MINISTÉRIO DA CIÊNCIA, TECNOLOGIA, INOVAÇÕES E COMUNICAÇÕES
MUSEU PARAENSE EMILIO GOELDI
PROGRAMA DE PÓS-GRADUAÇAO EM BIODIVERSIDADE E EVOLUÇÃO

CINTIA OLIVEIRA CARVALHO

TAXONOMIA INTEGRATIVA DO COMPLEXO DE ESPÉCIES *Haemulon steindachneri* (JORDAN & GILBERT, 1882) (EUPERCARIA; HAEMULIDAE) COM A DESCRIÇÃO DE UMA NOVA ESPÉCIE PARA O ATLÂNTICO

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Dissertação apresentada ao Museu Paraense Emilio Goeldi, como parte das exigências do Curso de Mestrado do Programa de Pós-Graduação em Biodiversidade e Evolução, Área de Concentração Evolução e Dinâmica da Diversidade Biológica para obtenção do Título de Mestre.

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ADVERTÊNCIA

Essa dissertação não é válida como publicação, conforme capítulo 3 do CÓDIGO INTERNACIONAL DE NOMENCLATURA ZOOLOGICA. Portanto, os novos nomes e mudanças taxonômicas propostos aqui não têm validade para fins de nomenclatura ou prioridade.

WARNING

This dissertation is not valid as publication, as described in the chapter 3 of the INTERNATIONAL CODE OF ZOOLOGICAL NOMENCLATURE. Therefore, taxonomic changes and new names proposed here are not valid for nomenclature or priority purposes.

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RESUMO

Haemulon steindachneri (Jordan & Gilbert, 1822), popularmente conhecida como cocoroca-de-boca-larga, latin grunt e burro latino, possui uma ampla distribuição, ocorrendo na costa Ocidental do Atlântico e na costa Oriental do Pacífico. A espécie é bastante abundante em áreas rasas e em costões rochosos, apresentando uma grande importância na manutenção da integridade destes ecossistemas. Filogenias moleculares recentes, baseadas em DNA mitocondrial e nuclear, sugerem que *H. steindachneri* é representada por duas linhagens distintas na costa do Atlântico e do Pacífico das Américas. O presente estudo procura definir o *status* taxonômico das linhagens do Atlântico e do Pacífico de *H. steindachneri*, tanto com base em evidências morfológicas como moleculares, incluindo exemplares de áreas geográficas ainda não examinadas. Associado à evidência molecular, nossos resultados mostram diferenças morfológicas que distinguem a linhagem do Atlântico e do Pacífico: nadadeira anal geralmente com oito raios (vs. geralmente nove raios no Pacífico); 13−15 escamas abaixo da linha lateral, raramente 12 (vs. 12 escamas abaixo da linha lateral, raramente 13 no Pacífico), margem posterior da maxila robusta com extremidade suavemente angulada (vs. maxila menor com extremidade moderadamente convexa), e a presença de uma mancha sobre o pré-opérculo larga e robusta, sem formato definido (vs. mancha estreita, com extremidade anterior afilada e posterior reta, assemelhando-se a um triângulo no Pacífico). Adicionalmente, a análise de DNA barcode gerou dois *clusters* distintos, com o primeiro incluindo exemplares do Brasil e Caribe e o segundo, exemplares do Pacífico, com distância genética de 7,4%, separadas por 35 pares de base. Desta forma, tanto evidências moleculares como morfológicas sustentam que as linhagens de *H. steindachneri* do Atlântico e do Pacífico representam espécies distintas. Considerando a localidade-tipo de *H. steindachneri*, Panamá e Mazatlán no Pacífico, reconhecemos que a linhagem do Atlântico é uma nova espécie.

Palavras-chave: biogeografia marinha, redescrição, reef fish, Western Atlantic, Eastern Pacific

ABSTRACT

Haemulon steindachneri (Jordan & Gilbert, 1822), popularly known as cocoroca-de-boca-larga, latin grunt and burro latino, is widely distributed, occurring on the Western Atlantic coast and on the Eastern Pacific coast. The species is abundant in shallow areas and in rocky shores, presenting a great importance in the maintenance of the integrity of these ecosystems. Recent molecular phylogenies, based on mitochondrial and nuclear DNA, suggest that *H. steindachneri* is represented by two distinct lineages on the Atlantic and Pacific coasts of the Americas. The present study seeks to define the taxonomic status of *H. steindachneri* of the Atlantic and Pacific lineages, based on morphological and molecular evidence, including specimens from geographical areas not yet examined. Associated with molecular evidence, our results show morphological differences that distinguish the Atlantic and Pacific lineages: anal fin usually with eight rays (vs. generally nine rays in the Pacific); 13−15 scales below the lateral line, rarely 12 (vs. 12 scales below the lateral line, rarely 13 in the Pacific), posterior margin of the maxilla, robust with smoothly angled end (vs. maxilla with moderately convex extremity), and presence of a spot on the pre-operculum, large and robust, with no definite shape (vs. narrow spot, with anterior extremity narrow and posterior straight, resembling a triangle in the Pacific). In addition, DNA barcode analysis generated two distinct clusters, with the first one including Brazil and Caribbean specimens and the second one Pacific specimens, with genetic distance of 7.4%, separated by 35 base pairs. In this way, both molecular and morphological evidence support that the lineages of *H. steindachneri* from the Atlantic and the Pacific represent distinct species. Considering the typical locality of *H. steindachneri*, Panama, and Mazatlan in the Pacific, we recognize that the Atlantic lineage is a new species.

Key words: marine biogeography, redescription, reef fish, Western Atlantic, Eastern Pacific

INTRODUÇÃO GERAL

Os peixes representam mais da metade das espécies de vertebrados viventes, o que equivale a aproximadamente 31.000 espécies válidas e cerca de 300 novas espécies descritas anualmente (HELFMAN et al., 2009; ESCHMEYER et al., 2010). A diversidade de espécies marinhas é relativamente bem conhecida em algumas regiões do mundo, principalmente em relação às espécies com interesse comercial. Entretanto, lacunas importantes no conhecimento da ictiofauna de peixes marinhos estuarinos ainda existem (MARCENIUK et al, 2012). Estima-se que o número de espécies de peixes marinhos não descritos chegue a 5.000, número duas vezes maior em relação ao número de espécies descritas nos últimos 19 anos (ESCHMEYER et al., 2010). O conhecimento taxonômico deficitário representa um dos principais obstáculos ao conhecimento biogeográfico e a tomada de medidas para conservação da diversidade no ambiente marinho. Nesse contexto, muitas espécies de peixes marinhos precisam ser reavaliadas taxonomicamente, para se obter mais informações sobre suas histórias evolutivas, bem com suas definições e amplitude de distribuição (MARCENIUK 2008, 2012, 2016).

A especiação, ainda hoje, é um dos tópicos mais controversos da biologia evolutiva. Quando se diz respeito aos organismos marinhos, mais desafios ainda são encontrados pelos cientistas para explicar os modelos de especiação, já que nesses ambientes muitas das espécies possuem amplas faixas geográficas, grandes tamanhos populacionais e longos estágios de fase larval que permitem a conectividade genética entre áreas biogeográficas distantes (LESSIOS & ROBERTSON, 2006; BOWEN et al., 2013). No entanto, fatores como extinção, recolonização, fragmentação da população e expansão de área, originados pelos eventos históricos de isolamentos, também podem ter influenciado a estrutura genética populacional desses organismos marinhos (FAUVELOT; PLANES, 2002). A glaciação que ocorreu durante o período do Pleistoceno, por exemplo, ocasionou mudanças periódicas no nível do mar, acarretando uma série de alterações na estrutura dos oceanos, como: a exposição dos recifes de corais, alteração nas direções das correntes da superfície do mar e até mesmo isolamento de oceanos inteiros, tais mudanças afetaram a disponibilidade de hábitat nesses ambientes, levando o possível surgimento de ciclos de isolamento populacional, contato secundário e subsequente especiação nas espécies de peixes dessas áreas (BENZIE, 1999; VORIS, 2000; LERAY et al., 2010).

Um segundo evento histórico responsável por grandes mudanças no ambiente marinho do Novo Mundo, assim como sua biota foi o soerguimento do Istmo do Panamá (O'DEA et al.,

2016). Este iniciou há mais de 30 milhões de anos, acarretando na separação do mar do Caribe do leste tropical do oceano Pacífico (O'DEA et al., 2016). No final do Plioceno, até pelo menos 3,2 milhões de anos atrás, ocorreu o cessamento do soerguimento do istmo, o que levou a interrupção dos canais interoceânicos, que antes possuíam um corpo contínuo de água, com fluxo gênico entre os membros das populações existentes (LEIGH et al., 2014; O'DEA et al., 2016). Este evento paleogeógrafico ocasionou a divergência de muitas linhagens, originando assim espécies pares reconhecidas como "espécies geminadas" por Jordan (1908). Como exemplo, estudos moleculares com a subfamília de arraias Styracurinae (CARVALHO et al., 2016) têm evidenciado que *Styracura schmardae* (Werner), que ocorre na costa do Atlântico, e *S. pacifica* (Beebe & Tee-Van), costa do Pacífico, são espécies irmãs (geminadas) (LOVEJOY, 1996; DE CARVALHO; ASCHLIMAN et al., 2012; NAYLOR et al., 2012; LAST et al., 2016). Jordan (1908) mostrou a presença de algumas supostas espécies geminadas nos Haemulídeos do Novo Mundo, como o *Haemulon album* Cuvier / *H. sexfasciatum* Gill, *H. parra* (Desmarest) / *H. Scudderii* Gill, entre outros (JORDAN, 1908). Dessa forma, o gênero *Haemulon* (Haemulidae) se mostra um excelente grupo para se estudar a história evolutiva e a dinâmica de especiação nos trópicos do Novo Mundo, já que possui espécies distribuídas em ambos os oceanos Atlântico e Pacífico.

Com o intuito de contribuir para o melhor conhecimento da biodiversidade de peixes marinhos, diversas ferramentas moleculares vêm sendo utilizadas, dentre elas, o "DNA barcoding", proposto por Hebert et al. (2003). Esta é uma técnica importante para a taxonomia molecular, já que visa a identificação das espécies com base na diversidade do gene mitocondrial citocromo c oxidase subunidade I (COI). Este gene é um importante marcador molecular por ser espécie-específico, onde suas sequências tendem a variar entre espécies, porém são relativamente constantes entre os indivíduos da mesma espécie (HEBERT et al., 2003). Muitos trabalhos têm demonstrado que a metodologia DNA barcode é eficiente até mesmo para a fauna largamente diversa dos peixes da região neotropical e regiões marinhas (CARVALHO et al., 2011, PEREIRA et al., 2011, 2013, MARCENIUK et al., 2016).

A taxonomia de algumas espécies de Haemulidae ainda carecem de revisão taxonômica (TAVERA et al., 2012), condição relacionada a grande diversidade ecológica e morfológica da família, bem como à semelhança morfológica e de padrão de coloração que muitas espécies, estreitamente relacionadas, compartilham (LIANG et al., 2012). Haemulidae Gill, 1885 é uma das famílias mais diversas da subordem Percoidei, com aproximadamente 145 espécies

distribuídas em 19 gêneros nominais (TAVERA et al., 2018). Seus representantes estão distribuídos nos oceanos Atlântico, Índico e Pacífico, com maior ocorrência nas regiões costeiras entre os trópicos (NELSON et al., 2016). A maioria de suas espécies é marinha, porém algumas delas podem ser encontradas em águas salobras (ESCHMEYER et al., 1983). Vivem em ambientes recifais, praias arenosas, estuários e bancos de fanerógamas, utilizando estes ambientes para proteção, alimentação e reprodução (LINDEMAN; TOXEY, 2002; APPELDOORN et al., 2009). Sua dieta é constituída principalmente por microcrustáceos nos primeiros estágios de vida, e quando atingem a maturidade sexual, apresentam hábitos generalistas (COCHERET de la MOROINIERE et al., 2003; FLORES-ORTEGA et al., 2014). Os haemulídeos são chamados de "roncadores", por possuírem a habilidade de produzirem sons, provocados pelo atrito dos dentes faringeanos e amplificados pela bexiga natatória (BURKENROAD, 1930). Muitos representantes da família têm uma grande importância comercial para pesca em todo o mundo (LIANG et al., 2012).

O gênero *Haemulon* Cuvier, 1829 é um excelente grupo para se estudar evolução e especiação nos trópicos do novo mundo, já que possui 19 espécies válidas, distribuídas em ambientes de recifes tropicais rasos do Atlântico e Pacífico Oriental (COURTENAY, 1961; ROCHA et al., 2008). As espécies deste gênero formam cardumes com até milhares de indivíduos, sendo considerados um dos mais importantes grupos de peixes recifais devido a sua abundância, valor comercial e importância trófica como presa/predador (LINDEMAN; TOXEY, 2002; FERREIRA et al., 2004). Apresentam diferenças morfológicas significativas entre os adultos, porém, os jovens são muito semelhantes entre si (COURTENAY, 1961). No oeste do Atlântico, são encontradas 15 espécies de *Haemulon*, e no Pacífico Oriental cinco, com a espécie *Haemulon steindachneri* (JORDAN; GILBERT, 1822) nominalmente compartilhada nas duas regiões (LINDEMAN, 1986; ROCHA; ROSA, 1999).

Haemulon steindachneri (Figura 1), conhecida vulgarmente como cocoroca-de-boca-larga, latin grunt e burro latino, é encontrada na costa do Atlântico Ocidental (Costa Rica até Santa Catarina-Brasil) e no Pacífico Oriental (Golfo da Califórnia até o Peru) (MENEZES; FIGUEIREDO, 1980; LINDEMAN; TOXEY, 2002; LINDEMAN et al., 2016). A espécie possui grande abundância em áreas rasas e em costões rochosos, sendo reconhecida como de grande importância na manutenção da integridade destes ecossistemas (FURIA, 1996; ROCHA, 1997). *Haemulon steindachneri* (JORDAN; GILBERT, 1882) foi descrita inicialmente como *Diabasis steindachneri* Jordan e Gilbert, 1882, tendo como características

diagnósticas: uma barra preta azulada na parte anterior inferior do opérculo, parcialmente oculta pelo ângulo do pré-opérculo; uma grande mancha arredondada e enegrecida na extremidade do pedúnculo caudal e base da nadadeira caudal. Posteriormente, Jordan e Swain (1884) revisaram novamente *D. steindachneri*, alterando o gênero, já que o nome *Diabasis* Desmarest, 1823 era pré-ocupado em Coleoptera.

Figura 1. Vista lateral esquerda de *Haemulon steindachneri*. A. Linhagem do Brasil MPEG 35753 Amapá.

Quase oitenta anos depois, Courtenay (1961) revisou as espécies do gênero *Haemulon* do Atlântico Ocidental e, em suas diagnoses, mostrou que *H. steindachneri* possuía 51 ou 52 escamas na linha lateral; nadadeira dorsal contendo usualmente 12 espinhos, 15−17 raios; nadadeira anal com três espinhos e oito ou nove raios; nadadeira peitoral, 17 ou 18 raios; e 22−25 raios branquiais. Courtenay (1961) também afirmou que os espécimes que revisou possuíam uma grande mancha preta abaixo da margem do pré-opérculo e uma mancha negra na base da nadadeira caudal. Hong (1977), revisando espécies de *Haemulon* do Pacífico Oriental, comparou seus dados de *H. steindachneri* do Atlântico e Pacífico com os de Courtenay (1961) e sugeriu que os espécimes de *H. steindachneri* do Atlântico e do Pacífico são similares, exceto pelo comprimento do focinho e comprimento do maxilar superior que são 1 a 2% SL maiores nos espécimes do Atlântico.

