diversity

MDPI

Article

Mayamysis bacalarensis n. gen., n. sp. (Crustacea: Malacostraca: Mysida) from Lake Bacalar (Quintana Roo State), an Oligotrophic Ecosystem in Southern Mexico

Diana Estela González-Trejo [1] and Manuel Elías-Gutiérrez [2,*,†]

[1] Tecnológico Nacional de Mexico—I. T. Chetumal, Av. Insurgentes 330, Chetumal 77013, Quintana Roo, Mexico; degt16739@gmail.com
[2] El Colegio de la Frontera Sur, Chetumal, Av. Centenario Km 5.5, Chetumal 77079, Quintana Roo, Mexico
* Correspondence: melias@ecosur.mx
† urn:lsid:zoobank.org:pub:DF423248-2CC8-4988-8E56-4F3960CC4676.

Abstract

Lake Bacalar is an oligotrophic body of water considered an extreme environment due to its unusual chemistry, with many species yet to be discovered. Mysids were recently collected in this system using light traps. Most previous records of these organisms in the continental Yucatán Peninsula are from closed cenotes (sinkholes or dolines) and caves. We applied integrative taxonomy, considering morphological characters of mature males and females, their geographical distribution, and mitochondrial cytochrome C oxidase subunit I (COI) gene sequences to describe *Mayamysis bacalarensis* n. gen., n. sp. The morphological analyses included scanning electron microscopy (SEM) and bright-field microscopy observations. We found that *M. bacalarensis* n. gen., n. sp. has a unique combination of morphological characters within the tribe Gastrosaccini, such as the absence of rostrum, the terminal structure of the third pair of pleopods in males, which are highly developed and have a reduced endopod. Both sexes have a lamellar projection on the fifth abdominal segment and numerous setae (35–40) and spines (22) on the margins of the exopod and endopod of the uropods. In addition, the telson has a posterior cleft margin, bearing a total of 32 spines, the apical and lateral ones being the largest. Furthermore, the genetic sequences of these mysids indicate that the organisms from Bacalar are unique, showing an average of 15% genetic divergence from the closest relative, represented by *Chlamydopleon dissimile*, and over 20% to more than 50% divergence with other mysids sequenced from Mexico. The geographic range of *M. bacalarensis* n. gen., n. sp. includes the northern part of Chetumal Bay, a brackish oligotrophic lagoon, where organisms identical to those from Lake Bacalar were collected. This finding indicates a possible permanent connectivity through subterranean water flows. Based on the evidence, we conclude that this is a new genus and species, possibly microendemic to southeastern Quintana Roo, Mexico.

Keywords: integrative taxonomy; DNA barcoding; morphology; freshwater; COI; DNA barcodes

Academic Editor: Simon Blanchet

Received: 16 March 2026
Revised: 1 May 2026
Accepted: 3 May 2026
Published: 7 May 2026

1. Introduction

Lake Bacalar is a tropical karst lake located in the southern part of the state of Quintana Roo. It is considered the largest freshwater lake in the Yucatán Peninsula [1], and the second largest in Mexico, with an average length of approximately 50 km, a mean depth of 8.85 m,

and an average width of 1.54 km [1]. The lake is located approximately 17 km away (straight-line distance) from Chetumal Bay. The latter is an oligotrophic estuary [2,3] and is indirectly connected to Lake Bacalar through the Hondo River, forming, together with Lake Bacalar, part of a complex underground system [3] that remains poorly understood.

Lake Bacalar is also renowned for harboring the largest living microbialites in the world [4]. It is characterized as an oligotrophic lake with waters supersaturated with $CaCO_3$ and sulfates [5] and is therefore considered an extreme environment [6].

The diversity of freshwater zooplankton is high, but biogeographically restricted, and remains understudied; consequently, many species that have only recently been observed still require formal description [7]. The best known and most extensively studied are rotifers, cladocerans, and copepods [7].

Efforts are currently underway to apply integrative taxonomy to recognize previously unknown groups, including arrenurid water mites. In this context, the species *Arrenurus* (*Megaluracarus*) *ecosur* n. sp. has already been described from Lake Bacalar and other aquatic systems in the southern Yucatán Peninsula [8].

The identification of zooplanktonic organisms using integrative taxonomy incorporates morphological, biogeographical and molecular traits, among others [9]. However, these studies require specific methodologies for the fixation and preservation of biological material [10]. Morphological characters used to determine taxonomic identity are based on traits that exhibit sexual dimorphism, as well as other structures that allow differentiation between organisms, such as antenna 1, the antennal scale, telson, uropod, and eyes [11].

On the other hand, molecular approaches commonly involve the analysis of the first half of the mitochondrial cytochrome C oxidase (COI) gene, which is widely used as a DNA barcode also known as the "Barcode of Life". According to Hebert et al. [12], the intraspecific divergence is usually less than 2%, but this figure largely depends on the rate of evolution, which is related to the genetic variation in the species [13].

Among the groups not previously recorded in the region are mysids. These organisms were collected for the first time using non-conventional sampling methods, such as light traps [10]. This method has proven to be an effective alternative for collecting zooplanktonic organisms [14], as it does not produce disturbances that can be detected by these organisms and cause them to evade the sampling net [15].

Mysids are of considerable scientific and applied interest, as they are regarded as bioindicators and play an important role as intermediate components of aquatic food webs [16]. Consequently, they have been widely used as food for other organisms, including crustaceans, and fish as seahorses [17,18].

Records of mysids are more common in marine habitats than in brackish, freshwater, or groundwater environments [19]. More than 90% of species occur in marine waters [16,20], whereas approximately 118 species have been reported from freshwater or brackish environments [16]. Until recently, freshwater mysids had only been reported from closed cenotes on the Yucatán Peninsula.

The closest previous record of mysids in the southern Yucatán Peninsula comes from Dos Hermanos, within Chetumal Bay, approximately 35 km from Lake Bacalar, which represents a different environment [21]. These organisms were identified as *Bowmaniella floridana* Holmquist, 1975, and are currently deposited in the Reference Collection of the Zooplankton Laboratory in ECOSUR. Their identification was based on morphology, and the samples were initially preserved in formalin.

The aim of this work is to elucidate the taxonomic identity of the mysids found in Lake Bacalar using an integrative taxonomic approach and to compare them with previous regional records.

2. Materials and Methods

We worked with samples previously collected by El Colegio de la Frontera Sur (ECOSUR), Chetumal Unit, at three sites in Lake Bacalar, Quintana Roo, which correspond to the following coordinates: site 1, (Cenote Cocalitos: 18.6509 N, 88.4099 W), site 2 (near Buenavista town: 18.8650 N, 88.2460 W), and site 3 (Near the town Pedro A. Santos: 18.9360 N, 88.1570 W) (Figure 1). The samples were collected from 2015 to 2020 at depths ranging from 5 to 15 m.

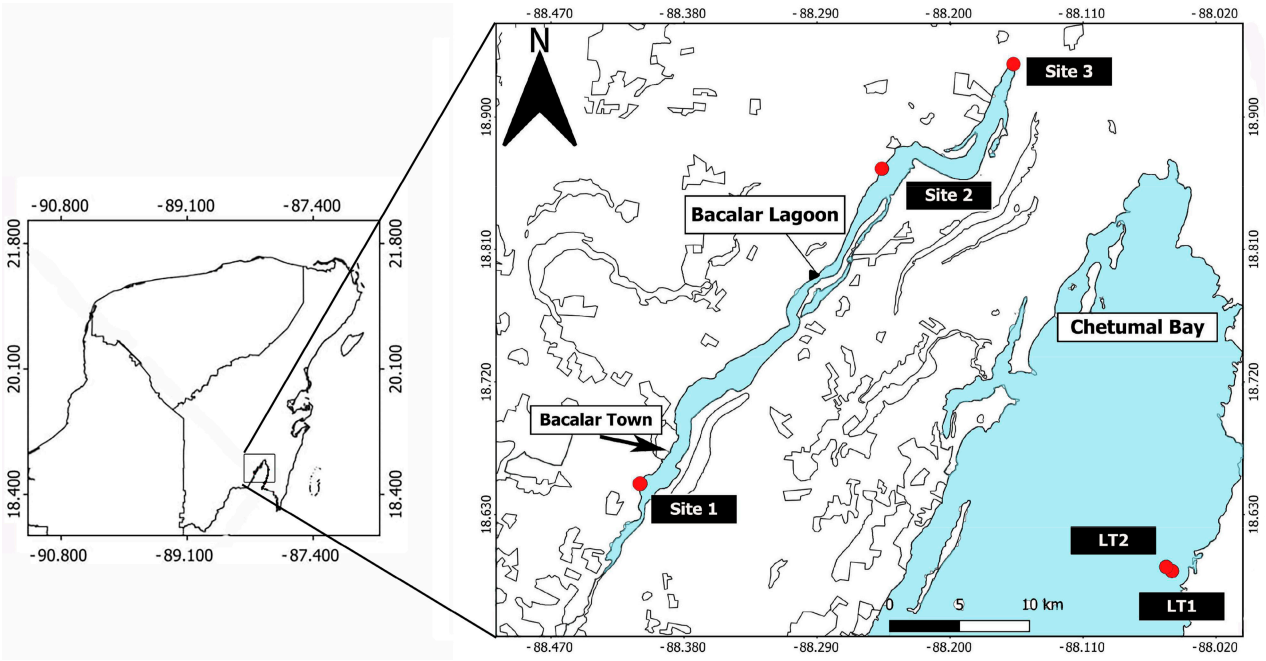

Figure 1. Collection sites in Lake Bacalar and sampling sites with light traps (LT), Dos Hermanos-Chetumal Bay, Quintana Roo, January 2025.

The specimens were separated from the zooplankton samples using fine-tipped tweezers and placed in vials with 96% alcohol for preservation.

2.1. Morphological Analysis

For the morphological description, bright-field microscopy was performed using two techniques: differential interference contrast (DIC; Nomarski) and phase-contrast. A scanning electron microscope (SEM; Jeol Model JSM6010, Tokyo, Japan) was also used. Two SEM observation modes were applied: low vacuum with cooled stage (−26 °C) for two specimens and high vacuum using 15 specimens dehydrated through a graded ethanol series, critical-point dried, and sputter-coated with gold. All observation settings are included in each photograph.

The morphological description of these organisms was compared with previously described mysids from the Yucatán Peninsula inhabiting freshwater, marine, and brackish environments, including material from Chetumal Bay deposited in the ECOSUR Zooplankton Reference Collection.

Photographs and diagrams obtained were processed using Adobe Photoshop (v25.12.3).

Subsequently, the mysids from Lake Bacalar were identified to the lowest possible taxonomic level based on morphological descriptions, taxonomic keys, and identification guides, among which the works of Price [11], Johnson & Allen [22], Ortiz & Cházaro-Olvera [23], Ortiz & Lalana [19], and Wittman et al. [24].

The genetic sequences of the Bacalar mysids, previously uploaded by Elías-Gutiérrez et al. [10], were obtained from the Barcode of Life Database (BOLD) platform.

Additionally, specimens were collected and prepared for DNA extraction and sequencing for cytochrome C oxidase subunit I (COI) gene from Chetumal Bay, at the locality known as Dos Hermanos, where these organisms had previously been recorded [21,25]. Sampling was conducted from 17–18 January 2025. Two light traps were used for the collection near the Dos Hermanos viewpoint from 17:20 and 17:53 until 9:00 on the following day. The coordinates of the sampling sites were: Light Trap 1 (LT1) (18.5916 N, 88.0497 W) and Light Trap 2 (LT2) (18.5944 N, 88.0536 W) (Figure 1).

