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Morphological and molecular evidence for a new species of longnose skate (Rajiformes: Rajidae: *Dipturus*) from Argentinean waters based on DNA barcoding

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Abstract

A new species of *Dipturus* is described from ten specimens collected off Patagonia, Argentina. Morphological and molecular approaches were used to compare among specimens of recognized *Dipturus* species. By comparing morphometric, meristic and mitochondrial cytochrome *c* oxidase I (COI) sequence data, specimens referred to as longnose skate and originally regarded as *D. chilensis* were shown to be a discrete species as distinguished from both the Yellownose skate, *D. chilensis* and the Roughskin skate, *D. trachyderma*. *Dipturus argentinensis* **n. sp.** can be distinguished from all other southwestern Atlantic longnose skate species by its color pattern, lack of squamation on both upper and lower surfaces of the disc, and a long, thin tail that is approximately half the total length. The new species has one median row of 10 to 24 small caudal thorns, one or two interdorsal thorns and 35 to 40, and 34 to 43 tooth rows on upper and lower jaws, respectively. The 648 base pair COI mitochondrial DNA "barcodes" derived from specimens of *D. argentinensis* are identical to each other and exhibit greater than 3% sequence divergence from all other *Dipturus* species similarly characterized to date. Taken together, these independent morphological and molecular observations serve to corroborate one another and thus provide strong evidence for the recognition of *D. argentinensis* as a new species.

Key words: *Dipturus argentinensis*, **n. sp.**, COI, DNA barcode, Argentina, Rajidae

Introduction

Longnose skates of the genus *Dipturus* Rafinesque have a worldwide distribution occurring mostly in cool-temperate to tropical seas, continental shelves and slopes except the Eastern North Pacific (Compagno 1999), from 25 to approximately 1150 m depth (Mc Eachran & Miyake 1990). Ebert & Compagno (2007) recognized 31 nominal species of *Dipturus*, two of which have been reassigned to the genus *Zearaja* (Last & Gledhill 2007). Since then 12 more new nominal species have been described for Australian waters (Last 2008, Last *et al*. 2008, Séret & Last 2008), and another one for the south China Sea (Jeong & Nakabo 2008). In the southwest Atlantic the genus is represented by five valid species: the South American Yellownose skate *D. chilensis* (Guichenot 1848), the Thorny tail skate *D. diehli* Soto & Mincarone 2001, the Thintail skate *D. leptocauda* (Krefft & Stehmann 1975), the South Brazilian skate *D. mennii* Gomes & Paragó 2001 and the Roughskin skate *D. trachyderma* (Krefft & Stehmann 1975). The Florida skate *D. teevani* (Bigelow & Schroeder 1951) although currently known from tropical waters of the Caribbean Sea (Jacob & McEachran 1994) has been

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recently recorded in the southwest Atlantic (Gomes & Picado 2001). Very recently, Last & Gledhill (2007) have reassigned *D. chilensis* to the genus *Zearaja* Whitley based mainly on clasper morphology. However, in the present paper we continue using the original designation until further studies of the skeletal morphologies of the other nominal species of *Dipturus* occurring in the south-west Atlantic will be compared.

Species of *Dipturus* are commonly recognized by possessing long-hard rostral cartilages, rhomboid discs and nearly smooth skin with few thorns on the disc (Compagno *et al.* 1989). In the Argentine Sea, species of *Dipturus* (mainly *D. chilensis*) are usually caught as by-catch in the bottom trawling fishery for Argentine hake *Merluccius hubbsi* (García de la Rosa *et al*. 2000). Skates are increasingly exploited as target species because traditional bony fish stocks for the commercial fisheries are being drastically depleted (García de la Rosa *et al*. 2004). The increase in commercial pressure suggests that better management tools are needed, particularly with respect to the identification of processed products that lack diagnostic morphological features. Molecular characterization can be extremely helpful in this regard and the 5' region of the mitochondrial cytochrome *c* oxidase I gene has been recommended by Hebert *et al*. (2003) as a standard molecular marker or "DNA barcode" for species identification. This proposal derives particular merit when sequence profiles are paired with more traditional sources of information (DeSalle *et al*. 2005), as recently demonstrated with dogfishes by Ward *et al*. (2007). Indeed, a new paradigm in ichthyological taxonomic research advocates the inclusion of a DNA barcode in the formal description of a new species (e.g. Victor 2007, 2008, Pyle *et al*. 2008). Herein we describe a new species of *Dipturus* from the southwest Atlantic using both morphological and molecular barcode data, following the recent paradigm shift toward the inclusion of the barcode as part of the species description.