Posteriormente, filogenias moleculares de Haemulidae e do gênero *Haemulon* reconheceram duas formas distintas de *H. steindachneri* para o Atlântico e Pacífico (ROCHA et al., 2008; TAVERA et al. 2012; 2018). Rocha et al. (2008) mostraram que as populações de

H. steindachneri do Brasil, Caribe e Pacífico possuem alta variação intraespecífica; as formas do Pacífico e Atlântico mostraram diferenças tanto em relação ao DNA mitocondrial, quanto com DNA nuclear, levando os autores a sugerir que essas duas formas devem ser tratadas como duas espécies distintas. Rocha et al. (2008) também sugeriram uma espécie emergente entre as formas brasileira e venezuelana, já que estas apresentaram uma divergência de sequência de 0.7% no conjunto de dados do DNA mitocondrial. Tavera et al. (2012; 2018) também sugeriu que *H. steindachneri* possui duas formas distintas, uma no Pacífico e outra no Atlântico. Apesar de esses trabalhos sugerir diferenciações em nível de espécie em *H. steindachneri*, em nenhum estudo subsequente foi realizado um estudo taxonômico com objetivo de revisar a nomenclatura adotada e/ou as relações entre as linhagens do Pacífico e do Atlântico. Nesse sentido, o presente trabalho visa elucidar a sistemática do complexo de espécies *Haemulon steindachneri* através da combinação de caracteres morfológicos e moleculares.

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Integrative taxonomy of the species complex *Haemulon steindachneri* (Jordan and Gilbert, 1882) (Eupercaria; Haemulidae) with a description of a new species from the western Atlantic

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Abstract

Haemulon steindachneri (Jordan and Gilbert) (Haemulidae), popularly known as "cocoroca-de-boca-larga", "latin-grunt" or "latin-burro", represents a species complex found on the Atlantic western coast and on the Pacific eastern coast, condition confirmed recently by molecular phylogenies. In the present study, DNA barcoding analysis recognizes two distinct clusters, the first includes Brazil and Caribbean, and the second is composed of Pacific specimens, with genetic distance of 7.4%, differentiated by 35 base pairs. In addition to the molecular evidence, our results show morphological differences that distinguish the Atlantic lineage from that of the Pacific: anal fin, usually, with eight rays (vs. generally nine rays in Pacific); 13–15 scales below the lateral line, rarely 12 (vs. 12 scales below the lateral line, rarely 13 in Pacific), posterior margin of the maxilla robust with a slightly angled end (vs. smaller maxilla with moderately convex extremity), and presence of a spot on the pre-operculum, broad and robust, with no definite shape (vs. narrow spot, with anterior extremity

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tuned and posterior straight, resembling a triangle in Pacific). Therefore, based on both molecular and morphological evidences, *H. steindachneri* is redescribed for the Pacific coast while a new species is described for the Atlantic coast.

Key words: marine biogeography, DNA barcode, morphology, Western Atlantic, Eastern Pacific, biodiversity

1. Introduction

The different processes of species formation are still one of the most controversial topics in evolutionary biology (Gaither et al., 2015). When it comes to marine organisms, the challenges encountered in the understanding of the speciation process are even greater, as many marine species have wide distribution, large populations and larval stages that provide genetic connectivity between different geographic areas (Lessios and Robertson, 2006; Bowen et al., 2013). Factors such as extinction, recolonization, fragmentation of populations, and area expansion resulting from different historical events may also influence the population genetic structure of marine organisms (Fauvelot and Planes, 2002). In this sense, the rise of the Isthmus of Panama was an important historical event, responsible for major changes in the marine environment of the New World, as well as its biota (O'Dea et al., 2016).

Beginning over 30 million years ago, the rise of the Isthmus of Panama was responsible for the separation of the Caribbean Sea from the tropical east of the Pacific Ocean (O'Dea et al., 2016). At the end of the Pliocene to at least 3.2 million years ago, the cessation of this uplift occurred, which led to discontinuation of interoceanic channels that previously had a continuous body of water with gene flow between existing populations (Leigh et al., 2014; O'Dea et al., 2016). This paleogeographic event led to the divergence of many lineages, thus giving rise to some species pairs recognized as "geminate species" by Jordan (1908), which showed the presence of some supposed geminate species in haemulids of the New World, such as *Haemulon album* Cuvier/*H. sexfasciatum* Gill, or *H. parra* (Desmarest)/*H. scudderii* Gill, among others (Jordan, 1908). Considering this, the genus *Haemulon* (Haemulidae) is an excellent group to study the evolutionary history and dynamics of speciation in the tropics of the New World, since it has species distributed in both Atlantic and Pacific Oceans (Rocha et al., 2008).

The genus *Haemulon* Cuvier, 1829 has 19 valid species found in shallow tropical reef environments of the Atlantic and Eastern Pacific (Courtenay, 1961; Rocha et al., 2008). Species of this genus form schools with up to thousands of individuals, with commercial value and great trophic importance as prey/predators (Lindeman and Toxey, 2002; Ferreira et al., 2004). In the western Atlantic 15 species of *Haemulon* are found and in the Eastern Pacific five species, with *H. steindachneri* cited both for the Western Atlantic (Costa Rica to Santa Catarina, Brazil), as well as the Eastern Pacific (Gulf of California to Peru) (Lindeman and Toxey, 2002; Lindeman et al., 2016). The species is abundant in shallow areas and in rocky shores, with recognized importance in maintaining the integrity of these ecosystems (Furia, 1996; Rocha, 1997).

Recently, molecular phylogenies of Haemulidae and *Haemulon* have recognized two distinct forms of *H. steindachneri* from the Atlantic and Pacific (Rocha et al., 2008; Tavera et al., 2012; Tavera et al., 2018). Rocha et al. (2008) showed that the populations of *H. steindachneri* from Brazil, Caribbean and Pacific have high intraspecific variation, with the Pacific and Atlantic forms showing differences in both mitochondrial and nuclear DNA, suggesting that the two recognized lineages could be treated as distinct species. Tavera et al. (2012; 2018; 2019) also concludes that *H. steindachneri* is represented by two distinct lineages, one in the Pacific and one in the Atlantic. Despite recent molecular studies showing species-level differences in *H. steindachneri*, no taxonomic study has reviewed the nomenclature adopted and/or the relationships between the Pacific and Atlantic lineages. In this sense, based on the combination of morphological and molecular characters, the present study redescribes *H. steindachneri* as a species restricted to the Eastern Pacific while a new species belonging to the *H. steindachneri* species complex is described for the Western Atlantic.

2. Material and Methods

2.1. Morphological data and analysis

The meristic and morphometric data were obtained following Courtenay (1961), with the following additions: HCP, height of the caudal peduncle: shorter distance between the dorsal and ventral margins of the caudal peduncle; WCP**,** width of the caudal peduncle: on the same axis as the HCP; CPL, caudal-peduncle length: between the posterior margin of the base of the last ray of the anal fin at the end of the hypural plate; DSPf, distance between the snout and the pelvic fin: from the tip of the snout to the base of the first ray of the pelvic fin; PvL,

pelvic-fin length: from the base to the end of the longest ray; DL, dorsal-fin height: from the base of the fourth ray to its extremity; PtL, pectoral-fin length: from the base to the end of the longest ray; AH, anal-fin height: from the base of the second ray to its extremity; PPL, pre-pectoral length: from the tip of the snout to the base of the first ray of the pectoral fin; HH, head height: the highest vertical height of the head on the posterior margin of the operculum; ID, Interorbital distance: shorter distance between the inner margins of the orbits; POL, post-orbital length: distance from the posterior margin of the orbit to the end of the posterior membranous part of the operculum; DBL, dorsal-fin base length: from the anterior margin of the base of the first ray to the posterior margin of the base of the last ray; ABL, anal-fin base length: from the anterior margin of the base of the first ray to the posterior margin of the base of the last ray; WM, width of the mouth: distance between the inner margins of the angles of the mouth; PJW, pre-jaw width: greater distance between the outer side edges of the pre-jaw, considering the two plates; LCL, lower caudal-fin lobe length: from the base of the first ray to the end of the longer ray; UCL, upper caudal-fin lobe length: from the base of the first ray to the end of the longer ray.

The morphological characters, measurements and counts were examined, preferably, on the left side of the specimens. The measurements were obtained with assistance of an ichthyometer with precision of 1 mm and a digital caliper with an accuracy of 0.1 mm. The counts were made with the assistance of stylet and fine-tipped tweezers under a stereoscopic microscope. The spines are presented as upper-case Roman numerals, branched rays as Arabic numerals and the unbranched rays as lower-case Roman numerals. Values observed in the holotype are represented by asterisks "*". Osteological information such as number of vertebrae, pleural ribs, and accessory rays of the anal and dorsal fin were also collected from digital radiographs. The measures are presented as a percentage of the standard length, aiming for a comparative analysis between the lineages and geographic areas examined. The meristic and morphometric characters were compared by areas and presented in the form of tables.

We analyzed 115 specimens belonging to the *H. steindachneri* species complex, deposited in the following ichthyological collections: Academy of Natural Sciences of Drexel University (ANSP), Philadelphia; Zoological Collection of Santa Cecilia (AZUSC) at the University of Santa Cecília, Santos; California Academy of Sciences (CAS), San Francisco; Natural History Museum of Los Angeles County (LACM), Los Angeles; Museu Paraense Emílio Goeldi (MPEG), Belém; Museu de Zoologia da Universidade de São Paulo (MZUSP),

São Paulo; Coleção do Laboratório de Biologia e Genética de Peixes (LBP) da Universidade Estadual de São Paulo (UNESP), Botucatu; National Museum of Natural History (USNM), Washington.

2.2. Molecular data and analysis

A total of 29 partial sequences of COI (517 bp) were obtained from *Haemulon* sp. n., which will be deposited in GenBank. The vouchers were deposited in the ichthyological collection of MPEG. In addition, seven additional COI sequences from *Haemulon* sp. n. and 12 of *H. steindachneri* were obtained from GenBank and BOLD.

Molecular techniques were performed at the LBP-UNESP, Botucatu and at the MPEG. Total DNA was obtained using a DNeasy Blood and Tissue kit (Qiagen, Hilden, Germany) by the mouse-tail protocol. The 517 bp fragment at the 5 ' end of the mitochondrial COI gene was amplified by PCR using the primers FishF1 and FishR1 (Ward et al., 2005).

The reactions were performed in a reaction volume of 12.5 μL: 0.5 μl of DNTP (1.25 mM), 1.25 μl of buffer (10x), 0.5 μl of MgCl2 (50 mM), 0.25 μl of each primer (200 ng/μl), 0.5−2.0 μl of total DNA (100 ng/μl), 0.2 μl of Taq DNA Polymerase (5U/μl) and purified water to complete the final volume of the reaction. The amplification protocol consisted of initial denaturation at 95ºC for two minutes, followed by 35 cycles of denaturation at 94ºC for 30 seconds, hybridization at 64ºC for one minute, extension at 72ºC for one minute, and final extension at 72ºC for ten minutes. PCR products were visualized on 1% agarose gel, and further purified using the ExoSap-IT kit (USB Europe GmbH, Staufen, Germany) at 37ºC for 60 min followed by 15 min at 80ºC. Sequencing reactions were conducted using a BigDye Terminators cycle kit (Applied Biosystems, California, USA) according to the manufacturer's instructions. Sequences were generated on ABI 3130-Genetic Analyzer (Applied Biosystems, California, USA) automated DNA sequencer at LBP and MPEG.

The bidirectional sequence "contigs" was assembled, and the sequences were aligned using Geneious v.5.6 (Kearse et al., 2012) to obtain consensus sequences and to check for indels or possible stop codons. The MEGA v 7.0 program (Kumar et al., 2016) was used to construct neighbor joining (NJ) dendrograms (see Supplement I Appendix A) and to estimate the Kimura 2-parameter (K2P) (Kimura, 1980) genetic divergence within and between clades. In addition, a cladistic haplotype analysis (Brower, 1999) was made based on a network

created in Haploviewer (Salzburger et al., 2011) following the parameters recommended by the software's authors.

The construction of an ultrametric tree with a strict clock was performed using a log-normal time distribution model through the BEAUTi and BEAST programs (Drummond et al., 2012). The evolutionary nucleotide model used to estimate the ultrametric tree was the HKY model with Gamma correction. We used a Yule prior speciation method, which is the most suitable for species-level phylogenies (Drummond and Rambaut, 2007). In total, 30,000,000 trees per 10,000 generations were sampled. The data was checked through the TRACER v.1.7 program (Rambaut et al., 2018) in order to evaluate if extra races would be necessary to achieve convergence. All topologies sampled below the asymptote (10,000 generations) were discarded as part of a burn-in procedure. The remaining trees were used to build a consensus tree by majority in the Tree Annotator program.

Additionally, species delimitation analyses (see Supplement I Appendix A) were made to see if its results corroborate the separation between the species of the *Haemulon steindachneri* species complex. Automatic Barcode Gap Discovery analysis (ABGD) (Puillandre et al., 2012), was processed using the "graphic" web version available at http://wwwabi.snv.jussieu.fr/ public/abgd/abgdweb.html, under the default parameters of *P*min = 0.001 to *P*max = 0.1, steps = 10, X (relative gap width) = 1.5, Nb bins (for distance distribution) = 20, and the Kimura (K80) molecular model.

For the GMYC method, an ultrametric tree was estimated in Beast v1.8.2 (Drummond et al., 2012), employing an uncorrelated lognormal relaxed clock and speciation Yule process, and the General Time Reversible (GTR) model (Lanave et al., 1984; Tavaré, 1986). The Bayesian topology reconstruction started with a UPGMA tree and the Markov Chain Monte Carlo (MCMC) method was performed for 500 million generations; a tree was sampled for every 20,000 generations. We used the software Tracer v1.7 (Rambaut et al., 2018) to check the convergence of the values. All sampled topologies beneath the asymptote (20,000,000 generations) were discarded as part of a burn-in procedure, and the remaining trees were used to construct a 90% majority-rule consensus tree using Tree Annotator v1.8.2 (Drummond et al., 2012). The GMYC analysis was performed with the package Species Limits by Threshold Statistics ("splits") (Fujisawa and Barraclough, 2013) using R v 3.0.0 (R Development Core Team, 2014). For the Bayesian Poisson tree process (bPTP) analyses, the same ultrametric input tree as used in the GMYC was used. The analyses were run on the web server

(http://species.h-its.org/ptp) with 100,000 MCMC generations, a burn-in of 0.1 and other parameters left as default.

2.3. *Comparative material*

Haemulon aurolineatum Cuvier. MPEG 34253 (4, 115–127), Alagoas; MPEG 34268 (2, 117–126), Alagoas; *H. melanura* (Linnaeus). AZUSC 4504 (4, 145–164 mm SL), Ceará; MZUSP 53092 (1, 113 mm SL), Maranhão; *H. parra*. MPEG 34588 (6, 148–188 mm SL), Ceará; MPEG 34571 (1, 189 mm SL), Ceará; MPEG 34531 (4, 169–215 mm SL), Ceará; MZUSP 46479 (2, 131–136), Fernando de Noronha; MZUSP 65671 (1, 169 mm SL), Pernambuco; *H. plumierii* (Lacepède). MPEG 34549 (5, 136–147 mm SL), Ceará; MPEG 34629 (7, 141–156), Ceará; USNM 398053 (2, 119–166 mm SL), Mexico; USNM 37089 (4, 147–166 mm SL), Mexico; USNM 80598 (2, 129–139 mm SL), Panama; *H. squamipinna* Rocha and Rosa. MZUSP 65676 (3, 128–133 mm SL), Pernambuco; MZUSP 65665 (2, 127–132 mm SL), Pernambuco; AZUSC 4491 (1, 132 mm SL), São Paulo; *H. striatum* (Linnaeus). AZUSC 82203 (1, 147 mm SL), Bahia.

3. Results

3.1. Haemulon sp. n.

(Fig. 1, Tables 1 and 2)

Haemulon steindachneri — Jordan and Swain, 1884: 299–300 [in part, revision].— Courtenay, 1961:86 [in part, revision].— Uyeno et al., 1983:356 [Fishes trawled off Suriname and French Guiana, description].— Cervigón, 1992:335 [in part, description, distribution].— Cervigón, 1993:209 [in part; los peces marinos de Venezuela; description, illustrated].— Robins and Ray, 1986:180 [in part, description, distribution].— Rocha and Rosa, 1999:448 [identification key].— Lindeman and Toxey, 2002:1545 [in part, description, distribution, illustrated].— Menezes et al., 2003:85 [in part; catalog of marine fishes of the Brazilian coast].— Rocha et al., 2008:921 [in part, Historical biogeography and speciation in the reef fish genus Haemulon].— Tavera et al., 2012:6 [in part, Molecular phylogeny].— Marceniuk et al., 2017:40 [identification key,

photography, error in Figure 14-F, the correct is figure 14-H].— Tavera et al., 2018:214 [in part, Multilocus phylogeny].

