A new species of *Lebbeus* (Crustacea: Decapoda: Caridea: Hippolytidae) from the Von Damm Vent Field, Caribbean Sea

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A new species of the hippolytid shrimp genus Lebbeus *White, 1847 is described from the Von Damm Vent Field (VDVF) on the Mid-Cayman Spreading Centre, Caribbean Sea, at 2294 m water depth.* Lebbeus virentova *sp. nov. is defined and illustrated from seven specimens, with brief notes on its distribution and habitat. Molecular phylogenetic data from the COI mDNA region are used to analyse the species' phylogenetic position, and its morphology is compared with previously described species. This new species represents the second family of caridean shrimp to be reported from the VDVF.* Lebbeus virentova *sp. nov. is the eighth member of the genus to be described from hydrothermal vents and appears to be the first hippolytid shrimp at a vent field known from outside the Pacific Ocean.*

INTRODUCTION

Following the discovery of aggregations of alvinocaridid shrimp at hydrothermal vents in the Pacific (Williams, 1980; Williams & Chace, 1982), and at cold seeps in the Gulf of Mexico (Williams, 1988), a substantial research effort has addressed the taxonomy, phylogeny, ecology, physiology and distribution of caridean shrimp from chemosynthetic ecosystems in the deep sea (e.g. Van Dover *et al.*, 1988, 1989; Gebruk *et al.*, 1993; Segonzac *et al.*, 1993; Shank *et al.*, 1998, 1999; Copley & Young, 2006; Komai *et al.*, 2010; Teixeira *et al.*, 2010).

At least fifty species from nine caridean families have been recorded from deep-sea vents and seeps (Martin & Haney, 2005; De Grave & Fransen, 2011). Members of the family Alvinocarididae Christoffersen, 1986 appear to be endemic to deep-sea chemosynthetic environments, and form a dominant component of the biomass at several vent fields in the Atlantic and Indian Oceans (see Nye *et al.*, 2012 for recent review). In contrast, the presence of other caridean families at vents and seeps is considered to be opportunistic (e.g. Martin & Haney, 2005; Desbruyères *et al.*, 2006).

The genus *Lebbeus* White, 1847, is composed of sixty species (Komai *et al.*, 2012), and represents the most diverse genus within the Hippolytidae Spence-Bate, 1888. Species of *Lebbeus* are found from shallow to deep waters (e.g. Chang *et al.*, 2010). The genus exhibits a cosmopolitan distribution from the tropics to high latitudes, but its species generally have narrow geographical ranges (Komai *et al.*, 2004).

The majority of *Lebbeus* species are described from the western North Pacific (e.g. Komai & Takeda, 2004; Komai *et al.*, 2004; De Grave & Fransen, 2011). *Lebbeus* is the only hippolytid recorded from deep-sea chemosynthetic environments, with several species documented from hydrothermal vents in the Pacific (see Table 1 and references therein).

Two high-temperature hydrothermal vent fields were discovered recently at the Mid-Cayman Spreading Centre (MCSC), Caribbean (Connelly *et al.*, 2012). The ultraslow-spreading MCSC is located in a deep trough, tectonically and geographically isolated from other mid-ocean ridges (Ballard *et al.*, 1979; German *et al.*, 2010). The Von Damm Vent Field (VDVF) is located away from the axis of the spreading centre on the upper slopes of an oceanic core complex at ~2300 m water depth (Connelly *et al.*, 2012). The VDVF is a sulphide mound approximately 100 m in diameter and 30 m high, venting clear, buoyant, high-temperature fluids at its peak, visually dominated by swarming shrimp (Connelly *et al.*, 2012). Investigations of the fauna inhabiting vent fields on the MCSC have the potential to enhance current understanding of the dispersal and evolution of vent taxa, and vent biogeography of the region (Van Dover *et al.*, 2002).

In this study, a new species of *Lebbeus* from the VDVF is described and illustrated. *Lebbeus virentova* sp. nov. belongs to the second family (Hippoloytidae) of caridean shrimp to be reported from the VDVF, the other being Alvinocarididae. In addition to enhancing existing knowledge about biodiversity in the deep sea, this appears to be the first record of a hippolytid shrimp from a vent field outside the Pacific Ocean.

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Table 1. Summary of geographical distribution and bathymetric range of *Lebbeus* species from hydrothermal vents.

Species	Site(s)	Depth (m)	Primary references
Lebbeus bidentatus +	South-eastern Pacific: southern East Pacific Rise	Not available	Martin & Haney (2005)
*Lebbeus laurente**	North-eastern Pacific: East Pacific Rise 13°N	2618–2640	Wicksten (2010); Komai *et al.* (2012)
Lebbeus manus	South-western Pacific: Manus Basin	1540–1577	Komai & Collins (2009)
Lebbeus pacmanus	South-western Pacific: Manus Basin	1662	Komai *et al.* (2012)
*Lebbeus shinkaiae***	North-western Pacific: Okinawa Trough	691–1491	Komai *et al.* (2012)
Lebbeus thermophilus	South-western Pacific: Manus & Lau Basins	1512–1842	Komai *et al.* (2012)
Lebbeus virentova sp. nov.	Caribbean: Von Damm Vent Field, Mid-Cayman Spreading Centre	2294–2375	This study
Lebbeus wera	South-western Pacific: Brothers Caldera, Kermadec Ridge	1208–1336	Ahyong (2009); Komai *et al.* (2012)

+ , record unverified. Known also from its type location, off Chile at 1680 m depth (Zarenkov, 1976); *, a replacement name for *L. carinatus* De Saint Laurent, 1984, a junior homonym of *L. carinatus* Zarenkov, 1976; **, recorded as *L. washingtonianus* (Kikuchi & Ohta, 1995; Martin & Haney, 2005; Komai & Collins, 2009) before being referred to *L. shinkaiae* Komai, Tsuchida & Segonzac, 2012.

MATERIALS AND METHODS

Specimens were collected from the VDVF (2294–2375 m) at the MCSC, Caribbean, during the 18th voyage of the RV 'Atlantis' (16th leg, January 2012). All specimens were collected using a suction sampler attached to the remotely operated vehicle (ROV) 'Jason II', together with still photographs and video recordings of them *in situ*. Material for morphological study was fixed immediately in 10% neutralized formalin and subsequently transferred to 75% industrial methylated spirits. Material for molecular analyses was immediately placed in 95% ethanol.

