BRILL

Crustaceana 85 (6) 685-712

CRUSTACEANA

MORPHOLOGICAL AND GENETIC VARIABILITY IN *HIPPOLYTE OBLIQUIMANUS* DANA, 1852 (DECAPODA, CARIDEA, HIPPOLYTIDAE) FROM BRAZIL AND THE CARIBBEAN SEA

BY

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ABSTRACT

Hippolyte obliquimanus is a marine shrimp reported from the Caribbean Sea and Brazil. The literature provides indications for morphological variation between populations from those regions and the species has a troubled taxonomic history. The aims of this study were to analyse morphological and genetic variation in the populations of *H. obliquimanus* from Brazil and the Caribbean Sea and to verify if those might support separation of *H. obliquimanus* into two or more species. This hypothesis was tested with the analysis of morphological and genetic data (mitochondrial gene 16S and the barcode region Cytochrome Oxidase I). The material analysed was obtained from samples and from loans of zoological collections. The rostrum as well as pereiopods 3, 4, and 5 were the adult morphological characters that showed variation, but this occurred in samples from both regions, Brazil and the Caribbean Sea. The sequences of the 16S gene were identical among all specimens analysed. There was, however, variation among the sequences of the barcoding gene COI (<2.0%); this divergence separated the specimens into two groups (Brazil versus the Caribbean) and these groups did not share haplotypes. In conclusion, specimens from the regions analysed showed both morphological and genetic variation, but these did not support the separation of *H. obliquimanus* into two or more species.

RESUMEN

Hippolyte obliquimanus es una especie de camarón marino reportada para el Mar del Caribe y Brasil, en la literatura existen reportes que indican la presencia de variación morfológica entre las poblaciones de estas regiones, además esta especie presenta una historia taxonómica problemática. De esta manera el objetivo de este estudio fue analizar la variación morfológica y genética de las poblaciones de *H. obliquimanus* de Brasil y del Mar del Caribe y verificar si estas variaciones soportan la separación de *H. obliquimanus* en dos o más especies. Esto fue realizado por medio

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DOI:10.1163/156854012X643762

de análisis de datos morfológicos y genéticos (genes mitocondriales 16S y Citocromo Oxidasa I). El material analizado fue obtenido por medio de muestreos y prestamos de colecciones zoológicas. Los caracteres morfológicos de los adultos que presentaron variación fueron el rostro y los pereiópodos 3, 4 y 5. Estas variaciones fueron observadas en las dos regiones analizadas (Brasil y Mar del Caribe). Las secuencias del gene 16 S fueran idénticas entre todos los especímenes de *H. obliquimanus* analizados. Hubo variación entre las secuencias del gene COI (<2,0%); esta divergencia separó los especímenes en dos grupos (Brasil y Mar del Caribe) y estos grupos no compartieron haplótipos. Como conclusión, *H. obliquimanus* presentó variación morfológica y genética entre las regiones analizadas (Brasil y Mar del Caribe), pero estas variaciones no soportan la separación de *H. obliquimanus* en dos o más especies.

INTRODUCTION

The genus *Hippolyte* Leach, 1814 comprises 32 species with global distribution except in extremely cold waters, and is considered taxonomically complicated due to the small morphological variation among the species (d'Udekem d'Acoz, 1996, 2007; De Grave et al., 2009). The genus is represented by eight species in America (d'Udekem d'Acoz, 2007): three species occur along the Pacific coast: *H. californiensis* Holmes, 1895, *H. williamsi* Schmitt, 1924, and *H. clarkii* Chace, 1951; and five species are reported from the Atlantic coast: *H. coerulescens* (Fabricius, 1775), *H. pleuracanthus* (Stimpson, 1871), *H. zostericola* (Smith, 1873), *H. obliquimanus* Dana, 1852, and *H. nicholsoni* Chace, 1972.

The only species occurring in Brazil is *Hippolyte obliquimanus*, which is endemic to the western Atlantic coast and restricted to shallow waters of the Caribbean and Brazil, from the States of Ceará to Santa Catarina (Fausto-Filho, 1975; d'Udekem d'Acoz, 1997; Christoffersen, 1998). This species has a troubled taxonomic history: based on specimens from Rio de Janeiro (Brazil), Dana (1852a) described *H. obliquimanus* and *H. exilirostratus*, and based on individuals from West Punt (Curaçao), Schmitt (1924a) described *H. curacaoensis*. Superficially, Chace (1972) suggested that those three species could have been synonyms and that, actually, *H. obliquimanus* and *H. exilirostratus* were male and female, respectively, of the same species. Christoffersen (1980), in a taxonomic revision of the superfamily Alpheoidea from Brazil, considered *H. curacaoensis* the only species of the genus *Hippolyte* occurring in Brazil. However, based on morphological data of adult individuals, d'Udekem d'Acoz (1997) reported that *H. obliquimanus* was a senior synonym of these three species: *H. exilirostratus*, *H. curacaoensis*, and *Virbius gracilis* var. *brasiliensis* Czerniavsky, 1884.

In addition, d'Udekem d'Acoz (1997) also reported some differences of *H. obliquimanus* specimens from the Brazilian and Caribbean regions in relation to the number of spines on the merus of pereiopods 3 and 4: from two to five spines on pereiopod 3 in Brazilian specimens (few specimens with five spines), and five

to six in the Central American specimens; two or three spines on pereiopod 4 of the Brazilian specimens, and four or five in the Central American specimens.

In view of these findings, the aim of this study was to analyse morphological and genetic variation in the populations of *H. obliquimanus* from the Atlantic in Brazil, as well as further away, from the Caribbean Sea, and to verify if those variations might support separation of *H. obliquimanus* into two or more species. Another goal was to re-describe *H. obliquimanus*.

MATERIAL AND METHODS

Some specimens were obtained from field collections, and different sampling methods were used in Brazil and the Caribbean Sea because the substrate was different: in Brazil (states of São Paulo and Santa Catarina), the specimens were obtained from seaweed (*Sargassum* sp.), which was lifted by hand and stored in plastic bags, in a water depth of approximately one metre; in the Caribbean Sea (Costa Rica and Panama, see table I), specimens were obtained with a push net, from the seagrass meadows dominated by turtle grass (*Thalassia testudinum* Banks ex König, 1805). At all sampling locations, after collection of the material the individuals were separated, immediately preserved in ethanol (80%) and deposited in the Crustacean Collection of the Department of Biology (CCDB), Faculty of Philosophy, Sciences and Letters of Ribeirão Preto (FFCLRP), University of São Paulo (USP), Brazil. Complementary specimens for analysis were obtained by donation, visit, or loan from the following crustacean collections: Brazil: CCDB, Museu Nacional do Rio de Janeiro (MNRJ), Museu de Zoologia da Universidade Estadual de Santa Cruz (MZUESC), Museu de Zoologia da Universidade de São Paulo – São Paulo (MZUSP); France: Muséum National d'Histoire Naturelle, Paris (MNHN); Germany: Senckenberg Museum, Frankfurt (SMF); Mexico: Colección Nacional de Crustáceos, Mexico City (CNCR); U.S.A.: University of Louisiana – Lafayette Zoological Collections (ULLZ).

Morphological data

Species identifications were confirmed on the basis of morphological characters given by Chace (1972) and d'Udekem d'Acoz (2007). All measurements, observations, and drawings were performed with a stereomicroscope and a compound microscope equipped with a camera lucida. Sexes were distinguished by the presence or absence of the appendix masculina on the second pleopod (Terossi et al., 2008).

The majority of *Hippolyte* species are 7 to 20 mm in total length, but some European species reach from 30 to 50 mm (Chace, 1972; d'Udekem d'Acoz, 1996,

TABLE I

Specimens (adult females) used in genetic analyses. CCDB: Crustacean Collection of the Department of Biology (FFCLRP-USP), Brazil; CNCR: Coleción Nacional de Crustáceos, Mexico; MZUESC: Museu de Zoologia da Universidade Estadual de Santa Cruz, Brazil; MZUSP: Museu de Zoologia da Universidade de São Paulo, Brazil; ULLZ: University of Louisiana – Lafayette Zoological Collections, U.S.A.