Material and methods

Specimens examined were collected by the R/V Dr. Eduardo Holmberg of the Instituto Nacional de Investigación y Desarrollo Pesquero (INIDEP), from different localities of the southwest Atlantic. Methods for making measurements and counts follow Hubbs & Ishiyama (1968), Menni (1973) and Krefft & Stehmann (1975). The holotype and 9 paratypes are deposited in the fish collection of the Instituto Nacional de Investigación y Desarrollo Pesquero, Mar del Plata, Argentina (INIDEP).

Samples of muscle tissue were collected from seven individuals and preserved in 99.5 % ethanol at ambient temperature for genetic analysis. Genomic DNA was extracted according to the protocol of Ivanova *et al*. (2006). DNA barcodes were generated for the mitochondrial 5' COI gene as follows: PCR products corresponding to base positions 6474-7126 of the *Danio rerio* mitochondrial genome were amplified using fish barcode primer cocktails developed by Ivanova *et al*. (2007). PCR reaction mixtures consisted of 6.25 µl of 10% trehalose, 3.0 µl of ultrapure ddH2O, 1.25 µl of 10X PCR buffer, 0.625 µl of 50 mM MgCl2, 0.125 µl of each forward and reverse primer cocktail (10 µM), 0.0625 µl of 10 mM dNTP mix, 0.06 µl of Platinum® Taq DNA polymerase (Invitrogen, Inc.), and 1.0 µl of template DNA. PCR amplification reactions were conducted on Eppendorf Mastercycler® gradient thermal cyclers (Brinkmann Instruments, Inc.) with the following reaction profile: 2 min. at 94ºC, followed by 35 cycles of 30s at 94ºC, 40s at 52ºC, and 1 min. at 72ºC, followed by 10 min. at 72ºC and then held at 4ºC. PCR products were visualized on 2% agarose E-gel® 96 plates (Invitrogen, Inc.) and labeled using the BigDye® Terminator v.3.1 Cycle Sequencing Kit (Applied Biosystems, Inc.). Bi-directional sequencing reactions were carried out using an ABI3730 capillary sequencer. Sequences were aligned using SeqScape 2.1.1 (Applied Biosystems, Inc.). Specimen provenance and sequence data was organized and analyzed using the Barcode of Life Data Systems (BOLD, Ratnasingham & Hebert 2007) in cooperation with the Fish Barcode of Life Initiative (see: www.FISHBOL.org). Mitochondrial COI barcode sequences were deposited in GenBank and were subsequently annotated with the reserved keyword BARCODE by the International Nucleotide Sequence Database Collaboration (www.INSDC.org)

owing to the fact that the sequences are associated with morphological voucher specimens held in the INIDEP collection and because they meet the BARCODE "data standard" established by the Consortium for the Barcode of Life in collaboration with the INSDC (see: http://barcoding.si.edu/PDF/DWG_data_standards-Final.pdf).

Results

Dipturus argentinensis n. sp.

(Figure 1, Table 1)

Holotype.—INIDEP 793, 765 mm TL, juvenile male, off central Patagonian continental shelf, 45º38'S, 64º08'W, 98 m, 21 January 2006, R/V DR. EDUARDO HOLMBERG, cruise H-01/06, sta. 38. GenBank Accession No. EU074410.

Paratypes.— INIDEP 794, 935 mm TL, female, 46° 20´ S, 64° 09´ W, 95 m, 03 September 2002, R/V DR. EDUARDO HOLMBERG, cruise H-04/02, sta. 363. INIDEP 795, 632 mm TL, immature female, off south Patagonian continental shelf, 50º 15' S, 63º 35' W, 140 m, 14 February 2006, R/V DR. EDUARDO HOLMBERG, cruise H-02/06, sta. 28. GenBank Accession No. EU074411. INIDEP 796, 710 mm TL, immature male, off south Patagonian continental shelf, 47º 45' S, 61º 28' W, 142 m, 21 February 2005, R/V DR. EDUARDO HOLMBERG, cruise H-02/05, sta. 45. INIDEP 797, 617 mm TL, immature male, off south Patagonian continental shelf, 45º 18' S, 64º 40' W, 87 m, 20 January 2005, R/V DR. EDUARDO HOLMBERG, cruise H-01/05, sta. 34. INIDEP 798, 555 mm TL, immature male, off south Patagonian continental shelf, 45º 47' S, 64º 47' W, 96 m, 19 January 2006, R/V DR. EDUARDO HOLMBERG, cruise H-01/06, sta. 26. GenBank Accession No. EU074409. INIDEP 799, 403 mm TL, immature male, off south Patagonian continental shelf, 46º 03' S, 66º 46' W, 91 m, 15 January 2006, R/V DR. EDUARDO HOLMBERG, cruise H-01/06, sta. 7. GenBank Accession No. EU074405. INIDEP 800, 690 mm TL, immature male, off south Patagonian continental shelf, 45º 54' S, 63º 10' W, 97 m, 16 January 2005, R/V DR. EDUARDO HOLMBERG, cruise H-01/05, sta. 13. GenBank Accession No. EU074406. INIDEP 802, 522 mm TL, immature female, off south Patagonian continental shelf, 45º 54' S, 63º 10' W, 97 m, 16 January 2005, R/V DR. EDUARDO HOLMBERG, cruise H-01/05, sta. 13. GenBank Accession No. EU074408. INIDEP 803, 670 mm TL, immature female, off south Patagonian continental shelf, 45º 54' S, 63º 10' W, 97 m, 16 January 2005, R/V DR. EDUARDO HOLMBERG, cruise H-01/05, sta. 13. GenBank Accession No. EU074407.