Measurements of specimens were taken to the nearest 0.1 mm using Vernier callipers. Postorbital carapace length (CL) was measured from the posterior margin of the orbit to the posterior margin of the carapace and is used herein as an indication of specimen size. Individuals were sexed under a dissecting microscope.

Illustrations were prepared with the aid of a camera lucida mounted onto a Leica MZ8 steromicroscope, scanned and inked digitally using a WACOM™ digitizer and Adobe® Illustrator® software, as described by Coleman (2003, 2009). Specimens are deposited in the invertebrate collection at the Smithsonian Institution, National Museum of Natural History (USNM), Washington, DC. Morphological terminology generally follows Komai *et al.* (2012).

Abdominal muscle for DNA extraction was cut from ethanol-preserved specimens and the carapace removed. Total genomic DNA was extracted using the CTAB (cetyltrimethyl ammonium bromide) procedure (Doyle & Dickson, 1987). Regions of mitochondrial Cytochrome *c* Oxidase subunit I (COI) DNA and 16S ribosomal DNA were amplified by performing polymerase chain reactions (PCR).

The COI region was amplified with the universal primers LCO1490 and HCO2198 described by Folmer *et al.* (1994). The 20 μl amplification mixture contained 1X buffer reagent (200 mM Tris pH 8.8, 500 mM KCL, 0.1% Trixton X-100, 2 mg/ml bovine serum albumen), 2 mM MgCl$_2$, 0.2 mM of each dNTP, 0.5 mM of each primer, 1 U Taq DNA polymerase (Bioline), 5 μl of template DNA and sterile H$_2$O to final volume. The thermal cycling conditions were: 94°C/2 minutes; followed by 5 cycles at (94°C/35 seconds; 45°C/35 seconds; 72°C/1:20 minutes) and 35 cycles at (94°C/35 seconds; 50°C/35 seconds; 72°C/1:20 minutes) with a final extension of 72°C/10 minutes.

Amplifications of the 16S region were performed using the universal primers 16Sar and 16Sbr described by Palumbi (1996). The 20 μl amplification mixture contained: 1X reaction buffer (same as for COI), 2.5 mM MgCl$_2$, 0.13 mM of each dNTP, 0.38 mM of each primer, 1 U Taq DNA polymerase (Bioline), 2.5 μl of template DNA and sterile H$_2$O to final volume. The thermal cycling conditions were: 94°C/4 minutes; 30 cycles at (94°C/30 seconds; 52°C/1 minute; 72°C/2 minutes) and 72°C/5 minutes.

The PCR products were purified with the ExoAP treatment by adding the following ExoAP mixture to 15 μl PCR product: 0.2 μl 10X ExoAP buffer (50 mM Bis-Tris, 1mM MgCl$_2$, 0.1 mM ZnSO$_4$), 0.05 μl 5000 U/ml Antarctic Phosphatase (New England Biolabs: Ipswich, MA), 0.05 μl 20000 U/ml Exonuclease I, and 3.7 μl sterile H$_2$O) and thermal-cycler incubation (37°C/60 minutes; 85°C/15 minutes). Sequencing reactions were performed using BigDye Terminator Reactions following the manufacturer's protocol (Applied Biosystems: Foster, CA) with the primer sets used for amplifications. For COI, the thermal-cycler reaction was performed as: 94°C/30 seconds followed by 25 cycles (94°C/15 seconds; 50°C/15 seconds; 60°C/3 minutes). The PCR conditions for 16S were identical to those described for COI, but with the use of 52°C and 64°C annealing temperatures. The sequencing reaction products were purified with the AMPure magnetic bead system following the manufacturer's protocol (Agencourt: Morrisville, NC) and were subsequently run on an ABI 3730x1 DNA Analyzer (Applied Biosystems International).

The sequence strands for each gene were proofread and assembled with CodonCode Aligner, version 3.7.1 (CodonCode Corporation, Dedham, MA, USA), to produce a continuous fragment. Sequences were compared with those in GenBank using the nucleotide BLAST program (NCBI Basic Alignment Search Tool) and manually aligned in BioEdit (Hall, 1999). Phylogenetic trees were constructed with *MEGA*5 (Tamura *et al.*, 2011) using the neighbour-joining (NJ) (Saitou & Nei, 1987) and maximum-likelihood (ML) (Kimura, 1980) methods on a 588-base pair (bp) alignment for COI. The bootstrap values were calculated on 1000 re-sampling replicates.

GenBank accession numbers for partial sequences of the COI and 16S regions are JQ837265 and JQ837266 respectively.

RESULTS

SYSTEMATICS

Order DECAPODA Latreille, 1802
Infraorder CARIDEA Dana, 1852
Superfamily ALPHEOIDEA Rafinesque, 1815
Family HIPPOLYTIDAE Spence Bate, 1888
Genus *Lebbeus* White, 1847
Lebbeus virentova sp. nov. (Figures 1 – 5)

MATERIAL EXAMINED

Holotype: female, CL 15.4 mm. VDVF, MCSC, Caribbean Sea; co-ordinates: 18°37.661′N 81°79.81′W; water depth: 2294 m [USNM 1183692]. Collected on the 18th voyage (16th leg) of RV 'Atlantis', on 19 January 2012.

Paratypes: six females, CL 11.12 – 15.6 mm [USNM 1183693 – 1183698]. Same data as holotype.

DESCRIPTION

Body moderately robust; integument glabrous.

Rostrum (Figures 1, 2A, B) straight, directed forward, 0.30 – 0.48 CL; reaching beyond mid-length but not to distal margin of first segment of antennular peduncle; laterally compressed, tapering to bifurcate apex; dorsal margin armed with 4 – 6 teeth (2 – 3 widely spaced teeth on rostrum proper; 2 – 3 larger, widely spaced postrostral teeth), posteriormost tooth arising at 0.16 – 0.26 CL; ventral margin armed with 2 – 4 teeth in distal 0.25, ventral lamina poorly developed. Carapace (Figures 1, 2A, B) with low but distinct median portrostral carina extending to posterior two-thirds of carapace; dorsal profile in lateral view gently convex. Supraorbital tooth strong, arising level with posterior margin of orbit, directed forward, reaching tip of suborbital lobe and antennal tooth; deep V-shaped notch inferior to base of supraorbital tooth. Orbital margin weakly concave; suborbital lobe bluntly triangular. Antennal tooth well-developed, acute, reaching tip of suborbital lobe. Pterygostomial tooth small, not reaching antennal tooth. Anterolateral margin between antennal tooth and pterygostomial tooth strongly sinuous with deep excavation below antennal tooth.