Species	Locality	Catalogue no.	GenBank accession numbers	
			16S	COI
Hippolyte acuta (Stimpson, 1860)	Japan	–	HQ315561	–
Hippolyte bifidirostris (Miers, 1876)	New Zealand	–	EU920927	–
Hippolyte inermis Leach, 1815	Plataria – Greece	CCDB 2783	JF794703	JF794740
	Lacco Ameno d'Ischia – Italy	CCDB 2383	JF794702	JF794739
Hippolyte obliquimanus Dana, 1852	Mahahual, Quintana Roo – Mexico	CNCR 21855	JF794688	–
	Twin Keys – Belize	ULLZ 8869	JF794689	JF794709
	Dorado – Puerto Rico	CCDB 2747	JF794690	–
	Playa Cahuita, Limón – Costa Rica	CCDB 2032	JF794691	JF794704; JF794705; JF794706; JF794707; JF794708
	Bocas del Drago, Bocas del Toro – Panama	CCDB 2580	JF794692	JF794710; JF794711; JF794712; JF794713; JF794714
	Boca Chica, Isla Margarita – Venezuela	CCDB 2381	JF794693	JF794715; JF794716; JF794717
	Porto Seguro, Bahia (BA) – Brazil	MZUESC 903	JF794694	JF794718; JF794719; JF794720; JF794721; JF794722
	Ilha Grande, Rio de Janeiro (RJ) – Brazil	MZUSP 18820	JF794695	JF794723; JF794724; JF794725; JF794726; JF794727
	Ubatuba, São Paulo (SP) – Brazil	CCDB 2147	JF794696	JF794728; JF794729; JF794730; JF794731; JF794732
	Florianópolis, Santa Catarina (SC) – Brazil	CCDB 2033	JF794697	JF794733; JF794734; JF794735; JF794736; JF794737
Hippolyte varians Leach, 1814	Netherlands	CCDB 2781	JF794700	–
	Normandy – France	CCDB 2778	JF794701	–
Hippolyte williamsi Schmitt, 1924	Puerto Aldea, Coquimbo – Chile	CCDB 2382	JF794699	JF794738
Hippolyte zostericola (Smith, 1873)	Playa Privada El Indio, Quintana Roo – Mexico	CCDB 2642	JF794698	–

2007; Espinoza-Fuenzalida et al., 2008), and in most species, females reach a larger size than males (d'Udekem d'Acoz, 1996 for European species; Terossi et al., 2008 for *H. obliquimanus*). The taxonomy of the genus *Hippolyte* is based on morphological data from adult females, because females have more readily useful characters than males (d'Udekem d'Acoz, 1996, 2007). In the present study, all morphological characters were analysed and described only from females, except the morphology of the rostrum. Five specimens were always measured to calculate the proportion among the appendages.

The mouth appendages were dissected only in specimens from CCDB. The carapace length (maximum length from the posterior margin of the ocular orbit to the posterior margin of the carapace) and the rostrum length (maximum length from the anterior apex to the posterior margin of the ocular orbit) of the specimens were measured.

Genetic data

Specimens of *H. obliquimanus* from ten localities (six from the Caribbean Sea and four from Brazil) (fig. 1, table I) were used to assess the genetic data. In addition, we used eight specimens from six other *Hippolyte* species (table I) to compare the genetic divergence among *H. obliquimanus* and these other species. All sequences used in this study were generated from our own extractions, and two additional comparative sequences were retrieved from GenBank (table I). Genetic vouchers, from which tissue samples were obtained, were deposited in the appropriate collections under the accession numbers stated (table I).

DNA extraction, amplification, and sequencing protocols followed Mantelatto et al. (2007, 2009) and Pileggi & Mantelatto (2010). DNA extraction from tissues that had been fixed in formalin followed the protocol of Pileggi (2009). Total genomic DNA was extracted from the muscle tissue of the abdomen. Muscle was ground and then incubated for 1-12 h in 600 ml of lysis buffer at 65°C; protein was separated by the addition of 200 ml of 7.5 M ammonium acetate before centrifugation. DNA was precipitated by the addition of 600 ml of cold absolute isopropanol followed by centrifugation; the resultant pellet was washed with 70% ethanol, dried, and resuspended in 10-20 ml of TE buffer. An ~600-bp region of the 16S mtDNA gene and ~700-bp region of the barcode of cytochrome oxidase subunit I (COI) gene were amplified from diluted DNA by means of a polymerase chain reaction (PCR) in a Thermo® (Portsmouth, NH) PxE 0.2 Thermal Cycler (thermal cycles: initial denaturing for 2-5 min. at 95°C; annealing for 35-40 cycles: 30-45 sec. at 95°C, 30-45 sec. at 48-50°C, 1 min. at 72°C; final extension of 3 min. at 72°C) with universal 16S mtDNA primers 1472 (5′-AGATAGAAACCAACCTGG-3′)/16SL2 (5′-TGCCTGTTTATCAAAAACAT-3′) (Schubart et al., 2000, 2002) and COI mtDNA

Fig. 1. Map with distribution of *Hippolyte obliquimanus* Dana, 1852 (ellipses) and sites of origin of the samples analysed for morphology (solid circles), or for both morphology and genetics (solid triangles). Distribution according to Fausto-Filho (1975), d'Udekem d'Acoz (1997), and Christoffersen (1998).

primers COL6b (5′-ACAAATCATAAAGATATYGG-3′)/COH6 (5′-TADACTTC DGGRTGDCCAAARAAYCA-3′) (Schubart & Huber, 2006). PCR products were purified using the kit of purification Sureclean® and were sequenced with the ABI Big Dye® Terminator Mix (Applied Biosystems, Carlsbad, CA) in an ABI Prism 3100 Genetic Analyzer® (Applied Biosystems automated sequencer) following Applied Biosystems protocols. All sequences were confirmed by sequencing both strands. A consensus sequence for the two strands was obtained using the computational program Bioedit 7.0.5 (Hall, 2005). All sequences were submitted to GenBank (table I).

Phylogenetic and distance analyses (genes 16S and barcoding COI). — The sequences were aligned using the computational program Bioedit 7.0.5 (Hall, 2005) and were first analysed in the program Modeltest 3.7 (Posada & Crandall, 1998) to find the best substitution model. Phylogenetic reconstructions of populations of *H. obliquimanus* were conducted using PAUP 4.0 b 10 (Swofford, 2003) for Maximum Likelihood analyses. The consistency of topologies was measured using a bootstrap method (1000 replicates), and only the confidence values $> 50\%$ were reported. Inter- and intraspecific genetic distances were calculated using PAUP 4.0 b 10 (Swofford, 2003) from all taxa by pairwise comparison, using p-distance as the substitution model. The distance matrix shows the proportion of divergent residues between all of the sequences in the alignment. All positions were compared directly for each pair of sequences, one at a time.

Population analyses (gene barcoding COI). — The analyses followed the methods used by Vergamini et al. (2011), except the Mantel test. The haplotype number was calculated in DnaSP 4.10.9 (Rozas & Rozas, 1999). Each population's haplotype and nucleotide diversities were calculated using Arlequin 3.1 (Excoffier et al., 2005). A haplotype network was constructed by the Median-Joining method in Network (Bandelt et al., 1999). Genetic variation was studied by Analyses of Molecular Variance (AMOVA) (Excoffier et al., 1992), and was computed in Arlequin. Analyses were run based on the haplotype frequencies with no hierarchical structure (all populations in a single group) and with regional subdivisions defined according to the results of the distance and the haplotype network. The significance was tested using a non-parametric permutation procedure (Excoffier et al., 1992), incorporating 10 000 permutations. The Mantel test (Mantel, 1967) was performed incorporating 1000 permutations in Arlequin to identify the relationship between genetic and geographic distance.

RESULTS

Hippolyte obliquimanus Dana, 1852
(figs. 2, 3, and 4)

Hippolyte obliquimanus Dana, 1852a: 24; Chace, 1972: 113; d'Udekem d'Acoz, 1996: 114, 115; d'Udekem d'Acoz, 1997: 470, figs. 1-2; Mantelatto et al., 1999: 691; McLaughlin et al., 2005: 222; Wicksten, 2005: 112, fig. 12; Coelho Filho, 2006: 10; Coelho et al., 2006: 53; De Grave et al., 2006: 1421; Almeida et al., 2007: 15; d'Udekem d'Acoz, 2007: 201, 205; Almeida et al., 2008: 28; Espinoza-Fuenzalida et al., 2008: 623, 631, 632; Terossi et al., 2008: 127, fig. 2; Bracken et al., 2009: 285-293; Román-Contreras & Martínez-Mayén, 2009: 120, 121, 124, 125; Terossi & Mantelatto, 2010: 213; Terossi et al., 2010a: 54; Terossi et al., 2010b: 571, figs. 1-4; Terossi et al., in press.

Hippolyte exilirostratus Dana, 1852a: 24; Dana, 1852b: 563; Dana, 1855, fig. 2a-d; Schmitt, 1924a: 69; d'Udekem d'Acoz, 1996: 115.