***Diagnosis*.** *Dipturus argentinensis* is characterized by the combination of the following characters: dorsal surface of disc brown purplish with no distinct ocelli or blotches margined with dark brown on pectoral and pelvic fins. Upper surface of disc smooth except few small spinules scattered on tip of snout. Ocular thorns present, with scapular thorns absent. A single nuchal thorn either present or absent. One median row of 10 to 24 small caudal thorns. Dorsal and caudal fins scattered with very few spinules. One or two interdorsal thorns. Relatively long and thin tail, approximately half the total length. Ventral surface of disc as dark as the upper side, smooth except few small spinules scattered on tip of snout. Interbranchial space with no prickles.

***Description*.** Measurements and counts are given in Table 1. Differing values of the paratypes following those for the holotype are in parentheses. Disc rhombic, 1.24 times as broad as long (1.22-1.27); snout greatly elongated, 4.35 times in total length (4.54 to 5.26); Anterior margin of disc concave and posterior margin convex with rounded inner corner to level of pelvic fins. Orbit length 0.62 times (0.5 – 0.66) of interorbital length and 1.09 times spiracle length (1.14–1.58). Distance between spiracles 1.18 times that between orbits (1.16 – 1.36). Preorbital length 4.49 times (3.88 to 4.33) interorbital width. Tail very slender and relatively long with a thin lateral tail fold. Its length from center of cloaca to tip 0.82 times that from tip of snout to center of cloaca (0.84 to 0.95). Space between dorsal fins, 0.28 times (0.19–0.38) base of first dorsal fin. Height of dorsal