Abdomen (Figure 1) rounded dorsally. Second somite with transverse groove on tergum, bordered posteriorly by low ridge; posterodorsal margin of third somite produced; pleura of anterior three somites unarmed marginally, posteroventral margin rounded; fourth pleuron with posteroventral tooth (Figure 2C); fifth pleuron bearing moderately strong posteroventral tooth and numerous long setae on ventral margin (Figure 2C). Sixth somite 1.35 – 1.95 times longer than fifth somite; armed with small posteroventral tooth; posterolateral process terminating in acute tooth.

Telson (Figures 1, 2D, E) length 3.10 – 4.31 times anterior width, 1.25 – 1.48 times longer than sixth abdominal somite in dorsal midline; lateral margins parallel in anterior third, tapering posteriorly to convex posterior margin, bearing 3 – 6 (usually 4) dorsolateral spines on each side; posterior margin with 2 pairs of lateral spines (mesial pair longer), 4 – 6 median spiniform setulose setae and several longer thin plumose setae (Figure 2E).

Uropods (Figures 1, 2D) with broad rami exceeding distal margin of telson; exopod with distinct transverse suture and small spine at distolateral angle; endopod shorter and narrower than exopod; posterolateral projection of protopod triangular with acute tip.

Eyes (Figures 1, 2A) subpyriform with stalk narrowing proximally; cornea distinctly wider than stalk, its maximum width 0.13 – 0.15 times CL; ocellus absent.

Antennular peduncles (Figures 1, 2A) extending approximately to distal 0.2 of antennal scale. First segment as long or slightly longer than distal two segments combined, not quite reaching mid-length of antennal scale, dorsodistal margin armed with 2 or 3 (sometimes bifid) slender teeth; stylocerite reaching or slightly exceeding dorsodistal margin of first peduncular segment, terminating in acute point, mesial margin sinuous. Second segment approximately 0.4 length of first segment; bearing strong distolateral tooth. Third segment less than half as long as second; with small dorsodistal tooth. Lateral flagellum with thickened aesthetasc-bearing portion approximately 0.3 times CL.

Antenna (Figures 1, 2F) with basicerite bearing small, acute ventrolateral tooth; carpocerite reaching to approximately distal 0.3 – 0.4 of antennal scale. Antennal scale 0.64
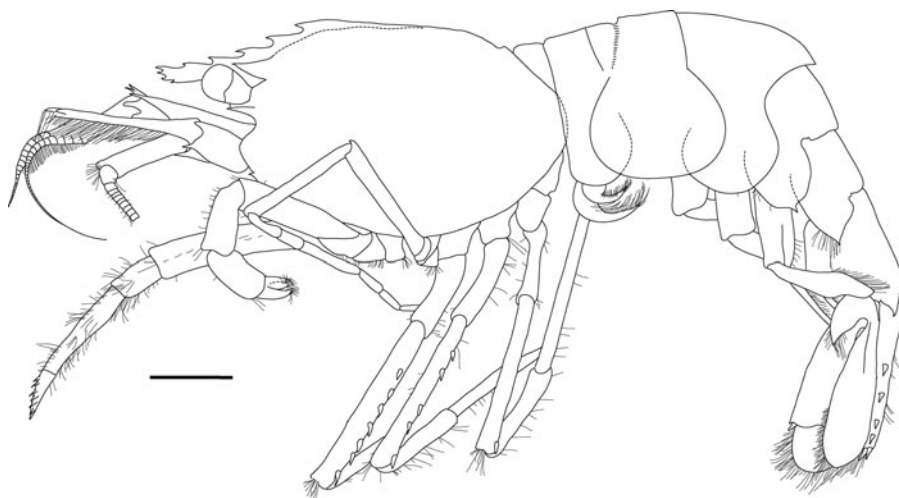

Fig. 1. *Lebbeus virentova* sp. nov., holotype, female (carapace length 15.4 mm), [USNM 1183692], from the Von Damm Vent Field, Mid-Cayman Spreading Centre: entire animal, lateral view. Scale bar = 5 mm.
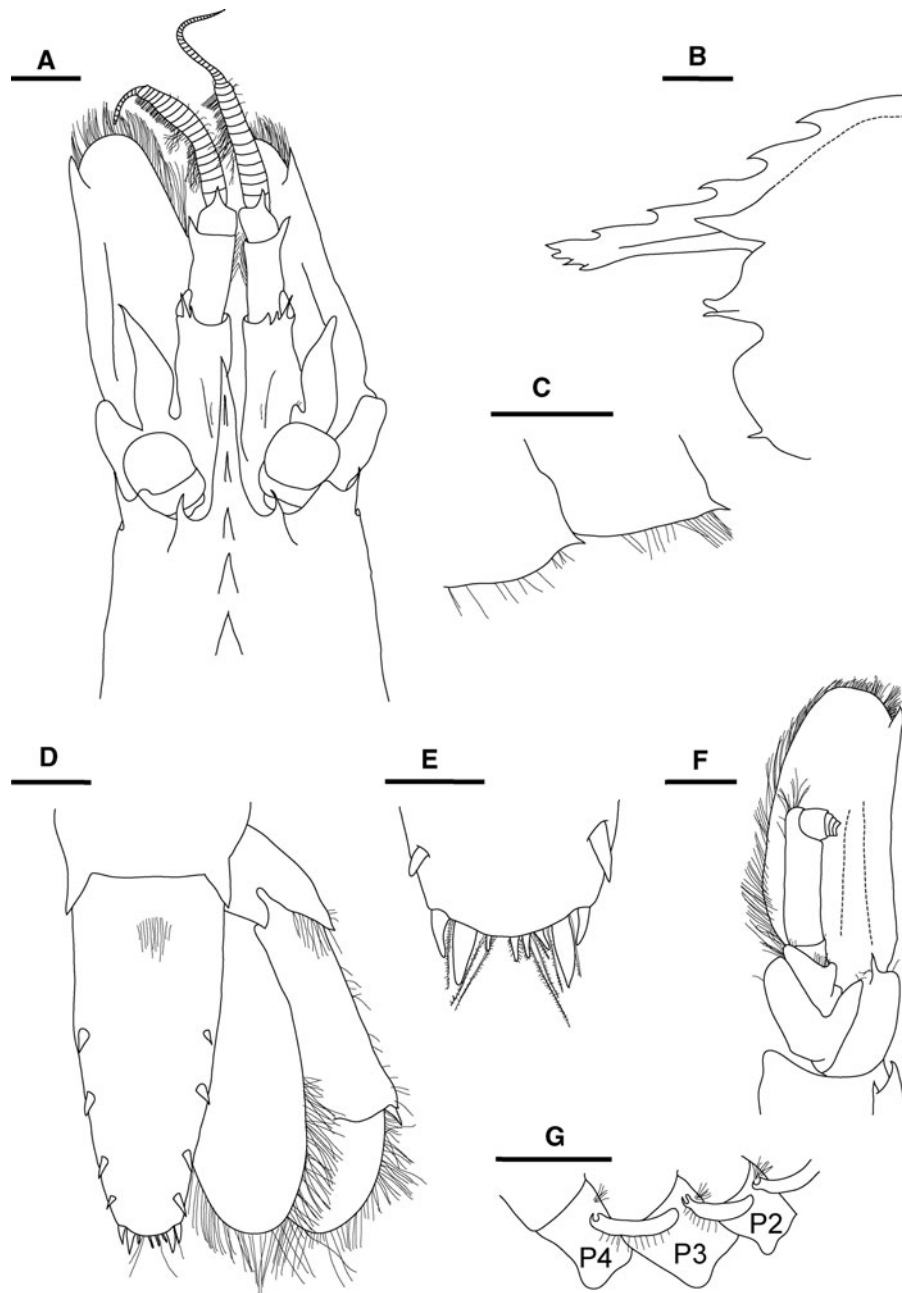
Fig. 2. *Lebbeus virentova* sp. nov., holotype, female (carapace length 15.4 mm), [USNM 1183692], from the Von Damm Vent Field, Mid-Cayman Spreading Centre: (A) anterior part of carapace and cephalic appendages, dorsal view; (B) anterior part of carapace, lateral view; (C) posterolateral margins of left pleura of fourth and fifth abdominal somites, lateral view; (D) telson and right uropod, dorsal view; (E) posterior part of telson, dorsal view; (F) Left antennal peduncle and scale, ventral view; (G) coxae of right second to fourth pereopods, showing presence of epipod on third pereopod and corresponding setobranch on fourth pereopod, lateral view. Scale bars: A–D, G, H = 2 mm; E = 1 mm.