*Holotype***.** MPEG 35708, 152 mm SL, Amapá, Brazil, 3º47'N, 50º22'W

Paratypes: MPEG 35753 (2, 143–153 mm SL), 3º47'N, 50º22'W, Amapá; AZUSC 4655 (5, 180–192 mm SL), 23°54'53"S, 46°37'19"W, Caete river, Bragança, Pará; LBP 21368 (1, 192 mm SL), 00°54'54.0"S, 46°38'22.0"W, Furo da ostra, Bragança, Pará; MPEG 33357 1, 211 mm SL, Ajuruteua, Bragança, Pará; MPEG 32859 (1, 201mm SL), Ajuruteua, Bragança, Pará; MPEG 33240 (1, 186 mm SL), Ajuruteua, Bragança, Pará; MPEG 34530 (3, 192–215 mm SL), Bragança, Pará; MZUSP 68041 (2, 143–165 mm SL), 0°33'0.0"S, 46°35'0.0"W, Pará; LBP 20027 (1, 106 mm SL), Fortaleza, Ceará; LBP 20014 (1, 95 mm SL), 03°41'33.0"S, 38°30'18.0"W, Enseada do Mucuripe, Fortaleza, Ceará; MZUSP 46479 (2, 131–136 mm SL), Fernando de Noronha, Pernambuco; MPEG 34269, 172 mm SL, Jaraguá, Maceió, Alagoas; MPEG 34251 (3, 148–154 mm SL), Jaraguá, Maceió, Alagoas; LBP 23833 (1, 127 mm SL), 13°22'22.1"S, 38°58'43.0"W, Valença, Bahia; MZUSP 68048 (1, 105 mm SL), Salvador, Bahia; MZUSP 51544 (2, 137–156 mm SL), 20°21'0.0"S, 40°15'0.0"W, Ilha dos pacotes, Vila Velha, Espírito Santo; LBP 22158 (2, 123–131 mm SL), 22°56'59.0"S, 43°47'40.0"W, Baia de Guanabara, Rio de Janeiro; LBP 22124 (1, 180 mm SL), 22°56'59.0"S, 43°47'40.0"W, Baia de Guanabara, Rio de Janeiro; MZUSP 68037 (3, 130–155 mm SL), Angra dos Reis, Rio de Janeiro; MZUSP 2438 (1, 159), Ilha Grande, Rio de Janeiro; AZUSC 3307 (3, 86–145 mm SL), 23°27'5"S, 45°2'48"W, Ubatuba, São Paulo; AZUSC 192 (1, 164 mm SL), 24°0'29"S, 46°19'26"W, Guarujá, São Paulo; AZUSC 1977 (1, 204 mm SL), 24°16'49"S, 46°10'37"W, Guarujá, São Paulo; AZUSC 2216 (3, 99–155 mm SL), 23°49'42"S, 45°26'15"W, São Sebastião, São Paulo; AZUSC 447 (1, 164 mm SL), 24°0'35"S, 46°19'30"W, Guarujá, São Paulo; AZUSC 1913 (1, 208 mm SL), 24°7'37"S, 46°31'56"W, Praia Grande, São Paulo; AZUSC 628 (1, 136 mm SL), 24°0'31"S, 46°19'24"W, Guarujá, São Paulo; AZUSC 4453 (2, 119–153 mm SL), 23°54'37"S, 46°23'49"W, Cubatão, São Paulo; AZUSC 4077 (2, 198–214 mm SL), 24°37'8"S, 46°58'58"W, Peruíbe, São Paulo; AZUSC 461 (1, 176 mm SL), 24°0'30"S, 46°19'26"W, Guarujá, São Paulo; AZUSC 2002 (4, 162–214 mm SL), 24°27'43"S, 46°42'56"W, Itanhaém, São Paulo; LBP 21326 (1, 170 mm SL), 23°49'25.0"S, 45°32'11.0"W, São Sebastião, São Paulo; LBP 10058 (2, 184–205 mm SL), 23°51'38.7"S, 46°09'10.5"W, Bertioga, São Paulo; LBP 3537 (2, 118–127 mm SL),

23°26'10.7"S, 45°02'58.9"W, Ubatuba, São Paulo; LBP 20691 (1, 224 mm SL), 25°21'53.0"S, 47°39'11.0"W, Cananéia, São Paulo; LBP 21614 (1, 192 mm SL), 25°10'51.0"S, 47°35'49.0"W, Cananéia, São Paulo; LBP 21359 (1, 180 mm SL), 23°49'25.0"S, 45°32'11.0"W, São Sebastião, São Paulo; MZUSP 68024 (1, 201 mm SL), São Sebastião, São Paulo; MZUSP 2439 (1, 177 mm SL), São Sebastião, São Paulo; MZUSP 1153 (1, 166 mm SL), São Sebastião, São Paulo; LBP 23329 (1, 114 mm SL), 27°07'12.05"S, 48°31'10.17"W, Porto Belo, Santa Catarina; LBP 23404 (1, 125 mm SL), 27°01'27.83"S, 48°34'35.22"W, Balneário Camboriú, Santa Catarina; LBP 23641 (1, 114 mm SL), 27°12'32.88"S, 48°28'11.01"W, Bombinhas, Santa Catarina; MZUSP 49094 (1, 106 mm SL), 27°7'0.0"S, 48°31'0.0"W, Porto Belo, Santa Catarina; ANSP 121585 (15, 55–132 mm SL), Playa Aquica, Peninsula de Araya, Golfo do Cariaco, Estado Sucre, Venezuela; ANSP 105304 (1, 129 mm SL), Quetepe, Golfo do Cariaco, Estado Sucre, Venezuela; ANSP 105164 (2, 91–94 mm SL), Punta Horno, Peninsula de Araya, Estado Sucre, Venezuela; ANSP 121391 (2, 84–102 mm SL), Punta Horno, Peninsula de Araya, Estado Sucre, Venezuela; ANSP 120214 (2, 73 ou 74 mm SL), Estado Sucre, Venezuela; ANSP 104645 (2, 117 mm SL), Laguna Chica, Peninsula de Araya, Golfo do Cariaco, Estado Sucre, Venezuela.

Non-type material: USNM 289563, 1, Colombia (photographic image and X-ray); USNM 361931, 3, Colombia (photographic image and X-ray); USNM 398161, 1, Colombia (photographic image and X-ray); USNM 398162, 1, Colombia photographic image and X-ray); USNM 389901, 2, Bocas Del Toro, Laguna de Chiriqui, Panama (photographic image and X-ray); USNM 148671, 3, Coco Solo, Panama (photographic image and X-ray).

3.1.1. Morphological diagnosis

Haemulon sp. n. differs from all congeners of the Western Atlantic by presenting a distinct color pattern composed of silvery and silver gray body without stripes, with a bluish black bar in the lower anterior part of the operculum, partially hidden by the angle of the pre-operculum, and a large spot rounded and blackish at the end of the caudal peduncle and base of the caudal fin. In addition, *Haemulon* sp. n. can be differentiated from: *H. album*, from Florida to Brazil, by presenting 30.7–40.5% SL of pre-dorsal length (PDL) (vs. 44–48% SL); of *H. aurolineatum*, from Chesapeake Bay to Brazil, by containing seven to nine scales above the lateral line (vs. six), 19–24 rakers in the 1st branchial arch (vs. 25–28, rarely 24), body

silver to silver gray without stripes (vs. white-silver body with two yellow stripes on the dorsolateral side); of *H. bonariense* Cuvier, from South Florida and Antilles to southern Venezuela, by having 50–56 scales on the lateral line (vs. 44–49), large mouth with maxillary tip reaching the center of the eye (vs. small mouth with tip of the jaw reaching the anterior border of the eye); of *H. boschmae* (Metzelaar), from Mexico to Chinchorro Bank and South American coast of Barranquilla, Colombia to French Guiana, by presenting 31.6–43.9% SL of body height (BH) (vs. 26–30 % SL) and 13.8–21.4% SL of premaxillary length (PL) (vs. 10–13% SL); of *H. carbonarium* Poey, from South Florida to Brazil, including the Gulf of Mexico and the entire Caribbean Sea, by presenting 30.7–40.5% SL of PDL (vs. 40–46% SL, rarely 40), seven to nine scales above the lateral line (vs. six), body silver to silver gray without stripes (vs. silvery gray body dorsally, dark to black ventrally, with at least ten yellowish stripes on the sides of the body); of *H. flavolineatum* (Desmarest), from South Carolina to Trinidad, by having 11–16 scales below the lateral line (vs. nine or ten), scales above and below the lateral line with same size (vs. scales below the lateral line twice the size of those above) and, body silver to silver gray without stripes (vs. silvery gray body with at least 12 yellowish stripes on the sides of the body); of *H. macrostoma* Günther, from South Florida to Suriname, by containing 19–24 rakers in the 1st branchial arch (vs. 26–28), 15–17 (rarely 18) pectoral-fin rays (vs. 17), body silver to silver gray without stripes (vs. white-silvery body with dark brown to black stripes), soft dorsal-fin rays relatively larger than spines (vs. soft rays twice the size of spines); of *H. melanura*, from Florida to Brazil, by presenting seven to nine scales above the lateral line (vs. six), spots below the pre-operculum and at the end of the caudal peduncle and base of the caudal fin (vs. black spot extending from the upper back and lower back of the dorsal fin joining a horizontal black V at the caudal fin); of *H. parra*, from the Gulf of Mexico to Brazil, by presenting 30.7–40.5% SL of PDL (vs. 41–49% SL), seven to nine scales above the lateral line (vs. six), serrated pre-operculum (vs. non-serrated in adults) and scales throughout the body with gray-brown centers (vs. scales with black centers); of *H. plumierii*, from Chesapeake Bay to Brazil, by presenting seven to nine scales above the lateral line (vs. five), scales above the lateral line of the same size as below (vs. scales above lateral line greater than those below) and body silver to silver gray without stripes (vs. body and head with blue and yellow stripes); of *H. sciurus* (Shaw), from South Carolina to the Guianas, by presenting 30.7–40.5% SL of PDL (vs. 42–46% SL), 19–24 rakers in the 1st branchial arch (vs. 26–31), white to gray membranes between the dorsal-fin spines (vs. yellow) and body silver to silver gray without stripes (vs. yellowish-bronze body

with blue stripes on the head and body to the base of the caudal fin); of *H. squamipinna*, from Fortaleza to Alagoas, by having seven to nine scales above the lateral line (vs. six), 19–24 rakers in the 1st branchial arch (vs. 25–27, rarely 24), and body silver to silver gray without stripes (vs. silvery white body with 10–12 yellow bands); of *H. striatum*, from North Carolina to southern Brazil, by presenting 19–24 rakers in the 1st branchial arch (vs. 28–34), XII spines in the dorsal fin (vs. XIII) and 15–17 dorsal-fin rays (vs. 13 or 14, rarely 12), oblong and compressed body (vs. elongated body not laterally compressed) and body silver to silver gray without stripes (vs. gray to bluish body dorsally, silvery ventrally, with five yellow stripes along the body); of *H. vittatum* (Poey), from northern Florida to Trinidad, by having XII spines in the dorsal fin (vs. IV to XVIII) and 15–17 dorsal-fin rays (vs. ten), 15–17 (rarely 18) rays in the pectoral fin (vs. 19); III spines in the anal fin and eight (rarely nine) soft rays (vs. II, 9), oblong and compressed body (vs. fusiform body, elongate and rounded), operculum without spine (vs. operculum with broad spine and flat at angle posterior), and body silver to silver gray without stripes (vs. blue-green metallic body dorsally, ventrally bluish white, a broad greenish band from the eye to the base of the caudal, plus three brown stripes above).

Haemulon sp. n. differs from its eastern Pacific congeners as follows: of *H. flaviguttatum* Gill, from Southern California to Peru, by having 19–24 rakers in the 1st branchial arch (vs. 26–31), eight (rarely nine) soft rays in the anal fin (vs. 10 or 11), large mouth with maxilla reaching the center of the eye (vs. small mouth, with maxilla reaching the anterior border of the eye); of *H. maculicauda* (Gill), from southern Baja California to northern Peru, by its scales on the sides of the body with grayish/silver centers forming oblique lines to the axis of the body (vs. scales with white and black centers forming longitudinal stripes parallel to the axis of the body), dorsal and anal fin yellow (vs. gray); of *H. scudderii*, from Southern Baja and Central Gulf of California to Ecuador, by presenting seven to nine scales above the lateral line (vs. five or six), scales all over the body with gray-brown centers (vs black spots on each scale); of *H. sexfasciatum*, from Baja California to Ecuador, by containing eight (rarely nine) anal fin soft rays (vs. nine or ten), body silver to silver gray without bars (vs. six or seven bars in the dorsolateral surface of the body) and head without spots (vs. dark spots on the side of the head); of *H. steindachneri*, from the Gulf of California to northern Peru, by presenting an anal fin usually with eight rays (vs. usually nine rays); 13–15 scales below the lateral line, rarely 12 (vs. 12 scales below the lateral line, rarely 13), posterior margin of the maxilla robust with a smoothly angled end (vs. smaller maxilla with moderately convex extremity), a spot on the pre-operculum, broad and robust, with no

definite shape (vs. narrow spot, with anterior extremity tuned and posterior straight, resembling a triangle) (Fig. 2).

3.1.2. Molecular diagnosis

Haemulon sp. n. forms a distinct group (Fig. 3) with genetic distance (K2P) of other representatives ranging from 7.4% (*H. steindachneri*) to 15.5% (*H. melanura*) (Table 3). The haplotypes of *Haemulon* sp. n. differ from their congeners by 35 (*H. steindachneri*) to 68 bases (*H. melanura*) (Table 4). The molecular separation of *Haemulon* sp. n. and *H. steindachneri* is corroborated by the species delimitation analysis, as well as in the haplotype network (see Supplement I Appendix A). In addition, two distinct molecular lineages were recognized in *Haemulon* sp. n. along the Atlantic coast, one comprising of specimens from Amapá on the coast of Brazil and the Caribbean coast and the other from additional coastal areas of Brazil (see Supplement I Appendix A). Although these lineages are differentiated by two exclusive COI mutations (see haplotype network in Supplement I Appendix A), the genetic distance between them is very small (0.013 ± 0.005). The GMYC and bPTP analyses recognized these two lineages as distinct species, however, the ABGD analysis did not support this recognition (see Supplement I Appendix A), and no morphological differences were found separating them.

3.1.3. Description

Morphometric data in Table 1 and meristic data in Table 2. Body elongated, compressed laterally, greater height under vertical through origin of dorsal fin. Dorsal profile of body, convex from end of snout to base of first ray of dorsal fin, posterolaterally inclined at dorsal-fin base, approximately straight from last ray of dorsal to caudal peduncle. Ventral profile of body, straight from anterior end of maxilla to base of pelvic fin, slightly convex from this point to origin of anal fin, posterodorsally at base of anal fin and approximately straight from last ray of anal to origin of inferior branch of caudal fin. Head longer than high, pointed lateral and dorsally. Pointed snout, with length about one third of length of head. Big eye, ellipsoid, elevated slightly before middle of length of head. Small nostrils located laterally on head, anterior broad in drop shape and posterior oval, about half size of first; posterior nostril in front and slightly above horizontal line of center of eye, anterior nostril just above horizontal of inferior margin of eye. Large mouth, posterior margin of robust

maxilla with slightly angled extremity, situated vertically through center of orbit; fleshy and thick lips; upper lip slightly beyond tip of jaw. Conical teeth, in narrow band on each jaw, extended outer series. Chin with two pores and median groove. Short and thin gill rakers on first branchial arch.

Operculum without spine, covered with five or six vertical lines of ctenoid scales. Pre-operculum with slightly concave and serrated posterior margin. Ctenoid scale (rough to touch), small or moderate, extending overhead (except in front of snout, lips and chin) to caudal fin; longitudinal scales above oblique lateral line along axis of body; those below in horizontal series. Continuous lateral line gently curved posteriorly, following dorsal contour of body and becoming straight on peduncle. Soft rays of dorsal and anal fin densely scaled almost to edge of fin. Scaled pectoral fin only at base.