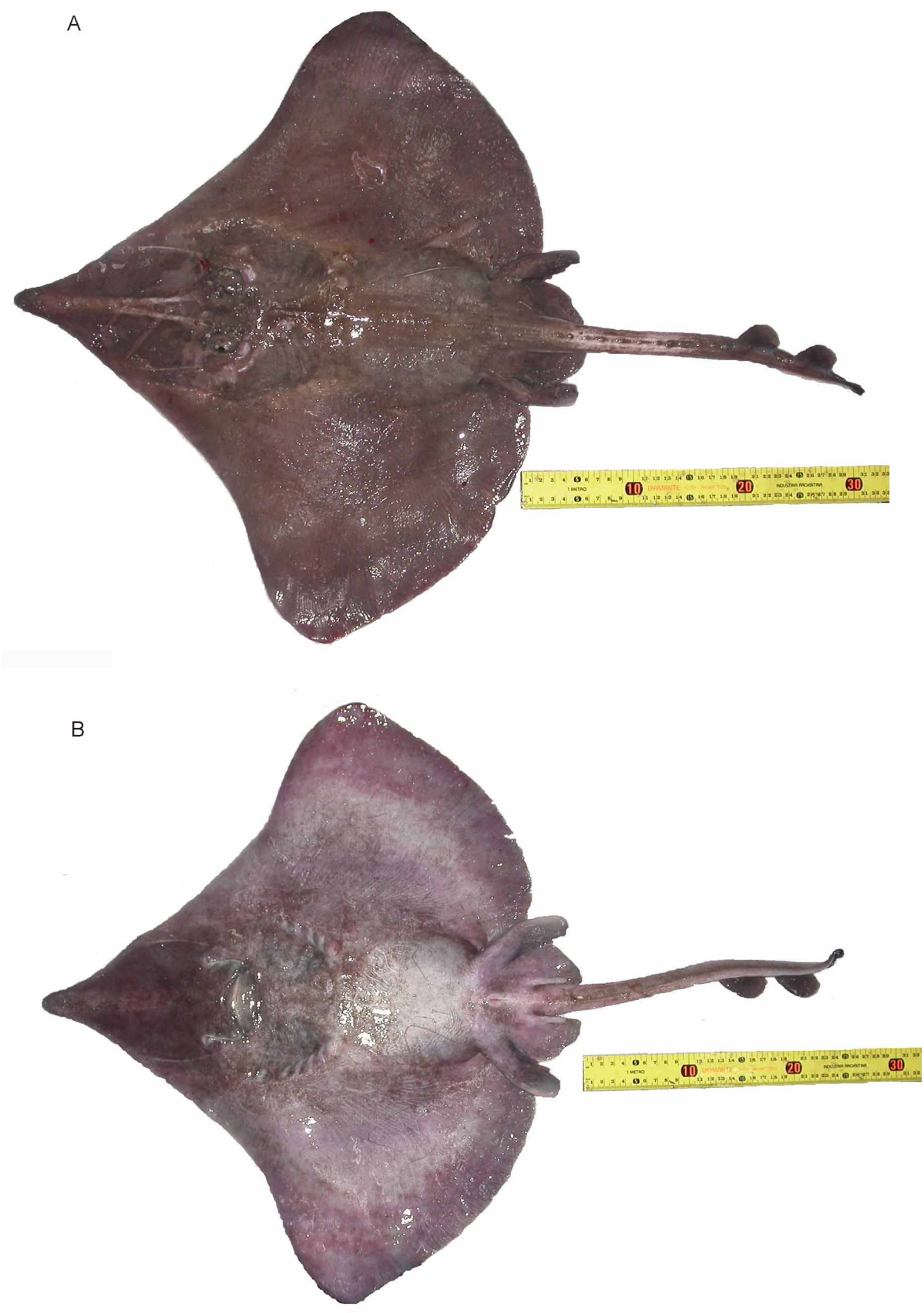

FIGURE 1. *Dipturus argentinensis* **n. sp.**, holotype, INIDEP 793, 765 mm TL, juvenile male, off central Patagonian shelf, Argentina. a–dorsal view; b– ventral view.

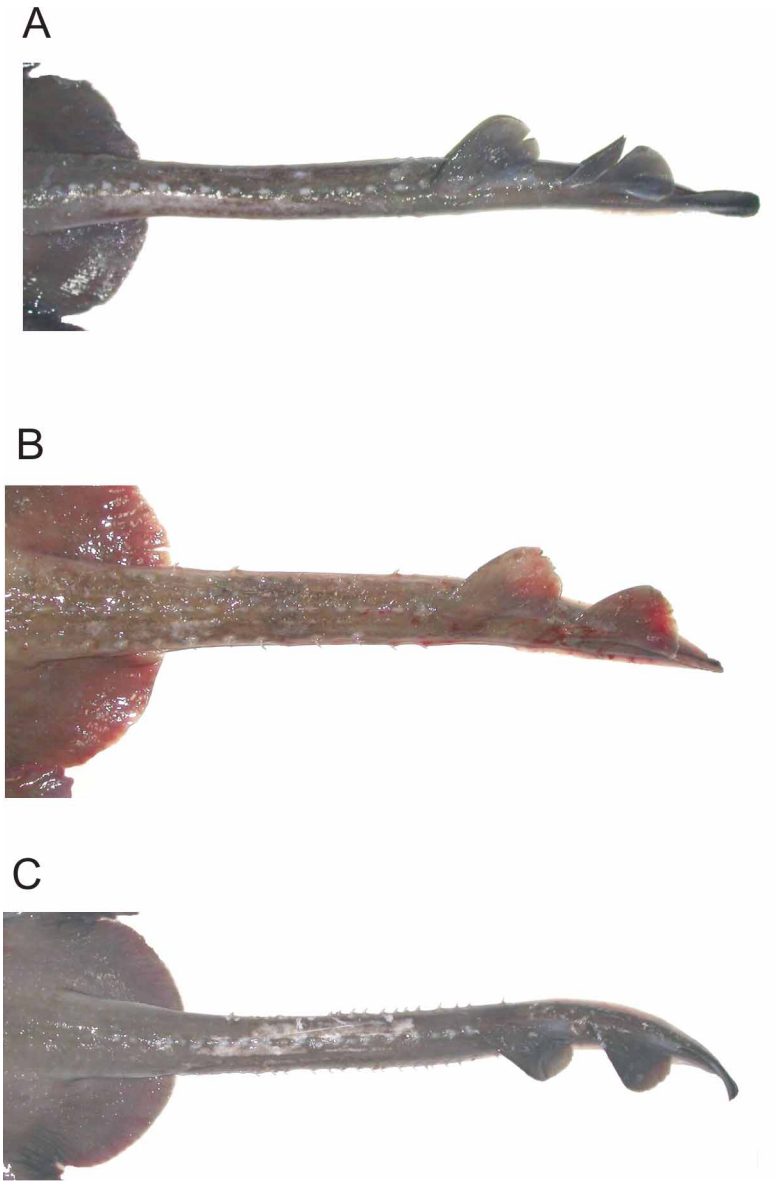

FIGURE 2. Tail thorns of *Dipturus argentinensis* **n. sp**., immature male paratype (INIDEP 797, 617 mm TL) (A), *Dipturus chilensis*, immature female (INIDEP 547, 715 mm TL) (B), and *Dipturus trachyderma*, immature male (INIDEP 789, 1211 mm TL) (C).