Hippolyte obliqui-manus — Dana, 1852b: 564; Dana, 1855, fig. 3a-f; Schmitt, 1924b: 165.
Virbius gracilis var. *brasiliensis* Czerniavsky, 1884 (apud d'Udekem d'Acoz, 1997); Holthuis, 1947: 15; d'Udekem d'Acoz, 1996: 115.
Hippolyte curacaoensis Schmitt, 1924a: 68-69, fig. 4; Schmitt, 1936: 370; Chace, 1937: 129; Chace, 1972: 111, 113, figs. 44-45; Camp et al., 1977: 27; Shield, 1978: 1; Rodriguez, 1980: 166, fig. 45; Christoffersen, 1980: 215-218; Tsukamoto, 1981: 394; Escobar, 1984: 51-52; Williams, 1984: 117, fig. 81; Bauer, 1985: 152; Abele & Kim, 1986: 231, 236, 237 (figs. i-k); Bauer, 1989: 179; Bauer, 1991: 184; d'Udekem d'Acoz, 1996: 115; Hernández Aguilera et al., 1996: 39-40; Christoffersen, 1998: 355; García-Madrigal et al., 2002: 146; Wehrtmann & Vargas, 2003: 270; Wehrtmann & Cortés, 2009.
Hippolyte exilirostris — Chace, 1937: 129.
Hippolyte curaçaoensis — Holthuis, 1947: 15.
Hippolyte exilirostrata — Holthuis, 1947: 15; Chace, 1972; Coelho & Ramos, 1972: 152; d'Udekem d'Acoz, 1996: 114.
Hippolyte obliquimana — Holthuis, 1947: 15; Coelho & Ramos, 1972: 152.
Hippolyte zostericola — Williams, 1965: 82, fig. 66; Rodriguez, 1980: 167, fig. 46.
Hippolyte zoostericola — Fausto-Filho, 1975: 79.

Material. — U.S.A. — Florida, Jim Island, Indian River, 27°28′40″N 80°18′30″W, 27/v/1995, P. Y. Noël coll., 2 ♀♀ (MNHN 12972). MEXICO, Quintana Roo — Puerto Morelos, 28/i/1988, 11 ♀♀ (CNCR 21313) — Mahahual, 20/v/1988, 21 ♀♀ (CNCR 21855); Mahahual, 20/iv/1988, 7 ♀♀ (CNCR 9099) — Chetumal, 19/iv/1988, 13 ♀♀ and 1 ♂ (CNCR 9081). BELIZE — Twin Keys, 08/iv/2007, D. Felder et al. coll., 1 ♀ (ULLZ 8869). PUERTO RICO — Dorado, 18°29′N 66°15′W, iv/1982, R. Bauer coll., 21 ♀♀ (CCDB 2895). GUADELOUPE — Sant Français, xi/1998, A. Anker coll., 1 ♀ (MNHN 13751). COSTA RICA, Limón — Playa Cahuita, 09°39′43″N 82°45′37″W, 06/iv/2007, F. L. Mantelatto & I. Wehrtmann coll., 1 ♀ (CCDB 2032); 2 ♀♀ and 2 ♂♂ (CCDB 2894); Playa Cahuita, 09°39′43″N 82°45′37″W, 14/ii/2009, M. Terossi, F. L. Mantelatto, I. Miranda & I. Wehrtmann coll., 5 ♀♀ and 3 ♂♂ (CCDB 2892); Playa Cahuita, 09°39′43″N 82°45′37″W, 23/v/2010, M. Terossi, F. L. Mantelatto & I. Wehrtmann coll., 12 ♀♀ and 3 ♂♂ (CCDB 2896); Playa Cahuita, 09°39′43″N 82°45′37″W, 17/ix/2005, F. L. Mantelatto & I. Wehrtmann coll., 3 ♀♀ (CCDB 1728) — Puerto Viejo, 09°39′30.4″N 82°45′16.3″W, 05/iv/2007, F. L. Mantelatto & I. Wehrtmann coll., 10 ♀♀ and 9 ♂♂ (CCDB 2527); Puerto Viejo, 09°39′30.4″N 82°45′16.3″W, 16/ix/2005, F. L. Mantelatto & I. Wehrtmann coll., 2 ♂♂ (CCDB 1699). COLOMBIA — Magdalena, Parque Nacional Tayrona N.E. Santa Marta, Bahia Gairaca, 23/x/1978, 5 ♀♀ (SMF 9913). PANAMA — Bocas del Toro, Bocas del Drago, 09°24′52″N 82°19′53″W, 17/ii/2009, M. Terossi, F. L. Mantelatto, I. Miranda & A. Baeza coll., 17 ♀♀ and 12 ♂♂ (CCDB 2890). VENEZUELA — Boca Chica, Isla Margarita, 10°57′42.52″N 64°21′21.41″W, 31/vii/2006, C. L. Lira coll., 4 ♀♀ (CCDB 2381). BRAZIL, Bahia — Itapagipe, Praia do Bugari, 11/vii/1976, V. Almeida coll., 2 ♀♀ and 1 ♂ (MZUSP 4889). — Município de Vera Cruz, Ilha de Itaparica, Barra do Gil, 04/ii/1993, P. S. Young & M. C. B. Pereira coll., 4 ♀♀ and 1 ♂ (MNRJ 15390). — Itacaré, Praia da Ribeira, 02/ii/1994, P. S. Young & M. C. B. Pereira coll., 4 ♀♀ and 1 ♂ (MNRJ 4195). — Porto Seguro, Praia do Mutá, 17/v/2007, A. O. Almeida & L. E. A. Bezerra coll., 8 ♀ and 4 ♂ (MZUESC 903); Porto Seguro, Arraial d'Ajuda, Praia de Mucugê, 01/i/1992, P. S. Young coll., 7 ♀♀ and 13 ♂♂ (MNRJ 3200). — Recife de Nova Viçosa, Abrolhos, 28/ix/1992, P. S. Young & C. B. Castro coll., 1 ♀ and 2 ♂♂ (MNRJ 4048). BRAZIL, Rio de Janeiro — Arraial do Cabo, Praia Grande, 22/iv/1993, P. S. Young et al. coll., 9 ♀♀ (MNRJ 15389). — Ilha Grande, Praia de Araçatiba, 31/i/2005, W. L. R. Barreto coll., 5 ♀♀ and 5 ♂♂ (MZUSP 18819), 1 ♀ (MZUSP 18820). — Praia da Figueira, Angra dos Reis, 04/xii/1993, L. Santi coll., 8 ♀ and 2 ♂ (MNRJ 4044). BRAZIL, São Paulo, Ubatuba — Praia do Itaguá, 23°27′24″S 45°03′03″W, v/2007, M. Terossi, L. A. G. Pileggi & F. G. Vergamini coll., 12 ♀♀ and 8 ♂♂ (CCDB 2523); Praia do Itaguá, 23°27′24″S 45°03′03″W, iii/2005, M. Terossi, Vergamini, F. G., I. Miranda & L. A. G. Pileggi coll., 17 ♀♀ and 10 ♂♂ (CCDB 1825); Praia do

Itaguá, 23°27′24″S 45°03′03″W, v/2005, M. Terossi, L. A. G. Pileggi & I. Miranda coll., 20 ♀♀ and 12 ♂♂ (CCDB 1826). — Praia do Codó, 23°29′57″S 45°07′03″W 02/vi/2008, F. L. Mantelatto coll., 4 ♀♀ and 3 ♂♂ (CCDB 2378). BRAZIL, Santa Catarina — Bombinhas, Praia da Conceição, 27°12′10.3″S 48°29′22.0″W, 20/iv/2007, F. L. Mantelatto, L. A. G. Pileggi, L. S. Torati & E. C. Mossolin coll., 15 ♀♀ and 12 ♂♂ (CCDB 1874). — Bombinhas, Praia dos Ingleses, 27°08′39.7″S 48°28′38.2″W, 20/iv/2007, F. L. Mantelatto, L. A. G. Pileggi, L. S. Torati & E. C. Mossolin coll., 3 ♂♂ (CCDB 1881). — Florianópolis, Ponta Norte, Praia Sambaqui, 27°29′12.7″S 48°32′20.0″W, 16/iv/2007, F. L. Mantelatto, L. A. G. Pileggi, L. S. Torati & E. C. Mossolin coll., 17 ♀♀ and 4 ♂♂ (CCDB 1875), 1 ♂ (CCDB 2033), 5 ♀♀ and 3 ♂♂ (CCDB 2891).