2.2. DNA Extraction

The procedure for extracting DNA from specimens collected in Chetumal Bay was as follows:

DNA was extracted from abdominal muscle tissue from 12 specimens, and the DNA was extracted according to Ivanova et al. [26]. From each DNA extract, 3–4 µL was added to a PCR mixture consisting of 2 µL of Hyclone ultrapure water, 6.25 µL of 10% D-(+)-trehalose dihydrate, 1.25 µL of 10× Platinum Taq buffer (Invitrogen, CA, USA), 0.625 µL of 50 µmol/L MgCl2 (Invitrogen), 0.0625 µL of 10 µmol/L dNTP, 0.125 µL of each 10 µmol/L primer (Zplank primers) [27], and 0.06 µL of PlatinumTaq (Invitrogen).

An Eppendorf thermocycler, model 6321, was used. The cycles were as follows: the temperature was increased to 95 °C for 1 min, followed by 5 cycles of 94 °C for 40 s, 45 °C for 40 s, 72 °C for 1 min, followed by 35 cycles at 94 °C for 40 s, 51 °C for 40 s, 72 °C for 1 min, and finally 72 °C for 5 min. The PCR products were visualized on a 2% agarose gel using a 95-well E-Gel Precast Agarose Electrophoresis System (Invitrogen), and those positive for sequencing were then selected [10].

The bidirectional sequencing reaction was performed at Macrogen (Seoul, Republic of Korea) using M13F and M13R primers [27]. The reactions were then sequenced on an ABI3730XL automated sequencer (Thermo Fisher Scientific, Waltham, MA, USA). The sequences obtained were edited with CodonCode v3.0.1 and uploaded to the BOLD platform database [28] (https://boldsystems.org) and GenBank.

All sequences were uploaded and examined using BOLD analytical tools to detect stop codons or insertions. Sequences that met minimum quality criteria (>500 bp, <1% ambiguous bases, and free of stop codons and contaminants) were assigned a Barcode Index Number (BIN) [29]. All sequences of *Mayamysis bacalarensis* n. gen, n. sp. are included in the dataset DS-MYSID (Mysidae from Mexico), with the following DOI: https://dx.doi.org/10.5883/DS-MYSID (created by us on 2 April 2026). For each specimen, the accession number from the Reference Collection at El Colegio de la Frontera Sur (ECOSUR), Chetumal Unit, as well as complete sampling details, are provided.

2.3. Molecular Data Processing

Each sequence represents a Molecular Operational Taxonomic Unit (MOTU) that was assigned a Barcode Index Number (BIN) if it met the minimum quality standards proposed by Ratnasingham & Hebert [29].

All mysid sequences from Mexico registered on the BOLD platform were downloaded in FASTA format. Prior to maximum likelihood analysis, sequences were aligned using the MUSCLE algorithm implemented in MEGA (v12.0) [30], resulting in a final alignment of 530 bp per sequence. The sequences were subsequently analyzed to determine the most appropriate model of molecular evolution. The best-fit model was selected using the Akaike Information Criterion (AICc). The model that best fits our dataset, as implemented in Mega

(v12.0) was the General Time Reversible + Gamma (GTR + G). Maximum likelihood analysis was conducted in Mega (v12.0), with 500 bootstrap replicates.

Additionally, using the same dataset, we generated a Bayesian inference tree in MrBayes v3.2.6 [31] by running 1,000,000 MCMC generations. This tree was subsequently used as input for the bPTP analysis implemented on the bPTP web server (https://species.h-its.org), which was run for 100,000 MCMC generations [32].

Due to the complexity of mysid systematics, we conducted a global comparative analysis. A worldwide search was performed for all mysid sequences available in BOLD, including public records, with sequence lengths $\geq$ 500 bp. This search yielded a total of 2562 sequences. From these, after filtering, 1929 were used to construct an identification tree using the analytical tools provided by BOLD. Sequences were aligned and genetic distances were calculated using the Kimura two-parameter (K2P) [33] model with 500 replications in the bootstrap method. The tree was generated using the nearest-neighbor method and subsequently simplified using the Compress/Expand Subtree tool in MEGA (v12.0) [30].

These results were contrasted with species delimitation outcomes obtained using the ASAP (Assemble Species by Automatic Partitioning) method [34] implemented in the Spart Explorer platform (https://spartexplorer.mnhn.fr/). MOTUs were inferred using the program's default settings after sequence alignment with the MUSCLE algorithm using default settings in MEGA (v12.0), resulting in 1873 aligned sequences.

Finally, the haplotype variability of *Mayamysis bacalarensis* n. gen., n. sp. was analyzed using DnaSP v6.12 [35]. A statistical parsimony approach was used to construct a minimum spanning haplotype network in PopART [36]. To assess whether the population from Bacalar has undergone expansion, we applied Tajima's neutrality test in DnaSP v6.12 [35].

2.4. Nomenclatural Acts

The electronic version of this article in Portable Document Format will represent a published work according to the International Commission on Zoological Nomenclature (ICZN), and hence the new names contained in the electronic version are effectively published under that Code from the electronic edition alone. This published work and the nomenclatural acts it contains have been registered in ZooBank, the online registration system for ICZN. The ZooBank Life Science Identifiers (LSIDs) can be resolved and the associated information viewed through any standard web browser by appending the LSID to the prefix http://zoobank.org/. The LSID for this publication is: urn:lsid:zoobank.org:pub:DF423248-2CC8-4988-8E56-4F3960CC4676. The online version of this work is archived and available from the following digital repositories: Diversity-Basel.

3. Results

Systematics

Subphylum Crustacea Brünnich, 1772

Superclass Multicrustacea Regier, Shultz, Zwick, Hussey, Ball, Wetzer, Martin & Cunningham, 2010

Class Malacostraca Latreille, 1802

Subclass Eumalacostraca

Superorder Peracarida Calman, 1904

Order Mysida Boas, 1883

Family Mysidae Haworth, 1825

Subfamily Gastrosaccinae Norman, 1892

Tribe Gastrosaccini Norman, 1892

Mayamysis n. gen.

Mayamysis bacalarensis n. sp.

3.1. Morphological Analysis

Material examined. Type material: Holotype. One adult male, (total length (L) = 6 mm), deposited in the ECOSUR Zooplankton Reference Collection, ECO-CH-Z-12640, Mexico, Cenote Cocalitos, Bacalar, Quintana Roo (18°39′3.6″ N, 88°24′33.8″ W), collected on 11 July 2015 (Figure 2).

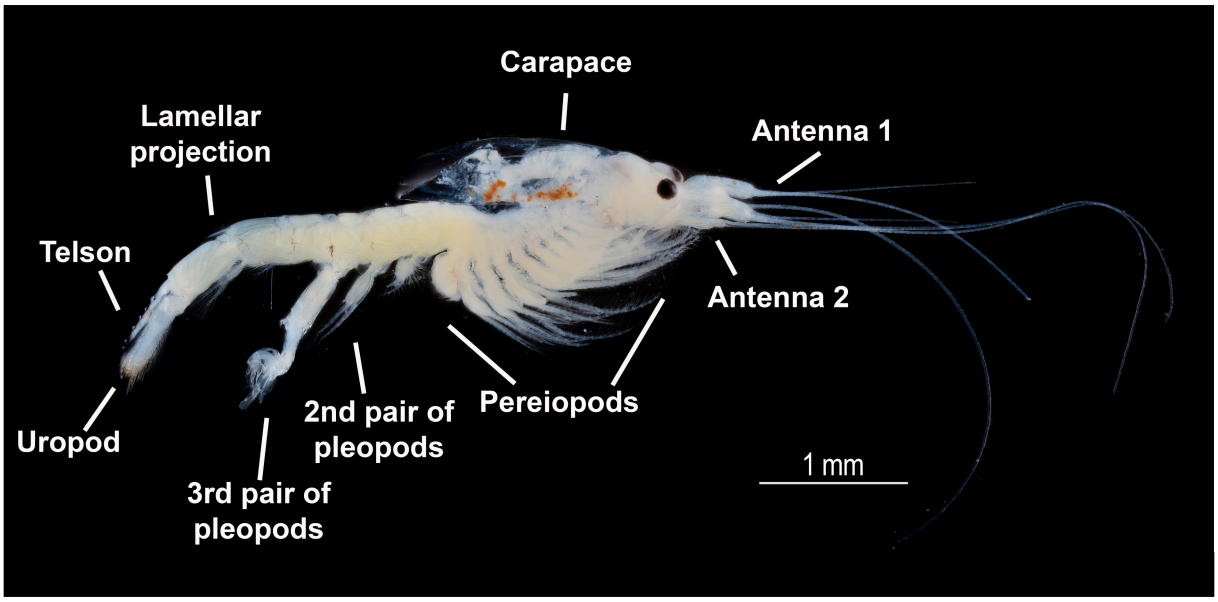

Figure 2. Holotype: adult male mysid from Lake Bacalar, lateral view (Photographed by Humberto Bahena Basave, 2025).

Allotype: One adult female, (L = 6.2 mm), deposited in the ECOSUR Zooplankton Reference Collection, ECO-CH-Z-12641. Collected on 11 July 2015 (Figure 3). Same collection data as the holotype.

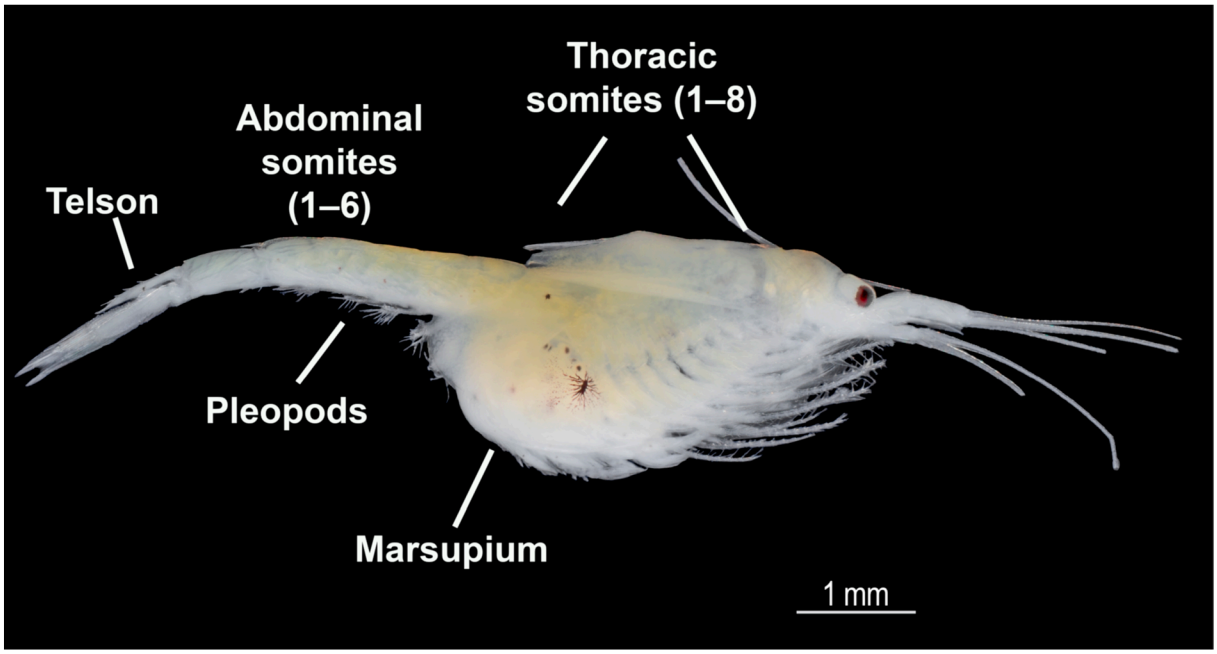

Figure 3. Allotype: adult female mysid in lateral view (Photographed by Humberto Bahena Basave, 2025).

