***Int. J. Morphol.*,**
35(1):42-46, 2017.

Meristic and Morphometric Analysis of Two Hagfish Species (*Myxine affinis* and *Notomyxine tridentiger*) from the Magellan Strait, Chile

Análisis Merístico y Morfométrico de dos Especies de Anguila Babosa (*Myxine affinis* y *Notomyxine tridentiger*) en el Estrecho de Magallanes, Chile

Mª. C. Pérez-Cuesta[1,3]; J. Del Campo[1]; G. Aedo[2]; C. Oyarzún[2] & E. Daza[3]

PÉREZ-CUESTA, M. C.; DEL CAMPO, J.; AEDO, G.; OYARZÚN, C. & DAZA, E. Meristic and morphometric analysis of two hagfish species (*Myxine affinis* and *Notomyxine tridentiger*) from the Magellan Strait, Chile. *Int. J. Morphol., 35(1)*:42-46, 2017.

SUMMARY: Myxinoids in Chile are represented by the subfamilies Eptatretinae and Myxininae, with a total of 14 species, the identification is complex due to the low level of morphological differentiation that characterizes this taxonomic group. Worldwide, hagfish are species of commercial value, and in Chile many attempts have been reported to initiate small-scale fisheries. The aim of the present study is describe the hagfish species caught in an incipient fishery of the Magellan Strait. Samples were collected in the Magellan Strait during eight fishing expeditions from June 2009 to October 2010 in Bahía Lomas (53º48`S; 70°46'W) and Agua Fresca (53º23`S; 70°45'W). The samples were taken at two depths, 0-70 meters and 71-140 meters. Taxonomic keys were used to identify the species. All specimens were individuals from the Myxininae subfamily. From a total of 3946 hagfishes, 99 % (n=3905) were the species *Myxine affinis* and the remaining 1 % were *Notomyxine tridentiger*, both reported for Chilean and Argentinean Patagonia. The range of variation for meristic variables recorded in this research was wider than those reported in the literature. This could be explained by differences in sample size between the present study and those previously published. Body proportions and meristic variables were not species specific in Myxine sp, so there is a large overlap of ranges between species, which makes their diagnostic use not applicable. This research updates the information and extends the meristic ranges for both species. The esophageocutaneous duct (in *N. tridentiger*) and the number of fused teeth (bicuspid in *M. affinis* and tricuspid in *N. tridentiger*) are the morphological characters that allow a clear identification in the field of the two species.

KEY WORDS: Hagfish; Magellan strait; *Myxine affinis*; *Notomyxine tridentiger*; Mertistic; Morphologic variables.

INTRODUCTION

The Agnatha is a superclass of jawless fish dating to the Ordovician Period (Carroll, 1998) that includes two orders: Myxiniformes (Myxinidae family, hagfishes) and Petromyzoniformes (Petromyzontidae family, lampreys). These taxonomic categories are known as Cyclostomes (Nelson, 2006). The hagfish has a cartilaginous skeleton and the notochord continues throughout life. It is characterized by double-edged fins and the absence of mandibles. The body is tubular, scaleless, with slime pores from head to tail. The species is adapted to live as a parasite and scavenger.

There are two subfamilies of Myxinoid in Chile: Eptatretinae and Myxininae, which differ mainly by the number of gill openings. Eptatretinae has a single genus (*Eptatretus*) with four species, *E. laurahubbsae* McMillan & Wisner, 1984; *E. polytrema* (Girard, 1854); *E. bischoffii* (Schneider, 1880) and *E. nanii* (Wisner & McMillan, 1988), while Myxininae is represented in Chile by the species *Myxine circifrons* (Garman, 1899); *M. hubbsoides* (Wisner & McMillan, 1994); *M. hubbsi* (Wisner & McMillan, 1994); *M. pequenoi* (Wisner & McMillan, 1994); *M. fernholmi* (Wisner & McMillan, 1994); *M. dorsum* (Wisner & McMillan, 1995); *M. debueni* (Wisner & McMillan, 1994); *M. australis* (Jenyns, 1842); *M. affinis* (Günter, 1870) and *Notomyxine tridentiger* (Garman, 1899) (Wisner & McMillan, 1988; Pequeño, 1989, 1997).