Dorsal fin with small notch in middle; origin at vertical of posterior margin of operculum; high, strong spines, fourth more prominent, about one-third greater than soft rays; rays, XII, 15 or 16 (rarely 17*), first unbranched, branched remnants. Anal fin origin below base of 3rd or 4th ray of dorsal fin; rays, III, eight* or nine, all branched; three strong spines, second most prominent; long soft rays, first ray almost reaching tip of first spine. Caudal fin emarginated to forked, lobes approximately equal in size; principal rays ten + eight, upper and lower rays unbranched. Pectoral fins moderately long, not reaching or reaching tip of pelvic fin, fifth ray longer; rays 15(5), 16(48 *), or 17(28) (rarely 18), first shorter, unbranched, second ray about two times longer, unbranched, remaining rays branched. Origin of pelvic fins below lower base of pectoral fins, vertically on base of 2nd or 3rd dorsal spine; first longest ray (second equal to first); rays, I, five, all branched. Caudal peduncle longer than high.

Lateral line scales 50(3), 51(3), 52(33), 53(20*), 54(11), 55(8), or 56(5); scales above lateral line to base of first spine eight or nine* (rarely seven); scales below lateral line to first spine of anal fin 12(3), 13(29), 14(33), 15(13*), or 16(4) (rarely 11); rakers on first branchial arch 19(1), 20(2), 21(6), 22(10), 23(21*), or 24(13); rakers on second branchial arch 15(10*), 16(17), 17(18), 18(35), or 19(5) (Table 2).

Supraneural 3. Vertebrae 11+15. First pterygophore of dorsal fin inserted above second neural spine; last pterygophore of dorsal fin inserted in front of neural spine of 19th vertebra. First pterygophore of anal fin inserted below first hemal spine; last pterygophore of anal fin inserted in front of sixth hemal spine. Caudal skeleton with one ural center; five

autogenic hypurais; three epurais; 17 (nine + eight) major caudal rays, with simple procurrent rays (see Supplement II Appendix A).

3.1.4. Coloration in life

Body gray dorsally, silver to dark gray ventrally; scales with gray/silver centers, forming oblique lines along rows of scales; dark gray head at top of snout to vertical passage through pre-operculum border, light gray/silver in infraorbital region; distinct blackish spot on lower anterior part of operculum, partially covered by pre-operculum angle; dark, large, distinct and rounded spot at end of caudal peduncle and at base of caudal fin. Dorsal and anal fins gray with lighter tonality on interradial membranes; pectoral and pelvic gray to yellowish; dark caudal fin.

3.1.5. Coloration of preserved specimens

Upper dorsal and lateral surface of body relatively light brown or gray. Ventral surface of body pale yellowish-brown coloration. Distinctively blackish spot on lower anterior part of operculum, partially hidden by pre-operculum angle; dark, large, distinct and rounded spot at end of caudal peduncle and at base of caudal fin.

3.1.6. Distribution and habitat

Haemulon sp. n. occurs on the western Atlantic coast of Costa Rica to Santa Catarina-Brazil (Fig. 4), inhabiting estuarine and coastal marine waters up to 30 m deep. The species is associated with coral reefs and can be found on sand and rubble substrates.

3.1.7. Remarks

Haemulon steindachneri was described by Jordan and Gilbert (1882), as *Diabasis steindachneri*, based on specimens collected on the Pacific coast. The same authors, when reviewing the type material of *H. caudimacula* Cuvier (Type locality: Acapulco, Rio de Janeiro, Rio Grande do Sul, Maranhão), recognized that *H. caudimacula* represents a junior synonym of *H. parra*, showing that *H. caudimacula* was erroneously identified as synonymous of *H. steindachneri* because it contained 15 dorsal-fin rays (vs. 16) and seven anal-fin rays (vs. 8) (Jordan and Gilbert, 1882).

Hong (1977) was the first author to discuss possible differences among populations of *H. steindachneri*, comparing specimens from the Eastern Pacific with specimens examined by Courtenay (1961) in the Western Atlantic. According to Hong (1977) the Atlantic and Pacific

specimens would be similar except for the length of the snout and length of the maxillary jaw that were 1 to 2% SL larger in the Atlantic specimens. These differences were not found in the present study; however, differences in the shape of the maxillary margin of these pairs of species were found.

Regarding nominal species available for the Western Atlantic, we consider that *Haemulon* sp. n. is not related to *Haemulon fur* Poey, by possessing a silver to silver gray body without stripes (vs. steel gray body with golden yellow bands); *H. helenae* Boulenger, by containing III spines in the anal fin and eight (rarely nine) soft rays (vs. III, 12); *H. hians* Haly, by having body silver to silver gray without stripes (vs. longitudinally striped body); *H. jaguanum* Poey, by having a large rounded blackish spot at the end of the caudal peduncle and base of the tail fin (vs. spot in the caudal absent); *H. melanopterum* Ranzani, by presenting 15–17 rays in dorsal fin (vs. 10–12); *H. modestum* Tschudi, by containing III spines in anal fin and eight (rarely nine) soft rays (vs. IV, 13); *H. schranki* Agassiz, by presenting 15–17 rays in dorsal fin (vs. 18); *H. serrula* (Cuvier), by having 15–17 rays in dorsal fin (vs. 13) and *H. similis* Castelnau, by possessing body silver to silver gray without stripes (vs. body with yellow longitudinal bands).

3.2. Haemulon steindachneri (Jordan and Gilbert, 1822)

(Fig. 1; Tables 1 and 2)

Haemulon steindachneri (Jordan and Gilbert, 1884) [Redescription]. — Hong, 1977:496 [in part, revision]. — Allen and Robertson, 1994:149 [Fishes of the tropical eastern Pacific, description; photograph]. — Bussing and López, 1994:120 [in part; description; illustrated]. — McKay and Schneider, 1995:1157 [in part; identification key; description; illustrated]. — De La Cruz Agüero et al., 1997:183 [in part, Catalogo de los peces marinos de Baja California Sur]. — Chirichigno and Vélez, 1998:375 [in part; identification key; distribution, Mexico to Peru]. — Castro-Aguirre et al., 1999:338 [in part, identification key; distribution, Panama to Brazil in Atlantic and California Golf to Panama in Pacific]. — Rocha et al., 2008:921 [in part, Historical biogeography and speciation in the reef fish genus Haemulon]. — Tavera et al., 2012:6 [in part, Molecular phylogeny]. — Galván-Villa et al., 2016:147 [in part, checklist]. — Tavera et al.,

2018:214 [in part, Multilocus phylogeny]. — González-Murcia et al., 2019:304 [in part, ichthyology collection at the Natural History Museum of El Salvador].

Diabasis steindachneri Jordan and Gilbert, 1882:322 [original description; type locality: Panama, Mazatlán. Syntypes: BMNH 1895.5.27.55–57 (3) Mazatlán; USNM 28172 (1), 29226 (1), 29305 (1, not found in 1980), 29387 (?), 29634 (1, lost), 29759 (1), 29778 (1), 29795 (0).

Syntypes: USNM 28172 (1, 175 mm SL) , Mazatlan, Sinaloa, Mexico (photographic image and X-ray); USNM 29226 (1, 196 mm SL), Mazatlan, Sinaloa, Mexico (photographic image and X-ray); USNM 29759 (1, 160 mm SL), Mazatlan, Sinaloa, Mexico (photographic image and X-ray); USNM 29778 (1, 164 mm SL), Mazatlan, Sinaloa, Mexico (photographic image and X-ray); USNM 29795 (1, 160 mm SL), Mazatlan, Sinaloa, Mexico (photographic image and X-ray).

Additional material: LACM 9476.1 (1, 115 mm SL), Rancho El Tule, Gulf of California, Mexico (photographic image and X-ray); LACM 30383.2 (2, 187–194 mm SL), Cabo San Lucas, Mexico (photographic image and X-ray); LACM 32155.2 (1, 170 mm SL), Point Mita, Mexico (photographic image and X-ray); LACM 34079.2 (2, 206–214 mm SL), Gulf of California, Mexico (photographic image and X-ray); LACM 35744.23 (1, 193 mm SL), Guaymas, Gulf of California, Mexico (photographic image and X-ray); LACM 35745.13 (1, 154 mm SL), Guaymas, Gulf of California, Mexico (photographic image and X-ray); LACM 38.098.1 (1, 157 mm SL), Magdalena Bay, Mexico (photographic image and X-ray); MZUSP 79692 (1, 172 mm SL), Cabe San Lucas, Mexico; SU 2825 (1, 191 mm SL), Gulf of California, Sonora, Mexico; SU 2858 (1, 121 mm SL), Sinaloa, Mazatlan, Mexico; SU 55462 (1, 198 mm SL), Manzanillo, Colima, Mexico; LACM 30723.3 (2, 169–184 mm SL), Gulf of Nicoya, Puntarena, Costa Rica (photographic image and X-ray); LACM 32499.17 (1, 65 mm SL), Punta Santa Helena, Costa Rica (photographic image and X-ray); LACM 35493.5 (1, 103 mm SL), Gulf of Nicoya, Costa Rica (photographic image and X-ray); ANSP 86216 (1, 174 mm SL), Pearl Island, Panama; CAS 234330 (1, 150 mm SL) Taboguila, Panama; SU 6879 (3, 133–164 mm SL), Panama; USNM 398152 (1, 137 mm SL), Mexico; USNM 396688 (1, 98 mm SL), Panama; USNM 404497 (1, 178 mm SL), Sonora, Mexico.

3.2.1. Morphological diagnosis

Haemulon steindachneri differs from its eastern Pacific counterparts as follows: from *H. flaviguttatum*, by presenting 19–24 rakers in the 1st branchial arch (vs. 26–31), nine soft rays in the anal fin (vs. 10 or 11), large mouth with maxillary tip reaching the center of the eye (vs. small mouth, with maxillary end reaching the anterior border of the eye); from *H. maculicauda*, by their scales on the sides of the body having grayish/silver centers forming oblique lines to the axis of the body (vs. scales with white and black centers forming longitudinal stripes parallel to the axis of the body); dorsal and anal fin yellow (vs. gray); from *H. scudderii*, by presenting 31.4–37.0% SL of PDL (vs. 38–45% SL), seven or eight scales above the lateral line (vs. five or six), nine soft rays in the anal fin (vs. seven or eight) and scales all over the body with gray-brown centers (vs. black spots on each scale); from *H. sexfasciatum*, by presenting 31.4–37.0% SL of PDL (vs. 40–46% SL), silver to silvery gray body without bars (vs. six or seven bars on the dorsolateral surface of the body) and head without spots (vs. dark spots on the side of the head).

Haemulon steindachneri differs from its western Atlantic counterparts as follows: *H. album*, by presenting 31.4–37.0% SL of PDL (vs. 44–48% SL), 9.8–13.7% SL of snout length (SnL) (vs. 14–17% SL), 12 or 13 scales below the lateral line (vs. 14); of *H. aurolineatum*, by containing seven or eight scales above the lateral line (vs. six), 19–24 rakers in the 1st branchial arch (vs. 25–28, rarely 24), body silver to silver gray without stripes (vs. silvery-white body with two yellow stripes on the dorso-lateral portion); of *H. bonariense*, by having 50–53 scales on the lateral line (vs. 44–49), large mouth with jaw end reaching the center of the eye (vs. small mouth, with jaw end reaching the anterior edge of the eye); of *H. boschmae*, by presenting 32.8–38.9% SL of BH (vs. 26–30% SL), and 14.4–19.2% SL of PL (vs. 10–13% SL), and XII spines on the dorsal fin (vs. XIII or XIV), 16 or 17 rays in the dorsal fin (vs. 11–15) and seven or eight scales above the lateral line (vs. five or six); of *H. carbonarium*, by presenting 31.4–37.0% SL of PDL (vs. 40–46% SL, rarely 40), 9.8–13.7% SL of SnL (vs. 14–17% SL), body silver to silver gray without stripes (vs. body dorsally silvery gray, dark to black ventrally, with at least ten yellowish stripes on the sides of the body); of *H. flavolineatum*, by possessing 16 or 17 dorsal-fin rays (vs. 14 or 15), 12 or 13 scales below the lateral line (vs. nine or ten), scales above and below the lateral line of the same size (vs. scales below lateral line two times the size of those above), and body silver to silver gray without stripes (vs. silvery gray body with at least 12 yellowish stripes on the sides of the body); of *H. macrostoma*, by containing 19–24 rakers in the 1st branchial arch (vs. 26–28), 16 or 17 (rarely 18) rays in the pectoral fin (vs. 18, rarely 17), body silver to silver gray

without stripes (vs. white-silvery body with dark brown to black stripes), soft dorsal-fin rays relatively larger than spines (vs. soft rays twice the size of spines); of *H. melanura*, by presenting seven or eight scales above the lateral line (vs. six), spots below the pre-operculum, at the end of the caudal peduncle and at the caudal-fin base (vs. black spot extending from the upper back and lower dorsal fin joining a horizontal black V at the caudal fin); of *H. parra*, by presenting 31.4–37.0% SL of PDL (vs. 41–49% SL), seven or eight scales above the lateral line (vs. six), pre-operculum serrated (vs. non-serrated in adults) and scales throughout the body with gray-brown centers (vs. scales with black centers); of *H. plumierii*, by presenting 31.4–37.0% SL of PDL (vs. 39–48% SL), seven or eight scales above the lateral line (vs. five), scales above the lateral line of the same size as below (vs. scales above the lateral line larger than those below) and silver to silver gray body without stripes (vs. body and head with blue and yellow stripes); of *H. sciurus*, by presenting 31.4–37.0% SL of PDL (vs. 42–46% SL), 19–24 rakers in the 1st branchial arch (vs. 26–31), membranes between the dorsal-fin spines white to gray (vs. yellow) and silver to silver gray body without stripes (vs. yellowish-bronze body with blue stripes on the head and body to the base of the caudal fin); of *Haemulon* sp. n., by presenting anal fin usually with nine rays (vs. usually eight rays); 12 scales below the lateral line, rarely 13 (vs. 13–15 scales below the lateral line, rarely 12), posterior margin of the maxilla with moderately convex extremity (vs. maxilla robust with anterior gently angulated), narrow spot on the pre-operculum, with anterior extremity tuned and posterior straight, resembling a triangle (vs. broad and robust spot, with no definite shape (Fig. 2); of *H. squamipinna*, by having seven or eight scales above the lateral line (vs. six), 19–24 rakers in the 1st branchial arch (vs. 24–27, rarely 24) and silver to silver gray body without stripes (vs. silvery white body with 10–12 yellow bands); of *H. striatum*, by presenting 19–24 rakers in the 1st branchial arch (vs. 28–34), XII spines in the dorsal fin (vs. XIII) and 16 or 17 rays in the dorsal fin (vs. 12–14), body oblong and compressed (vs. elongated body not laterally compressed) and silver to silver gray body without stripes (vs. gray to bluish body dorsally, silvery ventrally, with five yellow stripes along the body); of *H. vittatum*, by presenting XII spines in the dorsal fin (vs. IV to XVIII) and 16 or 17 rays in the dorsal fin (vs. ten), 16 or 17 (rarely 18) rays in the pectoral fin (vs. 19); III spines in the anal fin (vs. II), oblong and compressed body (vs. fusiform body, elongate and rounded), operculum without spine (vs. operculum with broad and flat spine in posterior angle) and silver to silver gray body without stripes (vs. blue-green metallic body

dorsally, ventrally bluish white, a broad greenish band from the eye to the base of the caudal, plus three brown stripes above).

3.2.2. Molecular diagnosis

Haemulon steindachneri forms a distinct group (Fig. 3) with genetic distance (K2P) of other *Haemulon* representatives ranging from 7.4% (*Haemulon* sp. n.) to 13.8% (*H. melanura*) (Table 3). The haplotypes of *H. steindachneri* differ from their congeners by 35 (*Haemulon* sp. n.) to 64 bases (*H. flavolineatum and H. melanura*) (Table 4). The molecular separation of *H. steindachneri* and *Haemulon* sp. n. is corroborated by the species delimitation analysis, as well as the haplotype network (see Supplement I Appendix A).