Paratypes: Ten adult females: (L = 5.1–6.7 mm) (ECO-CH-Z-12642); 2 subadult males: (L = 4.8–5.4 mm); 1 adult male, 6 mm (ECO-CH-Z-12643). Same collection data as the holotype.

Additional material: Slides with 1 juvenile male: 3.8 mm; and 2 juvenile females L = 1.8–2.9 mm; deposited in the ECOSUR Zooplankton Reference Collection, ECO-CH-Z-12644, Mexico, Cenote Cocalitos, Bacalar, Quintana Roo (18°39′3.6″ N, 88°24′33.8″ W), 18 April 2015. One slide with one adult female: 5.6 mm deposited in the ECOSUR Zooplankton Reference Collection, ECO-CH-Z-12645, Mexico, Cenote Cocalitos, Bacalar, Quintana Roo, (18°39′3.6″ N, 88°24′33.8″ W), 11 July 2015. One vial with 1 adult male: 5.4 mm; 2 adult females: 7.6 mm and 6.5 mm; deposited in the ECOSUR Zooplankton Reference Collection, ECO-CH-Z-12646 Mexico, Cenote Cocalitos, Bacalar, Quintana Roo (18°39′3.6″ N, 88°24′33.8″ W), 11 July 2015. Vial with 1 juvenile female, 2.8 mm, deposited in the ECOSUR Zooplankton Reference Collection, ECO-CH-Z-12647, Mexico, Cenote Cocalitos, Bacalar, Quintana Roo, (18°39′3.6″ N, 88°24′33.8″ W), 1 December 2015.

Etymology and gender. The generic name is derived from "Maya" referring to the Yucatán Peninsula, a region historically associated with the Mayan culture, and "mysis". Gender is neutral.

Mayamysis n. gen.

Diagnosis: Normal eyes with numerous pigmented and pedunculated ommatidia. Second segment of antenna 1 with two lateral spines and a smaller spine on the third segment. The basal part of the external antennular flagellum is smooth and unisegmented. External margin of the antennal scale smooth with a distal tooth, while the rest of the margin is covered with long and medium setae. Posterodorsal carapace straight; abdominal segments fully covered. Exopod of the pereiopods multisegmented, while the endopod of the third to eighth has a multisegmented and densely setose carpus. The marsupium encompasses the first abdominal segment supported by pleural plates. Pleopods are biramous in both sexes; endopod is short and unisegmented with setae in both sexes; exopod is multisegmented in males and unisegmented in females, both with setae. Third male pleopod has a pronounced and distally complex exopod. The fifth abdominal segment has a caudally projected lamella. The exopod of the uropod has spiny setae on the outer margin, while the rest is covered with long setae; the endopod has short setae near the basal part, while those on the distal part are larger. Statocyst in the endopod. The telson has marginal spines; apical cleft with a lamina with small spines.

Mayamysis bacalarensis n. gen., n. sp.

Diagnosis: All characteristics described above in the genus description, plus the following. Concave face extending to less than 10% of eye length. Carapace covers all thoracic segments; posterodorsal margin straight without median or submedian lobes. In adult males, the distal structure of the exopod of the third pair of pleopods is characterized by the presence of a bow lacking apophysis. The apical slit of the telson covers 10–11% of the total length.

Etymology: The term "*bacalarensis*" refers to Lake Bacalar, where the specimens were collected by the first time.

Male: a single morphotype was observed, represented in Figures 2 and 4. This morphotype is described below.

The head of the organism has a pair of developed, separated, pigmented pedunculated eyes with numerous ommatidia. Rostrum absent. Two pairs of antennae, one pair unirameous (antenna 1) and the other birrameous (antenna 2). Antenna 1 is divided into three segments, with two small lateral spines on the second segment, while the third segment consists of two flagellar extensions for both sexes (Figure 5A). As for antenna 2, the endopod is divided into three segments, while the exopod is an antennular scale extending toward

the second antennular segment. It has a smooth margin ending in a toothlike structure followed by approximately 13 setae with hairs (Figure 5B).

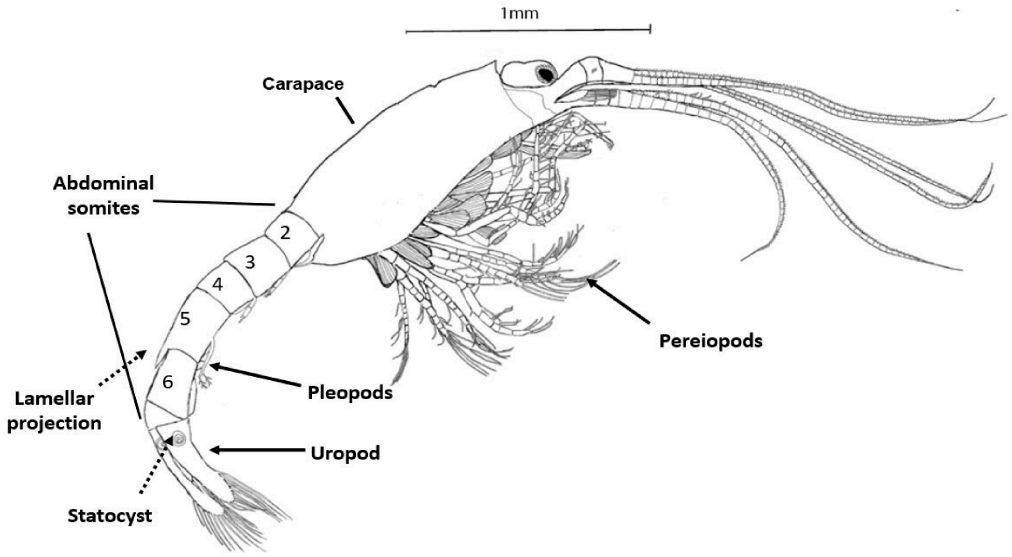

Figure 4. Juvenile mysid collected in Lake Bacalar on 18 April 2015, from Cenote Cocalitos. Lateral view.

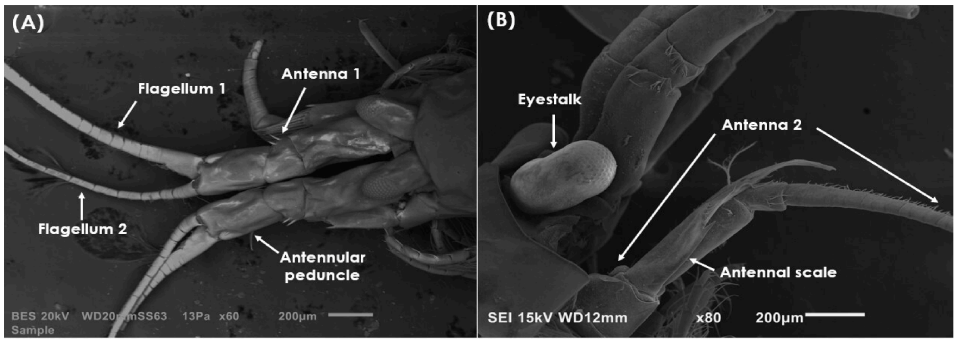

Figure 5. (**A**): Anterior part in dorsal view seen under a scanning electron microscope (SEM). Note the absence of rostrum. (**B**) Anterior part in lateral view (SEM).

The thorax is completely covered by the carapace, which forms a short, concave face toward the anterior side in dorsal view, with a straight edge in posterodorsal view without medial or submedial lobes. It has eight pairs of biramous thoracic appendages (pereiopods), of which the first two are considered maxillipeds; from the third to the eighth pair, the endopod is characterized by being longer than the exopod, where the latter is divided into two segments, the first segment having a broad laminar shape from which the second segment protrudes, which has multiple divisions with setae (Figure 6). The endopod is formed by five segments, of which the last (carpus) is subdivided into five to eight subsegments. Each of these has a pair of small lateral spines in its apical region (Figure 6).

Males have a pair of short penises located on the last pereonal segment [19] (Figure 7).

The abdomen is divided into six segments, where the first segment is partially covered by the carapace, the fifth has a protruding lamellar projection (Figure 8B), while the sixth is characterized by being the largest, as can be seen in Figure 8A.

The first five abdominal segments have a pair of biramous pleopods with fine setae.

The exopod is multisegmented with various setae, while the endopod is shorter and unisegmented. The third pair of pleopods is characterized by being a distally widened

copulatory organ [19]. The exopod is distinguished by its large size and by being divided into five segments, the last of which is a complex structure with an outer branch, apical and sub-apical spine, two long projections, one finger-like and the other tipped, a blade, bow without apophysis, inner branch, distal lobe, and accessory lobe (Figure 9). The endopod is smaller and has two processes, each bearing a seta. Both setae are unequal in length and size (Figure 9E). The development of the third pair of pleopods is present in adult males, whereas in juvenile males they are usually reduced or semi-developed (Figure 10A).

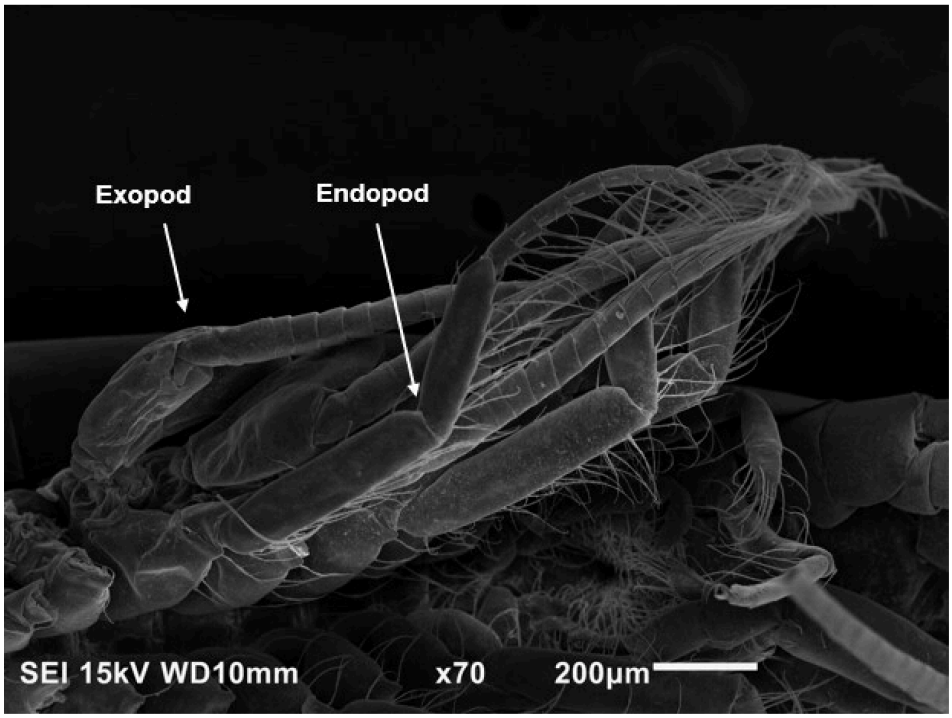

Figure 6. Pereiopods in ventral view, taken under a scanning electron microscope.