[1] Universidad de Concepción, Facultad de Ciencias Naturales y Oceanográficas, Concepción, Chile.
[2] Universidad de Concepción, Departamento de Oceanografía, Concepción, Chile.
[3] Instituto de Fomento Pesquero (IFOP), Valparaíso, Chile.

Between 2004 and 2006 hagfishes were fished commercially in the Magellan Strait, but without identifying or biologically describing captured species. Because of interest in this type of species as a potential fishing resource, a biological study was carried out in the waters of the Magellan Strait in 2009-2010. The aim of the present study was to provide a morphometric and meristic analysis of hagfish species captured in an incipient fishery in that region.

MATERIAL AND METHOD

Samples were collected from two fishing grounds in the south-eastern of the Magellan Strait: Bahía Lomas (53º48`S; 70°46'W) and Agua Fresca (53º23`S; 70°45'W) (Fig. 1). Individuals were collected with baited traps (< 300 g lamb entrails per trap), during eight fishing expeditions: winter and spring 2009, summer and autumn 2010. Hagfish specimens were identified with the following protocols: i) the Myxininae subfamily was determined using the key by Wisner (1999), which primarily considers the number of gill pouches and gill apertures, ii) the description proposed by Nani & Gneri (1951), which refers mainly to the number of fused teeth and the opening of the esophageocutaneous duct. This allowed distinguishing specimens from the genus Myxine and Notomyxine (specifically *N. tridentiger*), iii) the identification of *M. affinis* at species level was carried out considering the key and diagnosis offered by Wisner & McMillan (1995). Eleven specimens of *M. affinis* collected on October 2009 in Agua Fresca (53º48'44" ºS; 70°46'36" ºW) at depth of 105 meters, were deposited at the Natural History Museum in Santiago, Chile (MNHNCL). These samples can be found under the MNHNCL code P.7280 with the numbers 85, 90, 91, 96, 97, 98, 106, 108, 110, 111 and 112. Another 26 specimens were deposited at the Zoological Museum of the University of Concepción (MZUC-UCCC) with codes MZUC No 40262 and 40263 (*N. tridentiger*), and eight lots with codes MZUC No 40265, 40266, 40354, 40355, 40356, 40357, 40358 and 40359 (*M. affinis*). The protocols of Wisner & McMillan (1995) were followed for the meristic counts and measurements of morphometric variables of *N. tridentiger* and *M. affinis*. These protocols consider three body zones: a) prebranchial, from the edge of the nasal openings to the gill apertures; b) trunk, from the gill apertures to the cloaca; c) tail, from the cloaca to the end of the tail.

Morphometric characterization of the specimens considered the following variables: total length (TL), prebranchial length (PL), trunk length (TRL), tail length (TAL), body width (BW), body height with ventral fold (HVF) and without (HWF), and height over the cloaca (HC). Meristic counts considered the number of slime pores per body zone and the number of unicusp teeth. In both cases only the left side is represented because no differences between right and left sides have been found in these groups (Wisner & McMillan, 1995).