times CL, 3 times longer than wide; lateral margin straight; distolateral tooth nearly reaching rounded distal lamella of blade.

Mouthparts (Figure 3) similar to those of other species of the genus. Mandible (Figure 3A) composed of flattened incisor, stout molar and biarticulate palp; incisor process bearing 2 acute distal teeth and 2 fine setae on mesial margin; molar process subcylindrical with obliquely truncate grinding surface and area of dense setae distally; palp curved, basal article broad with few short setae, distal article bearing many long setae.

Maxillule (first maxilla) (Figure 3B) with well-developed endites; coxal endite bearing numerous long setae; basial endite with row of stiff setae and row of spines along the mesial margin; palp curved weakly, slightly bilobed, bearing several distal setae.

Maxilla (second maxilla) (Figure 3C) with bilobed upper endite, fringed with many setae and flanked by well-developed palp with two distal setae; lower endite reduced, bearing several long setae; scaphognathite well developed, with rounded posterior lobe fringed with numerous setae on all margins.

First maxilliped (Figure 3D) with well-developed endites fringed with setae; palp biarticulate; exopod with caridean lobe; epipod large, bilobed.

Second maxilliped (Figure 3E) with broad ultimate segment fringed with stiff setae; ischial segment with excavated mesial margin; exopod and epipod well-developed.
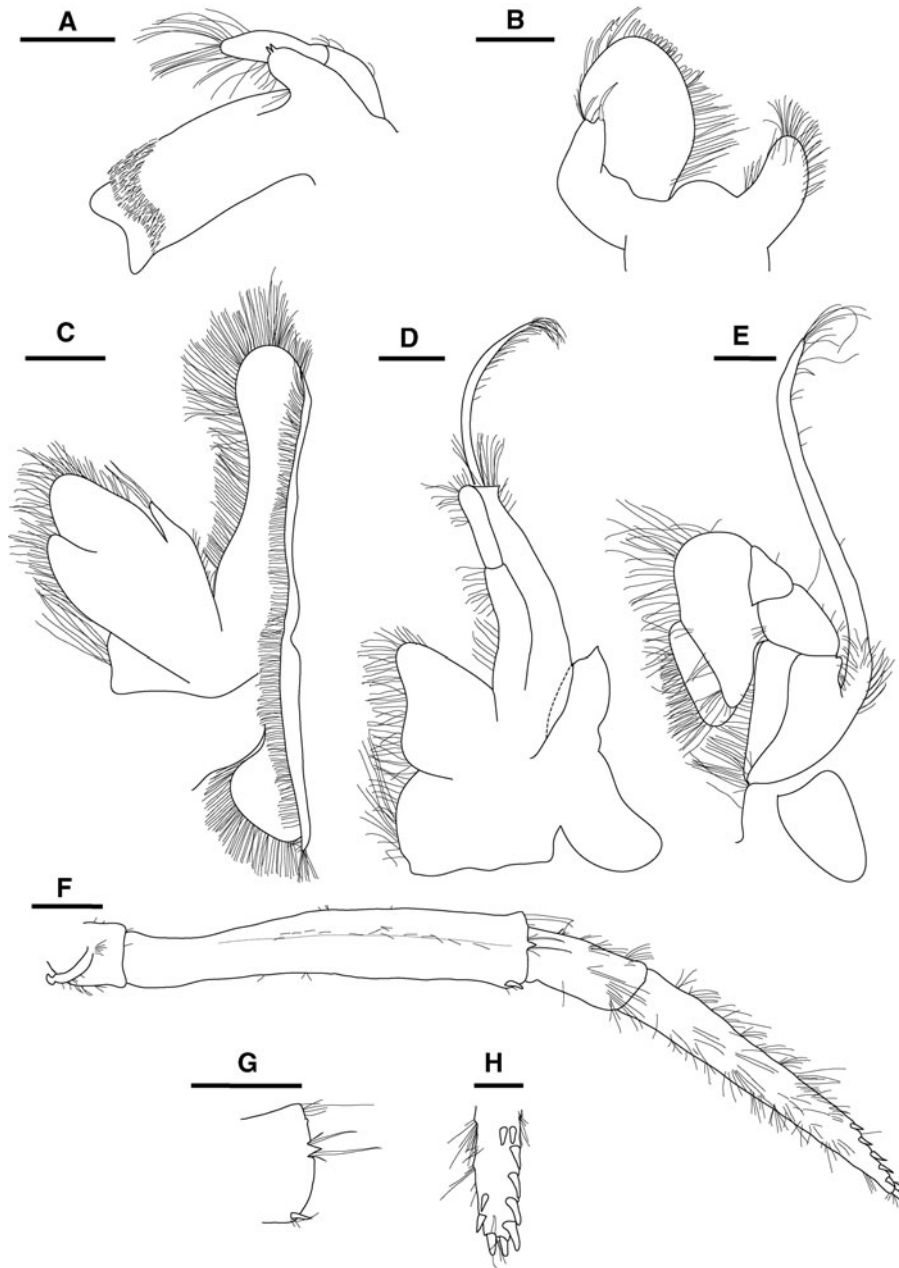
Fig. 3. *Lebbeus virentova* sp. nov., holotype, female (carapace length 15.4 mm), [USNM 1183692], from the Von Damm Vent Field, Mid-Cayman Spreading Centre: (A) left mandible, ventral view; (B) left maxillule (first maxilla), dorsal view; (C) left maxilla (second maxilla), ventral view; (D) left first maxilliped, ventral view; (E) left second maxilliped, ventral view; (F) right third maxilliped, lateral view; (G) distal part of antepenultimate segment of right third maxilliped, dorsal (extensor) view; (H) distal part of ultimate segment of right third maxilliped, dorsal view. Scale bars: A – E, H = 1 mm; F, G = 2 mm.