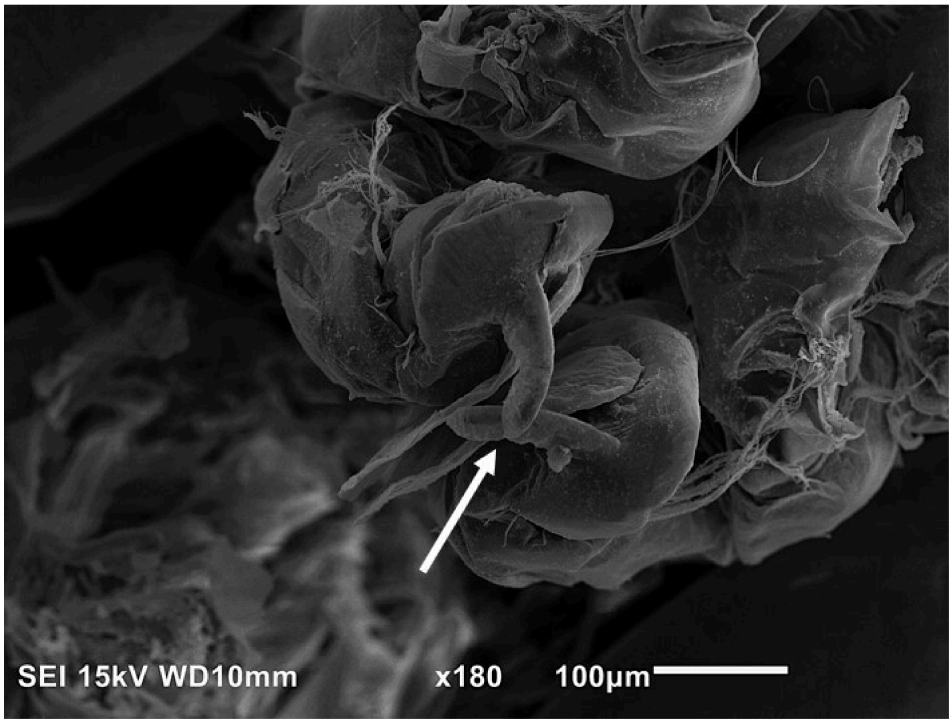

Figure 7. Scanning electron microscope photograph in ventral view where the arrow shows a pair of short penises in the males.

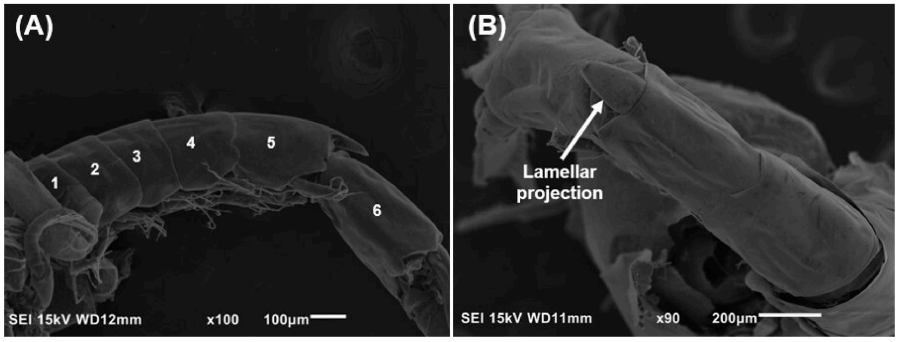

Figure 8. (**A**) Abdominal segments in lateral view (SEM); (**B**) lamellar projection of the fifth abdominal segment in dorsal view (SEM).

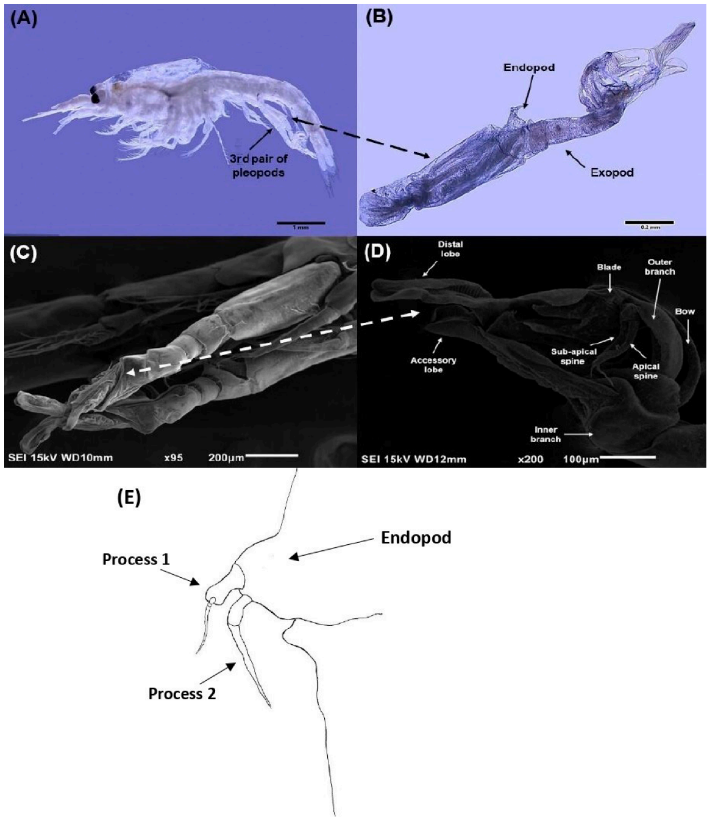

Figure 9. (**A**) Third pleopod in lateral view (MO). (**B**) Endopod of the third pleopod in lateral view (MO). (**C**) Pair of pleopods in ventral view under a scanning electron microscope. (**D**) Terminal portion of the exopod in lateral view (SEM). (**E**) Detail of endopod of third pleopod.

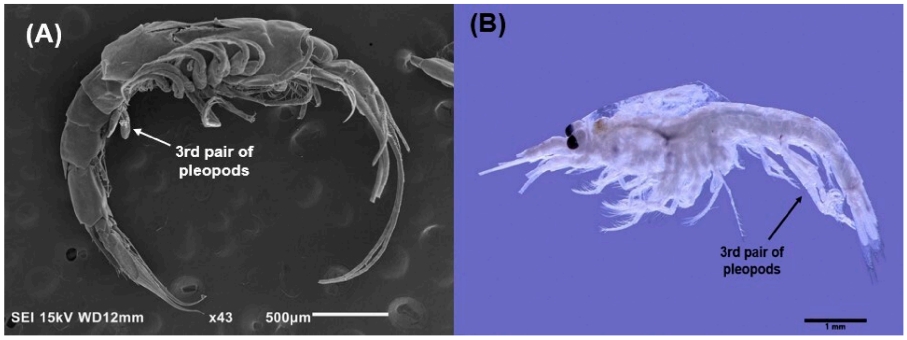

Figure 10. (**A**) Juvenile male mysid in lateral view, photographed under SEM. (**B**) Adult male mysid under light microscope. Collected from Lake Bacalar.

The telson has a cleft at the rear with 18 spines, while each side of the external margin has seven spines (not counting those in the cleft), four of which are uniform in size and two distal ones which are more pronounced (Figure 11).

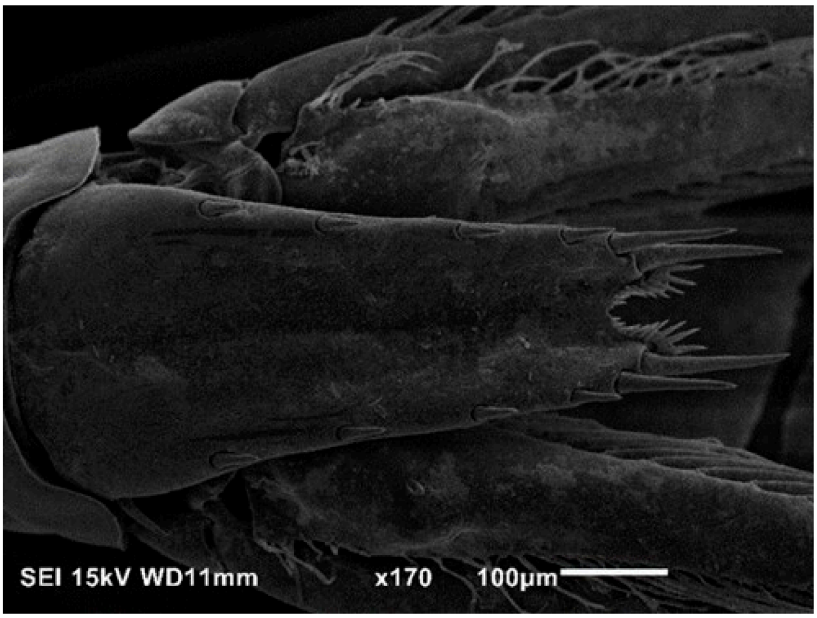

Figure 11. Telson taken in SEM in dorsal view.

The uropod is biramous. A statocyst (balance organ) is present in the base of each endopod (Figure 12A). It bears four spines on the left lateral margin and 35 to 40 setae distributed along the entire margin. The exopod bears 22 spines on the lateral margin and 7–10 setae on the apical part of the margin (Figure 12B).

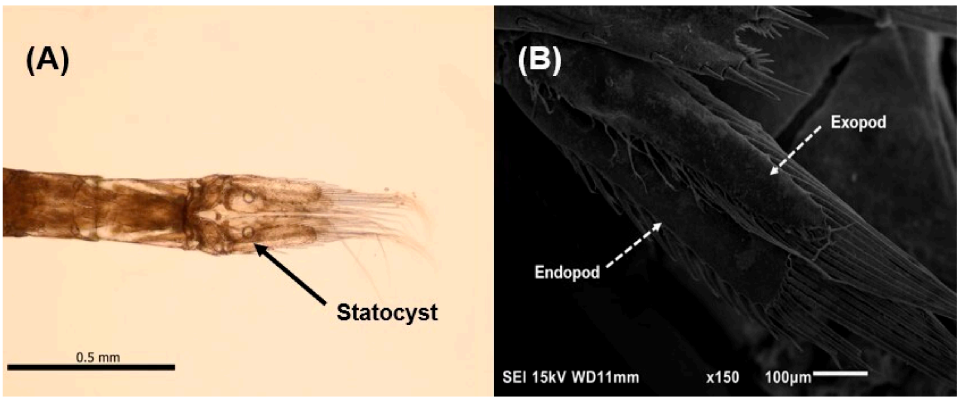

Figure 12. (**A**) Ventral view photograph of the uropod as observed under a bright-field optical microscope (20×). (**B**) Dorsal view of the endopod and exopod of the uropod (SEM).

Female: Similar to males, except for the following characteristics. Females are larger and have a brood pouch that extends from the posterior margin of the thorax to the anterior margin of the abdomen (Figure 13). The pouch is supported by a pair of pleural plates protruding from the first abdominal segment.

The pleopods are reduced relative to those of males; both the exopod and endopod are unisegmented, with the fifth pleopod pair being the most pronounced. The exopod shorter and bears two elongated, pointed, stiletto-shaped extensions, whereas the endopod bears six long setae on its distal portion (Figure 14).