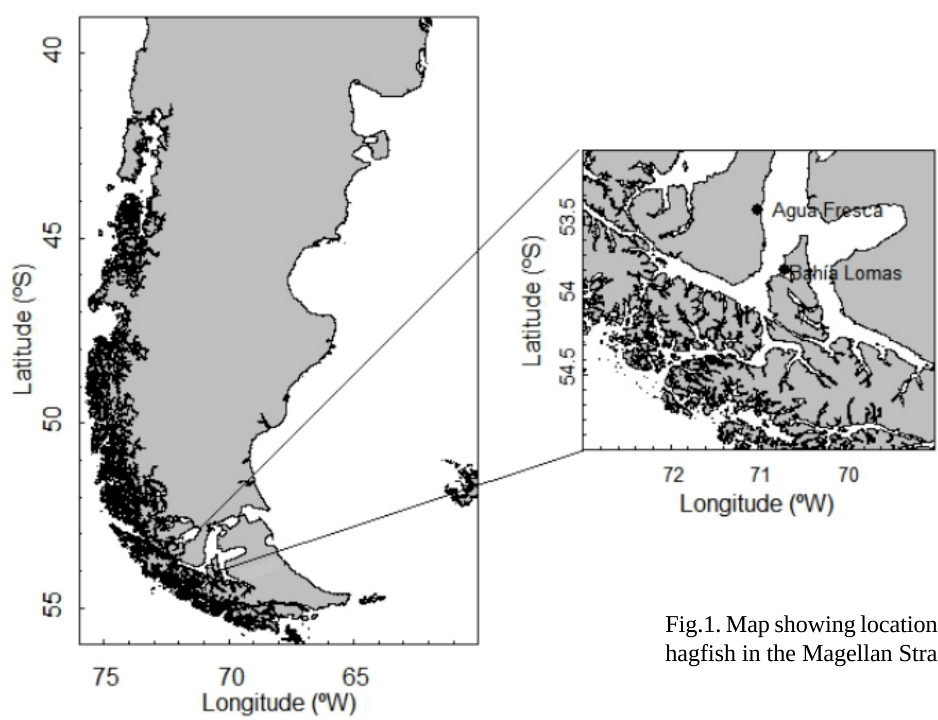

Fig.1. Map showing location of sampling sites of hagfish in the Magellan Strait.

Both meristic and morphometric variables are presented in their mean values and variation ranges, the latter expressed as percentage of TL. These variables are compared in terms of averages and ranges to data for *N. tridentiger* reported by Nani & Gneri. In the case of *M. affinis*, significant differences in slime pore counts (prebranchial, trunk, tail and total) were assessed by a frequency test (Chi-square), with Infostat statistic software version 2009e and Microsoft Excel 2010 software. Sample sizes were previously standardized for ranges of slime pores (E_i) from this research with respect to those presented by Wisner & McMillan (1995), according to the following expression:

$$E_i = \frac{F_i}{N_T} * N_t$$

Where F_i = is the number of specimens in this study in the range of pores i, N_T = total number of specimens in this study N_t= total number of individuals from Wisner & McMillan (1995).

RESULTS

A total of 3496 specimens of hagfish were collected from the Magellan Strait; 99 % (n=3905) were *M. affinis* and the remaining 1 % (n=41), *N. tridentiger*. 1242 specimens of *M affinis* were randomly selected to record the following variables: total length, total weight and sex, as well as for meristic and morphometric analyses. Only 37 specimens of *N. tridentiger* were used because the remaining four were severely damaged.

Meristic and morphometric description of *N. tridentiger.* Table I summarizes the morphometric measurements and the meristic counting of the samples identified as belonging to *N. tridentiger*. The species is characterized by a pair of gill apertures; behind which there is a third auxiliary opening called the esophageocutaneous duct (Wisner & McMillan, 1995). In addition, the first three teeth of the each anterior plates left and right are fused (tricuspid teeth), whereas on both posterior plates only the first two teeth are fused (bicuspid teeth).

Total length of the 37 specimens ranged from 34 and 66 cm (mean = 47 cm TL). The number of prebranchial slime pores varied between 17 and 32; slime pores of the trunk varied between 8 and 12; and the total number of slime pores between 82 and 113. In all cases, the upper limit was higher than reported by Nani & Gneri. The number of unicusp teeth on anterior and posterior plates varied between 7 and 10, with a total number of teeth in the range of 38-50 (Table I), which is more than indicated by Nani & Gneri (op. cit.).

Table I. Body proportions (mean and total length ranges in percentages), and pore and tooth counts for *T. tridentiger* recorded in the present study by Nani & Gneri (1951).