3.2.3. Redescription

Morphometric data in Table 1, meristic data in Table 2. Body elongated, compressed laterally, greater height under vertical through dorsal-fin origin. Dorsal profile of body convex from snout's extremity to base of first ray of dorsal fin, posterolaterally inclined at dorsal-fin base, approximately straight from last ray of dorsal to caudal peduncle. Ventral profile of body straight from anterior end of maxilla to base of pelvic fin, slightly convex from this point to anal-fin origin, posterodorsally at base of anal fin and approximately straight from last ray of anal to origin of lower lobe of caudal fin. Head longer than high, pointed lateral and dorsally. Pointed snout, about one-third length of head. Big eye, ellipsoid, raised slightly before middle length of head. Small nostrils located laterally on head, anterior broad in drop shape and posterior oval, about half size of first; posterior nostril in front and slightly above horizontal line of center of eye, anterior nostril just above horizontal of lower margin of eye. Large mouth, posterior margin of maxilla with moderately convex extremity, located vertically through center of orbit; fleshy and thick lips, upper lip slightly beyond tip of jaw. Conical teeth, in narrow band on each jaw, extended outer series. Chin with two pores and median groove. Short and thin gill rakers on first branchial arch.

Operculum without spine, covered with five or six vertical lines of ctenoid scales. Pre-operculum with slightly concave and serrated posterior margin. Ctenoid scale (rough to touch), small or moderate, extending overhead (except in front of snout, lips and chin) to caudal fin; longitudinal scales above oblique lateral line along axis of body; those below in horizontal series. Continuous lateral line, gently curved posteriorly, following dorsal contour

of body, becoming straight on peduncle. Soft rays of dorsal and anal fin densely scaled almost to edge of fin. Pectoral fin scaled only at base.

Dorsal fin with small notch in middle, origin at vertical of posterior margin of operculum; high, with strong spines, fourth spine more prominent, about one-third greater than soft rays; rays, XII, 15 or 16 (rarely 17*), first unbranched, branched remnants. Anal-fin origin below base of 3rd or 4th rays of dorsal fin; rays, III, nine, all branched; three strong spines, second most prominent; long soft rays, first ray almost reaching tip of first spine. Caudal fin emarginated to forked, lobes approximately equal size; principal rays ten + eight, upper and lower rays unbranched. Pectoral fins moderately long, not reaching or reaching tip of pelvic fin, fifth ray longer; rays 17(10) (rarely 16), first shorter, unbranched, second ray about two times longer, unbranched, remaining rays branched. Pelvic-fins origin below lower base of pectoral fins, vertically on base of 2nd or 3rd dorsal spine; first longest ray (second equal to first); rays, I, five, all branched. Caudal peduncle longer than high.

Lateral line scales 50(1), 51(2), 52(5), 53(4); scales above lateral line to base of first spine, seven to nine; scales below lateral line to first spine of anal fin, 12(9) or 13(3); rakers on first branchial arch, 19(1), 20(1), 21(1), 22(2), 23(3) or 24(2); rakers on second branchial arch, 14(1), 15(2), 16(3), 17(5) or 18(1) (Table 2).

Supraneural 3. Vertebrae 11+15. First pterygophore of dorsal fin inserted above second neural spine and last pterygophore of dorsal fin inserted in front of neural spine of 19th vertebra. First pterygophore of anal fin inserted below first hemal spine and last pterygophore of anal fin inserted in front of sixth hemal spine. Caudal skeleton with one ural center; five autogenic hypurais; three epurais; 17 (nine + eight) major caudal rays, with simple procurrent rays (see Supplement II Appendix A).

3.2.4. Coloration in life

Body gray dorsally, silver to dark gray ventrally; scales with gray/silver centers, forming oblique lines along rows of scales; dark gray head at top of snout to vertical through pre-operculum border, light gray/silver in infraorbital region; distinct blackish spot on lower anterior part of operculum, partially covered by pre-operculum angle; dark, large, distinct and rounded spot at end of caudal peduncle and at base of caudal fin. Dorsal and anal fins gray with lighter tonality on interradial membranes; pectoral and pelvic gray to yellowish; dark caudal fin (Fig. 1).

3.2.5. Coloration of preserved specimens

Upper dorsal and lateral surface of body relatively light brown or gray. Ventral surface of body pale yellowish-brown. Distinctively blackish spot on lower anterior part of operculum, partially hidden by pre-operculum angle; dark, large, distinct and rounded spot at end of caudal peduncle and at base of caudal fin.

3.2.6. Distribution and habitat

Haemulon steindachneri occurs in the eastern Pacific between the Gulf of California and Peru (Fig. 4), inhabiting estuarine and coastal marine waters up to 30 m deep. The species is associated with coral reefs and can be found on sand and rubble.

3.2.7. Remarks

Jordan and Gilbert (1882) described *Haemulon steindachneri* as *Diabasis steindachneri* Jordan and Gilbert, 1882, based on two specimens collected in Panama and six specimens collected in Mazatlan, Mexico, from the Pacific. Jordan and Swain (1884) reviewed *D. steindachneri*, altering the generic nomenclature, since the name *Diabasis* Desmarest was pre-occupied in Coleoptera. The examination of the type specimens indicated that *H. steindachneri* is a valid species of *Haemulon* (Fig. 1) based on a bluish black bar in the lower anterior part of the operculum, partially hidden by the angle of the pre-operculum; a large blackish rounded spot at the end of the caudal peduncle and the base of the caudal fin.

According to Courtenay (1961), specimens of *H. steindachneri sensu lato* present a large black spot below the margin of the pre-operculum and a black spot at the base of the caudal fin, whereas Hong (1977) when comparing their *H. steindachneri* data from the Atlantic and the Pacific with data from Courtenay (1961), suggested that the specimens from both oceans were similar, except for the snout length and upper jaw length that were 1 to 2% SL larger in the Atlantic specimens. This difference was not observed in the present work.

In addition, *H. steindachneri* differs from *Haemulon* sp. n. by presenting the anal fin usually with nine rays (vs. generally eight rays); 12 scales below the lateral line, rarely 13 (vs. 13−15 scales below the lateral line, rarely 12), posterior margin of the maxilla with moderately convex extremity (vs. maxilla robust with anterior gently angulated), narrow spot on the pre-operculum, with anterior extremity tuned and posterior straight, resembling a triangle (vs. broad and robust spot, with no definite shape) (Fig. 1−2).

4. Discussion

The examination of the morphology of *Haemulon* sp. n. confirms that the new species shares the diagnostic characteristics of the genus *Haemulon*, such as the presence of serrated pre-operculum, the last dorsal spine associated with dorsal rays, and scaly dorsal and anal fins (Jordan and Swain, 1884; Courtenay, 1961; Hong, 1977).

The genus *Haemulon* is a group that presents pairs of closely related sister species that have totally or partially overlapping distributions or with specimens found on both sides of the Isthmus of Panama (Rocha et al., 2008; Tavera and Wainwright, 2019). Tavera and Wainwright (2019) recognized two groups of allopatric species pair within the *Haemulon*, which are *H. aurolineatum* Western Atlantic / *H. sp. A* Gulf of Mexico and *H. steindachneri* Western Atlantic / *H. steindachneri* Eastern Pacific. Classical taxonomic assessments considered *H. steindachneri* as a single species composed of two populations, one in the Western Atlantic and one in the Eastern Pacific (Courtenay, 1961; Hong, 1977). However, recent molecular studies have recognized two distinct lineages of *H. steindachneri*, one for the Atlantic and one for the Pacific (Rocha et al., 2008; Tavera et al., 2012; Tavera et al., 2018).

On the other hand, our molecular analysis showed the presence of an emergent species in the Atlantic coast from Amapá and Caribbean. Although this lineage and the other lineage from the Brazilian coast are differentiated by two exclusive COI mutations and by the GMYC and bPTP analysis (see haplotype network in Supplement I Appendix A), the genetic distance between them is very small (0.013 ± 0.005) and the ABGD and morphological analysis did not support this recognition. Given those results, we consider the two Atlantic lineages of *Haemulon* sp. n. to be an example of gray zone, where alternative species concepts come into conflict (De Queiroz, 2007).

Menezes et al. (2003) showed that the majority of species found in Brazil also occurred in the Caribbean coast (Menezes et al., 2003). This situation can be determined by the absence of any major physical barriers between these two regions (Marceniuk et al., 2019). For the provinces of the Greater Caribbean, variations in shelf environments could be responsible for the substantial differences in its faunas (Robertson and Cramer, 2014). Similarly, the differentiation encountered on the Brazilian coast fauna, may be caused by the considerable heterogeneity of its environments (Silva et al., 2016). Considering these variations in its environments, further taxonomic reviews of the marine-estuarine fish fauna of

the Brazilian and the Caribbean coasts are necessary to better understand how the biogeographic patterns are changing the species in these areas.

The molecular analyses of this work showed that the genetic distance (K2P) among *Haemulon* sp. n. and other species of the genus varies from 7.4% (among *H. steindachneri*) and 15.5%, which is compatible with the divergences of other trans-isthmian twin pairs, such as the pairs of the genus *Chromis* (Pomacentridae, 3.1–3.5 million years ago), and *Chaetodon* (Chaetodontidae, 3.4 Ma), whose divergences vary from 9.35% to 10.40% when using the K2P model (Coates et al., 1992; Bermingham et al., 1997; Domingues et al., 2005). The separation time estimated from our data, 3.2±0.7 mya, is compatible with the final data of closure of the Panama Isthmus (Figure 3). Minor divergence rates, such as that found between the *Haemulon* sp. n. and *H. steindachneri* pairs, are related to trans-isthmian species that have a greater affinity with coastal and estuarine waters; this prediction correlates with the geological history of the isthmus closure, where the only connections between the Caribbean and the eastern Pacific prior to the final closure were probably coastal areas with estuarine conditions (Knowlton et al., 1993; Coates and Obando, 1996; Tringali et al., 1999; Bernardi and Lape, 2005). Both *Haemulon* sp. n. and *H. steindachneri* occur in coastal areas and are often found in low salinity waters (Raz-Guzman and Huidobro, 2002), with juveniles in shallow areas and in rocky shores, and are recognized as crucial to maintaining the integrity of these ecosystems (Furia, 1996; Rocha, 1997).

As previously proposed, there are great evidences that the rise of the Isthmus of Panama influenced the speciation of *H. steindachneri* and *Haemulon* sp. n. The cessation of the uplift of the isthmus occurred about 3 million years ago at the end of the Pliocene (O'Dea et al., 2016). Tavera et al. (2012; 2018; 2019) in its molecular analyses with the Haemulidae showed that the separation of the lineages of *H. steindachneri* occurred in the Pliocene, around 3 million years ago. Time of divergence similar to this was found in our analyses (Fig. 3), corroborating the fact that the Isthmus of Panama was responsible for the allopatric speciation between the species pairs *Haemulon* sp. n. and *H. steindachneri*.

Haemulon sp. n. and *H. steindachneri* have a conserved morphology, being considered very morphologically similar. In this sense, distinct species identified as a single species but with genetic divergence are designated as cryptic species (Bickford et al., 2007). Cryptic species are common among metazoans and can be found in all types of biogeographic zones (Bickford et al., 2007; Pfenninger and Schwenk, 2007). The identification of cryptic species is

of fundamental importance to understand diverse evolutionary, biogeographic and ecological processes (Bickford et al., 2007; Pfenninger and Schwenk, 2007). Many studies have demonstrated that molecular techniques associated to the traditional taxonomy are efficient for the identification of these enigmatic lineages (Neusser et al., 2011; Jörger and Schrödl, 2013; Souza et al., 2017). Based only on the morphological criteria, *Haemulon* sp. n. and *H. steindachneri* were during many times recognized as a single species. Molecular studies have come to recognize that *H. steindachneri* has two distinct forms, one for the Atlantic and one for the Pacific (Rocha, 2008; Tavera et al., 2012; Tavera et al., 2018). Additionally, these studies showed that *Haemulon* sp. n. and *H. steindachneri* are sister species, a condition corroborated in the present study.

In this way, the combination of the DNA barcode with the examination of the available specimens in the ichthyological collections was essential for the recognition of a new species of the genus *Haemulon* for the Atlantic coast. Showing the effectiveness of an integrated approach, DNA barcode was confirmed as an important complementary tool in the recognition of cryptic species.

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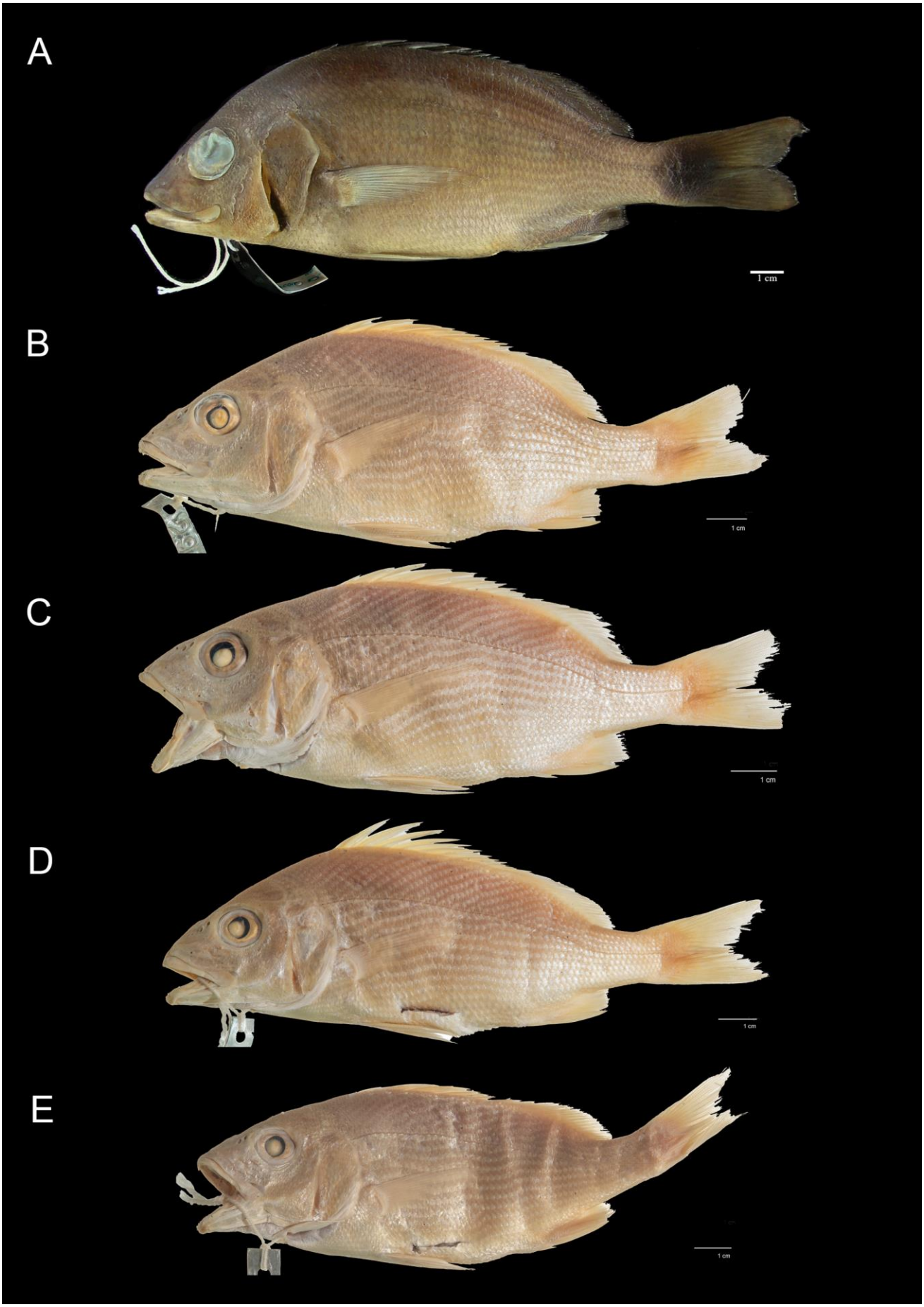
Figure captions

FIGURE 1. Body of *Haemulon* sp. n. and *Haemulon steindachneri* in lateral view. (A) *Haemulon* sp. n. holotype, MPEG 35708, 152 mm SL. *Haemulon steindachneri* syntypes. (B) USNM 29226, Mexico. (C) USNM 29759, Mexico. (D) USNM 29778, Mexico. (E) USNM 29795, Mexico.