Third maxilliped (Figure 3F) exceeding antennal scale by half length of ultimate segment. Antepenultimate segment approximately 0.8 times as long as two distal segments combined; armed with a small tooth and two long spiniform setae on distolateral margin and a small spine at ventrodistal angle (Figure 3G); lateral surface bearing row of spiniform setae on blunt ridge parallel to dorsal margin. Ultimate segment approximately three times longer than penultimate segment, with dense tufts of setae; tapering distally, with short row of corneous spines distomesially and distolaterally (Figure 3H).

Strap-like, terminally hooked epipods present on third maxilliped to third pereopod; corresponding setobranchs on first to fourth pereopods (Figure 2G).

First pereopod (Figure 4A) moderately stout, extending to distal margin of antennal scale. Chela (Figure 4B – E) approximately 1.6 as long as carpus; dactylus approximately 0.6 times as long as palm, strongly curved distally, terminating in two corneous claws with two smaller corneous claws arising inferior to terminal claws; fixed finger terminating in one corneous claw flanked by two smaller corneous claws. Carpus bearing grooming apparatus (a feature widely spread in the Hippolytidae, e.g. Bauer, 1978), comprising a dense patch of serrate setae arising from a recessed area on mesial face.

Second pereopod (Figure 4F) distinctly more slender than first, overreaching antennal scale by approximately 0.33 length of carpus when extended. Chela (Figure 4G) small; dactylus
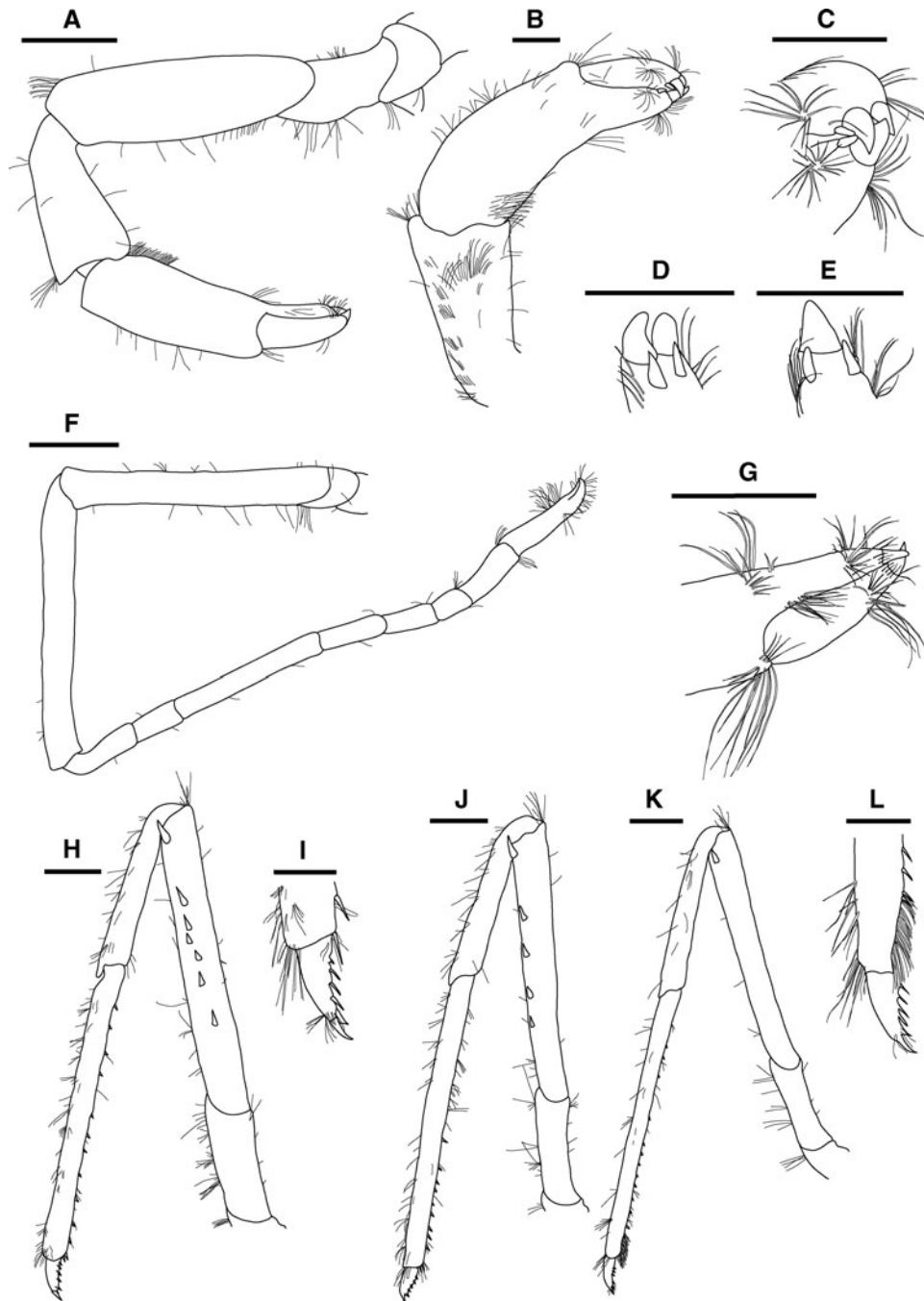
Fig. 4. *Lebbeus virentova* sp. nov., holotype, female (carapace length 15.4 mm), [USNM 1183692], from the Von Damm Vent Field, Mid-Cayman Spreading Centre: (A) left first pereopod, lateral view; (B) chela and carpus of left first pereopod, mesial view; (C) distal part of chela of left first pereopod; (D) same, tip of dactylus, inner view; (E) tip of fixed finger, inner view; (F) left second pereopod, lateral view; (G) chela of left second pereopod, lateral view; (H) left third pereopod, lateral view; (I) same, dactylus and distal part of propodus, lateral view; (J) left fourth pereopod, lateral view; (K) left fifth pereopod, lateral view; (L) same, dactlyus and distal part of propodus, lateral view. Scale bars: A, F, H, J, K = 2 mm; B–E, G, I, L = 1 mm.