Redescription. — Ratio rostrum/carapace average 0.85, range from 0.3 to 1.4, equal for males and females; rostrum straight and moderately long, with dorsal teeth in proximal portion when in number equal to or lower than 2, or equally distributed when in number larger than or equal to 3; with ventral teeth in distal portion; females with high rostrum, always reaching to and sometimes overreaching antennular peduncle, from 1 to 5 dorsal teeth, from 1 to 4 ventral teeth, one ventral tooth very close to apex; males with slender rostrum, reaching antennular peduncle, from 2 to 4 dorsal teeth, from 0 to 2 ventral teeth, one ventral tooth very close to apex, except in some individuals.

Carapace with supraorbital, antennal, and hepatic spines present, no postrostral teeth, postorbital ridge present. Hepatic spine reaching or overreaching margin of carapace. First segment of antennule 2 times as long as wide (stylocerite excluded); second segment 1.4 times as wide as long; third segment 1.3 times as wide as long; first segment 3 times as long as second segment; second segment 2 times as long as third segment; first segment with 3 distal outer spines (2 in some individuals); stylocerite reaching 88% of first segment (outer spines excluded); inner flagellum 1.1 times as long as antennular peduncle; outer flagellum wider and shorter than inner flagellum. Stylocerite of antenna 2.2 times as long as wide; distolateral spine of scaphocerite far from reaching tip of the blade; flagellum 5.5 times as long as scaphocerite. Mouthparts as illustrated. P1: coxa 1.8 times as wide as long; basis 4 times as wide as long; ischium 2.1 times as long as wide; merus 1.1 times as long as wide; carpus 1.7 times as wide as long; propodus 1.5 times as long as wide; dactylus 1.5 as long as wide; distal portion of dactylus overreaching distal portion of propodus; distal apex of propodus and dactylus with robust spines and inner margin serrate. P2: carpus with three segments; coxa 1.5 times as wide as long; basis 1.7 times long as wide; ischium 2.6 times as long as wide; merus 3.8 as long as wide; proximal segment of carpus 2 times as long as wide; median segment of carpus 1.2 times as wide as long; distal segment of carpus 1.8 times as long as wide; propodus 2.4 times as long as wide; dactylus 3 times as long as wide; propodus as long as sum of distal and median segments of carpus; dactylus 0.5 times as short as propodus; distal segment of carpus 2.4 times as long as median segment; proximal segment of carpus 2.1 as long as median segment; merus as

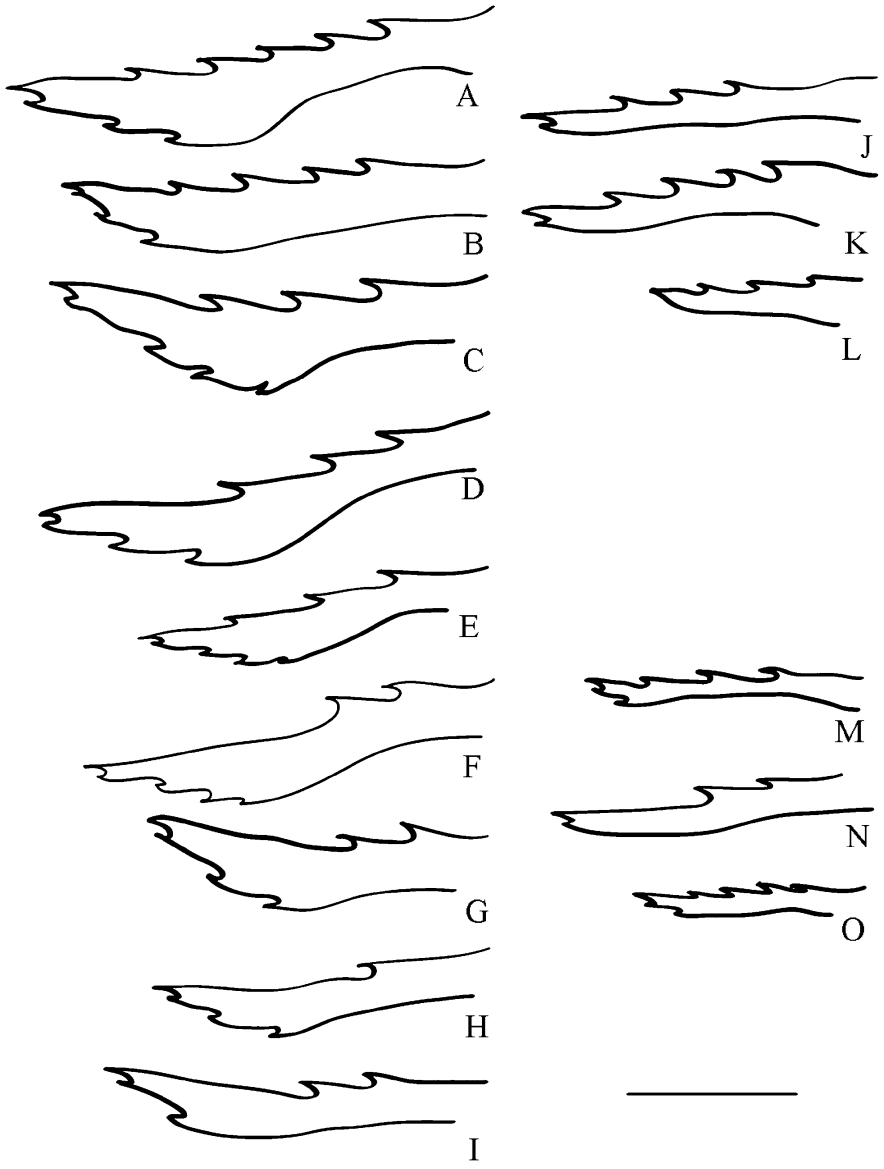

Fig. 2. *Hippolyte obliquimanus* Dana, 1852. Variability of rostrum morphology in specimens from Brazil and the Caribbean Sea. A-I, rostrums only observed in females; J-L, rostrums only observed in males; M-O, rostrums observed in both females and males. Scale: 0.5 mm.

long as carpus (sum of three segments); ischium 0.5 times as short as merus; coxa and basis as long as proximal segment of carpus; propodus and dactylus with 3 robust spines on flexor margin. P3: coxa 1.7 times as wide as long; basis 1.7 times as long as wide; ischium 3 times as long as wide; merus 5.3 times as long as wide;