FIGURE 2. Head and maxilla of *Haemulon* sp. n. and *H. steindachneri* in lateral view. (A) *Haemulon* sp. n. 192 mm SL, Pará (MPEG 34530). (B) *H. steindachneri*, 191 mm SL, Mexico (SU 2825). (C) Right jaw of *Haemulon* sp. n., 172 mm SL, Alagoas, Brazil (MPEG 34269). (D) Right jaw of *H. steindachneri*, 178 mm SL, Mexico, Panama (USNM 404497).

FIGURE 3. Time-calibrated tree of New World *Haemulon* obtained from COI data. White circles indicate 0.75−1 posterior probability.

FIGURE 4. Map of the locations of *Haemulon* sp. n. and *H. steindachneri* reviewed.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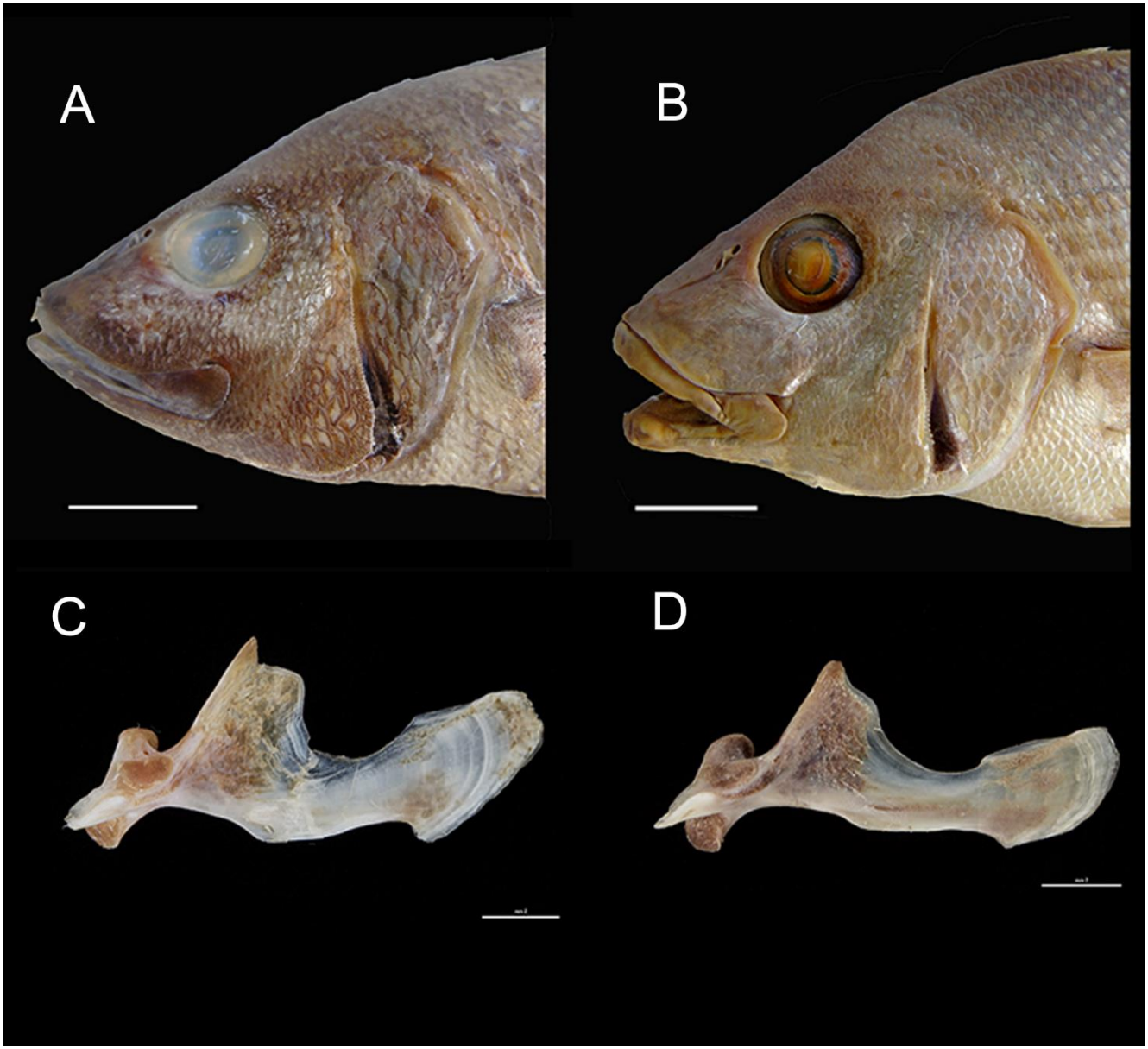
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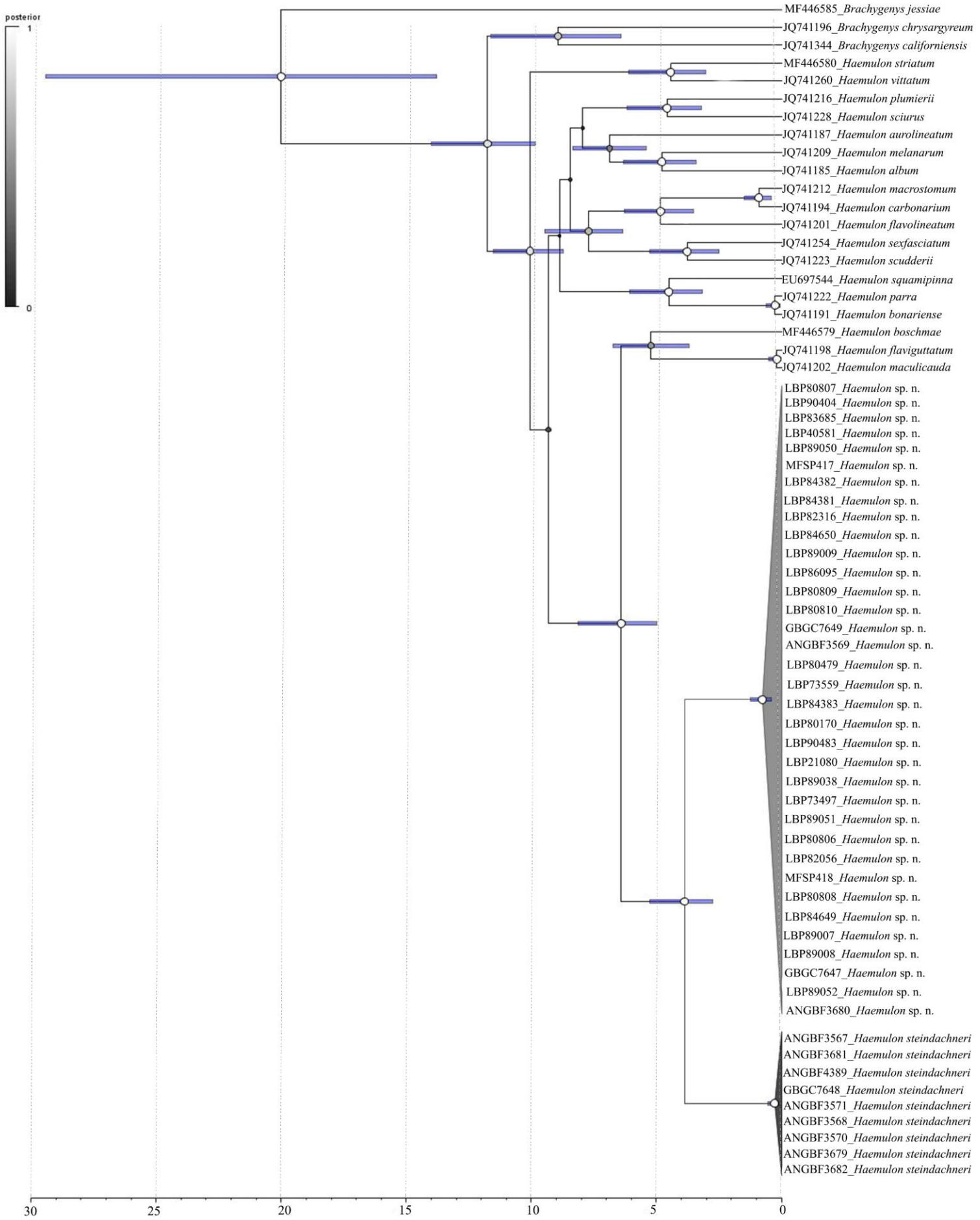
posterior
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JQ741196_Brachygenys chrysargyreum
JQ741344_Brachygenys californiensis
MF446580_Haemulon striatum
JQ741260_Haemulon vittatum
JQ741216_Haemulon plumierii
JQ741228_Haemulon sciurus
JQ741187_Haemulon aurolineatum
JQ741209_Haemulon melanarum
JQ741185_Haemulon album
JQ741212_Haemulon macrostomum
JQ741194_Haemulon carbonarium
JQ741201_Haemulon flavolineatum
JQ741254_Haemulon sexfasciatum
JQ741223_Haemulon scudderii
EU697544_Haemulon squamipinna
JQ741222_Haemulon parra
JQ741191_Haemulon bonariense
MF446579_Haemulon boschmae
JQ741198_Haemulon flaviguttatum
JQ741202_Haemulon maculicauda
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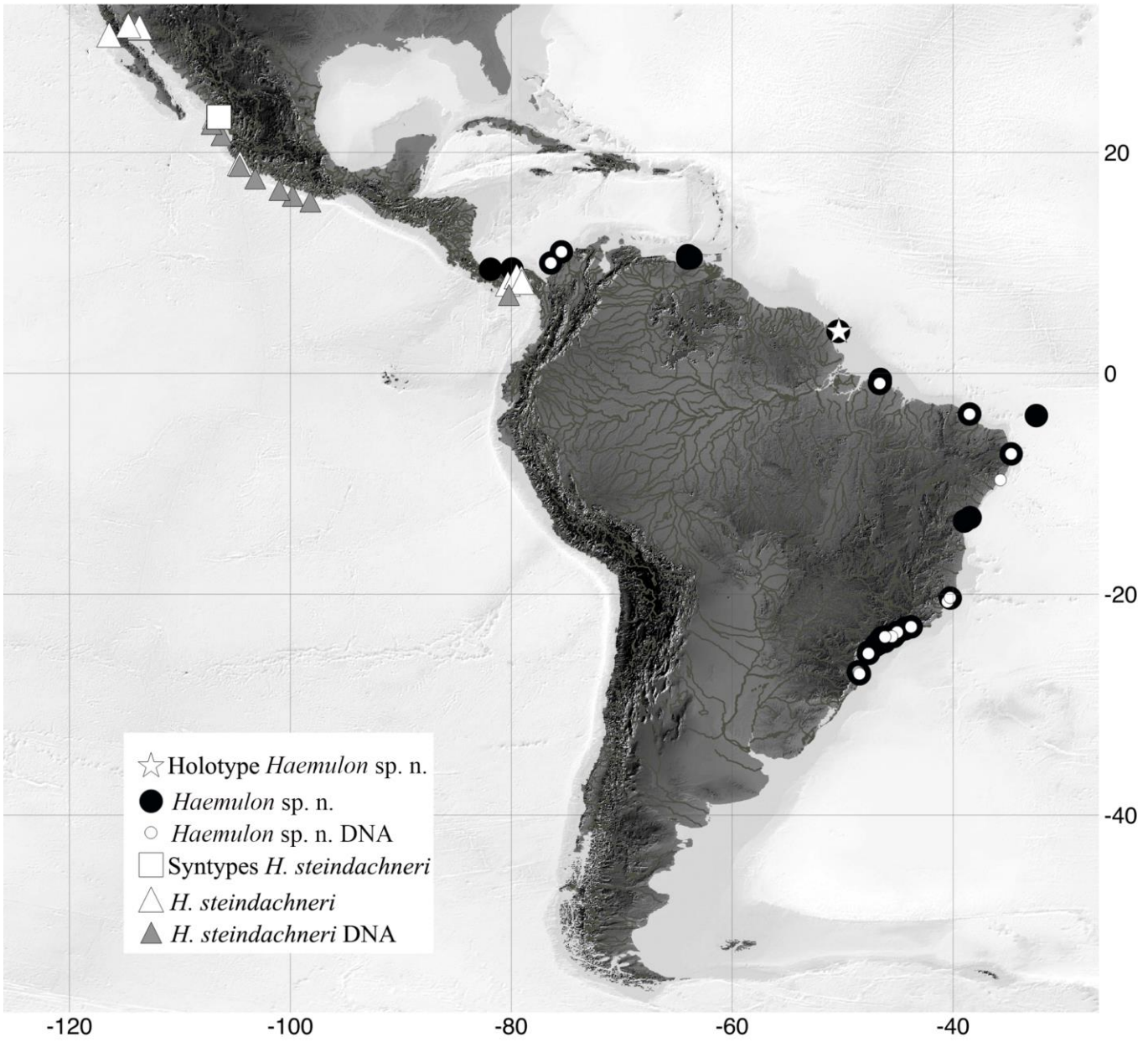
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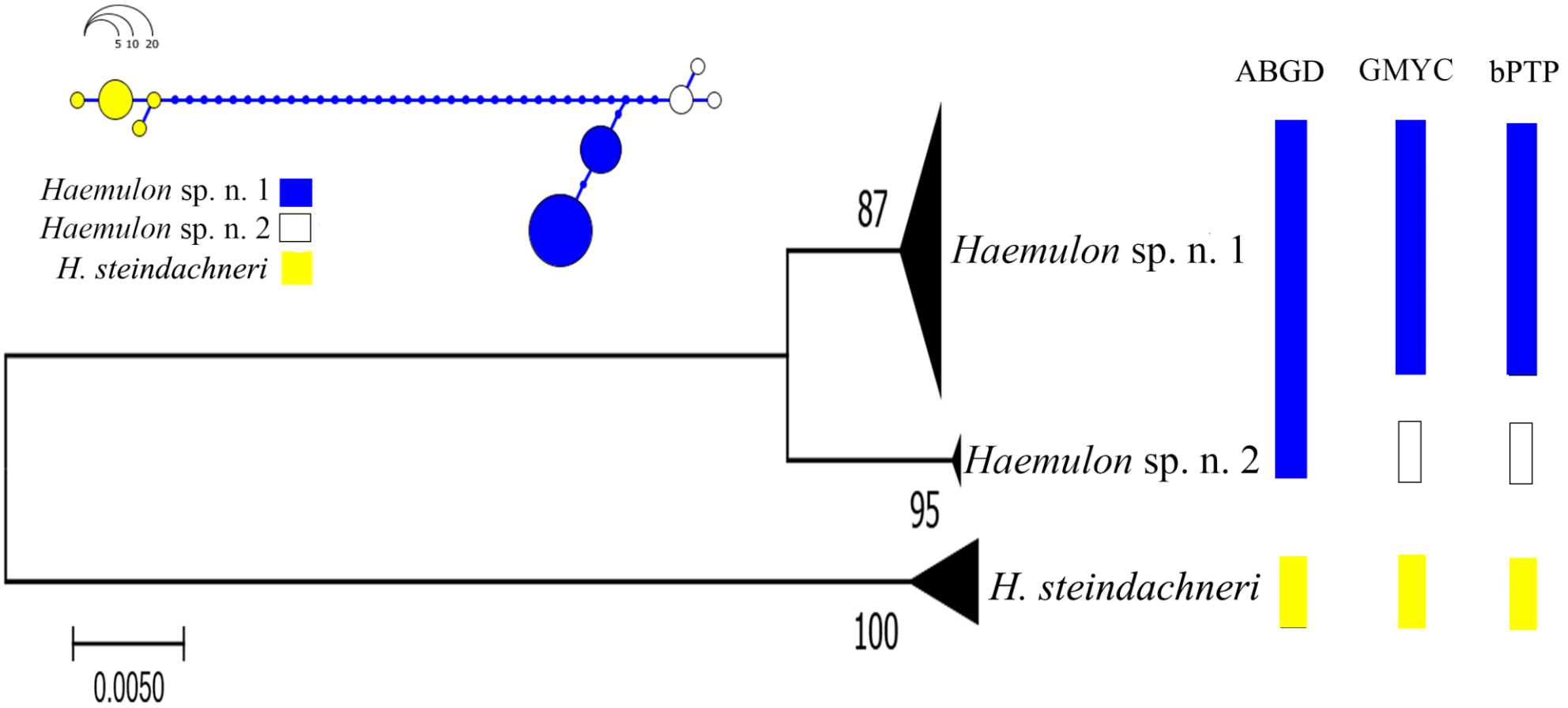
Holotype *Haemulon* sp. n.
Haemulon sp. n.
Haemulon sp. n. DNA
Syntypes *H. steindachneri*
H. steindachneri
H. steindachneri DNA

Supplementary material captions

Supplement I. NJ tree based on K2P distances, ABGD, GMYC and bPTP of all species analyzed. Haplotype network of the *H. Steindachneri* species complex.