	N. tridentiger this study		*N. tridentiger* Nani & Gneri
Variable	Mean (***n***)	Range	Range (***n***)
Length			
Prebranchial	25 (37)	22 – 30	
Trunk	65 (37)	59 – 73	
Tail	11 (37)	10 – 14	
Total (cm)	47 (37)	34 – 66	
Body width	4 (37)	3 – 6	
Body depths			
with ventral finfold	5 (37)	4 – 7	
without ventral finfold	5 (37)	4 – 7	
over cloaca	4 (37)	3 – 5	
tail	4 (37)	3 – 6	
Slime pores			
Prebranchial	21 (37)	17 – 32	17 – 23
Trunk	64 (37)	52 – 84	57 – 65
Tail	10 (37)	8 – 12	8 – 11
Total	94 (37)	82 – 113	82 – 99
Unicusp teeth			
Anterior (c/u)	8 (24)	7 – 10	7 – 8 (**12**)
Posterior (c/u)	8 (24)	7 – 10	8 – 10 (**12**)
Total	41 (24)	38 – 50	40 – 46 (**12**)

n, effective size sample on which the record is based.

Meristic and morphometric description of *M. affinis.* Average values for all morphometric variables of *M. affinis* were similar to those reported by Wisner & McMillan (1995), although the ranges were higher (Table II). The same applied to the ranges for meristic variables (Table III), with the exception of the upper limit in the number of trunk slime pores and total slime pores, which were lower than those recorded by Wisner & McMillan (op. cit). There were significant differences in the number of slime pores in the prebranchial zone (Chi-square test, $c^2 = 32.69$, $P<0.05$) and tail ($c^2 = 37.43$, $P<0.001$), as well as the total body ($c^2 = 84.35$, $P<0.001$) between the samples reported by Wisner & McMillan (1995) and those in this study (Table IV). However, no differences were found in the number of slime pores from the trunk ($c^2 = 33.87$, $P>0.05$).

Table II. Body proportions (mean and total length ranges in percentages) for *M.affinis* recorded in the present study and by Wisner & McMillan (1995).

Variable	*M. affinis* this study mean (range)	*M. affinis* Wisner & McMillan mean (range)
Prebranchial length	30 (18 – 40)	28 (26 – 31)
Trunk length	60 (37 – 88)	59 (56 – 62)
Tail length	12 (7 – 16)	13 (11 – 15)
Total length (cm)	46 (26 – 84)	* (14 – 66)
Body width	4 (2 – 7)	4 (3 – 5)
Body depths		
with ventral finfold	5 (3 – 8)	5 (4 – 7)
without ventral finfold	4 (2 – 7)	5 (3 – 7)
over cloaca	3 (2 – 7)	4 (4 – 5)
tail	3 (2 – 6)	5 (4 – 6)
N	1242	271

n, effective sample size on which the record is base; *, not reported.

Table III. Slime pore and tooth counts (mean and range) of *M.affinis* recorded in the present study and according to Wisner & McMillan (1995).

	M. affinis this study mean (range) *n*	*M. affinis* Wisner & McMillan mean (range) *n*	*M. australis* Wisner & McMillan mean (range) *n*
Nº of slime pores			
Prebranchial	32 (18 – 44) 1242	37 (26 – 42) 288	31(22-42) 106
Trunk	65 (54 – 76) 1242	66 (57 – 79) 295	59 (51-68) 98
Tail	11(7 – 17) 1242	12 (9 – 14) 280	11(8-14) 88
Total	108 (91 – 122) 1242	112 (99 – 124) 279	102(86-119) 88
Nº of unicusp teeth			
Anterior (c/u)	8 (6 – 11) 1242	9 (7 – 10) 512	6(4-7) 125
Posterior (c/u)	8 (6 – 12) 1242	9 (7 – 10) 512	7(5-8) 125
Total	41 (32 – 54) 1242	42 (38 – 46) 256	34(29-38) 131

n, effective sample size on which the record is based; *, not calculated because of small sample size.

Table IV. Chi-square test to determine differences among the frequencies in the number of slime pores among specimens of *M. affinis* identified in the present study and according to Wisner & McMillan (1995).