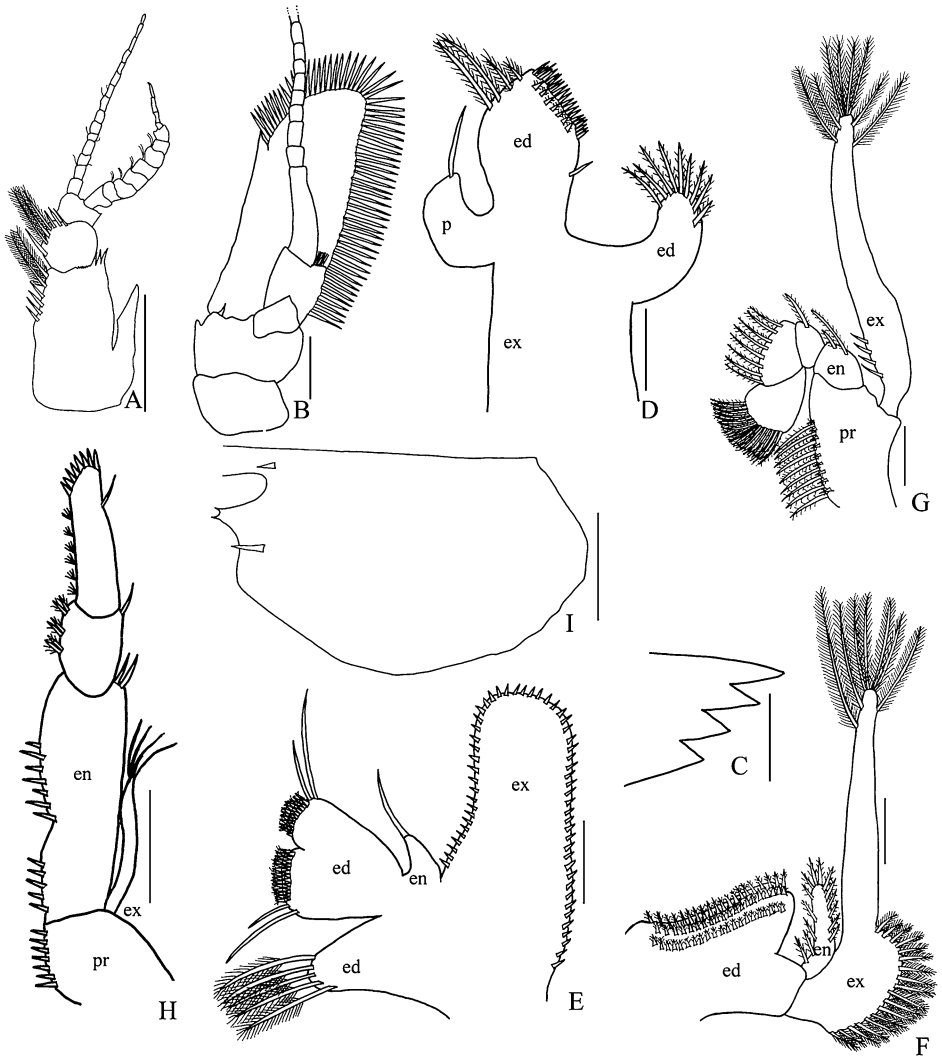

Fig. 3. *Hippolyte obliquimanus* Dana, 1852. Brazil, São Paulo, Ubatuba, ♀ (CCDB 2523). A, right antennule in dorsal view; B, right antenna in ventral view; C, pars incisiva of mandible; D, right maxillule; E, left maxilla; F, left maxilliped 1; G, left maxilliped 2; H, left maxilliped 3; I, carapace in lateral view (ed, endite; en, endopodite; ex, exopodite; p, palp; pr, protopodite). Scales: A, B, 0.5 mm; C, 0.05 mm; D, E, F, G, H, 0.2 mm; I, 0.7 mm.

carpus 2.7 times as long as wide; propodus 6.1 times as long as wide; dactylus 3 times as long as wide; ischium 0.5 as short as merus; basis and coxa as long as dactylus; merus 2.2 times as long as carpus; propodus 1.8 times as long as carpus; dactylus 0.3 times as short as propodus; merus with 2 to 6 lateral spines; carpus with one lateral proximal spine; propodus with 6 or 7 pairs of ventral spines; dactylus with 3 spines on flexor margin; distal spine shorter than others;

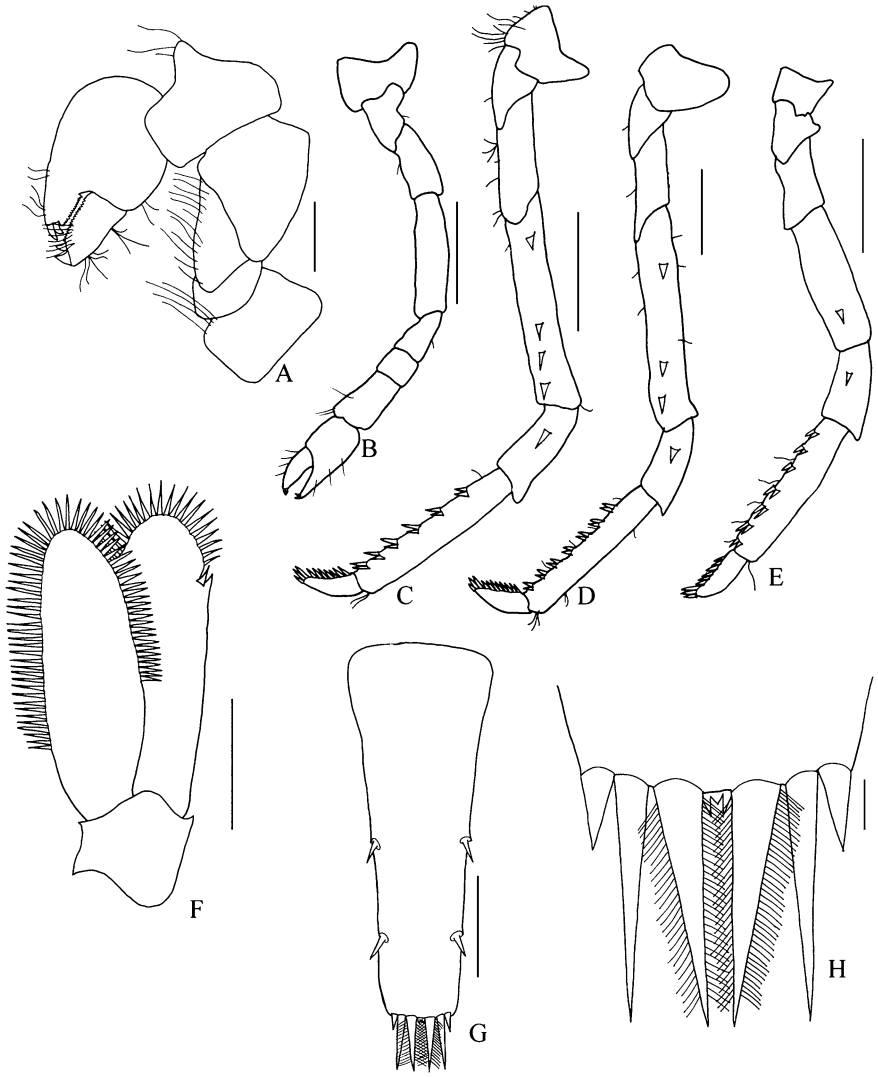

Fig. 4. *Hippolyte obliquimanus* Dana, 1852. Brazil, São Paulo, Ubatuba, ♀ (CCDB 2523). A, left pereiopod 1; B, right pereiopod 2; C, right pereiopod 3; D, right pereiopod 4; E, right pereiopod 5; F, left uropod in dorsal view; G, telson in dorsal view; H, apex of telson in dorsal view. Scales: A, 0.3 mm; B, C, D, E, F, 0.7 mm; G, 0.5 mm; H, 0.05 mm.

7 to 9 ventral spines that decrease gradually in proximal direction. P4: similar to P3; merus with 2 to 5 lateral spines; carpus with one lateral proximal spine; propodus with 7 pairs of ventral spines; dactylus with 3 spines on flexor margin; distal spine shorter than others; 8 ventral spines that gradually decrease in size in proximal direction. P5: similar to P3; merus with one lateral distal spine; carpus with one lateral proximal spine; propodus with 7 pairs of ventral spines; dactylus

with 3 spines on flexor margin, distal spine shorter than other two, 6 to 8 ventral spines that gradually decrease in size in proximal direction. Endopod and exopod of uropod of approximately same length and shape; exopod a little longer than endopod; exopod with a spine and a mobile seta shaped like a spine close to distal margin of blade. Telson 3.8 times as long as wide; 2 pairs of dorsolateral spines, first pair in the median portion and second pair in distal portion of telson; 6 strong spines on the posterior extremity (apex), the two pairs of inner spines are 3.5 times as long as the pair of outer spines, the pairs of inner spines have many setae; three spinules in the middle of the posterior extremity, the inner spinule shorter than the two outer spinules.

Size. — Carapace length, females: 0.6-3.1 mm; males: 0.8-2.4 mm.

Type-locality. — Rio de Janeiro, Brazil.

Distribution. — Western Atlantic – U.S.A., North Carolina (Chace, 1972; Williams, 1984; Abele & Kim, 1986) and Florida (d'Udekem d'Acoz, 1997); Mexico (García-Madrigal et al., 2002; Wicksten, 2005; Román-Contreras & Martínez-Mayén, 2009); Belize (present study); Honduras (De Grave et al., 2006); Cuba, Saint Christopher, Antigua, Guadeloupe, Carriacou, Cuba, Tobago (Chace, 1972; Williams, 1984); Puerto Rico (Bauer, 1989); Costa Rica (Wehrtmann & Vargas, 2003; Wehrtmann & Cortés, 2009); Panama, Colombia (present study); Curaçao (Schmitt, 1924a); Bonaire (Schmitt, 1936); Venezuela (Rodriguez, 1980); Brazil, from Ceará to Santa Catarina (Dana, 1852a; Fausto-Filho, 1975 as *H. zostericola*; Christoffersen, 1980, 1998; d'Udekem d'Acoz, 1997; Coelho Filho, 2006; Coelho et al., 2006; Almeida et al., 2007, 2008).

Coloration. — The animal's body can show plain coloration in brown, red, green, or greenish-blue, or disruptive coloration; in this case, they are transparent with pigmented bands (Christoffersen, 1980 as *H. curacaoensis*; Terossi & Mantelatto, 2010). Some specimens can present scattered tufts of plumose setae (d'Udekem d'Acoz, 1997; Terossi & Mantelatto, 2010).

