Sardinella aurita (Clupeidae) in Mar Chiquita coastal lagoon: morphological and DNA barcoding identification approaches

by

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RÉSUMÉ. - *Sardinella aurita* (Clupeidae) dans la lagune côtière de Mar Chiquita : approche morphologique et identification par ADN barcoding.

Deux spécimens de Sardinella aurita ont été capturés dans la lagune côtière de Mar Chiquita, Argentine, et identifiés sur des bases morphologique et moléculaire. Le statut taxinomique du genre dans l'océan Atlantique ouest reste encore incertain. Les résultats présentés dans cette étude, basés sur l'analyse par ADN barcoding, sont en accord avec ceux qui ont été obtenus par d'autres marqueurs moléculaires et suggèrent que les espèces S. aurita et S. brasiliensis sont conspécifiques.

Key words. - Clupeidae - *Sardinella aurita* - Argentina - Mar Chiquita - Morphology - DNA Barcode.

Two clupeids of the genus *Sardinella* were caught in Mar Chiquita coastal lagoon (MCH) at the end of December 2008, with a beach seine net (Fig. 1A-C). MCH is a temperate shallow coastal lagoon located in Buenos Aires province, Argentina (37°32′-37°45′S; 57°19′-57°26′W). The fish fauna of this lagoon is composed by approximately 28 species (González-Castro *et al.*, 2009), and no records of the genus *Sardinella* were previously reported.

Specimens were morphologically identified based on Whitehead (1985) and Munroe and Nizinski (2002). A sample of white muscle tissue was excised from each individual and preserved in 100% ethanol at -20°C for genetic analysis. The specimens were deposited in the fish collection of Universidad Nacional de Mar del Plata, Argentina, under the catalogue number UNMDP 1310 and UNMDP 1311. Individuals were measured following Cervigón (1982). Meristic characters for pair and unpaired fin rays and anterior gill rakers on lower limbs were recorded. DNA extraction, polymerase chain reaction (PCR), and sequencing of COI gene were performed following standard DNA barcoding methods (Haijbabaei *et al.*, 2005).

Morphometric and meristic characters of collected specimens are shown in table I. Body somewhat compressed and moderately elongate, abdomen rounded with scutes, posterior end of the gill opening with fleshy outgrowths and caudal fin deeply forked; colour in fresh light bluish dorsally and lateral surfaces silvery grey; anterior gillrakers on lower limbs of second and third gill arches relatively flattened (Fig. 1D).

A 652 base pair amplicon for the 5' region of the mitochondrial COI gene was bidirectionally sequenced in both specimens (GenBank Accession numbers: HM421919 and HM421920). The individuals differed by only two nucleotides. Using the library of sequences collected in BOLD (Barcode of Life Data Systems) the

Table I. - Morphometric (in mm) and meristic measurements for the two *Sardinella aurita* collected in Mar Chiquita coastal lagoon (Argentina).

Measurements	UNMDP 1310	UNMDP 1311
Total length, TL	215.0	191.0
Standard length, SL	185.0	153.0
Weight	80.0	50.0
Head length	40.5	39.5
Preorbital length	12.0	12.1
Postorbital length	18.5	17.7
Horizontal eye diameter	10.0	9.7
Body height	48.7	41.3
Dorsal fin base length	22.7	18.9
Anal fin base length	26.9	24.1
Pectoral fin length	26.6	25.4
Dorsal-fin rays	15	14
Pectoral-fin rays	17	19
Pelvic-fin rays	8	8
Anal-fin rays	17	16
Gill rakers	116	111

closest matches of our sequences were located. Considering the "Species level Barcode Database" (includes every COI Barcode record with a species level identification as a minimum sequence length of 500 bp), UNMDP 1311 matched with *S. aurita* Valenciennes, 1847 in 98.82 to 99.85% (n = 10) and with *S. janeiro* (Eigenmann, 1894) in 99.16 to 99.85% (n = 5), whereas UNMDP 1310 matched 99.1 to 100% with *S. aurita* (n = 9) and 99.49 to 100% with *S. janeiro* (n = 5).

Two species of Sardinella occurred almost sympatrically in the western Atlantic Ocean, S. aurita and S. janeiro, but only the former was registered in Argentine waters (Whitehead, 1985; Munroe and Nizinski, 2002). The latter species was recently renamed (Figueiredo et al., 2010) as S. brasiliensis (Steindachner, 1879). Nonetheless, the validity of both species is somewhat questionable. Whitehead (1970) removed *S. brasiliensis* from the synonym of *S*. aurita based on the difference on gill-rakers counts in the syntypes of S. brasiliensis from off Brazil, and concluded that those with low gill-rakers counts (80-120) were S. aurita whereas those with high gill-rakers counts (over 150) were S. brasiliensis (Steindachner, 1879). On the other hand, he also proposed that both species might be distinguished from each other based on the shape of the gill rakers of the 2^{nd} and 3^{rd} branchial arch, being flattened in S. aurita and strongly curved in S. brasiliensis (Whitehead, 1985). However, the same author stated that "the separation of S. aurita and S. brasil-

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iensis by means of gillrakers shape and a higher gillraker count is tentative".

Several works showed no molecular differences between *S. aurita* and *S. brasiliensis* (Tringali and Wilson, 1993 and references therein; De Donato *et al.*, 2005). Tringali and Wilson (1993) employing analysis of restriction-site polymorphisms of mtDNA compared specimens with different counts in gillrakers. Their results suggest that both are conspecific. In accordance with these studies, each COI sequence of the specimens collected in our study revealed a high similarity with sequences of both *S. aurita* and *S. brasiliensis*. Moreover, Munroe and Nizinski (2002) describing the species of the western central Atlantic considered also that purported differences in gillraker morphology of both species need to be confirmed and suggest that *S. brasiliensis* may be a junior synonym of *S. aurita*.

Individuals collected in MCH presented low gill-raker counts and gill rakers of the 2nd and 3rd branchial arch slightly flattened. These characteristics are coincident with those reported for *S. aurita*. Molecular results also related our specimens with *S. aurita*. Taking into account previous evidences and those found in the present work, new studies employing morphological, meristic, morpho-

37°33'S

Mar Chiquita
Lagoon
ATLANTIC
OCEAN

Study
area

57°24'W
57°19'W

metrical and molecular techniques with specimens collected over a wide distribution range are needed to reinforce *S. brasiliensis* as a junior synonym of *S. aurita*.

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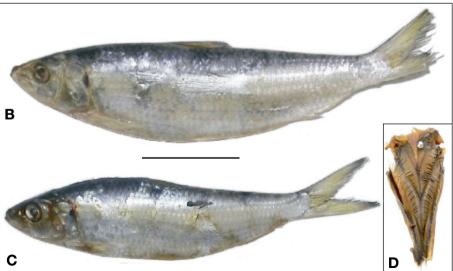
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Figure 1. - **A**: Map of Mar Chiquita coastal lagoon where the individuals of *Sardinella aurita* were captured. **B**: Specimen UNMDP 1311. **C**: Specimen UNMDP 1310. **D**: Detail of lower gill raker. Scale bar = 50 mm.



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