Variable	χ^2	d.f.	P-value
Nº of slime			
Prebranchial	32.69	20	<0.05
Trunk	33.87	23	>0.05
Tail	37.43	8	<0.0001
Total	84.35	32	<0.0001

DISCUSSION

The present study confirms the presence and relative abundance of *M. affinis* and *N. tridentiger* in waters of the Magellan Strait. The esophageocutaneous duct and the number of fused tricuspid teeth for *N. tridentiger* and the number of fused bicuspid teeth for *M. affinis*, are morphological characters that allow rapid and clear identification of these two species that can be applied even during fieldwork. Body proportions and meristic variables were not species-specific and overlapped between species, making their use difficult as diagnostic. The overlap in range of morphometric and meristic variables was reported by Wisner & McMillan (1995) for 14 species of *Myxine*, with distribution on the coasts of the Pacific and Atlantic Oceans of North and South America. On the other hand, Mincarone & Fernholm (2010), in a revision and updating of hagfish taxonomy, determined differences in the number of cusps and slime pores of prebranchial and total regions, among other diagnostic characters that allowed developing an identification key for the species of the genus *Eptatretus*.

Almost all the ranges for body proportions and the numbers of slime pores and unicusp teeth in *N. tridentiger* and *M. affinis* were higher than those reported by Nani & Gneri and Wisner & McMillan (1995), respectively. This could be a consequence of the greater number of specimens analyzed in the present study. The effect of sample size is particularly relevant for *N. tridentiger* since the study by Nani & Gneri (op. cit.) involved only 12 specimens, whereas the analyses of the samples from the Magellan Strait considered 37 specimens. Nevertheless, it is probable that analyses with larger samples would extend the limits for these variables.

There were significant differences in the slime pore counts for most of the examined body regions (Table IV) in Wisner & McMillan (1995) and in this study. These discrepancies could be the result of different origins and structure of the samples. The samples used by Wisner & McMillan (op. cit.) were specimens of *M. affinis* collected from several geographic sites and at different dates that had been stored in 24 museum collections. On the other hand, our samples come from two localities close to each other, visited in the course of one year. Therefore, the differences could be the result of geographic variations in the frequency of slime pores as the product of different phenotypic expression of morphological characters depending on the particularities of each environment. In relation to the cusps, the range for total cusps, in our case was from 32 to 54, whereas Wisner & McMillan (1995) reported values from 38 to 46 for *M. affinis* and 34 to 36 for *M. dorsum.*

The range in the number of total pores of *M. dorsum* (counting two specimens) fall within the values found for *M. affinis*, which did not allow for differentiating them. Thus, the main characteristic to separate the two species is the caudal fold, which reaches only to the level of the cloacae in *M. affinis* and somewhat beyond in *M. dorsum* (Wisner & McMillan, 1995).

The number of pores in all the body segments of *M. affinis* and *M. australis* is similar, while the number of unicuspid teeth in *M. australis* is lower than what has been registered in this study for *M. affinis* (Table III). Finally, the most significant morphological characteristic that distinguishes the two species is the presence of whitish spots in the ventral side, which in *M. affinis* do not extend above the line of pores on the trunk, while in *M. australis* they extend to the area between the gill opening and the face.

In this research 1242 specimens of *M. affinis* were used for quantitative analysis, which allowed extending the range of meristic characters and showing that overlapping levels are very high, especially when the sample size is large. Thus, it is not possible to select a single character as identifier. This situation should be considered in future analysis.

ACKNOWLEDGEMENTS. The authors thank the Fishing Research Fund (FIP) for funding this research (FIP 2008-44), the Fisheries Development Institute (IFOP) of Punta Arenas for transporting samples, and Jean Paul Navarrete for his valuable contribution and advice about the statistical analysis.

PÉREZ-CUESTA, M. C.; DEL CAMPO, J.; AEDO, G.; OYARZÚN, C. & DAZA, E. Análisis merístico y morfométrico de dos especies de anguila babosa (*Myxine affinis* y *Notomyxine tridentiger*) en el Estrecho de Magallanes, Chile. *Int. J. Morphol., 35(1)*:42-46, 2017.