fins greater than half their bases' lengths. Preoral length 2.79 times mouth width (2.36 to 2.75). Preorbital length 7.29 times orbit length (6.24 to 7.88). Nostril flaps are short, thick and tube -like. Anterior nasal flap (nasal curtain) well developed and fringed along distal margin. Posterior nasal flap poorly developed and smooth. Mouth slightly arched. Upper and lower jaws with 37 and 38 (35 to 40, and 34 to 43) tooth rows, respectively. Distance between first gill slits 1.86 times distance between nares (1.71 to 1.91). Distance between fifth gill slits 1.06 times internarial distance (1.03 to 1.28). First dorsal fin about equal in size and shape to second dorsal fin. Dorsal side of disc smooth except few small spinules scattered on tip of snout (in one specimen the spinules also placed on both sides of eyes as well as in front of ocular region). Ventral side of disc, pelvics, claspers and tail without dermal denticles except few small spinules scattered on tip of snout. Midline of tail with 13 (10 to 24) small thorns, with oval bases and backwardly directed crowns.

***Coloration when fresh*.** Upper surface of disc plain purplish brown and margined with dark brown on pectoral and pelvic fins with no distinct ocelli or blotches. Thorns marked off pale milky-white pigment. Lateral tail folds creamy white pigment. Dorsal fins uniformly brown.

TABLE 1. Morphometrics (in mm) and meristics of the Holotype (INIDEP 793) and 9 paratypes of *Dipturus argentinensis*. Range and mean values expressed in % of total length, except total length and disc width in mm. SD = standard deviation.

		Paratypes		
	Holotype	Range	Mean	SD
Total length	765	403–935		
Disc width	572	297–683		
Disc length	463	57.8–61.3	59.2	1.1
Snout length (preorbital)	175	19.1–22.4	20.8	1.0
Snout length (preoral)	173	19.3–23.7	21.5	1.5
Orbit diameter	24	2.7–3.2	3.0	0.2
Distance between orbits	39	4.5–5.4	5.0	0.3
Orbit and spiracle length	34	4.1–4.7	4.4	0.2
Spiracle length	22	1.9–2.6	2.2	0.3
Distance between spiracles	46	6.2–6.6	6.3	0.2
Mouth width	62	8.1–8.9	8.5	0.2
Distance between nostrils	67	8.1–9.2	8.7	0.3
Width: first gill openings	15	1.2–1.9	1.6	0.3
Width:second gill openings	16	1.4–2.1	1.8	0.2
Width: third gill openings	16	1.3–2.1	1.7	0.2
Width: fourth gill openings	15	1.3–2.2	1.8	0.3
Width: fifth gill openings	14	1.2–2.0	1.5	0.2
Distance: first gill openings	119	15.1–16.7	15.9	0.6
Distance: third gill openings	102	12.8–13.9	13.3	0.3
Distance: fifth gill openings	71	8.9–11.4	9.8	0.7
Height: 1st dorsal fin	27	3.4–4.1	3.7	0.3
Length: 1st dorsal fin base	43	5.0–6.5	5.9	0.5
Height: 2nd dorsal fin	27	2.9–4.0	3.5	0.4
Length: 2nd dorsal fin base	41	4.8–5.7	5.3	0.3
Height caudal fin	6	0.7–1.3	1.1	0.2
Length: caudal fin base	32	3.8–6.4	4.9	0.8
Interdorsal distance	12	1.2–2.3	1.5	0.4
Tail width at axil of pelvic fin	25	2.8–4.1	3.5	0.5
Anterior pelvic fin length	84	11.4–13.8	12.6	0.8
Distance: snout to 1st dorsal fin	18	1.5–2.7	1.9	0.8
Distance: snout to cloaca	420	51.4–54.5	52.9	0.9
Distance: cloaca to caudal tip	345	45.5–48.6	47.2	1.0
Distance: 2nd dorsal fin to caudal tip	35	4.6–17.6	7.7	4.4
Distance:cloaca to 1st dorsal fin	211	27.2–29.7	28.8	1.0
Distance:cloaca to 2nd dorsal fin	266	34.3–36.7	35.9	0.8
Inner side clasper length	36	3.7–4.1	4.0	0.2
Upper jaw tooth rows	37	35–40	37.6	1.7
Lower jaw tooth rows	38	34–43	37.8	3.1

Lower surface of the disc brownish on central part, becoming pale brown to outer parts of pectoral fins and with darker margins. Anterior lobes of pelvic fins dark brown whereas posterior ones are lighter and narrowly edged grey. Underside of tail uniformly brown with light margins at level of dorsal fins.