Supplement II. X-ray of *Haemulon* sp. n. and *Haemulon steindachneri* in lateral view. (A) *Haemulon* sp. n. holotype, MPEG 35708, 152 mm SL. *Haemulon steindachneri* syntypes. (B) USNM 29226, Mexico. (C) USNM 29759, Mexico. (D) USNM 29778, Mexico. (E) USNM 29795, Mexico.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H. steindachneri
ABGD
GMYC
bPTP
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Haemulon sp. n. 2
95
H. steindachneri
100
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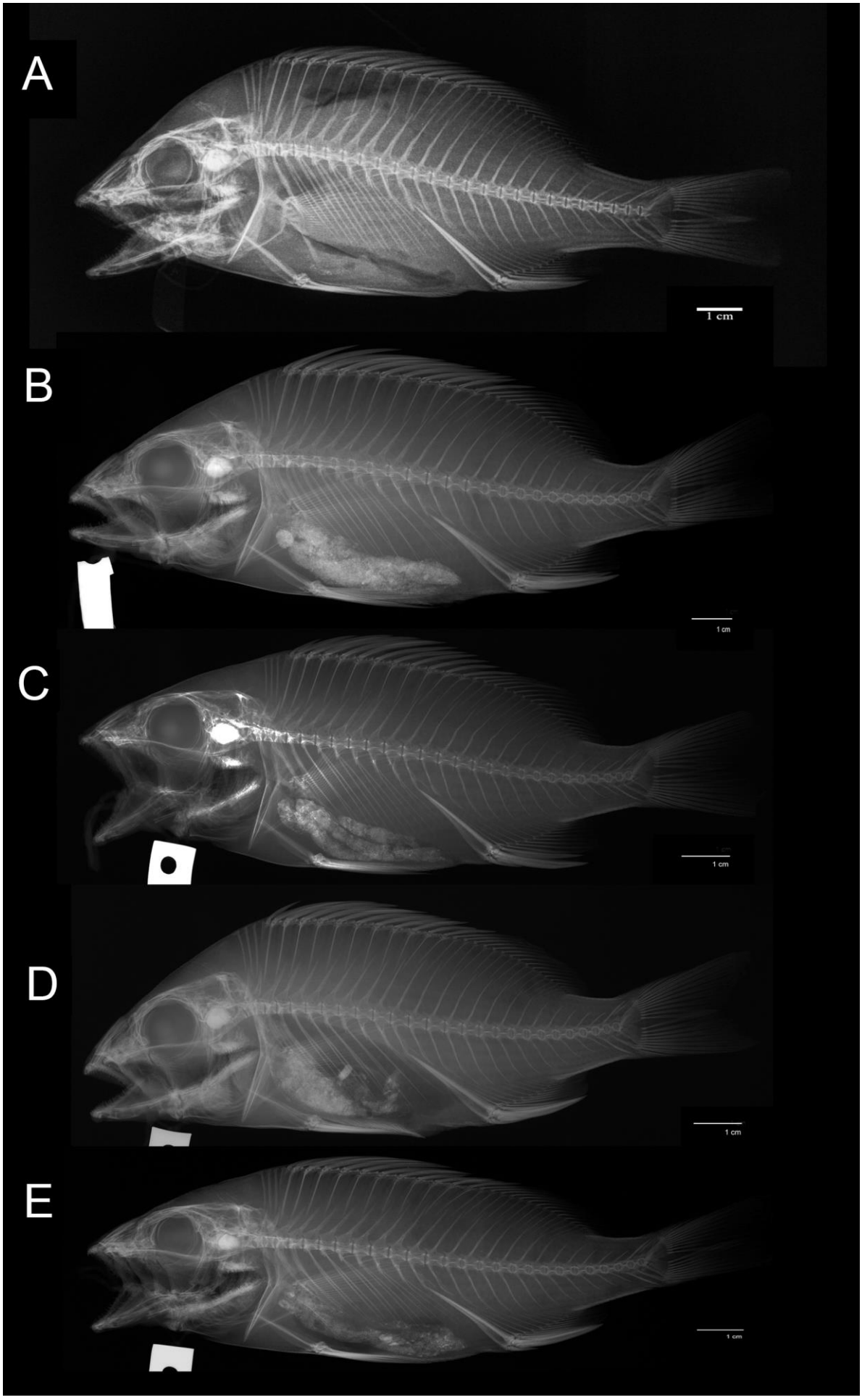
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Table 1

Measures of *Haemulon* sp. n. and *H. steindachneri* expressed as percentages of the standard length.

	Haemulon sp. n.				*Haemulon steindachneri*		
	N	**Holotype**	**Amplitude**	**Average**	**N**	**Amplitude**	**Average**
Standard length	101	152	55−224	-	12	98−201	-
Body height	101	39.5	31.6−43.9	36.5	12	32.8−38.0	35.1
Body width	101	19.2	9.8−21.3	16.7	12	12.3−18.7	15.1
Height of caudal peduncle	101	11.2	8.9−12.3	10.7	12	7.8−11.2	10.1
Width of caudal peduncle	101	3.7	2.7−5.6	3.6	12	2.8−4.1	3.5
Length of caudal peduncle	101	18.4	15.5−23.0	18.2	12	15.8−21.4	19.0
Distance between the snout and the pelvic fin	101	39.6	34.1−43.0	37.9	12	33.1−41.4	37.0
Length of the pelvic fin	101	21.9	18.0−24.3	20.4	12	17.8−21.0	19.8
Dorsal fin height	101	15.9	12.1−20.7	14.9	12	13.2−15.6	14.6
Pectoral fin length	101	29.4	22.2−32.8	27.0	12	22.8−29.5	26.8
Anal fin height	101	13.7	10.2−19.9	14.9	12	13.5−17.6	15.9
Pre-pectoral length	101	34.5	31.5−40.4	35.2	12	31.0−38.3	33.8
Pre-dorsal length	101	36.2	30.7−40.5	35.9	12	31.4−37.0	34.8
Pre-anal length	101	72.7	62.4−79.1	70.1	12	67.9−72.5	70.0
Head length	101	34.1	31.2−41.2	35.3	12	30.6−37.2	33.7
Head height	101	38.3	29.1−41.2	34.5	12	29.8−35.5	33.2
Interorbital distance	101	8.6	6.1−10.0	8.3	12	7.8−9.6	8.7
Post-orbital length	101	15.2	10.2−16.7	14.3	12	10.1−15.4	13.6
Snout length	101	12.6	8.4−15.1	12.1	12	9.8−13.7	11.5
Diameter of the orbit	101	10.1	8.6−17.2	10.7	12	8.6−10.9	9.4
Dorsal fin base length	101	56.6	45.7−58.5	52.3	12	49.4−57.0	52.4
Anal fin base length	101	14.2	11.8−17.2	14.1	12	12.6−15.2	13.8
Mouth width	101	13.3	8.4−15.4	12.6	12	9.1−14.5	11.9
Premaxillary length	101	18.4	13.8−21.4	18.0	12	14.4−19.2	16.5
Premaxillary width	101	2.9	2.4−4.3	3.2	12	2.4−3.3	3.0

Length of the lower lobe of the caudal fin	101	23.3	17.4−29.2	23.1	12	18.5−23.5	21.8
Length of upper lobe of caudal fin	101	26.3	16.8−26.5	21.3	12	17.6−22.8	21.0

Table 2

Meristic data of *Haemulon* sp. n. and *H. steindachneri*. Holotype represented by *.

Lateral-line scales	**50**	**51**	**52**	**53**	**54**	**55**	**56**
Haemulon sp. n.	3	3	33	20*	11	8	5
H. steindachneri	1	2	5	4			
Longitudinal series of scales above the lateral line	**7**	**8**	**9**				
Haemulon sp. n.	5	53	26*				
H. steindachneri	3	9					
Longitudinal series of scales below the lateral line	**11**	**12**	**13**	**14**	**15**	**16**	
Haemulon sp. n.	1	3	29	33	13*	4	
H. steindachneri		9	3				
Dorsal-fin rays	**XII**			**15**	**16**	**17**	
Haemulon sp. n.	85			34	47	4*	
H. steindachneri	12				11	1	
Anal-fin rays	**III**			**8**	**9**		
Haemulon sp. n.	85			59*	26		
H. steindachneri	12				12		
Pectoral-fin rays	**15**	**16**	**17**	**18**			
Haemulon sp. n.	5	48*	28	3			
H. steindachneri		2	10				
1st Branchial Arch	**19**	**20**	**21**	**22**	**23**	**24**	**24**
Haemulon sp. n.	1	2	6	10	21*	28	13
H. steindachneri	1	1	1	2	3	1	2
2nd Branchial Arch	**14**	**15**	**16**	**17**	**18**	**19**	
Haemulon sp. n.		10*	17	18	35	5	
H. steindachneri	1	2	3	5	1		

Table 3

Genetic distance K2P obtained among *Haemulon* clusters. Interspecific distance below the diagonal. Standard error above diagonal.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1. Haemulon sp. n.	*0,005*	0,012	0,017	0,016	0,015	0,017	0,019	0,015	0,019	0,017	0,015	0,020	0,016	0,019	0,019	0,019	0,017	0,018	0,017	0,019
2. H. steindachneri	0,074	0,002	0,017	0,016	0,018	0,018	0,018	0,013	0,017	0,018	0,014	0,019	0,018	0,016	0,018	0,018	0,015	0,019	0,017	0,017
3. H. album	0,122	0,125	0,002	0,017	0,016	0,018	0,017	0,016	0,016	0,017	0,016	0,015	0,016	0,018	0,016	0,017	0,015	0,019	0,017	0,017
4. H. aurolineatum	0,116	0,106	0,117	0,002	0,016	0,017	0,018	0,016	0,016	0,018	0,016	0,016	0,017	0,017	0,015	0,017	0,016	0,018	0,017	0,019
5. H. bonariense	0,107	0,130	0,107	0,112	0,000	0,018	0,017	0,017	0,017	0,018	0,017	0,017	0,004	0,018	0,017	0,017	0,015	0,014	0,017	0,020
6. H. boschmae	0,122	0,129	0,137	0,121	0,135	0,000	0,018	0,015	0,019	0,019	0,014	0,019	0,018	0,018	0,018	0,019	0,018	0,019	0,018	0,020
7. H. carbonarium	0,136	0,123	0,117	0,130	0,127	0,133	0,000	0,018	0,015	0,007	0,018	0,018	0,018	0,018	0,018	0,017	0,015	0,020	0,018	0,018
8. H. flaviguttatum	0,096	0,077	0,114	0,106	0,128	0,099	0,130	0,004	0,017	0,019	0,003	0,016	0,018	0,015	0,017	0,016	0,014	0,018	0,016	0,018
9. H. flavolineatum	0,139	0,115	0,118	0,107	0,122	0,148	0,088	0,130	0,000	0,015	0,017	0,017	0,018	0,018	0,019	0,017	0,016	0,020	0,018	0,021
10. H. macrostoma	0,123	0,123	0,120	0,130	0,135	0,138	0,024	0,142	0,090	0,000	0,019	0,020	0,018	0,018	0,019	0,019	0,017	0,021	0,020	0,020
11. H. maculicauda	0,096	0,082	0,109	0,108	0,125	0,099	0,133	0,004	0,133	0,142	0,006	0,016	0,018	0,016	0,017	0,016	0,014	0,018	0,016	0,018
12. H. melanura	0,155	0,138	0,091	0,108	0,120	0,150	0,140	0,113	0,113	0,157	0,111	0,000	0,017	0,017	0,018	0,017	0,016	0,020	0,017	0,018
13. H. parra	0,109	0,132	0,110	0,120	0,006	0,143	0,130	0,136	0,130	0,132	0,133	0,122	0,002	0,019	0,017	0,017	0,015	0,014	0,016	0,019
14. H. plumierii	0,133	0,100	0,125	0,124	0,131	0,132	0,128	0,101	0,128	0,133	0,106	0,124	0,134	0,011	0,014	0,016	0,016	0,020	0,016	0,019
15. H. sciurus	0,132	0,119	0,111	0,098	0,122	0,134	0,127	0,119	0,141	0,135	0,116	0,129	0,125	0,079	0,002	0,017	0,017	0,019	0,015	0,019
16. H. scudderii	0,138	0,126	0,116	0,129	0,120	0,156	0,127	0,103	0,128	0,140	0,101	0,114	0,123	0,101	0,114	0,002	0,012	0,016	0,017	0,018
17. H. sexfasciatum	0,117	0,098	0,101	0,113	0,100	0,139	0,107	0,091	0,109	0,117	0,088	0,113	0,102	0,100	0,113	0,062	0,000	0,017	0,015	0,017
18. H. squamipina	0,131	0,127	0,139	0,125	0,081	0,141	0,148	0,134	0,148	0,159	0,131	0,141	0,088	0,147	0,139	0,115	0,120	0,000	0,018	0,018
19. H. striatum	0,122	0,115	0,118	0,115	0,115	0,133	0,138	0,113	0,133	0,148	0,116	0,118	0,112	0,111	0,103	0,123	0,108	0,125	0,002	0,015
20. H. vittatum	0,145	0,128	0,118	0,147	0,149	0,157	0,132	0,136	0,165	0,150	0,139	0,139	0,146	0,137	0,142	0,129	0,124	0,133	0,089	0,000

Table 4

Nucleotide differences observed in the COI gene among the analyzed specimens.

	5	6	8	11	14	17	20	23	26	29	35	41	44	47	50	53	56	77	83	86	89	95	98	107	110	111	113	119	125	128	131	134	137
Haemulon sp. n.	C	C	T	G	C	T	A	C	T	C	T	T	G	T	G	C	A	G	T	C	C	G	T	A	C	G	T	A	C	A	G	C	T
H. steindachneri	.	.	C	.	.	.	.	.	.	.	.	.	.	.	.	.	G	.	.	.	.	A	.	.	.	.	.	.	.	G	.	.	.
H. album	.	.	.	.	T	.	G	.	C	.	.	.	.	.	A	.	.	A	.	.	T	.	.	.	T	.	C	.	.	.	A	.	.
H. aurolineatum	.	.	G	A	.	C	G	.	.	.	.	.	.	.	.	.	.	.	.	.	T	A	.	.	T	.	C	.	.	G	.	T	.
H. bonariense	.	.	G	.	.	C	G	.	.	.	.	.	.	C	.	.	.	.	.	.	T	.	.	G	.	.	.	.	.	G	.	.	.
H. boschmae	.	.	.	A	.	.	G	.	C	T	.	.	.	.	.	.	.	.	.	.	T	A	.	.	.	.	.	.	.	.	A	.	.
H. carbonarium	T	.	A	.	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	.	.	A	.	.	.	.	.	.	.	G	C	A	.
H. flaviguttatum	.	.	C	.	.	C	G	T	.	.	.	.	.	.	.	.	.	.	.	.	T	A	.	.	T	.	.	.	.	.	.	.	C
H. flavolineatum	.	.	G	.	.	.	G	.	.	.	.	.	A	.	.	.	G	.	.	.	T	A	.	.	T	.	C	G	.	G	C	A	.
H. macrostoma	T	.	.	A	.	.	G	.	.	.	.	.	.	.	.	.	.	.	.	.	.	.	C	.	.	.	.	.	.	G	C	A	.
H. maculicauda	.	.	.	.	.	C	G	T	.	.	.	.	.	.	.	.	.	.	.	.	T	A	.	.	T	.	.	.	.	.	.	.	C
H. melanura	.	.	G	.	.	C	G	T	.	.	.	A	A	.	.	.	.	.	.	T	T	A	.	.	T	.	C	.	T	G	.	T	.
H. parra	.	.	G	.	.	C	G	.	.	.	.	.	.	C	.	.	.	.	.	.	T	.	.	G	.	.	.	.	T	G	.	.	.
H. plumierii	.	.	C	.	.	C	.	.	.	.	.	.	.	.	.	.	.	.	C	.	T	A	.	.	.	.	.	.	T	G	.	.	C
H. sciurus	.	.	G	A	.	C	G	.	.	.	.	.	.	.	A	.	.	.	C	.	T	.	.	.	.	.	C	.	T	G	A	.	.
H. scudderii	.	.	A	.	.	C	.	T	.	.	.	.	.	.	.	.	.	.	.	.	T	A	C	.	.	.	C	.	T	.	.	.	C
H. sexfasciatum	.	.	A	.	.	C	G	T	.	.	.	C	.	.	.	.	.	.	.	.	T	A	.	.	.	.	.	.	T	G	.	.	C
H. squamipinna	.	T	G	.	.	C	G	.	.	.	.	.	.	.	.	T	.	.	.	.	T	A	.	.	.	A	C	.	.	.	.	.	.
H. striatum	.	.	G	.	.	C	G	.	.	.	C	C	.	.	.	.	G	.	C	.	T	A	.	.	T	.	C	.	.	G	.	.	.
H. vittatum	.	.	A	.	T	C	G	.	.	.	C	.	.	.	A	.	.	.	.	.	T	A	C	.	.	.	C	.	.	.	.	T	.