RESUMEN: Los Myxinoideos en Chile están representados por las subfamilias Eptatretinae y Myxininae, con un total de 14 especies, cuya identificación resulta compleja debido al bajo nivel de diferenciación morfológica que caracteriza a este grupo taxonómico. A nivel mundial las anguilas babosas constituyen especies de valor comercial, y en Chile se reportan varios intentos para iniciar pesquerías de pequeña escala. El presente estudio tuvo como propósito la descripción de las especies de anguila babosa capturadas en una pesquería incipiente del Estrecho de Magallanes. Los ejemplares fueron colectados durante ocho expediciones de pesca, desde junio de 2009 a octubre de 2010 en Bahía Lomas (53º48`S; 70°46'W) y Agua Fresca (53º23`S; 70°45'W). Las muestras fueron tomadas en dos rangos de profundidad, 0-70 metros y 71-140 metros. Todos los especímenes fueron pertenecientes a la subfamilia Myxininae. De un total de 3946 anguilas el 99 % (n=3905) pertenecieron a la especie *Myxine affinis* y el restante 1 % a la especie *Notomyxine tridentiger*, ambas reportadas para Chile y la Patagonia Argentina. El rango de variación para las variables merísticas, registradas en esta investigación, fue mayor a los reportados en literatura. Esto puede ser explicado por las diferencias en el tamaño de muestra entre el presente estudio y aquellos publicados previamente. Las proporciones corporales y las variables merísticas no fueron especie-específica en *Myxine* sp, por lo que existe una gran sobre posición de los rangos entre las especies, lo que hace que su uso como diagnóstico no sea aplicable. Esta investigación actualiza la información y extiende los rangos merísticos para ambas especies. El conducto esofágico-cutáneo (en *N. tridentiger*) y el número de dientes fusionados (bicúspide en *M. affinis* y tricúspide en *N. tridentiger*), son los caracteres morfológicos que permiten una identificación clara de ambas especies.

PALABRAS CLAVE: Anguila babosa; Estrecho de Magallanes; *Myxine affinis*; *Notomyxine tridentiger*; Variables merísticas y morfológicas.

REFERENCES

Carroll, R. L. *Vertebrate Paleontology and Evolution*. New York, W. H. Freeman & Co., 1998. pp.698.

Martini, F.; Heiser, J. B. & Lesser, M. P. A population profile for Atlantic hagfish, *Myxine glutinosa* (L.) in the Gulf of Maine. Part I: Morphometrics and reproductive state. *Fish. Bull., 95(2)*:311-20, 1997.

Mincarone, M. M. & Fernholm, B. Review of the Australian hagfishes with description of two new species of Eptatretus (Myxinidae). *J. Fish Biol., 77(4)*:779-801, 2010.

Nani, A. & Gneri, F. S. Introducción al estudio de los Myxinoideos sudamericanos I. Un nuevo género de "Babosa de Mar", "Notomyxine". *Zoolog. Rev. Mus. Argent. Cienc. Nat. Bernadino Rivadavia, 2(4)*:183-225, 1951.

Nelson, J. S. *Fishes of the World*. 4th ed. Hoboken, John Wiley & Sons, 2006. pp.622.

Pequeño, G. Peces de Chile. Lista sistemática revisada y comentada. *Rev. Biol. Mar. Oceanogr. (Valparaíso), 24(2)*:1-132, 1989.

Pequeño, G. Peces de Chile. Lista sistemática revisada y comentada: Addendum. *Rev. Biol. Mar. Oceanogr. (Valparaíso), 32(2)*:77-94, 1997.

Wisner, R. L. & McMillan, C. B. A new species of hagfish, genus Eptatretus (Cyclostomata, Myxinidae), from the Pacific Ocean near Valparaiso, Chile, with new data on *E. bischoffii* and *E. polytrema. Trans. San Diego Soc. Nat. Hist., 21*:227-44, 1988.

Wisner, R. L. & McMillan, C. B. Review of new world hagfishes of the genus Myxine (Agnatha, Myxinidae) with descriptions of nine new species. *Fish. Bull., 93(3)*:530-50, 1995.

Wisner, R. L. Descriptions of two new subfamilies and a new genus of hagfishes (Cyclostomata: Myxinidae). *Zool. Stud., 38(3)*:307-13, 1999.

Corresponding author:
María Cristina Pérez Cuesta
Instituto de Fomento Pesquero (IFOP)
Blanco #839
Valparaíso - CHILE

Email: mperezc@udec.cl

Received: 08-09-2016
Accepted: 28-11-2016