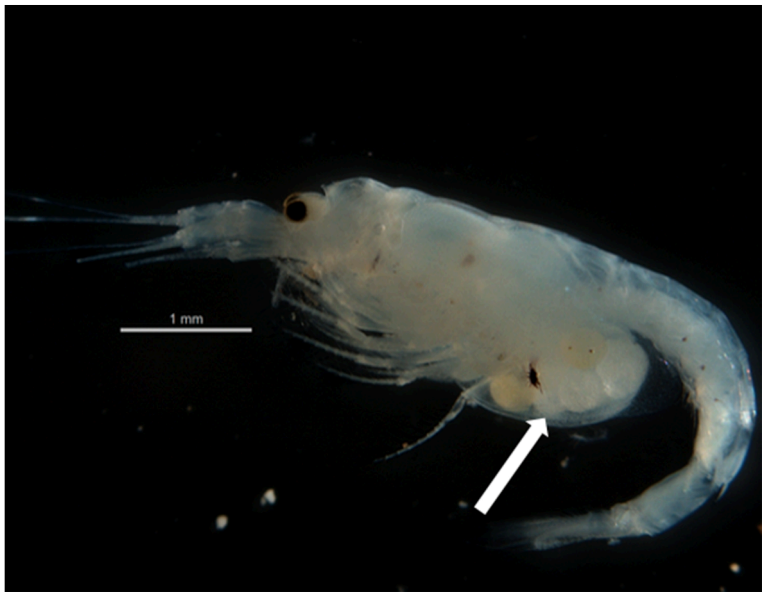

Figure 13. Habitus of female mysid where the arrow shows the marsupium with the embryos. Collected on 18 April 2015, in Bacalar, Quintana Roo (Photography by Martha Valdez Moreno).

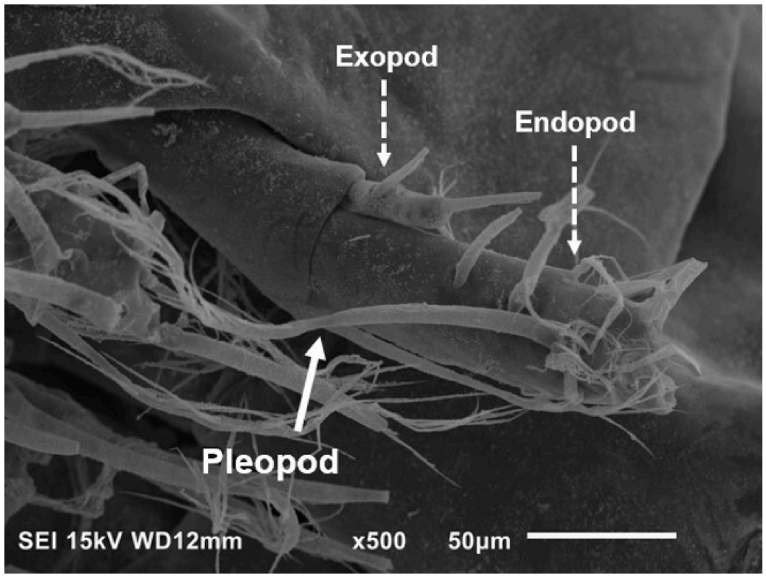

Figure 14. Left pleopod 3 of female mysid under SEM.

3.2. Morphological Comparison

The differences between the closest genera *Chlamydopleon* Ortmann, 1893, *Coifmanniella* Heard & Price, 2006, and the specimens from Lake Bacalar and Chetumal Bay are summarized in Table 1. The specimens from the two latter localities are identical.

Table 1. Comparative table of morphological differences between the *Mayamysis* n. gen., *Chlamydopleon* Ortmann, 1893 and *Coifmanniella* Heard & Price, 2006.

Characteristic	***Mayamysis* n. gen.**	***Chlamydopleon* Ortmann, 1893**	***Coifmanniella* Heard & Price, 2006**
Size	Small: males 5.8 ± 0.3 mm (N = 3); females 6.1 mm ± 0.8 mm (N = 10)	Big: males 7.7 mm (N = 1) (Yum Balam, Mexico); females 9.93 ± 0.5 mm (N = 3) (Wittmann (2009))	Big: males 8.93 ± 0.3 mm (N = 4); females 9.92 mm (N = 2)
Eyes	Normal eyes, oval-shaped and small	Robust	Robust

Table 1. *Cont.*

Characteristic	*Mayamysis* n. gen.	*Chlamydopleon* Ortmann, 1893	*Coifmanniella* Heard & Price, 2006
Antenna 1	Two small proximal spines on the second segment, and a smaller spine on the third segment of antenna 1	Two small spines on the second and third segments of antenna 1	Two spines, one big proximal and one short distal
Shell	The shell covers completely all thoracic segments. Posterodorsal margin without lobes.	The shell is shorter and does not cover completely the thoracic segments. The posterodorsal margin of the carapace exhibits a median lobe and two submedian lobes that extend caudally.	Does not cover completely the thoracic segments and posterodorsal margin with a concave mid-dorsal lobe and thin medial lobes, either reflected or not.
Rostrum	Absent	Rostrum with a long, smooth, anteriorly directed spiniform projection	Absent
Abdomen	Fifth segment with a dorsal lamellar projection	Fifth segment with a dorsal lamellar projection	Lamellar projection absent in fifth segment
Pleopods	In adult male more pronounced	Less pronounced	In adult male pronounced
3rd pair of pleopods (male)			
Exopod	Multisegmented, without setae, the last segment has the following structures: • Bow • Outer branch • Apical spine • Subapical spine • Blade • Inner branch • Medium-sized laminated accessory lobe • Distal laminated lobe	Multisegmented, without setae, the presence of the following structures distinguishes it from the genus *Mayamysis*: • Inner stylet • Outer stylet • Small accessory lobe, not laminated • Distal lobe not laminated	Multisegmented, the presence of the following structures distinguishes it from the genus *Mayamysis*: • Bow with an associated apophysis • Inner stylet • Outer stylet • Small accessory lobe (not laminated and a larger) • Laminated distal lobe
Endopod	A lobe, with two processes each with one seta of different size and distal process without spines	With two processes, each with one seta. Both seta of the same size and the distal process is covered with smaller spines	Triangle shaped, with two processes, each sit one seta. A medial small seta
Telson	With spines on each side of the margin of the telson, the cleft is shallow, lined with smaller spines.	The lateral margins of the telson bear numerous spines; additionally, a well-defined apical cleft is present, furnished with multiple laminae.	With spines on each side of the margin. The cleft varies in depth, ranging from about 10% to over 40% of the total telson length.
Uropod			
Exopod	Outer margin with spines	With spine-like setae on outer margin	
Endopod	Outer margin with short, thin setae, without spines near statocyst.	With a series of spines, mostly medium to large in size, along the inner margin; there are no smaller spines near the statocyst	Two spines and four small in medial position near statocyst. Other two more distal. Presence of small spinose setae located distal to the statocyst

3.3. Molecular Analysis

In total, we obtained sequences from 63 of the 67 processed specimens, representing a 94% success rate. Of these, 51 were previously obtained from Lake Bacalar and 12 were recently obtained from Chetumal Bay, representing a 100% success rate for the latter. Two sequences were discarded after quality evaluation using BOLD (v5.0) tools. The remaining 61 sequences were included in all subsequent analyses.

The identification trees obtained from BOLD and ASAP included all mysid sequences available worldwide, representing 207 and 175 Molecular Taxonomic Units (MOTUs), respectively. ASAP was more conservative than the Barcode Index Number (BIN), as the selected partition corresponded to the lowest score rank (7.5), resulting in a finer splitting of groups (Figures S1 and S2). Nevertheless, both approaches indicate that the Bacalar mysid sequences are almost identical to those from Chetumal Bay and were therefore

placed within the same clade (61 specimens, corresponding to a single BIN: ACX6091) (Figures S1 and S2). These results suggest that the specimens from this region represent a unique lineage distinct from all other sequenced mysids worldwide.

3.4. COI Comparison with Mexican Material

The analysis performed using only mysid records from Mexico, under a maximum likelihood framework and bPTP, yielded consistent results. The bPTP partitions supporting the clade of *Mayamysis bacalarensis* showed a support value of 0.98, while the support for the closest species, *Chlamydopleon dissimile*, was also 0.98. These results were congruent with the BIN assignments. We present here the resulting tree after ML, and consisted of 15 MOTUs, each represented by associated data such as its BIN, collection site, and the finest possible taxonomic assignment (Figure 15). Seven BINs correspond to marine mysids and are indicated in green (Figure 15). These specimens were collected in Yum Balam and the Caribbean Sea in the state of Quintana Roo, as well as offshore of Isla Partida in the Sea of Cortés, Baja California state (28.894 N, 113.043 W).

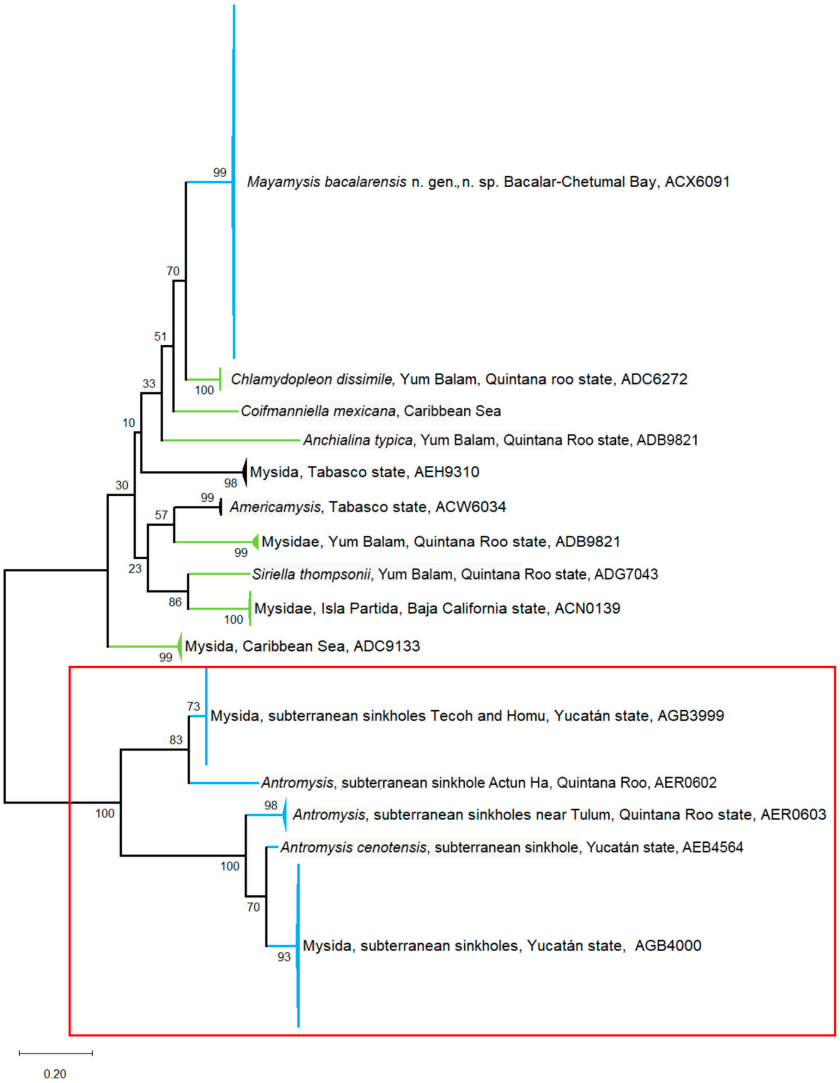

Figure 15. Maximum likelihood tree of mysids sequenced in Mexico. Blue branches: freshwater mysids; green branch: marine water; brown branch: brackish water mysids. The numbers following each locality correspond to the BINs defining each MOTU. Each clade terminates in a triangle, the length of which indicates the number of specimens collected and sequenced, while the width reflects haplotype divergence. A shorter sequence (450 bp) of *Coifmanniella mexicana* was included in the tree solely for comparative purposes, given its morphological proximity to *Mayamysis*.