terminating in two corneous claws; fixed finger terminating in one corneous claw. Carpus divided into seven articles.

Third to fifth pereopods (Figure 4H–L) similar in structure, long and slender, normally folded at mero-carpal articulation, decreasing in length and stoutness posteriorly. Third pereopod (Figure 4H, I) overreaching antennal scale by approximately 0.9 length of propodus; dactylus 0.14 length propodus, terminating in acute unguis and armed with 5 or 6 accessory spinules on flexor margin, distalmost spinule distinctly larger than others, making dactylus tip appear

biunguiculate; carpus approximately 0.6 as long as propodus; propodus with 2 rows of ventral accessory spinules; merus armed with 3–7 lateral spines.

Fourth pereopod (Figure 4J) overreaching antennal scale by approximately 0.6 length of propodus; dactylus with 5 or 6 accessory spinules on flexor margin; propodus with two rows of ventral flexor spinules; merus armed with 3–5 lateral spines.

Fifth pereopod (Figure 4K) overreaching antennal scale by approximately 0.2 length of propodus; dactylus with 5 or 6 accessory spinules on flexor margin (Figure 4L); propodus

with two rows of ventral flexor spinules; merus armed with 1 – 2 lateral spines.

Female pleopods similar to those of other species of the genus, without distinctive feature.

COLORATION IN LIFE (FIGURE 5)

Carapace bright red anteriorly, becoming paler distally; gonad green and visible through carapace. Abdomen pale, scattered with red chromatophores, making abdomen appear pinkish red. Rostrum and cephalic appendages translucent. Cornea darkly pigmented. Third maxilliped red with darkly pigmented corneous pinules on ultimate article. Pereopods red with thin white bands at joints; chelae of first two pereopods terminating in darkly pigmented corneous claws; corneous spines and ungui on dactyli of third to fifth pereopods also darkly pigmented. Gills typically bright white and visible through carapace.

Eggs green.

COMPARATIVE REMARKS

Lebbeus virentova sp. nov. belongs within the group of *Lebbeus* species characterized by the presence of epipods on the anterior three pairs of pereopods and absence of armature on the anterior three abdominal pleura. It is closest in morphology to the following species: *L. antarcticus* (Hale, 1941); *L. carinatus* Zarenkov, 1976; *L. cristatus* Ahyong, 2010; *L. formosanus* Chang, Komai & Chan, 2010; *L. kuboi* Hayashi, 1992; *L. microceros* (Krøyer, 1841); *L. pacmanus* Komai, Tsuchida & Segonzac, 2012; *L. polyacanthus* Komai, Hayashi & Kohtsuka, 2004; *L. shinkaiae* Komai, Tsuchida & Segonzac, 2012; *L. similior* Komai & Komatsu, 2009; *L. thermophilus* Komai, Tsuchida & Segonzac, 2012; *L. washingtonianus* (Rathbun, 1902); and *L. wera* Ahyong, 2009 (see Komai *et al.*, 2012 for updated distribution data on these species).

Characters shared between these species and *Lebbeus virentova* sp. nov. include: rostrum styliform, not reaching distal margin of second segment of antennular peduncle, armed with four or more dorsal teeth including postrostral teeth and more than one ventral tooth; distinct u- or v-shaped notch inferior to base of supraorbital tooth; sinuous anterolateral margin of carapace between antennal and pterygostomial teeth and deep excavation below antennal tooth; first segment of antennal peduncle bearing more than one tooth on dorsodistal margin; dactyli of posterior three pairs of pereopods distinctly biungulate.