Biology. — Shallow waters; in the Caribbean found in seagrass meadows dominated by turtle grass (*Thalassia testudinum*) (Bauer, 1989 as *H. curacaoensis*; Terossi et al., 2010b); along the Brazilian coast found in algae beds, particularly in association with seaweed of the genus *Sargassum* (cf. Mantelatto et al., 1999); females reach a larger size than males and the sexual system is gonochoristic (Terossi et al., 2008); sex ratio biased to females (Terossi & Mantelatto, 2010); seasonal to continuous reproduction (Bauer, 1989 as *H. curacaoensis*; Terossi & Mantelatto, 2010); fecundity can vary from 64 to 187 eggs and eggs recently extruded can vary from 0.005 to 0.020 mm^3 (Bauer, 1991 as *H. curacaoensis*; Mantelatto et al., 1999; Terossi et al., 2010b); reproductive output can vary from 0.03 to 0.31 (Terossi et al., 2010b); carapace length of zoea is 0.31 $\pm$ 0.02 mm (Terossi et al., 2010a);

sperm morphology is distinct from the "thumbtack" shaped spermatozoa observed in the majority of caridean shrimps (Terossi et al., in press); some Brazilian specimens are bopyrized (Chace, 1972 as *H. curacaoensis;* Tsukamoto, 1981 as *H. curacaoensis*; d'Udekem d'Acoz, 1997; Terossi & Mantelatto, 2010).

Genetic data

The multiple sequence alignment of the 16S gene obtained had 530 positions in 12 specimens of *H. obliquimanus* and 8 specimens from 6 other *Hippolyte* species. However, the sequences retrieved from GenBank (*H. acuta* and *H. bifidirostris*) were shorter than the sequences obtained in this study (409 and 453 bp, respectively).

The 10 sequences of *H. obliquimanus* were identical (16S gene), which means that they had 0% genetic divergence (table II); the same result occurred with morphologically well-defined species: two specimens of *H. inermis* (Italy and Greece) and two specimens of *H. varians* (France and the Netherlands) (table II). Genetic distances estimated among the *H. obliquimanus* specimens and other *Hippolyte* species ranged from 3.0 to 23.2%. The dendrogram generated by cluster analysis showed a clear separation of *H. obliquimanus* specimens from the other species analysed (fig. 5).

TABLE II

Genetic divergence matrix of the 16S gene among some specimens of *Hippolyte obliquimanus* Dana and other *Hippolyte* species

Specimens	1	2	3	4	5	6	7	8	9	10
1. *H. obliquimanus* – Brazil (4 specimens)	0.000									
2. *H. obliquimanus* – Caribbean Sea (6 specimens)	0.000	0.000								
3. *H. zostericola* (Smith) – Mexico	0.132	0.132	0.000							
4. *H. williamsi* Schmitt – Chile	0.030	0.030	0.135	0.000						
5. *H. varians* Leach – Netherlands	0.169	0.169	0.213	0.181	0.000					
6. *H. varians* – France	0.169	0.169	0.213	0.181	0.000	0.000				
7. *H. inermis* Leach – Italy	0.178	0.178	0.206	0.186	0.158	0.158	0.000			
8. *H. inermis* – Greece	0.178	0.178	0.206	0.186	0.158	0.158	0.000	0.000		
9. *H. bifidirostris* (Miers) – New Zealand	0.178	0.178	0.212	0.182	0.196	0.196	0.211	0.211	0.000	
10. *H. acuta* (Stimpson) – Japan	0.232	0.232	0.272	0.230	0.212	0.212	0.235	0.235	0.190	0.000

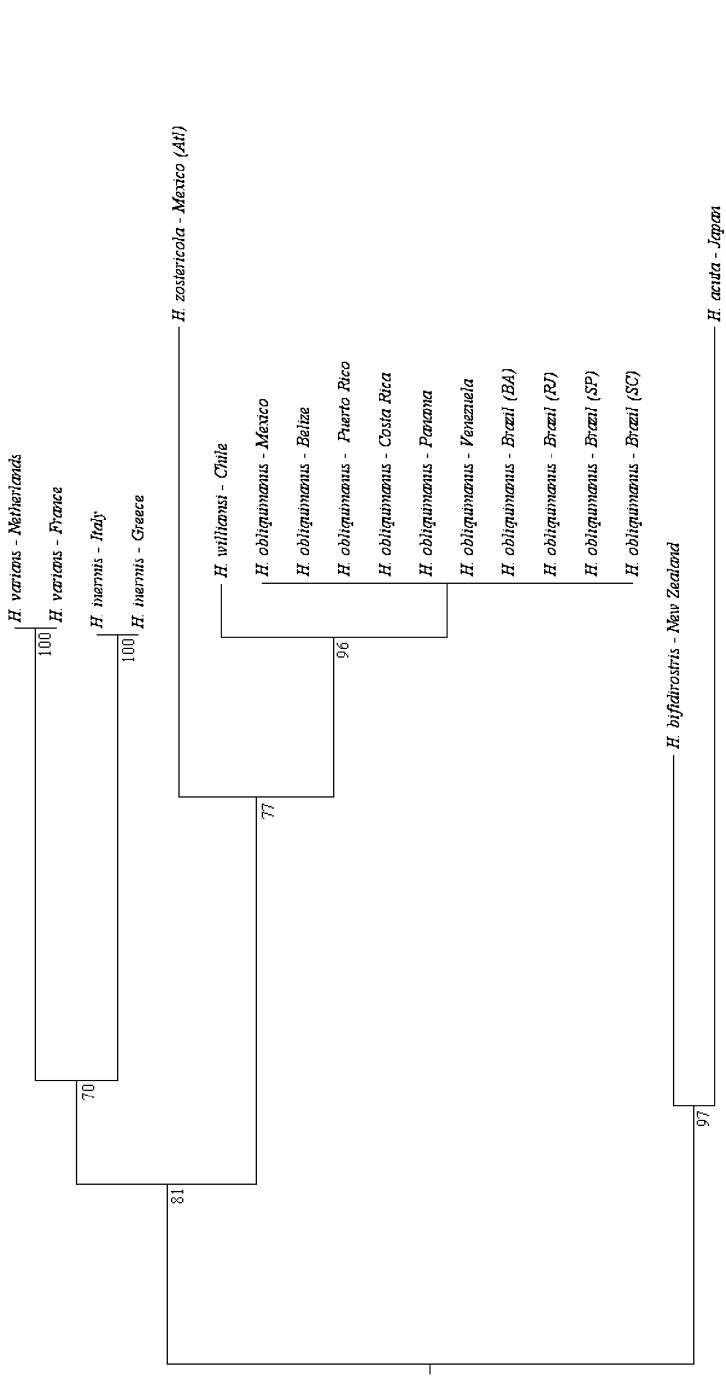

Fig. 5. Phylogenetic tree for populations of *Hippolyte obliquimanus* Dana, 1852 and other *Hippolyte* species using Maximum Likelihood analysis of 16S gene sequences. Numbers are support values for 1000 bootstraps; values < 50% are not shown. Locality abbreviations: Atl, Atlantic Ocean; BA, Bahia; RJ, Rio de Janeiro; SP, São Paulo; SC, Santa Catarina.

TABLE III
Genetic divergence matrix of the barcoding COI gene among some specimens of *Hippolyte obliquimanus* Dana and other *Hippolyte* species

Specimens	1	2	3	4	5
1. *H. obliquimanus* – Brazil (20 specimens)	0.000-0.018				
2. *H. obliquimanus* – Caribbean Sea (14 specimens)	0.005-0.020	0.000-0.014			
3. *H. williamsi* Schmitt – Chile	0.122-0.133	0.126-0.135	0.000		
4. *H. inermis* Leach – Italy	0.199-0.210	0.203-0.214	0.196	0.000	
5. *H. inermis* – Greece	0.203-0.212	0.206-0.217	0.210	0.034	0.000

The multiple sequence alignment of the barcoding COI gene obtained had 599 positions in 10 specimens of *H. obliquimanus* and three specimens from two other *Hippolyte* species. There was variation among the sequences of this gene from *H. obliquimanus* specimens (0 to 1.7%; table III); the variation separated the specimens into two groups (fig. 6): (1) the Caribbean specimens (Belize, Costa Rica, Panama, and Venezuela), and (2) the Brazilian specimens (states of Bahia, Rio de Janeiro, São Paulo, and Santa Catarina). However, the genetic distances estimated among the specimens of *H. obliquimanus* and the other *Hippolyte* species were higher than 1.7% (12.9 to 20.7%, table III).

The locality Belize was excluded from the haplotype analyses, since there was just one individual from this site. Based on partial fragments of the barcoding gene COI from 33 specimens from seven localities (three from the Caribbean Sea and four from Brazil) 18 haplotypes were identified (table IV), of which 14 (77%) represented single individuals. Populations from the Caribbean Sea did not share haplotypes with the populations from Brazil (table IV, fig. 7).

