/research and this is also highly commendable. I would like to add here, this is a real achievement.

The thesis could have been organized with three distinct results chapters which we often do from my laboratory but nevertheless the data are in place and the large results chapter does work – although it might have been an easier read if the various individual projects within the thesis had been divided to make reading the thesis slightly easier and each piece of work more defined, with its own specific discussion. However, this would not be a major criticism.

In the UK, at the university where I work, the thesis work would need to have at least three smaller projects within that could constitute the separate results chapters, be novel and be publishable. The work here is certainly all those things.

The research work is intricate and has taken a long-held hypothetical view points and theories of the presence of in and how rRNA is found in oocytes in Sauropsida - reptiles and birds. These notions are nicely demonstrated in figure 1. The candidate has readdressed the presence and distribution of rRNA in a range of members of Sauropsida with modern tools (indirect immunofluorescence combined with fluorescence in situ hybridization in 3D, qPCR and bioinformatics, combined with detailed and careful analyses of her findings, generating new unique information and knowledge and addressing dogma and hypothesis within the field. The aims and objectives and the findings and how the findings fit within the literature is very clearly put at the beginning of the introduction. The topic is very important to not only the basic understanding of how eggs of different species acquire and accumulate ribosomal RNA for development but will also be relevant to aspects of female fertility. However, the most significant finding is the novel discovery of 5S RNA within the intergenic spacer regions of the vertebrates turtles and crocodiles. This is a major finding within evolutionary biology and beyond and will be the beginning of a whole new raft of investigations.

The introduction (Chapter 1) is well written and well referenced and is of an equivalent size to one being presented in the UK. It is particularly commendable due its range of citations from the seminal early work to the very recent findings. The figures are informative, easy to decipher and attractive to look at.

The Materials and Methods Chapter (Chapter 2) is also well written with the relevant details of the methods described so that other scientists would be able to follow the methods and recapitulate the experiments. What is missing for me is the detail around the quantitation – how aspects of the results were scored and any statistical analyses performed.

The results chapter combines all the results together and it might have been nice to have them as separate chapters with a discussion after each set of data and findings. The work on the immature and mature hens is carefully performed and presented with beautiful images of the ovary sections combined with indirect immunofluorescence and fluorescence in situ hybridisation for relevant antigens and gene sequences and there is no argument about how well this analysis has been performed but I am missing the numbers, I know there are % mentioned in the text but how were they derived and from how many samples etc. Have statistical analyses be performed? I appreciate that the presence and finding of rRNA structures is important but an opportunity to quantitate further could have been done.

The red-eared turtle study looking at the nucleolar structure and the confirmation of a bipartite nucleolar structure is very interesting, as is the distribution of coilin. However, the tour de force in this thesis is the demonstration by bioinformatics of the presence of 5S NOR-5S RNA in turtles and crocodiles, which has led to rewriting of the evolutionary tree. Further, the candidate has gone beyond bioinformatics in this study and provided evidence of its actual presence and function in tissue.

The conclusion is well written and outlines the findings and importance of these findings well. This is an exciting and interesting body of work, more than adequate to meet the specifications outlined for a Doctorate of Philosophy (PhD) in the UK.

Dissertation of Asya Genrikovna Davidian on the topic: "FUNCTIONING OF THE NUCLEOLUS ORGANIZER REGION IN THE GROWING OOCYTES OF SAUROPSIDA" meets the basic requirements established by Order No.11181/1 dd. 19.11.2021 "On the procedure for awarding academic degrees at St. Petersburg State University". The applicant Asya Genrikovna Davidian deserves to be awarded the academic degree of candidate of Biological sciences in a scientific speciality 1.5.23. Developmental biology, embryology. Paragraphs 9 and 11 of the specified Order have not been violated.

Member of the Dissertation Council

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29th April 2022