Morphological differences between *Lebbeus virentova* sp. nov. and allied species are summarized below. The
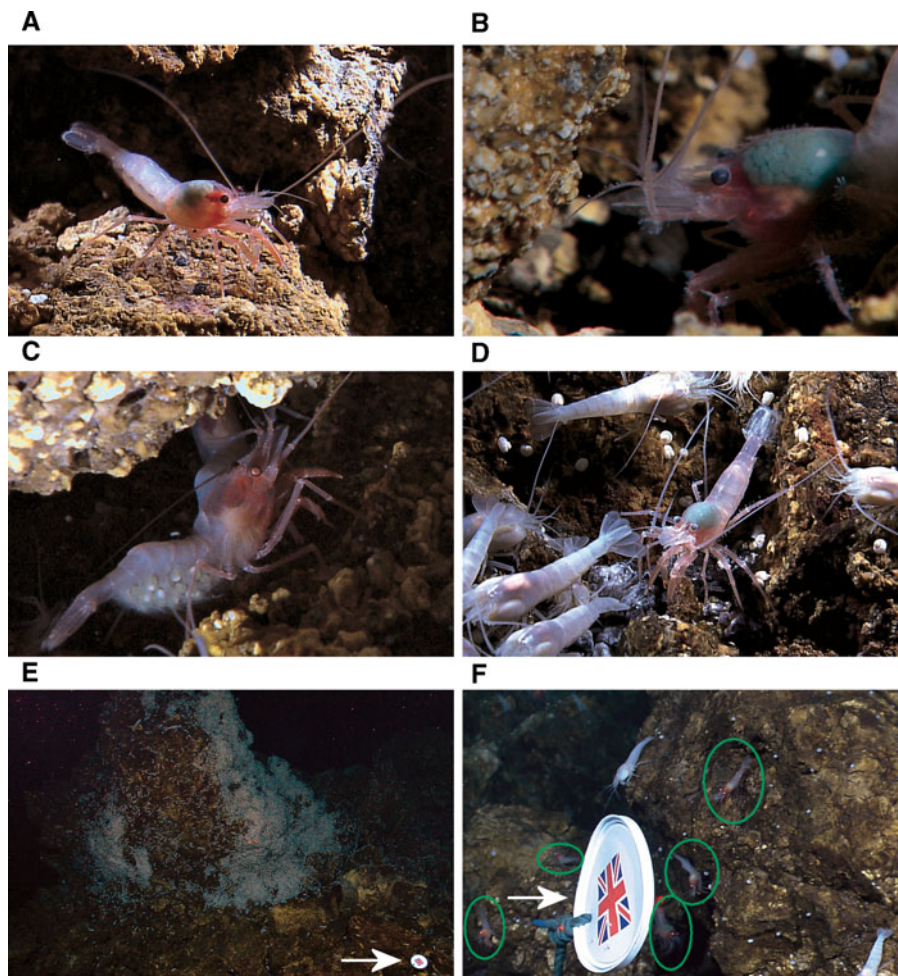

Fig. 5. *Lebbeus virentova* sp. nov., live *in situ*, the Von Damm Vent Field, Mid-Cayman Spreading Centre, ∼2300 m: (A, B) female with green eggs visible through carapace; (C) berried female; (D) *L. virentova* sp. nov. (centre) surrounded by specimens of the alvinocaridid shrimp *Rimicaris hybisae* Nye, Copley, Plouviez, 2012 and skeneid gastropods; (E) base of the spire, arrow points to JC44 navigational marker; (F) base of the spire (close-up), arrow points to JC44 navigational marker, green ellipses highlight occurrences of *L. virentova* sp. nov. Images A – D courtesy of the NOAA Okeanos Explorer Program, MCR Expedition 2011.

comparisons are limited to females because there is no information available on males of the new species.

Lebbeus virentova sp. nov. most closely resembles *L. carinatus*, *L. cristatus*, *L. formosanus*, *L. kuboi*, *L. microceros* and *L. thermophilus* in the stylocerite reaching or slightly overreaching the dorsodistal margin of the first segment of the antennular peduncle and having relatively few dorsal rostral teeth (six or less). *Lebbeus virentova* sp. nov. is separated from *L. carinatus* by the longer carpocerite (reaching to distal 0.3–0.4 of antennal scale versus reaching its midlength), longer third maxilliped (overreaching antennal scale by half length of ultimate segment versus reaching just beyond it) and longer first perereopod (reaching distal margin of antennal scale versus falling short of it).

The new species is distinguished from *Lebbeus cristatus*, *L. formosanus* and *L. kuboi* by the longer antennular peduncle (reaching base of distolateral tooth of antennal scale versus not reaching it), the shorter distolateral tooth of the antennal scale (not reaching the lamella versus reaching it), and number of meral spines on the posterior three pairs of pereopods. It is differentiated further from *L. cristatus* and *L. formosanus* by the shorter third maxilliped (overreaching antennal scale by half length of ultimate segment versus overreaching it by two-thirds and one-third respectively) and the presence of a posteroventral tooth on the fourth abdominal pleuron (versus absent and variable), and from *L. kuboi* by the straight (versus curving dorsally) rostrum and greater number of dorsal teeth (4–6 versus 2–4).

Lebbeus virentova sp. nov. is distinguished easily from the Atlantic species *L. microceros* by the shorter stylocerite (not reaching or slightly overreaching dorsodistal margin of second segment of antennular peduncle), the longer antennular peduncles (reaching base of distolateral tooth of antennal scale versus not reaching it), longer carpocerite reaching to distal 0.3–0.4 of antennal scale versus reaching its midlength), small tooth (versus strong, curved tooth) on the distolateral margin of the antepenultimate article of the third maxilliped, and fewer meral spines on the fifth pereopod (1–2 versus 3). The new species differs from *L. thermophilus* by the presence of a posteroventral tooth on the fourth abdominal pleuron (versus variable), the longer antennular peduncles (reaching base of distolateral tooth of antennal scale versus far falling short of it), the longer first pereopod (reaching distal margin of antennal scale versus falling short of it), and the presence of plumose setae on the posterior margin of the telson.

Lebbeus virentova sp. nov. is separated from *L. polyacanthus*, *L. shinkaiae* and *L. wera* by fewer dorsal rostral teeth (4–6, including 2–3 postrostral versus 6 or more, including 3 or more postrostral), the presence of plumose setae on the posterior margin of the telson, and number of teeth on the meri of the posterior three pereopods.

The new species is differentiated from *Lebbeus antarcticus*, *L. pacmanus*, *L. similior* and *L. washingtonianus* by the longer stylocerite (reaching or slightly overreaching the dorsodistal margin of the first segment of the antennular peduncle versus not) and the presence of plumose setae on the posterior margin of the telson. In addition, the first pereopod of *L. virentova* sp. nov. is longer than that of *L. pacmanus* and *L. similior*, and shorter than that of *L. antarcticus* (reaching distal margin of antennal scale versus falling short of it, overreaching it by length of fingers). Furthermore, *L. virentova* sp. nov. has a longer carpocerite than *L. antarcticus* and *L. similior*, and shorter carpocerite than *L. pacmanus*

and *L. washingtonianus* (reaching to distal 0.3–0.4 of antennal scale versus reaching its mid-length, reaching distal 0.2 of antennal scale).

DISTRIBUTION AND HABITAT

Presently known only from the type locality, the VDVF, MCSC, Caribbean Sea, in 2294–2375 m water depth. See Connelly *et al.* (2012) for a description of the geological, geochemical and biological setting of the VDVF.

Observed on the edifice spire in close proximity to actively venting orifices with high abundances of the alvinocaridid shrimp *Rimicaris hybisae* Nye, Copley & Plouviez, 2012, and below the spire on the sulphide mound with *R. hybisae*, *Alvinocaris* sp., skeneid gastropods, zoarcid fish and siboglinid polychaetes.