In contrast, two BINs originate from the lagoon and canal system of San Pedrito, Tabasco, which is considered a complex brackish water environment (brown branch, Figure 15). This system has multiple hydrological connections, including canals and rivers that discharge into the Gulf of Mexico, and therefore warrants further detailed study.

Finally, five clades marked in blue (within the red box in Figure 15) correspond to freshwater systems, specifically cenotes of the Yucatán Peninsula. These include the cenotes named Pol Box, Tecoh, Homun, Cuzamá, and Sacalum in the state of Yucatán, as well as two others, Actun Ha and Chac Mool in the state of Quintana Roo.

The clade that corresponds to *Mayamysis bacalarensis* (BIN ACX6091), includes the sequenced specimens collected from Lake Bacalar and Chetumal Bay, the latter considered a brackish water estuary [3]. The triangle representing this branch is the longest in the tree, indicating that it represents the number of sequenced specimens (49 sequences from Bacalar and 12 from Chetumal Bay). Furthermore, it is narrow, reflecting low haplotype divergence, with nearly identical sequences in both locations.

In Table 2, it is shown the evolutionary divergence of the specimens used to construct the maximum likelihood tree.

Table 2. Estimates of evolutionary divergence over sequence pairs between groups for all mysids sequenced in Mexico (132 in total). The number of base substitutions per site, averaged over all sequence pairs between groups, is shown (orange). Standard error estimates are shown above the diagonal (green) and were obtained by a bootstrap procedure with 500 replicates. The analysis was conducted using the Kimura 2-parameter model. After each name, the genetic sequence divergences and their respective BINs for mysids from Mexico are included. *Coifmanniella mexicana* was excluded because only a single short sequence was available (450 bp after alignment). * Indicates freshwater mysids from cenotes of the Yucatán Peninsula.

	Taxa and BIN	1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	*Mayamysis bacalarensis*_ ACX6091		0.03	0.03	0.03	0.02	0.02	0.03	0.04	0.03	0.02	0.04	0.05	0.04	0.05
2	Mysida_ ACN0139	0.28		0.03	0.03	0.03	0.02	0.04	0.04	0.03	0.03	0.05	0.05	0.04	0.05
3	Mysidae_ ADB9821	0.25	0.29		0.03	0.02	0.03	0.04	0.04	0.03	0.02	0.04	0.04	0.04	0.04
4	Mysida_ ADC9133	0.28	0.29	0.29		0.03	0.03	0.04	0.04	0.03	0.03	0.04	0.04	0.04	0.04
5	*Chlamydopleon dissimile*_ ADC6272	0.15	0.25	0.23	0.27		0.03	0.03	0.04	0.03	0.02	0.04	0.04	0.04	0.05
6	*Siriella_thompsonii* ADG7043	0.24	0.22	0.27	0.29	0.25		0.03	0.04	0.03	0.03	0.04	0.04	0.04	0.04
7	*Anchialina_typica* ADG7073	0.31	0.37	0.37	0.36	0.29	0.36		0.04	0.03	0.03	0.04	0.05	0.04	0.05
8	*Antromysis cenotensis*_ AEB4564 *	0.44	0.51	0.45	0.47	0.43	0.47	0.49		0.04	0.04	0.02	0.04	0.02	0.04
9	Mysida_ AEH9310	0.32	0.30	0.27	0.31	0.27	0.30	0.36	0.50		0.03	0.05	0.04	0.04	0.05
10	*Americamysis*_ ACW6034	0.22	0.25	0.23	0.28	0.21	0.23	0.31	0.42	0.28		0.04	0.05	0.04	0.04
11	*Antromysis*_ AER0603 *	0.46	0.52	0.49	0.47	0.45	0.46	0.49	0.15	0.54	0.45		0.04	0.02	0.03
12	*Antromysis*_ AER0602 *	0.50	0.51	0.50	0.44	0.51	0.46	0.54	0.44	0.47	0.51	0.43		0.04	0.02
13	Mysida_ AGB4000 *	0.42	0.48	0.47	0.46	0.44	0.48	0.49	0.11	0.49	0.42	0.18	0.41		0.04
14	Mysida_ AGB3999 *	0.49	0.50	0.48	0.46	0.50	0.45	0.54	0.37	0.52	0.48	0.37	0.19	0.38	

The following COI sequence from Bacalar mysids are compared with those of *Chlamydopleon dissimile* (Coifmann, 1937) collected in Yum Balam and can be considered as an additional character defining the species. The genetic divergence (p-distance) between these two is 15.0%:

```
                                                                    1-60
MysidaBacalar     GACTTTATATTTTATTTTTGGTGCTTGAGCAGGAATAGTAGGTTCTTCTTTAAGGGTTTT
Chlamydopleon     A**T****************A*********************************A**AC*
                                                                   61-120
MysidaBacalar     AATTCGTTTAGAATTAGGTCAACCTGGTAGTTTAATTGGGGATGATCAAATTTATAATGT
Chlamydopleon     ******A**************G**T**A**TT*G*****A**T**C**G**********
                                                                  121-180
MysidaBacalar     TATTGTGACAGCACATGCTTTTGTAATAATTTTTTTTATAGTTATGCCTATTATAATTGG
Chlamydopleon     ******A****************T******************G*****T**G*****
                                                                  181-240
MysidaBacalar     AGGATTTGGAAATTGGTTAGTACCCTTAATATTAGGTGCTCCTGATATAGCTTTCCCTCG
Chlamydopleon     G**T*****T**T**AC*A*****T**********G*****************T**A**
                                                                  241-300
MysidaBacalar     TATAAACAATATAAGATTTTGACTTTTACCTCCTGCTTTAACTTTATTGTTAAGAAGGGG
Chlamydopleon     ******T**************T*A**A******T*A*****A***C*T********A**
                                                                  301-360
MysidaBacalar     TTTAGTTGAGAGAGGAGTAGGAACAGGTTGAACTGTGTATCCTCCTTTAGCTGCTGGAAT
Chlamydopleon     *********A**A**G**T**G*****G********T***************AGA**G**
                                                                  361-420
MysidaBacalar     TGCTCATGTGGGAGCTTCTGTAGATTTAGGTATTTTTTCTTTACATTTAGCGGGAGCTTC
Chlamydopleon     T**A*****T*****A*****T***C*T**********************A**A*****
                                                                  421-480
MysidaBacalar     TTCAATTTTAGGTGCTGTTAATTTTATTTCTACTGTAATTAATATGCGAAGACCTGGTAT
Chlamydopleon     ***T**TC**********G***********A*****T********A*****G*****A**
                                                                  481-540
MysidaBacalar     AACTTGAGACCGACTACCTCTTTTTGTATGGTCAGTTTTTATTACTGCAGTACTTTTATT
Chlamydopleon     *********T**TT*******A*****T**GG*T**C**************T******C*
                                                                  541-600
MysidaBacalar     ATTGTCTTTGCCTGTTTTAGCGGGAGCTATTACTATGTTATTAACTGATCGAAACTTAAA
Chlamydopleon     TC*T*****A*****A*****A**************AC*T*****G*****T**T*****
                                                                  601-658
MysidaBacalar       TACGACATTTTTTGACCCAGTAGGAGGCGGAGATCCTATTTTATATCAACATCTATTT
Chlamydopleon       ***T**T********T**TAC***T**T**T**T**C***************T*****
```

Finally, after the analysis of haplotype diversity in *Mayamysis bacalarensis* n. sp., n. gen., we found 36 haplotypes with an Hd = 0.86, indicating a possible expansion from a marine environment. The haplotype network in Figure 16 indicates not a detectable geographic population structure between Bacalar and Chetumal Bay, indicating a continuous connectivity between both systems.

Tajima's D was significantly negative (D = −2.50, $p < 0.01$), indicating an excess of low-frequency polymorphisms. Given the low nucleotide diversity (π = 0.00301) and higher θ values (θ = 7.72 per sequence; θ = 0.01247 per site), these results suggest a recent demographic expansion, potentially associated with increased connectivity among populations.

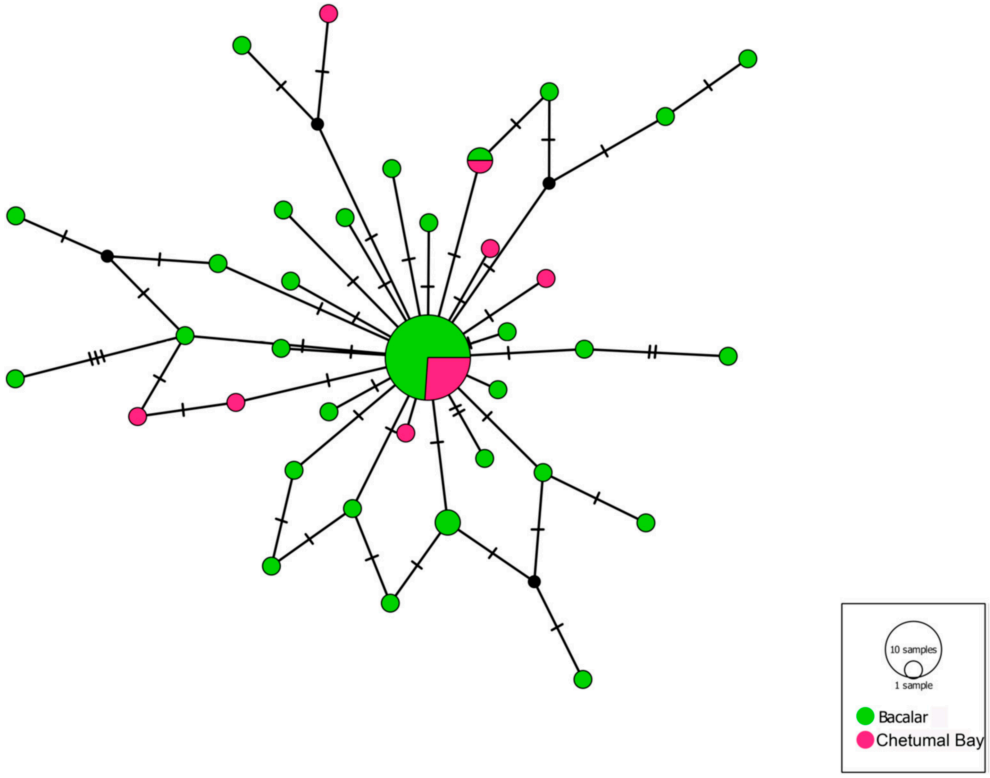

Figure 16. Haplotype network of *Mayamysis bacalarensis* n. gen., n. sp. Small lines on branches represent mutational steps. The size of circles is proportional to haplotype frequency. Color indicates haplotype location (Lake Bacalar and Chetumal Bay).

4. Discussion

The mysids found in Lake Bacalar and Chetumal Bay represent a notable new record. Given the low nutrient concentrations and hyper carbonate waters of the lake together with the low salinity in the Chetumal Bay, zooplankton diversity would be expected to be relatively low. However, the presence of this group suggests that a considerable proportion of the zooplanktonic fauna in these water bodies remains unknown, as previously suggested for Lake Bacalar by Elías-Gutiérrez et al. [10].

Mysids had not previously been collected in Lake Bacalar, likely due to water disturbance caused by the use of trawl nets, a traditional sampling method that may allow individuals to evade capture [37,38].

The mysids from Lake Bacalar and Chetumal Bay exhibit a statocyst and reduced pleopods, indicating that they belong to the order Mysida and the family Mysidae. According to Fernández [20], the absence of a statocyst in the uropod and the presence of highly developed pleopods are diagnostic characters of the order Lophogastrida. The third order, Stygiomysida, includes troglobitic species characterized by the absence of eyes [19], which is not the case for the specimens from Lake Bacalar and Chetumal Bay.

