

RE-EVALUATION OF THE TAXONOMIC STATUS OF THE THREE NOTOTHENIIDS FOUND ON BOTH SIDES OF ANTARCTIC POLAR FRONT

Tshoanelo Miya, O. Gon and M. Mwale

South African Institute for Aquatic Biodiversity (SAIAB), Research Division, Grahamstown

t.miya@saiab.ac.za

Background

Notothenioid species that are found south and north of the Antarctic polar front (APF) have previously been split into separate species and/or subspecies in their respective localities. This study re-evaluated taxonomic status of *Lepidonotothen squamifrons*, *L. larseni* and *Gobionotothen marionensis*, which were coined into 'squamifrons', 'larseni' and 'marionensis' groups consisting of geographically restricted species (Fig. 1). Taxonomic identifications of these species were largely based on morphological characteristics. However, the analyses of some recent morphological studies disagreed with some of these taxonomic acts. Molecular systematic studies conducted on notothenioid species so far have concentrated largely on the Atlantic Ocean sector. Therefore, this study re-evaluated the taxonomic status of these three nototheniids at the DNA level, by comparing specimens from different localities in the Southern Ocean.



Results

- In all tree genes the intra-specific variability (0.3-1.1%) was smaller than the interspecific variability (1.3-18%).
- The 'squamifrons' group lineage had >95% bootstrap support and genetic distance among individuals ranged from 0% to 0.9%. These results indicated that this group consist of one species, *Lepidonotothen squamifrons*.
- The 'larseni' group had >98% bootstrap support with genetic distance ranging from 0.2% to 0.6%. These results indicated that this group consist of one species, *L. larseni*.
- The 'marionensis' group had two lineages; the first one consisted of *Gobionotothen acuta* from Heard and MacDonald Islands, and the second one with *G. acuta* from Marion Island, *G. angustifrons* and *G. marionensis*. The genetic distance between these lineages ranged from 1.4% to 2.8%, indicating that these lineages consist of sister species. The second lineage had 98% bootstrap support and genetic distance among individuals ranged from 0.2% to 0.7%. These results indicated that this lineage consist of one species, *G. marionensis* and therefore *G. acuta* of Marion Island was misidentified.

Materials and methods

Tissue samples of *Lepidonotothen* and *Gobionotothen* species were collected in the Atlantic, Indian and Pacific sectors of the Southern Ocean (Fig. 1). DNA was extracted from these samples using the DNeasy QIAGEN tissue extraction kit. DNA sequencing was performed using three gene regions, including two mitochondrial coding genes, ND2 and COI and one nuclear DNA, S7 intron 1 gene. The estimates of evolutionary distance among sequences were analysed by pairwise genetic distance calculations implemented in Mega 5.05. These estimates were calculated for different taxonomic levels including family, genus and species, and at the individual level, where the average genetic distance between sister species. Maximum likelihood and Parsimony phylogenetic trees were constructed using PAUP.



Figure 2. Phylogenetic trees resulted from the two mitochondrial gene were similar and therefore only ND2 is shown. (a) ND2 and (b) S7 intron 1 gene maximum likelihood phylogenetic trees showing relationships within 'squamifrons', 'larseni' and 'marionensis' groups, respectively. The bootstrap support values are from parsimony analyses and defined as weak (50-69%), moderate (70-89%) and strong support (90-100%). The colour codes correspond with the species listed in Fig. 1.

Conclusion

- This study does not support the splitting of *L. squamifrons* and *L. larseni* into different species.
- It confirms that *G. marionensis* and *G. acuta* are different species.
- The species of the two studied genera were clearly separated, i.e.
 - *Lepidonotothen* consisted of *L. squamifrons*, *L. larseni*, *L. nudifrons* and *L. mizops* (not shown here); and *Gobionotothen* composed of *G. marionensis*, *G. acuta* and *G. gibberifrons*. (taxonomic status of *G. barsukovi* needs to be confirmed).

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