Total haplotype diversity was relatively high (0.90), the nucleotide diversity was higher in the specimens from the Caribbean Sea (5.34×10^{-3}) than in the specimens from Brazil (2.82×10^{-3}).

The analysis of molecular variance (table V) without hierarchical structure indicated that the highest percentage of variation (56.4%) was within *H. obliquimanus* populations. When the populations were structured according to the groups indicated by previous analyses (distance and haplotype network), a highest and significant percentage among the groups was detected (54.2%) and the percentage of variation among populations was low and not significant. The Mantel test identified a significant relationship between the genetic distance and the geographic distance (correlation coefficient: 0.926; $P = 0.036$) of the specimens analysed.

Fig. 6. Phylogenetic tree for populations of *Hippolyte obliquimanus* Dana, 1852 and other *Hippolyte* species, using Maximum Likelihood analysis of COI gene sequences. Numbers are support values for 1000 bootstraps; values $< 50\%$ are not shown. Locality abbreviations: BA, Bahia; RJ, Rio de Janeiro; SP, São Paulo; SC, Santa Catarina.

TABLE IV

Hippolyte obliquimanus Dana, 1852. Distribution of haplotypes detected in the various populations. N, number of individuals analysed in each population; Hd, haplotype diversity; Nd, nucleotide diversity; Sd, standard deviation

	Populations	Haplotypes (H)																		N	Hd	Nd + Sd ($\times 10^{-3}$)
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18			
Caribbean	Costa Rica	1	1	1	1	1														5	1.00	5.39 ± 3.93
	Panama		1	3			1													5	0.70	1.44 ± 1.43
	Venezuela							1	1	1										3	1.00	9.57 ± 7.90
Brazil	Bahia										3	1	1							5	0.70	1.79 ± 1.67
	Rio de Janeiro										4			1						5	0.40	2.15 ± 1.91
	São Paulo										2				1	1	1			5	0.90	4.67 ± 3.49
	Santa Catarina										1		2					1	1	5	0.90	3.23 ± 2.59
	Total	1	2	4	1	1	1	1	1	1	10	1	3	1	1	1	1	1	1	33	0.90	6.07 ± 3.54

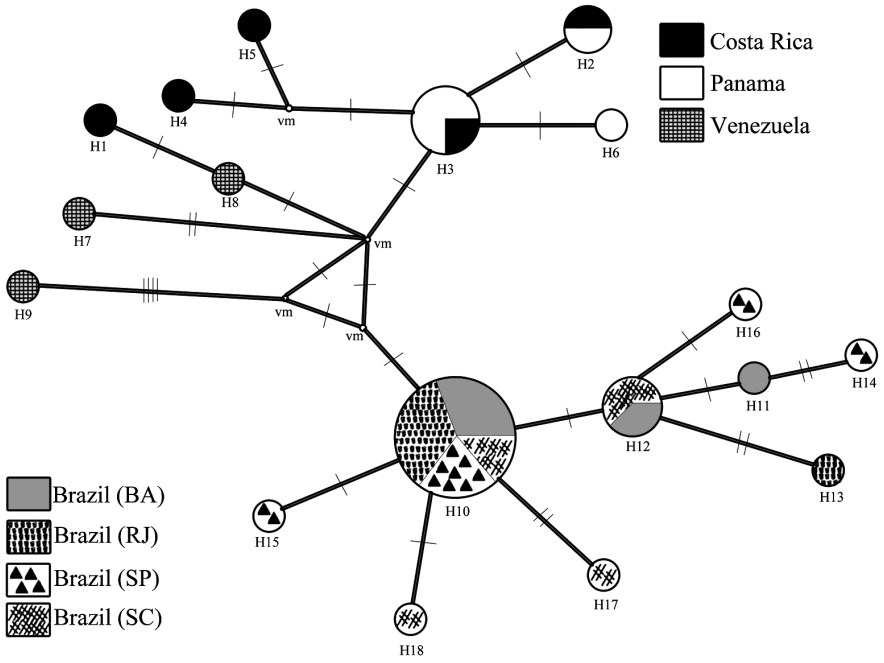

Fig. 7. *Hippolyte obliquimanus* Dana, 1852. Haplotype network based on Median-Joining analysis indicating the distribution of each haplotype (H) found in the populations (18 haplotypes in 33 specimens). The haplotype identification is below each circle. The circle size of each haplotype is proportional to the overall frequency in our sample. Each small trace represents a mutational step; vm, median vector. States of Brazil: BA, Bahia; RJ, Rio de Janeiro; SP, São Paulo; SC, Santa Catarina.

TABLE V

Hippolyte obliquimanus Dana, 1852. Analysis of molecular variance (AMOVA). *Significant values, $P < 0.05$

AMOVA structure	Source of variation	%	F_{st}/F_{ct}	*P*
Without	among populations	43.6%	FST: 0.436	0.000*
	within populations	56.4%		
Caribbean Sea (Costa Rica,	between groups	54.2%	FSC: 0.063	0.000*
Panama, and Venezuela)	among populations	2.9%	FST: 0.570	0.09
Brazil (Bahia, Rio de Janeiro,	within populations	42.9%	FCT: 0.542	0.000*
São Paulo, and Santa Catarina)				

DISCUSSION

Morphological remarks

There were no morphological differences between the Brazilian specimens and those from the Caribbean. Among the characters analysed, the rostrum and the number of merus spines of pereiopods 3, 4, and 5 showed the largest variation.

Fifteen different shapes of rostrum were observed, nine were present only in females (figs. 2A-I), three only in males (figs. 2J-L), and three in both sexes (figs. 2M-O). Three shapes are predominant (figs. 2D, J, and M), and five were observed in only one individual (figs. 2A, F, H, L and M).

According to the literature, there is variation in the numbers of the rostral teeth: *Hippolyte obliquimanus* described by Dana (1852a) had 4 dorsal and 2 ventral teeth; *H. curacaoensis* described by Schmitt (1924a) had 3 dorsal teeth and 1 small, distal ventral tooth; *H. curacaoensis* reported by Schmitt (1936) had 2-5 dorsal teeth and 1-3 ventral teeth; *H. curacaoensis* reported by Rodriguez (1980) had 1-3 dorsal teeth and 2 ventral teeth; *H. curacaoensis* reported by Williams (1984) had 3-4 dorsal teeth and 1-3 ventral teeth; d'Udekem d'Acoz (1997) redescribed *H. obliquimanus* and reported for the first time that the rostrum can vary according to sex; that author found 2-4 dorsal teeth in both sexes, 3-4 ventral teeth in females, and 1-2 ventral teeth in males.

In the present study, males of *H. obliquimanus* had 2-4 dorsal teeth and 0-2 ventral teeth, and females had 1-5 dorsal teeth and 1-4 ventral teeth. One shape of rostrum can be found in many localities (in Brazil and in Caribbean Sea), there was no shape exclusive for one locality. A high variation in rostrum shape was observed in the present study, probably due to the diversity of localities and the high number of individuals analysed. Based on this variation, it is possible to affirm that the rostrum is not an informative character to separate some species of the genus *Hippolyte*, since 15 different combinations of dorsal and ventral teeth were observed in the individuals of *H. obliquimanus* analysed.

About 15 years ago, d'Udekem d'Acoz (1997) proposed that *H. curacaoensis* was a junior synonym of *H. obliquimanus*, based on morphological analysis. Our molecular data confirm this synonymy, since we accessed some specimens previously as signed to *H. curacaoensis*. Actually, before the study of d'Udekem d'Acoz (1997), all specimens from the Caribbean Sea with 2 or 3 distal outer spines on the first segment of the antennule were previously identified as *H. curaoensis* (cf. Schmitt, 1936; Chace, 1937, 1972; Rodriguez, 1980; Escobar, 1984; Williams, 1984; Bauer, 1985, 1989, 1991; Abele & Kim, 1986). In addition, d'Udekem d'Acoz (1997) reported some differences between Brazilian and Caribbean specimens in relation to the number of lateral spines on the merus of pereiopods 3 and 4: 2-5 spines on pereiopod 3 in Brazilian specimens and 5-6 spines in Caribbean specimens; 2-3 spines on pereiopod 4 in Brazilian specimens and 4-5 spines in Caribbean specimens. The Caribbean specimens analysed by Chace (1972) had 6-7 spines on the merus of pereiopod 3, and 4 spines on the merus of pereiopod 4. In the present study, 2-6 spines on the merus of pereiopod 3 and 2-5 on the merus of pereiopod 4 were counted, but this variation was observed in both regions studied. All specimens analysed in the present study had one lateral spine on the merus of pereiopod 5, as was described by d'Udekem d'Acoz (1997). Nevertheless, Chace (1972) drew 2 lateral spines on the merus of pereiopod 5 in specimens from Tobago (Caribbean), which means that this character can be individually variable, too.