The mysids of Chetumal Bay were previously recorded as *Bowmaniella floridana* Holmquist, 1975 [21]. However, according to the World Register of Marine Species (WoRMS, https://www.marinespecies.org/index.php), this name is currently considered a junior synonym of *Chlamydopleon dissimile* (Coifmann 1937). The closest sequences to the Bacalar and Chetumal Bay specimens correspond to *Chlamydopleon dissimile* from Yum Balam (North of Yucatán Peninsula), with an average divergence of 15% (Table 2). This level of genetic divergence clearly suggests that the Bacalar–Chetumal mysids represent a distinct lineage.

When reviewing the genus *Chlamydopleon* Ortmann, 1893, Wittmann [39] noted that it is characterized by pigmented eyes, biramous pleopods in both sexes, a protruding spine on

the fifth abdominal segment, and a cleft telson bearing 15–17 marginal spines. In addition, species of this genus exhibit spines on only one side of the margin of both the endopod and exopod of the uropod.

Based on these diagnostic characters, *Chlamydopleon* represents the genus morphologically closest to the mysids from Lake Bacalar. Specimens from Chetumal Bay were initially identified as *Bowmaniella floridana* Holmquist, 1975, which is currently recognized as *Chlamydopleon dissimile* (Coifmann, 1937). This material was deposited under this latter name in the ECOSUR reference collection.

Among the tribe Gastrosaccini, we propose the establishment of the new genus *Mayamysis* on the basis of the following features:

Morphological differences.

The combination of characters distinguishes this taxon from the six genera currently recognized within the tribe Gastrosaccini. In Table 1, we summarize these characters and compare them with the two closest genera, *Chlamydopleon* and *Coifmanniella*, both recorded from the Mexican Caribbean Sea. The remaining four genera *Eurobowmaniella* Murano, 1995, *Gastrosaccus* Norman, 1868, *Haplostylus* Kossmann, 1880, and *Iiella* Băcescu, 1968 also differ in their combination of characters.

Eurobowmaniella has a rostrum with a pseudorostral process located anterior to the true rostrum. In males, the terminal segment of the exopod of the third pair of pleopods bears spines, and the endopod is multisegmented and the second to fifth female pleopods are uniramous [40]. *Gastrosaccus*, distributed in warm waters of the western Pacific, Indian Ocean, Mediterranean Sea, and eastern Atlantic, also has a rostrum. The third male pleopod has a simple styliform exopod bearing terminal spines [39].

Haplostylus presents a rostrum; in males, the endopod of the third pair of pleopods is small, unsegmented, and rudimentary, whereas the exopod is elongate and slender, extending beyond the middle of the telson, with the terminal segment bearing claws [41].

Iiella is distributed in the tropical and temperate western Pacific. It has a rostrum; in males, the endopod of the third pair of pleopods is multiarticulated, and the exopod is markedly elongate, reaching the base of the telson, with the terminal segment bearing apical claws. In females, the pleopods are uniramous [42].

Geographical distribution.

Mayamysis n. gen. has a restricted distribution in Lake Bacalar and Chetumal Bay. Despite recent extensive sampling across the Yucatán Peninsula and the Caribbean Sea, it has not been recorded from any other locality [43]. All other members of the tribe are marine.

Molecular evidence.

Although the COI marker is widely used for species-level identification, it can also provide phylogenetic signal at higher taxonomic levels, such as genus, in arthropods [44]. In the global comparison (Figure S1), the sequenced representatives of *Gastrosaccus*, *Haplostylus*, and *Iiella* form well-defined clades that are clearly separated from the clade formed by *Mayamysis* n. gen. However, further studies using more conserved markers are required to confirm the monophyly of Gastrosaccini and the phylogenetic positions of the different genera within this group.

It is evident that the sequences of freshwater mysids from other parts of the Yucatán Peninsula are not closely related to those from Bacalar and Chetumal Bay (Figure 15; Table 2). Moreover, these differences are also evident at the morphological level, including body size in both sexes (Table 1), which, together with habitat, constitutes stronger evidence supporting the establishment of the new genus.

Although it is now certain that the sequences of the Bacalar organisms are almost identical to those from Chetumal Bay, it was initially expected that they would be closely related to freshwater mysids collected from hypogean ecosystems of the Yucatán Peninsula. However, divergence estimates obtained from the pairwise genetic distances showed values ranging from 42% to 50% (Table 2). Therefore, they do not correspond to the same species or genus. This conclusion is consistent with the taxonomy and morphology of these stygobiont organisms, which belong to the subfamily Mysinae (Haworth, 1825) and the genus *Antromysis* (Creaser, 1936). These taxa are characterized by their distribution in wells and caves and the absence of ommatidia [21].

The average divergence with the closest sequence was 15%, corresponding to *Chlamydopleon dissimile* (Table 2), whereas the second closest divergence (19%) corresponded to *Coifmanniella mexicana*. This latter value should be interpreted with caution, as it is based on a single, relatively short sequence (450 bp). Both species are marine and have been previously recorded in Yum Balam, located in the northern part of the Yucatán Peninsula.

Another apparently close sequence match corresponds to *Boreomysis urospina* Daneliya, 2023, with 19% divergence. We did not include this species in our ML comparison because it is represented by only two sequences in GenBank (OQ699903 and OQ699904), with no chromatograms available, and because this species has been recorded only from the Tasman Sea at a depth of approximately 1000 m. Moreover, the morphology of these animals is completely different from that of *Mayamysis bacalarensis* n. gen., n. sp. [45].

Haplotype analyses of *Mayamysis bacalarensis* n. gen., n. sp. suggest a possible ancient marine expansion associated with multiple marine transgressions in the southern Yucatán Peninsula. The emergence of the Yucatán Peninsula is considered geologically recent [46]; however, its southern region has an older tectonic history [47]. Repeated marine transgressions and regressions throughout geological time likely promoted speciation by increasing ecological niche diversification.

Thus, the unique mysid recorded in this study may represent a lineage shaped by post-emergence processes affecting the terrestrial portion of the peninsula. Despite the presence of multiple haplotypes, there is evidence of continuous gene flow within a single population between Chetumal Bay and Lake Bacalar, as indicated by the haplotype network and Tajima's neutrality test. *Mayamysis bacalarensis* represents a well-defined group with a restricted distribution.

Final Remarks

Based on the information gathered, it can be inferred that there is a seemingly continuous hydrological connectivity between Lake Bacalar and Chetumal Bay, allowing a dynamic exchange of organisms. Juvenile stages appear to constitute the most mobile component of this exchange, whereas adults were scarce in Chetumal Bay.

We cannot rule out the possibility that these organisms remain undetected in other regional systems such as Muyil or nearby systems, particularly when conventional sampling methods such as trawling are employed. However, limited sampling using light traps in these systems did not reveal the presence of any mysids [43].

Integrative taxonomy has proven to be an effective approach for species identification. Although traditional taxonomy has been successful, it presents inherent limitations in species delimitation, particularly due to the exclusive reliance on morphological characters and the limited knowledge available for some groups. These limitations may lead to synonymies derived from confusion between juvenile and adult stages or from morphological convergence. Therefore, the integration of all possible characters constitutes a fundamental tool for accurately establishing the taxonomic identity of species alongside traditional taxonomy.

5. Conclusions

The restricted distribution of freshwater mysids in the Yucatán Peninsula, previously limited to cenotes and caves in its northern region, makes the record *of M. bacalarensis* n. gen., n. sp. in Lake Bacalar and Chetumal Bay particularly noteworthy. Although its apparent absence from other systems may reflect true rarity, it is also possible that conventional sampling methods have limited their detection elsewhere.

The combination of distinctive morphological characters, geographical distribution and molecular divergence from closely related taxa, particularly *Chlamydopleon*, supports the recognition of *Mayamysis bacalarensis* as a new genus and species. The integration of molecular tools provides additional support for the morphological and distributional evidence, reinforcing the taxonomic decision.

M. bacalarensis n. gen., n. sp. may represent a microendemic species with a yet unknown ecological role. Its occurrence in both Lake Bacalar and Chetumal Bay suggests hydrological connectivity between these systems and highlights its potential value as a bioindicator. Given the oligotrophic condition and exceptional ecological importance of both environments, continued integrative studies of their zooplankton communities are strongly recommended.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/d18050279/s1, Figure S1: Id tree of 2517 sequenced specimens worldwide, belonging to freshwater, marine, and brackish environments. Figure S2: Delimitation of species using ASAP method for worldwide mysids.

Author Contributions: D.E.G.-T. and M.E.-G. contributed equally for this study. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding. M.E.-G. personally funded the fieldwork and PCR costs. María A. Mendoza Becerril from Medusozoa, Mexico supported the sequencing costs of material from Chetumal Bay.

Data Availability Statement: All sequenced mysids (including those of *Mayamysis bacalarensis*) from Mexico are available in the public dataset "DS-MYSID Mysidae from Mexico" in the Barcode of Life Database (BOLD; boldsystems.org), and under the DOI: https://doi.org/10.5883/DS-MYSID. All data presented in this study are publicly available in the BOLD.

Acknowledgments: Alma E. García Morales from the Chetumal laboratory for DNA barcoding developed molecular procedures to obtain the sequences of mysids from Chetumal Bay. Humberto Bahena Basave assisted us with photographs in light microscopy. Wendy N. Reyes Monzón and José A. Cohuo Colli assisted us with field collections in Chetumal Bay. We appreciate all facilities given to us by Paula Dzul Canul and Victor Manuel Rojas for collecting specimens from Cocalitos sinkhole. The comments provided by two anonymous reviewers significantly improved the original manuscript.

Conflicts of Interest: The authors declare no conflicts of interest.

References

1. Carrillo, L.; Yescas, M.; Nieto-Oropeza, M.O.; Elías-Gutiérrez, M.; Alcérreca-Huerta, J.C.; Palacios-Hernández, E.; Reyes-Mendoza, O.F. Investigating the Morphometry and Hydrometeorological Variability of a Fragile Tropical Karstic Lake of the Yucatán Peninsula: Bacalar Lagoon. *Hydrology* **2024**, *11*, 68. [CrossRef]
2. Ruiz-Pineda, C.; Suárez-Morales, E.; Gasca, R. Copépodos planctónicos de la Bahía de Chetumal, Caribe Mexicano: Variaciones estacionales durante un ciclo anual. *Rev. Biol. Mar. Oceanogr.* **2016**, *51*, 301–316. [CrossRef]
3. Carrillo, L.; Palacios-Hernandez, E.; Yescas, M.; Ramirez-Manguilar, A.M. Spatial and Seasonal Patterns of Salinity in a Large and Shallow Tropical Estuary of the Western Caribbean. *Estuaries Coasts* **2009**, *32*, 906–916. [CrossRef]
4. Gischler, E.; Gibson, M.A.; Oschmann, W. Giant Holocene freshwater microbialites, Laguna Bacalar, Quintana Roo, Mexico. *Sedimentology* **2008**, *55*, 1293–1309. [CrossRef]