Etymology. The specific epitet *argentinensis* is named in reference to the Argentine Sea where the type material was collected.

***Common name*.** New English name: Argentine Skate; new Spanish name: Raya hocicuda de cola larga.

***Barcode sequence*.** A 651 base pair amplicon from the 5´ region of the mitochondrial COI gene was bi-directionally sequenced for the holotype and six paratypes (GenBank accession numbers EU074410, EU074405, EU074406, EU074407, EU074408, EU074409, EU074411, respectively). The holotype and five of the paratype sequences were virtually identical, while the seventh differed by only a single nucleotide (0.146 % sequence divergence). The mtDNA COI barcode profile of the holotype is reported herein as an aspect of the type description:

CCTTTACTTAATTTTTGGTGCCTGAGCAGGCATGGTCGGGACTGGCCTAAGTCTTTTAATCCGAGC
AGAACTAAGTCAACCCGGGACCCTCCTGGGTGACGATCAGATTTATAATGTCATTGTTACAGCCCA
TGCCTTTGTAATAATCTTTTTTATGGTTATACCAATTATAATCGGCGGGTTTGGTAATTGACTCGTCC
CTTTAATAATTGGCTCCCCCGACATGGCCTTCCCACGCATAAATAACATAAGTTTCTGACTTTTACC
CCCCTCTTTTCTCCTCCTCCTGGCCTCCGCTGGAGTTGAGGCCGGGGCCGGAACAGGTTGAACTG
TCTACCCCCCTCTGGCAGGAAATCTGGCCCACGCGGGGGCCTCCGTAGACTTAACAATTTTCTCT
CTTCACTTGGCAGGTGTTTCATCTATTCTAGCCTCCATTAACTTCATCACCACAATTATTAACATAA
AACCACCAGCAATCTCTCAATACCAGACACCCTTATTCGTGTGATCAATTCTTGTTACAACTGTTTT
ACTTCTTATGGCCCTCCCAGTTCTAGCAGCCGGCATCACTATACTACTCACGGACCGTAATCTCAA
CACAACTTTCTTTGACCCGGCTGGAGGGGGCGACCCCATTCTATACCAACACTT

Details of the individuals sequenced along with those congeneric species for making comparisons are provided in Table 2.

TABLE 2. Voucher specimens of *Dipturus* sequenced for the barcode region of mtDNA COI.

Species	Sample ID	GenBank No.	Voucher No.	Collection site
D. argentinensis **n.s.**	INIDEP-T 0019	EU074405	INIDEP 799	46º 03’ S, 66º 46’ W
D. argentinensis **n.s.**	INIDEP-T 0319	EU074406	INIDEP 800	45º 54’ S, 63º 10’ W
D. argentinensis **n.s.**	INIDEP-T 0318	EU074407	INIDEP 803	45º 54’ S, 63º 10’ W
D. argentinensis **n.s.**	INIDEP-T 0317	EU074408	INIDEP 802	45º 54’ S, 63º 10’ W
D. argentinensis **n.s.**	INIDEP-T 0020	EU074409	INIDEP 798	45º 47’ S, 64º 47’ W
D. argentinensis **n.s.**	INIDEP-T 0021	EU074410	INIDEP 793	45º38’S, 64º08’W
D. argentinensis **n.s.**	INIDEP-T 0127	EU074411	INIDEP 795	50º 15’ S, 63º 35’ W
D. chilensis	INIDEP-T 0016	EU074400	INIDEP	46° 47´S, 66° 37´W
D. chilensis	INIDEP-T 0337	EU074401	INIDEP	34° 57´ S, 52° 26´W
D. chilensis	INIDEP-T 0015	EU074402	INIDEP	46° 47´S, 66° 37´W
D. chilensis	INIDEP-T 0018	EU074403	INIDEP	46° 31´S, 66° 59´W
D. chilensis	INIDEP-T 0014	EU074404	INIDEP	46° 47´S, 66° 37´W
D. trachyderma	INIDEP-T 0364		INIDEP 789	45° 30´S, 66° 17´W