ETYMOLOGY

The species name, *virentova*, is the combination of the Latin, *vireo* (= be green), and *ova* (= eggs), in reference to the green eggs of the new species.

MOLECULAR PHYLOGENY

Partial sequences of the COI (683 bp) and 16S (523 bp) regions of *Lebbeus virentova* sp. nov. were consistent amongst specimens. Fixed and unique mutations were evident in the partial sequences of the COI and 16S regions in comparison with all other species in the GenBank database. The only partial sequence of the 16S region for *Lebbeus* in GenBank is from *L. virentova* sp. nov. [JQ837266].

Based on NJ and ML phylogenetic analyses for COI sequences available in GenBank, *Lebbeus virentova* sp. nov. exhibits the smallest evolutionary distance (6.5% divergence) to the species recorded therein as '*L. carinatus*' from 13° North on the East Pacific Rise (EPR) [AF125421.1 and AF125422.1]. *Lebbeus carinatus* Zarenkov, 1976 was described from off Peru and has not been recorded from the EPR, whereas *L. laurentae* is known only from 13° North at the EPR. *Lebbeus laurentae* is a replacement name for *L. carinatus* de Saint Laurent, 1984 (a junior homonym of *L. carinatus* Zarenkov, 1976). It is apparent therefore that '*L. carinatus*' [AF125421.1 and AF125422.1] is *L. laurentae*. Based on a 588-bp alignment, NJ and ML methods produced identical topologies and place the new species in the same clade as *L. laurentae* (100% and 96% bootstrap support for NJ and ML methods respectively) (Figure 6).

DISCUSSION

Morphological analysis of this hippolytid shrimp reveals it to be a new species in the genus *Lebbeus*. Based on morphology, the new species belongs to the species group characterized by the presence of epipods on the anterior three pairs of pereopods, stylocerite reaching or slightly overreaching the dorsodistal margin of the first segment of the antennular peduncle, and six or fewer dorsal rostral teeth. It is distinguished from other species by a combination of morphological features (see above). Consistency in partial sequences of the COI mDNA and 16S rDNA genes between specimens from the VDVF confirms that they belong to a single species, and the presence of unique and fixed mutations in the sequences indicate that they are genetically distinct from all other species in the GenBank database.
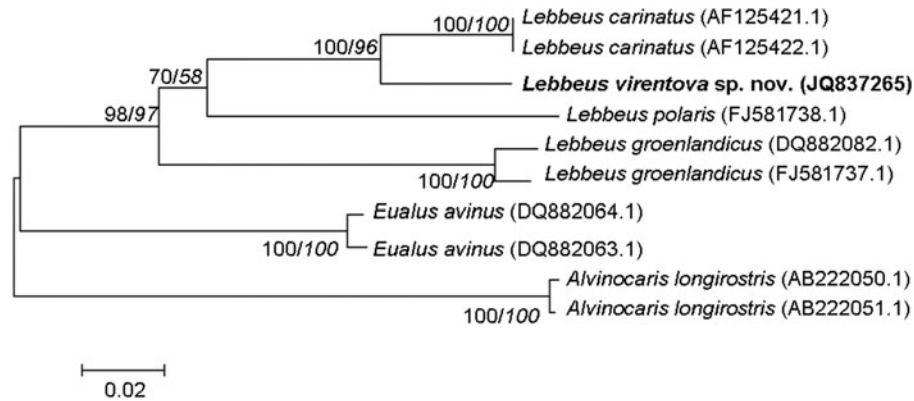
Fig. 6. Neighbour-joining tree of *Lebbeus* based on a 588-base pair alignment of partial nucleotide sequences from the mitochondrial COI DNA region with *Eualus avinus* (Rathbun, 1899) (Hippolytidae) and *Alvinocaris longirostris* Kikuchi & Ohta, 1995 (Alvinocarididae) as outgroups. Evolutionary distance computed using the Jukes–Cantor method (Jukes & Cantor, 1969) is represented by branch length; scale bar is proportional to inferred nucleotide divergence. Bootstrap support calculated on 1000 re-sampling replicates is shown by the numbers along the branches (neighbour-joining, roman text; maximum likelihood, italic text). GenBank accession numbers are given after species names.

Seven species of *Lebbeus* have been recorded previously from hydrothermal vents, six of which are only known from a vent environment (Table 1); they may be vent-endemic (Komai *et al.*, 2012). These six species are from vent fields on the EPR and in western Pacific back-arc basins at 691–2640 m water depth (Table 1). The new species therefore appears to be the first hippolytid shrimp to be described from a vent field outside the Pacific Ocean, and may be the first record of the genus in the Caribbean.

The recent discovery of hydrothermal vents and chemosynthetic assemblages on the MCSC has provided an opportunity to enhance existing knowledge about biodiversity in the deep sea. *Lebbeus virentova* sp. nov. is the third taxon described from the VDVF, where it co-occurs with the alvinocaridids *Rimicaris hybisae* and *Alvinocaris* sp. Based on observations made from two research cruises to two vent fields of the MCSC, the new species is so far only known from the VDVF. In contrast, *R. hybisae* is present and abundant at the VDVF and the Beebe Vent Field (BVF). The BVF is only 30 km from the VDVF, but is 2660 m deeper and has different geological and geochemical settings (Connelly *et al.*, 2012).

The genus *Rimicaris* Williams & Rona, 1986 is a deepwater (1700–4960 m) genus known exclusively from hydrothermal vents (Nye *et al.*, 2012). The genus *Lebbeus* exhibits a shallower bathymetric range, from the littoral zone to at least 2640 m, and is not endemic to hydrothermal vents (e.g. Squires, 1990; Hayashi, 1992; De Grave & Fransen, 2011). The presence of *L. virentova* sp. nov. at the VDVF, and its absence from the BVF, suggest that water depth and/or environmental conditions may determine its distribution among MCSC vent fields. Further characterization of the faunal composition of assemblages at the vent fields at the MCSC will elucidate the vent biogeography of this region.

Approximately half of all species of *Lebbeus* have been described from the north-west Pacific (De Grave & Fransen, 2011), suggesting a possible centre of radiation for the genus in that region (e.g. Vavilov, 1926). An extensive and comprehensive molecular phylogenetic analysis of the genus *Lebbeus* and higher taxa, requiring the collection and molecular analyses of further specimens, is a prerequisite for clarifying the phylogenetic relationships, evolutionary history and geographical distribution of this genus.

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