5. Matzuk, R. Water Chemistry and Lake Dynamics of Laguna Bacalar, Quintana Roo, Mexico. Master's Thesis, University of Wisconsin, Madison, WI, USA, 2020.
6. Perry, E.; Velazquez-Oliman, G.; Marin, L. The hydrogeochemistry of the karst aquifer system of the northern Yucatan Peninsula, Mexico. *Int. Geol. Rev.* **2002**, *44*, 191–221. [CrossRef]
7. Elías-Gutiérrez, M.; Montes-Ortiz, L. Present day kwnoledge on diversity of freshwater zooplancton (invertebrates) of the Yucatan Peninsula, using integrated taxonomy. *Teor. Prax.* **2018**, *14*, 31–48.
8. Montes-Ortiz, L.; Elías-Gutiérrez, M.; Ramirez-Sanchez, M.M. Checklist of Arrenurids (Acari: Hydrachnidia: Arrenuridae) of Mexico, with New Records from the Yucatan Peninsula, and the Description of Five New Species of the Subgenera *Megaluracarus* and *Dadayella*. *Diversity* **2022**, *14*, 276. [CrossRef]
9. Dayrat, B. Towards integrative taxonomy. *Biol. J. Linn. Soc.* **2005**, *85*, 407–415. [CrossRef]
10. Elías-Gutiérrez, M.; Valdez-Moreno, M.; Topan, J.; Young, M.R.; Cohuo-Colli, J.A. Improved protocols to accelerate the assembly of DNA barcode reference libraries for freshwater zooplankton. *Ecol. Evol.* **2018**, *8*, 3002–3018. [CrossRef]
11. Price, W.W. Mysida. In *Thor and Covich's Freshwater Invertebrates*; Thorp, J.H., Rogers, D., Eds.; Elsevier: London, UK, 2015; Volume V, pp. 972–981.
12. Hebert, P.D.N.; Ratnasingham, S.; DeWaard, J.R. Barcoding animal life: Cytochrome c oxidase subunit 1 divergences among closely related species. *Proc. R. Soc. Lond. Ser. B Biol. Sci.* **2003**, *270*, S96–S99.
13. Zhang, H.; Bu, W. Exploring Large-Scale Patterns of Genetic Variation in the COI Gene among Insecta: Implications for DNA Barcoding and Threshold-Based Species Delimitation Studies. *Insects* **2022**, *13*, 425. [CrossRef] [PubMed]
14. Davies, B.R. A trap for capturing planktonic chironomid larvae. *Freshw. Biol.* **1976**, *6*, 373–380. [CrossRef]
15. Gartz, R.G.; Miller, L.W.; Fujimura, R.W.; Smith, P.E. Measurement of larval striped bass (*Morone saxatilis*) net avoidance using evasion radius estimation to improve estimates of abundance and mortality. *J. Plankton Res.* **1999**, *21*, 561–580. [CrossRef]
16. Liévano-Beltran, L.A.; Simoes, N. Updated Distribution of the Mysid *Antromysis cenotensis* (Crustacea: Peracarida), a Protected Key Species in Yucatan Peninsula Cenotes. *Diversity* **2021**, *13*, 154. [CrossRef]
17. Neves, A.; Sequeira, V.; Paiva, R.B.; Vieira, A.R.; Gordo, L.S. Feeding habits of the bluemouth, *Helicolenus dactylopterus dactylopterus* (Delaroche, 1809) (Pisces: Sebastidae) in the Portuguese coast. *Helgol. Mar. Res.* **2012**, *66*, 189–197. [CrossRef]
18. Corse, E.; Valladares, S.; Planas, M.; Chamorro, A.; Pintado, J. Analysis of the diet of the long-snouted seahorse *Hippocampus guttulatus* by 18SrDNA amplification of prey in faeces. *Aquac. Nutr.* **2015**, *21*, 528–540. [CrossRef]
19. Ortiz, M.; Lalana, R. *Los Misidáceos (Lophogastrida, Stygiomysida y Mysida) de Cuba: Historia, Claves Dicotómicas, Figuras y Bibliografía*; Editorial Academia: Havana, Cuba, 2017.
20. Fernández, C.V. *Órdenes Mysida y Lophogastrida*; Universidad Nacional Autónoma de México: Ciudad de México, Mexico, 2015; Volume III.
21. Castellanos-Osorio, I. Estudios de zooplancton: Logros y retos. In *El Sistema Ecológico de la Bahía de Chetumal/Corozal: Costa Occidental del Mar Caribe*; Espinoza-Avalos, J., Islebe, G., Hernández Arana, H., Eds.; El Colegio de la Frontera Sur (ECOSUR): Ciudad de México, Mexico, 2009; pp. 61–67.
22. Johnson, W.S.; Allen, D.M. *Zooplankton of the Atlantic and Gulf Coasts: A Guide to Their Identification and Ecology*, 2nd ed.; Johns Hopkins University Press: Baltimore, MD, USA, 2012; p. 469.
23. Ortiz, M.; Winfield, I.; Cházaro-Olvera, S. Updated checklist and illustrated key to mysid (Crustacea, Peracarida) genera from the Intra-American Sea. *Rev. Mex. Biodivers.* **2012**, *83*, 983–1003.
24. Wittmann, K.J.; Ariani, A.P.; Lagardère, J.P. Orders Lophogastrida Boas, 1883, Stygiomysida Tchindonova, 1981, and Mysida Boas, 1883 (also known collectively as Mysidacea). In *Treatise on Zoology—Anatomy, Taxonomy, Biology–The Crustacea*; von Vaupel Klein, M., Charmantier-Daures, M., Schram, F.R., Eds.; Brill: Leiden, The Netherlands, 2014; Volume 4, pp. 189–396.
25. Gasca-Serrano, R.; Castellanos-Osorio, I.; Suárez-Morales, E. Análisis preliminar y perspectivas de una comunidad ecológica fronteriza: El zooplancton de la bahía de Chetumal. *Rev. Biol. Trop.* **1994**, *42*, 89–103.
26. Ivanova, N.V.; DeWaard, J.R.; Hebert, P.D.N. An inexpensive, automation-friendly protocol for recovering high-quality DNA. *Mol. Ecol. Notes* **2006**, *6*, 998–1002.
27. Prosser, S.; Martínez-Arce, A.; Elías-Gutiérrez, M. A new set of primers and some methodological improvements for COI amplification in freshwater microcrustaceans. *Mol. Ecol. Resour.* **2013**, *13*, 1151–1155.
28. Ratnasingham, S.; Hebert, P.D.N. BOLD: The Barcode of Life Data System (www.barcodinglife.org). *Mol. Ecol. Notes* **2007**, *7*, 355–364. [CrossRef] [PubMed]
29. Ratnasingham, S.; Hebert, P.D.N. A DNA-Based Registry for All Animal Species: The Barcode Index Number (BIN) System. *PLoS ONE* **2013**, *8*, e66213.
30. Kumar, S.; Stecher, G.; Suleski, M.; Sanderford, M.; Sharma, S.; Tamura, K. MEGA12: Molecular Evolutionary Genetic Analysis Version 12 for Adaptive and Green Computing. *Mol. Biol. Evol.* **2024**, *41*, msae263. [CrossRef]

31. Ronquist, F.; Teslenko, M.; van der Mark, P.; Ayres, D.L.; Darling, A.; Hohna, S.; Larget, B.; Liu, L.; Suchard, M.A.; Huelsenbeck, J.P. MrBayes 3.2: Efficient Bayesian Phylogenetic Inference and Model Choice Across a Large Model Space. *Syst. Biol.* **2012**, *61*, 539–542. [CrossRef]
32. Zhang, J.; Kapli, P.; Pavlidis, P.; Stamatakis, A. A general species delimitation method with applications to phylogenetic placements. *Bioinformatics* **2013**, *29*, 2869–2876. [CrossRef] [PubMed]
33. Kimura, M. A simple method of estimating evolutionary rate of base substitutions through comparative studies. *J. Mol. Evol.* **1980**, *16*, 111–120. [CrossRef]
34. Puillandre, N.; Brouillet, S.; Achaz, G. ASAP: Assemble species by automatic partitioning. *Mol. Ecol. Resour.* **2021**, *21*, 609–620. [CrossRef] [PubMed]
35. Rozas, J.; Ferrer-Mata, A.; Sánchez-DelBarrio, J.C.; Guirao-Rico, S.; Librado, P.; Ramos-Onsins, S.E.; Sánchez-Gracia, A. DnaSP 6: DNA Sequence Polymorphism Analysis of Large Data Sets. *Mol. Biol. Evol.* **2017**, *34*, 3299–3302. [CrossRef]
36. Leigh, J.W.; Bryant, D. POPART: Full-feature software for haplotype network construction. *Methods Ecol. Evol.* **2015**, *6*, 1110–1116. [CrossRef]
37. Gal, G.; Rudstam, L.G.; Greene, C.H. Acoustic characterization of *Mysis relicta*. *Limnol. Oceanogr.* **1999**, *44*, 371–381. [CrossRef]
38. Stehle, M.; Dos Santos, A.; Queiroga, H. Comparison of zooplankton sampling performance of Longhurst-Hardy Plankton Recorder and Bongo nets. *J. Plankton Res.* **2007**, *29*, 169–177. [CrossRef]
39. Wittmann, K.J. Revalidation of *Chlamydopleon aculeatum* Ortmann, 1893, and its consequences for the taxonomy of Gastrosaccinae (Crustacea: Mysida: Mysidae) endemic to coastal waters of America. *Zootaxa* **2009**, *2115*, 21–33. [CrossRef]
40. Murano, M. *Eurobowmaniella phuketensis* n. gen., n. sp. (Crustacea: Mysidacea) from the Indian coast of Thailand. *Phuket Mar. Biol. Cent. Res. Bull.* **1995**, *60*, 21–28.
41. Panampunnayil, S. Descriptions of five new species of *Haplostylus* (Mysidacea-Crustacea) from south west Australia. *J. Plankton Res.* **1997**, *19*, 1205–1233. [CrossRef]
42. Fukuoka, K.; Murano, M. Mysidacea from coastal waters of Iriomote Island, Ryukyu Islands, southwestern Japan, with descriptions of three new species. *J. Crustac. Biol.* **1997**, *17*, 520–537. [CrossRef]
43. Valdez-Moreno, M.; Mendoza-Carranza, M.; Rendon-Hernandez, E.; Alarcon-Chavira, E.; Elías-Gutiérrez, M. DNA Barcodes Applied to a Rapid Baseline Construction in Biodiversity Monitoring for the Conservation of Aquatic Ecosystems in the Sian Ka'an Reserve (Mexico) and Adjacent Areas. *Diversity* **2021**, *13*, 292. [CrossRef]
44. Unnikrishnan, U.A.; Sobczyk, T.; Jose, R.T.; Jose, J. *Capulopsyche keralensis* gen. et sp. nov., a new genus and species of the subfamily Taleporiinae (Lepidoptera: Psychidae) from Kerala, India. *Zootaxa* **2023**, *5258*, 270–284. [CrossRef]
45. Daneliya, M.E. Mysid Subfamily Boreomysinae (Crustacea: Mysida: Mysidae) in the Southeast Australian Deep-sea. *Rec. Aust. Mus.* **2023**, *75*, 87–124. [CrossRef]
46. Torrescano-Valle, N.; Folan, W.J. Physical Settings, Environmental History with an Outlook on Global Change. In *Biodiversity and Conservation of the Yucatán Peninsula*; Islebe, G., Calmé, S., León-Cortés, J.L., Schmook, B., Eds.; Springer International Publishing: Cham, Switzerland, 2015; pp. 9–37.
47. Stanek, K.P.; Maresch, W.V.; Scherer, E.; Krebs, M.; Berndt, J.; Sergeev, S.S.; Rodionov, N.; Pfänder, J.; Hames, W.E. Born in the Pacific and raised in the Caribbean: Construction of the Escambray nappe stack, central Cuba. *Eur. J. Mineral.* **2019**, *31*, 5–34. [CrossRef]