Discussion

Morphological comparisons. Six nominal species of *Dipturus* have been recorded in the southwestern Atlantic (Gomes & Picado 2001, Soto & Mincarone 2001, Gomes & Costa 2003). Of these, two (*D. chilensis* and *D. trachyderma*) are known from the Argentinean and Uruguayan continental shelves (Cousseau *et al*. 2000, Menni & Stehmann, 2000, Cousseau *et al*. 2007). *Dipturus argentinensis* is most similar to the Thintail skate *D. leptocauda* because of the thin and relatively long tail. *Dipturus leptocauda* has interbranchial space rough with small scattered prickles and numerous rounded whitish blotches on the dorsal surface of the disc. Mouth width is three times preoral length. *Dipturus argentinensis* has interbranchial space smooth with no prickles and the dorsal surface of disc purplish brown with no distinct ocelli or blotches. Mouth width is less than three times in preoral length. The pattern of caudal thorns distinguishes the new species from the Yellownose skate *D. chilensis*, the South Brazilian skate *D. mennii* , the Roughskin skate *D. trachyderma* and the Thorny tail skate *D. diehli*. These longnose skates have three irregular rows of thorns on dorsal and dorsolateral surfaces of tail. *Dipturus argentinensis* has only one row of thorns on tail (Fig. 2A). Coloration and tail distinguish *D. chilensis* from the new species. The former has the dorsal surface of disc light brown with an oval dark blotch near the middle of the base of each pectoral fin, ventral side of disc white greyish, snout translucid, both sides of rostrum marked yellowish-white, and short and wide tail with more than one row of tail thorns (Fig. 2 B). Tail length is 42 % of body length, and its width is 9.6 % of tail length. Conversely, *Dipturus argentinensis* has the ventral surface of disc as dark as the upper side, snout not translucid, and a longer (47 % body length) and more slender (7.5 % of tail length) tail. Lack of dermal denticles on both sides of disc distinguishes the new species from *D. menni*, *D. trachyderma*, *D. diehli* and *D. teevani*. Additionally, both *D. mennii* and *D. diehli* exhibit median thorn rows on disc, and along with *D. trachyderma* also have lateral caudal thorns (Fig. 2 C), however these characteristics are both missing in *D. argentinensis*.

DNA sequence. Noting the consistent relationship between morphological taxonomy and molecular divergence values for the COI gene, Lefébure *et al*. (2006) and Hebert *et al*. (2004) have proposed a molecular threshold of divergence to aid in highlighting potentially cryptic species worthy of further investigation. To this end, we attempted to sequence all sympatric forms of *Dipturus* found in Argentinean waters. The seven individual specimens of *D. argentinensis* examined in this study yielded identical sequences despite being collected across a wide geographic range. They possess a unique haplotype that is substantially divergent from all other *Dipturus* species represented in Barcode of Life Data Systems (n=15 described + 5 undescribed species). This haplotype is presumed to be unique to this species, although complete barcode coverage of the genus would be necessary to confirm this. We also generated bidirectional sequences from five *D. chilensis* specimens and attempted to sequence two specimens of *D. trachyderma* in order to directly compare the new species with all other species occurring in Argentinean waters. Nucleotide sequences from *D. argentinensis* and *D. chilensis* exhibited a striking level of pairwise sequence divergence (3.15%) well above the threshold proposed by Lefébure *et al*. (2006) and Hebert *et al.* (2004). This degree of separation is also in agreement with range of COI divergence values found among other congeneric species of skates (e.g. Speis *et al*. 2006). Interestingly, *D. trachyderma*, the only other sympatric species of *Dipturus* in Argentinean waters, would not amplify (n=2) following the protocols used in this study. Because the tissues were fresh, the failure of COI to amplify in *D. trachyderma* is likely the result of mutational differences within the primer site(s) of the COI gene occurring in this species, indicating that *D. trachyderma* possesses a significantly divergent COI haplotype of its own. Following this reasoning, it is logical to conclude that the haplotype for *D. argentinensis* is divergent from the other two longnose skates present in the Argentine Sea. Moreover, this haplotype is unique when compared against the barcode profiles of more than 20 other *Dipturus* species collected globally (R. D. Ward & P. Smith, unpublished).

Distribution. Type specimens of the Argentine longnose skate *Dipturus argentinensis* were collected off central and south Patagonian continental shelf at 87-142 m depth (Fig. 3). The new species seems to occur

sympatrically with those longnose skates primarly inhabiting the shelf and upper slope of southern southwest Atlantic. Of the five recognized species of *Dipturus* that have been reported to occur in the southwest Atlantic, the Yellownose skate *D. chilensis* and the Roughskin *D. trachyderma* overlap in depth and geographic distribution with *D. argentinensis*.

Dipturus chilensis has been recorded from southern Brazil (Gomes & Picado 2001) to the southern tip of Argentina (Menni & Stehmann 2000) on the Atlantic side, and in the Pacific from central to southern Chile (Leible 1987, Leible *et al*. 1990). It is the most abundant rajid species in Argentina (García de la Rosa *et al.* 2004).

The Roughskin skate *D. trachyderma* has been found in the southeastern Pacific (Leible & Stehmann 1987), and southern Patagonia, Argentina (Krefft & Stehmann 1975, Menni & Gosztonyi 1977). Recent records have extended its distribution range to waters off southeast Brazil (Gadig 1998, Menni & Stehmann 2000; Gomes & Picado 2001), but those references probably refer to *D. menni* (See Gomes & Parago 2001 and Lamilla & Massa 2007).