	134	137	140	143	149	152	155	158	161	170	176	179	182	185	188	194	197	200	203	206	209	212	215	218	221	224
Haemulon sp. n.	C	T	G	A	C	A	A	T	C	T	C	T	C	T	C	C	C	T	C	T	T	A	G	A	G	C
H. steindachneri	.	.	.	.	G	.	G	.	.	.	.	C	T	.	T	.	.	.	.	.	.	.	.	.	.	T
H. album	.	.	A	.	.	.	G	.	.	.	T	C	T	.	T	.	.	C	.	C	.	G	C	.	A	.
H. aurolineatum	T	.	.	.	A	.	G	C	T	.	.	.	T	A	T	T	.	.	T	C	.	.	C	.	A	T
H. bonariense	.	.	.	.	.	.	G	.	.	.	.	C	.	.	T	.	T	.	A	C	.	.	.	T	A	.
H. boschmae	.	.	.	G	.	.	G	C	.	.	.	C	T	G	T	T	.	A	.	C	.	.	.	.	.	T
H. carbonarium	A	.	.	.	.	.	G	.	.	.	G	C	.	.	T	A	.	C	.	C	.	.	.	.	A	.
H. flaviguttatum	.	C	.	.	.	.	G	C	T	.	.	C	T	.	T	T	.	.	.	C	.	.	.	.	.	T
H. flavolineatum	A	.	.	.	T	C	G	C	.	C	T	C	.	.	T	.	.	.	.	C	.	.	.	.	A	.
H. macrostoma	A	.	.	.	.	.	G	.	.	.	G	C	.	.	T	A	.	C	.	C	.	.	.	.	A	.
H. maculicauda	.	C	.	.	.	.	G	C	T	.	.	C	T	.	T	T	.	.	.	C	.	.	.	.	.	T
H. melanura	T	.	A	.	.	.	G	C	.	.	.	C	.	.	T	T	T	C	.	C	C	.	.	.	A	.
H. parra	.	.	.	.	.	.	G	.	.	.	.	C	.	.	T	.	T	.	A	C	.	.	.	T	A	.
H. plumierii	.	C	.	.	.	.	G	.	.	.	.	C	T	.	T	T	.	.	.	.	.	.	T	.	.	T
H. sciurus	.	.	.	.	.	.	G	.	.	.	.	C	T	C	T	T	.	.	.	C	.	.	C	.	.	T
H. scudderii	.	C	.	.	.	G	G	.	.	.	.	C	.	.	T	T	.	.	.	C	.	.	A	.	.	.
H. sexfasciatum	.	C	.	.	.	.	G	.	.	.	.	C	T	.	T	.	.	.	.	C	.	.	.	.	.	.
H. squamipinna	.	.	.	.	.	.	G	.	.	.	.	C	.	C	.	.	.	.	T	C	.	.	.	T	A	G
H. striatum	.	.	.	.	.	.	G	.	.	.	.	C	T	.	T	T	T	C	.	C	.	.	A	.	.	G
H. vittatum	T	.	.	.	G	.	G	.	.	.	.	C	T	.	.	.	.	C	.	.	.	.	.	.	.	A

	227	230	233	236	239	242	245	48	251	254	257	258	260	263	266	269	270	272	275	278	281	284	287	290	293	296
Haemulon sp. n.	G	T	C	A	A	A	T	T	C	C	T	C	G	T	G	T	C	G	A	C	C	G	G	G	T	T
H. steindachneri	A	.	A	G	.	.	.	C	.	.	.	.	.	C	A	.	.	.	.	.	.	.	A	.	.	.
H. album	.	.	A	T	G	G	.	.	.	.	.	.	.	.	.	C	T	.	.	T	.	A	.	A	.	C
H. aurolineatum	.	.	G	T	.	.	.	.	T	.	.	.	A	.	.	.	.	.	.	.	.	.	A	A	.	.
H. bonariense	.	.	G	T	.	.	.	.	.	T	.	.	A	.	.	C	.	C	.	.	.	A	.	C	A	C
H. boschmae	A	A	G	.	.	.	.	.	.	.	.	.	A	C	.	.	.	A	G	T	.	A	.	A	C	C
H. carbonarium	A	.	G	T	G	.	C	G	.	.	.	T	A	.	A	.	.	.	.	T	T	A	A	C	.	C
H. flaviguttatum	.	.	A	G	G	.	.	C	.	.	.	.	A	.	.	.	.	A	.	T	.	.	.	.	.	C
H. flavolineatum	.	.	T	T	.	.	.	A	.	.	.	T	A	C	A	.	.	A	.	T	.	.	A	C	.	C
H. macrostoma	A	.	G	T	.	.	C	G	.	.	.	T	.	.	A	.	.	.	.	T	T	.	A	C	.	C
H. maculicauda	.	.	A	G	G	.	.	C	.	.	.	.	A	.	.	C	.	A	.	T	.	.	.	.	.	C
H. melanura	.	.	A	T	G	G	.	C	.	.	.	.	A	C	.	C	T	.	.	T	.	A	.	A	.	C
H. parra	.	.	G	T	.	.	.	.	.	T	.	.	.	.	.	C	.	C	.	.	.	A	.	C	A	C
H. plumierii	.	C	A	T	G	G	.	.	.	.	.	.	A	C	A	.	.	A	.	T	T	.	A	A	.	C
H. sciurus	.	C	G	G	G	.	.	.	.	.	.	.	A	G	T	C	.	.	.	.	T	.	A	A	.	C
H. scudderii	.	.	G	T	G	.	.	C	.	T	C	T	A	.	T	C	.	A	G	.	.	A	A	A	.	.
H. sexfasciatum	.	.	G	T	.	.	.	C	.	.	.	T	A	.	A	C	.	.	.	T	.	C	A	A	.	.
H. squamipinna	A	.	G	T	.	.	.	C	.	T	C	.	A	.	A	C	.	C	.	.	.	A	.	A	A	C
H. striatum	.	.	G	T	.	.	.	C	T	.	.	.	.	A	.	.	.	.	.	.	.	A	.	A	C	C
H. vittatum	A	.	G	T	G	G	.	C	.	.	.	.	.	A	.	.	.	.	.	.	.	A	.	A	.	C

	299	302	305	308	311	314	318	320	323	326	329	332	335	341	344	347	350	353	359	362	365	368	371	374	377	380
Haemulon sp. n.	T	A	C	C	C	T	T	A	A	T	T	A	A	T	A	C	C	C	T	A	A	C	T	C	A	A
H. steindachneri	.	.	.	.	.	.	.	.	.	.	C	.	.	.	G	T	.	.	.	.	.	.	.	.	G	G
H. album	.	.	T	.	.	C	.	.	.	.	C	.	.	C	G	.	.	.	.	.	G	T	.	.	G	G
H. aurolineatum	.	.	.	.	.	C	.	.	.	.	.	.	.	.	G	.	.	.	.	.	.	.	.	T	G	G
H. bonariense	G	.	T	.	.	C	.	.	.	.	C	C	.	C	G	.	.	.	.	.	.	.	.	.	G	.
H. boschmae	.	.	.	.	.	C	.	.	G	.	C	.	G	.	.	.	.	.	.	G	.	T	.	.	G	.
H. carbonarium	.	.	T	.	.	C	.	.	.	.	C	.	G	.	G	.	.	.	C	.	G	T	.	T	G	G
H. flaviguttatum	.	.	.	.	.	C	.	.	.	.	C	.	G	.	.	.	.	.	.	.	.	T	.	.	G	.
H. flavolineatum	C	.	T	.	.	C	.	.	.	.	G	.	.	.	G	.	.	.	C	.	.	T	.	.	G	G
H. macrostoma	.	.	T	.	.	C	.	.	.	.	.	.	G	.	G	.	.	.	C	.	G	T	.	.	G	G
H. maculicauda	.	.	.	.	.	C	.	.	.	.	C	.	G	.	.	.	.	.	.	.	.	T	.	.	G	.
H. melanura	.	.	T	.	.	C	.	.	T	.	C	.	.	.	.	.	.	T	C	.	.	.	C	.	G	G
H. parra	G	.	T	.	.	C	.	.	.	.	C	C	.	C	G	.	.	.	.	.	.	.	.	.	G	.
H. plumierii	.	.	T	.	.	.	.	.	.	.	.	.	.	C	G	T	T	.	C	.	.	T	.	.	G	.
H. sciurus	.	.	.	T	.	.	.	.	.	.	C	G	.	C	G	.	.	.	C	.	.	T	.	T	G	G
H. scudderii	.	.	T	.	.	C	C	.	.	G	C	.	C	C	G	.	T	.	C	.	G	T	.	.	G	.
H. sexfasciatum	A	.	T	.	.	C	.	.	.	G	C	G	.	G	G	.	T	.	.	.	G	T	.	.	G	.
H. squamipinna	G	.	T	.	.	A	C	G	.	.	C	.	.	C	G	.	.	T	.	T	.	.	.	.	G	.
H. striatum	.	.	.	.	.	.	.	G	.	.	C	G	.	G	G	.	.	.	.	G	.	T	.	.	.	.
H. vittatum	.	G	T	.	T	C	.	G	G	G	C	G	.	G	G	.	.	.	C	G	G	.	.	T	G	.

	383	386	389	392	395	398	401	404	407	410	411	413	416	419	422	425	428	431	434	437	440	443	446	449	452	455
Haemulon sp. n.	T	T	C	C	G	A	C	G	C	C	C	G	C	A	A	A	T	C	C	T	C	T	T	T	T	A
H. steindachneri	.	.	.	.	.	G	.	A	T	G	.	A	.	.	.	.	C	.	.	.	.	C	.	.	C	G
H. album	.	C	T	.	.	G	.	.	.	G	.	.	.	.	.	.	C	.	T	.	.	G	C	.	.	G
H. aurolineatum	C	C	T	.	.	.	.	A	.	A	T	A	T	.	.	.	C	.	.	.	.	A	C	.	.	G
H. bonariense	.	C	T	.	T	.	.	.	.	T	.	.	T	G	.	.	G	.	.	C	.	G	C	.	.	.
H. boschmae	C	.	.	.	A	.	T	A	.	.	.	A	T	G	.	.	C	T	.	C	T	.	A	.	.	G
H. carbonarium	C	C	T	.	C	.	.	.	.	.	.	C	.	.	.	.	C	T	T	.	T	.	A	C	A	G
H. flaviguttatum	.	.	.	.	.	G	.	A	T	.	.	A	.	.	.	.	.	.	T	.	.	C	.	.	C	G
H. flavolineatum	C	C	T	.	.	G	.	.	.	A	.	.	.	.	.	C	C	T	.	.	T	A	C	.	.	G
H. macrostoma	C	C	T	.	.	.	.	.	.	.	.	C	.	.	.	.	C	T	T	.	T	.	A	C	A	G
H. maculicauda	.	.	.	.	.	G	.	A	T	.	.	A	.	.	.	.	.	.	T	.	.	C	.	.	C	G
H. melanura	.	C	T	.	.	G	.	A	.	G	.	.	T	.	G	C	C	.	A	.	.	A	C	.	.	G
H. parra	.	C	T	.	T	.	.	.	.	T	.	.	T	G	.	.	G	.	.	C	.	G	C	C	.	.
H. plumierii	.	C	T	.	C	G	T	A	.	.	.	A	.	G	G	.	C	.	T	.	T	A	C	.	.	C
H. sciurus	.	C	T	.	A	.	T	A	.	.	.	.	.	.	G	.	C	.	T	.	.	A	.	.	.	C
H. scudderii	.	C	T	.	.	G	.	A	T	.	.	.	.	.	.	G	C	.	A	.	.	A	C	.	.	G
H. sexfasciatum	C	.	T	.	.	G	.	.	T	.	.	.	.	.	.	.	C	.	A	.	.	G	C	.	.	G
H. squamipinna	.	C	T	T	.	.	.	A	T	.	.	A	T	G	.	.	C	T	.	.	.	G	.	.	.	T
H. striatum	.	C	T	.	C	G	.	A	.	.	.	.	.	C	.	C	C	.	T	.	T	A	.	C	.	.
H. vittatum	.	C	T	T	C	G	.	.	T	.	.	A	.	C	.	C	C	.	T	.	T	A	.	C	C	.

	458	461	464	467	470	473	476	479	482	485	488	491	494	497	500	501	503	505	506	507	509	510	512
Haemulon sp. n.	C	C	A	A	C	A	T	C	T	A	G	C	C	A	C	C	A	A	C	C	G	A	T
H. steindachneri	.	.	.	.	.	G	.	T	C	.	.	.	.	.	.	.	.	.	.	.	.	.	C
H. album	.	.	.	.	T	G	G	.	C	.	.	.	.	.	T	.	.	.	.	.	A	.	C
H. aurolineatum	.	.	.	C	T	G	.	.	C	.	.	.	.	.	.	.	.	.	.	.	A	.	C
H. bonariense	.	.	C	.	.	G	C	.	.	.	.	.	A	.	.	.	.	.	.	.	A	.	C
H. boschmae	.	.	.	.	T	.	.	.	C	.	.	T	.	.	.	.	.	.	T	.	A	.	.
H. carbonarium	.	.	C	T	.	G	C	.	.	.	.	T	.	.	.	.	.	.	T	.	.	.	C
H. flaviguttatum	T	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	A	.	.
H. flavolineatum	.	.	C	T	.	G	.	.	.	.	.	A	T	.	.	G	.	T	.	.	.	C	C
H. macrostoma	.	T	C	T	.	G	C	.	.	.	.	T	.	.	.	.	.	.	T	.	.	.	C
H. maculicauda	T	.	.	.	.	.	.	.	.	.	.	.	T	.	.	.	.	.	.	.	A	.	.
H. melanura	.	.	.	.	T	G	G	.	.	.	.	.	.	.	T	.	.	.	T	A	A	.	C
H. parra	.	.	C	.	.	G	C	.	.	.	.	.	A	.	.	.	.	.	.	.	A	.	C
H. plumierii	.	T	.	.	.	G	A	.	.	G	.	.	T	.	.	.	.	.	.	.	A	.	C
H. sciurus	.	.	.	G	.	G	.	T	.	G	.	T	.	.	.	.	.	.	.	.	A	.	.
H. scudderii	.	.	.	.	.	G	.	.	.	G	.	G	T	G	.	.	.	.	T	.	A	.	C
H. sexfasciatum	.	.	.	.	.	G	.	T	.	.	.	.	T	G	.	.	.	.	.	.	A	.	C
H. squamipinna	.	.	T	.	.	G	C	.	.	.	.	A	G	.	.	.	.	.	T	.	A	.	C
H. striatum	.	.	.	.	.	G	.	.	.	.	.	T	T	.	.	.	.	.	T	.	A	.	C
H. vittatum	.	.	.	.	T	G	.	.	.	.	A	T	T	.	.	.	G	.	T	.	A	.	C

ANEXO

Normas para formatação e submissão de artigos – Zoology ISSN 0944-2006

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The main conclusions of the study may be presented in a short Conclusions section, which may stand alone or form a subsection of a Discussion or Results and Discussion section.

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