Two or three distal outer spines on the first segment of the antennule were observed in all studies that reported *H. obliquimanus.* This may be a character that can separate this species from the others in the Atlantic Ocean and the Mediterranean Sea: *H. coerulescens* (Fabricius, 1775) has only one distal outer spine (d'Udekem d'Acoz, 2007) and other *Hippolyte* species described in those areas do not have spines on the first segment of the antennule (d'Udekem d'Acoz, 2007). Other *Hippolyte* species, mainly in the Pacific Ocean, had distal outer spines on the first segment of the antennule. One example is *Hippolyte williamsi* Schmitt, 1924 (three spines), that occurs in the eastern Pacific; that species was considered morphologically closer to *H. obliquimanus* than to other *Hippolyte* species from the western Atlantic (d'Udekem d'Acoz, 1997), similar to what was supported here by the molecular analysis (discussed below). According to Chace (1937), *H. mexicana* (junior synonym of *H. williamsi*) is apparently the Pacific representative of the group made up of *H. exilirostris* (junior synonym of *H. obliquimanus*) from Brazil and *H. curacaoensis* (junior synonym of *H. obliquimanus*) from Curaçao.

Additionally, this is the first record of *H. obliquimanus* from Belize, Panama, and Colombia.

Genetic data

Based on estimated genetic distances (16S gene) among *H. obliquimanus* specimens (0%), among *H. inermis* specimens (0%), among *H. varians* specimens (0%), and among the *H. obliquimanus* specimens and other *Hippolyte* species (3.0% to 23.2%), we can confirm that the 16S gene can be used to separate species in the genus *Hippolyte*.

The phylogenetic tree (16S gene) shows a clear separation of *H. obliquimanus* specimens from the other species analysed. However, we could not separate the specimens of *H. obliquimanus* into two or more different groups at species level. In relation to *H. obliquimanus* and other *Hippolyte* species, although *H. obliquimanus* is morphologically closer to *H. williamsi* (Pacific) than to the other *Hippolyte* species from the West Atlantic (d'Udekem d'Acoz, 1997), it also showed a lower genetic divergence from *H. williamsi* (3.0%) than from *H. zostericola* (Atlantic, 13.5%). More studies about the relationship among the species of the genus *Hippolyte* of America are necessary to clarify this question.

The analyses of the barcoding gene COI showed a separation of *H. obliquimanus* specimens into two groups: the Caribbean specimens and the Brazilian specimens. These groups were supported by all analyses (genetic distances, haplotypes network, and AMOVA).

The reduced genetic variation (COI barcoding gene) estimated among the specimens of *H. obliquimanus* ($<$2.0%) did not allow to make definitive conclusions, but was a good indication not to separate the specimens into more than one species. The genetic divergence between the *H. obliquimanus* group and the other congeners varied from 12.2 to 21.7%, and consequently more than 2% genetic divergence would be necessary to separate the species of *Hippolyte* by the barcoding gene COI. The AMOVA showed that the highest genetic variation existed between the large groups (the Caribbean Sea and Brazil); furthermore, the populations from the Caribbean Sea did not share haplotypes with those from Brazil.

These groups are separated geographically by a gap in the known distribution of *H. obliquimanus* between Venezuela and northern Brazil (fig. 1), but is that gap true, or the result of a lack of studies in those intermediate localities (Guyana, Suriname, French Guiana, and northern Brazil)? We believe that the absence of *H. obliquimanus* in those regions could be explained by the known geographic barrier formed by the rivers Orinoco (Venezuela) and Amazon (Brazil). The coastline between the mouths of those two rivers, with numerous additional freshwater effluents in the Guyana region, covers a length of about 2700 km (Collette & Rützler, 1977). The salinity of the coastal zone is drastically modified due to the large volumes of fresh water from those rivers; the Amazon River alone dumps approximately 190 000 m^3/s in to the Atlantic Ocean (Boltovskoy et al., 1999).

H. obliquimanus is a species of shallow water, where the salinity variation is larger; possibly, this species cannot establish itself in that region. Other studies with coastal invertebrates also suggested that gene flow was interrupted by the geographic barrier between the West Indies and Brazil (Collette & Rützler, 1977; Werding et al., 2003).

One species with geographically isolated populations, as the populations of *H. obliquimanus*, can show genetically distinct populations due to the absence of gene flow among them (Odinetz-Collart & Rabelo, 1996). If there is no continuing gene flow among populations, the genetic distance between a pair of populations presumably increases as the gene flow between them decreases (Slatkin, 1985). The fact that *H. obliquimanus* shares no haplotypes between the Caribbean Sea populations and those from Brazil evidences a genetic isolation, and indicates that gene flow is not happening among these populations.

Marine species with mobile phases in their life history are capable of a widespread dispersion and are, therefore, genetically homogeneous throughout their distribution (Gopurenko & Hughes, 2002); however, *H. obliquimanus* does not show such homogeneity. Possibly, the larvae of this species do not manage to cross the geographic barrier created by the Amazon River, or perhaps are restrained by other factors, such as the hydrodynamics of the marine currents, the dispersal limitation by larval behaviour, or the selection of local settlement (Broekaert, 2007). All these factors may act in the genetic divergence of the populations.

Conclusion

The differences found in the morphology of the adults are considered intraspecific variation, since they comprise small variations and were found in both regions analysed (Brazil and the Caribbean Sea). It was not possible to separate *H. obliquimanus* by the 16S gene and the genetic divergence found in the barcoding COI gene was probably a variation among different populations. The larval morphology also could not separate the species from Brazil and from the Caribbean Sea (Terossi et al., 2010a), the zoea I of *H. obliquimanus* showed little intraspecific variation between the two populations (Brazil and Costa Rica) studied by those authors: the first zoea from the Brazilian population had four denticles on the ventral margin of the carapace, while the specimens from the Costa Rican population had only three denticles. In conclusion, there are variations in the populations of *H. obliquimanus* from the Caribbean Sea and Brazil. Nevertheless, such variations are considered intraspecific and at the moment they do not support the separation of *Hippolyte obliquimanus* into two or more species.

ACKNOWLEDGEMENTS

This study is part of a Ph.D. Thesis by MT supported by a fellowship from FAPESP (Proc. 06/61771-0) and CAPES (Doctoral Sandwich Program – Proc. 1306-09-0). MT is grateful to FAPESP for a postdoctoral fellowship (2011/11901-3). Additional support to this project was provided by FAPESP (Grants: 2002/08178-9; Coleções Cientificas – 2009/54931-0; Biota – 2010/50188-8) and CNPq (Research Grants 473050/2007-2, 491490/2004-6 and 490353/2007-0; Research Scholarships PQ 301261/2004-0 and 302748/2010-5) to FLM. We are deeply grateful to many colleagues and friends (Alexandre Almeida, Antonio Baeza, Carlos Lira, Cédric d'Udekem d'Acoz, Cristiana Serejo, Darryl Felder, Fernando Alvarez, Ingo Wehrtmann, Jose Luis Villalobos, Juan Bolaños, Leonardo Pileggi, Marcos Tavares, Martin Thiel, Michael Türkay, Patrício Hernáez, Raymond Bauer, Regis Cleva, Valério Zupo) for their help in collecting, for making available some essential, fresh specimens, and for lending material from their collections used in our analysis. Thanks are due to Fernanda G. Vergamini and Leonardo G. Pileggi for their help during the obtaining and the analyses of the genetic data. We thank Alexandre Almeida, Antônio Castilho, Eduardo Almeida, and Irene Cardoso for commenting on an earlier version of the manuscript during the Doctoral Thesis defense, and anonymous reviewers for their suggestions and contributions toward improving this paper. We thank Nicole Olguín for his assistance with revision of the Spanish abstract and Julia Hetem for revision of the English. The support and assistance of the Postgraduate Program in Comparative Biology of FFCLRP/USP and CEBIMar/USP during the fieldwork are gratefully acknowledged. The collections of species conducted in this study complied with currently applicable state and federal laws of Brazil (DIFAP/IBAMA 121/05; permanent license to FLM for collection of Zoological Material No. 11777-1 MMA/IBAMA/SISBIO).

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First received 26 August 2011.
Final version accepted 14 February 2012.