The Thintail skate *D. leptocauda* was originally known only from the holotype collected off eastern Brazil at 500 m depth (Krefft & Stehmann 1975) but is currently known from scattered records off southeastern and southern Brazil (Gomes & Costa, 2003) though never reported to occur southwards (Cousseau *et al*. 2000).

The South Brazilian skate *D. mennii* is known from the upper slope of Rio Grande do Sul to the boundary between São Paulo and Rio de Janeiro, Brazil (Gomes & Paragó 2001). The Thorny skate *D. diehli* is known from a single specimen collected off southern Brazil at 480 m depth (Soto & Mincarone 2001).

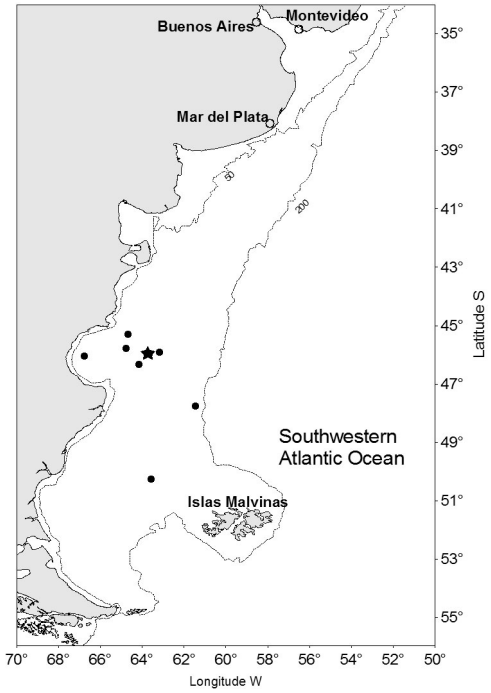

FIGURE 3. Geographic distribution of *Dipturus argentinensis* **n. sp**. based on material collected. Symbols represent more than one capture. Star indicates original locality of holotype.

According to the capture localities of the type specimens, *Dipturus argentinensis* appears to have a restricted range distribution occurring in the central and southern Patagonian region on the southwest Atlantic. No recorded specimens have been reported northwards 45º S. The region where the type specimens of the Argentine skate were collected is dominated by sub-Antarctic cold-temperate waters with low salinity. These waters have a predominantly NNE flow and form the Patagonian Current (Brandhorst & Castello 1971). Range temperatures and salinities of the localities where the specimens were collected were 5.9-9.38ºC, and

33.24-33.64 ups, respectively. These physical characteristics agree with the Magellanic Biogeographic Province where the specimens have been captured.

Two nominal species, *D. chilensis* and *D. nasuta* (Müller & Henle 1841), although considered belonging to the genus *Dipturus* (Mc Eachran & Dunn 1998) have been recently assigned to the resurrected genus *Zearaja* Whitley based on clasper morphology (Last & Gledhill 2007). According to said authors, this resurrected genus is similar to *Dipturus* in external morphology, neurocranium, pelvic girdle and scapulocoracoid, but has major differences in clasper morphology. Unfortunately all male specimens of *Dipturus argentinensis* available in this study were juveniles and therefore analysis of clasper morphology has not been made.

Key to species of *Dipturus* of the western South Atlantic

1a. Tail with more than one row of thorns 2
1b. Tail with a single median row of thorns 5
2a. Dorsal and ventral surface of disc rough, scattered with small prickles; ocular and scapular thorns present but median disc thorns absent; adults up to 200 cm TL ..... *D. trachyderma*
2b. Dorsal and ventral surface of disc relatively smooth, except interorbital region; scapular and median disc thorns either present or absent; adults less than 170 cm TL 3
3a. Scapular thorns and median disc thorns present; 5 or more nuchal thorns; interorbital region rough 4
3b. Scapular thorns and median disc thorns absent; a single nuchal thorn; interorbital region smooth ..... *D. chilensis*
4a. Dorsal surface of disc smooth except interorbital region; ventral surface of disc with few prickles in the interbranchial space; 5 or 6 nuchal thorns ..... *D. menni*
4b. Dorsal and ventral surface of disc with small scattered denticles; 8 nuchal thorns ..... *D. diehli*
5a. Interbranchial space rough with small scattered prickles; dorsal surface of disc with numerous rounded whitish blotches; mouth width three times preoral length; a pair of scapular thorns present *D. leptocauda*
5b. Interbranchial space smooth; dorsal surface of disc purplish brown with no distinct ocelli or blotches; mouth width less than three times preoral length; scapular thorns absent ..... *D. argentinensis* **n. sp.**

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