# Carnarvonis gen. nov. and Warregoensis gen. nov.: Two New Genera and Species of Subterranean Amphipods (Crangonyctoidea: Chillagoeidae) Described from North-eastern Australia

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ABSTRACT. Biological surveys are crucial and appropriate approaches for capturing invertebrate biodiversity data and specimens, particularly considering that a significantly high proportion of Australian invertebrates are thought to be undescribed. During the 2014 Carnarvon Station Reserve Bush Blitz Biological Survey, specimens of stygobiotic amphipod crustaceans were collected from a spring-fed groundwater site. Taxonomic analyses revealed that the specimens comprised two new genera and species of stygobiotic amphipods belonging to the family Chillagoeidae. Further examination required a revision of the family, which is presented herein along with the descriptions of *Carnarvonis katjae* gen. et sp. nov. and *Warregoensis lowryi* gen. et sp. nov. Importantly, the description of two new genera and species increases the total number of described stygobiotic amphipods in Queensland to three genera and species and indicates that additional and potentially diverse stygobiont taxa are likely to be discovered in the largely unexplored groundwater habitats across the region.

## Introduction

A biological survey of groundwater associated sites (springs, marshes, gravel beds) on Carnarvon Station Reserve, *ca.* 600 km west of Bundaberg in central Queensland, Australia, was carried out in October 2014 as part of a Bush Blitz Biological Survey (https://bushblitz.org.au). This survey was coordinated by Bush Blitz (an Australian Government, BHP, and Earthwatch Australia partnership), partnered with Bush Heritage Australia and the Queensland Museum (Commonwealth of Australia, 2017). From a single spring-fed groundwater site, approximately 20 specimens comprising two separate species of stygobiont crangonyctoid

amphipod crustaceans were collected. Initial identifications concluded that these were "paramelitid-like" amphipods based largely on the morphology of their gnathopods, gills, and uropods, but with a unique looking uniramous uropod 3.

The Paramelitidae comprise a significant component of the Australian freshwater amphipod fauna, with 14 currently described genera encompassing stygobiont and epigean taxa (Bradbury & Williams, 1999; Lowry & Myers, 2012). Yet, this family has historically remained ambiguously defined from at least two other existing freshwater amphipod families (Neoniphargidae Bousfield, 1977 and Perthiidae Williams & Barnard, 1988 (Bradbury & Williams, 1999)). Lowry & Myers (2012, 2013) attempted to extensively redefine and

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Keywords: Crustacea, subterranean Amphipoda, Carnarvonis, Warregoensis, stygobiotic amphipods, Queensland

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diagnose amphipod families using morphological cladistic analyses, presenting new diagnoses and arrangements for Paramelitidae, Neoniphargidae and Perthiidae, yet these families remain morphologically very similar, separated by a small selection of synapomorphic characters rather than being defined by unique characters. Importantly, Lowry & Myers (2012) also elevated three monotypic Australian stygobiont amphipod genera to their own new families (Chillagoeidae and Giniphargidae respectively for the former paramelitid taxa Chillagoe Barnard & Williams, 1995 from Queensland and Giniphargus Karaman & Barnard, 1979 from Victoria, and Uronyctidae for the former (though uncertainly placed) crangonyctid taxon Uronvctus Stock & Iliffe, 1990 from South Australia). Chillagoeidae, Giniphargidae, and Uronyctidae are currently the only Australian crangonyctoid families to possess a uniramous uropod 3.

The Chillagoeidae is represented by a single species, *Chillagoe thea* Barnard & Williams, 1995, and is the only described stygobiont amphipod from Queensland. It is found in the Chillagoe cave system in Far North Queensland, which is *ca.* 900 km north-northwest of Carnarvon Station Reserve. Given the geographic distances between Chillagoe Caves and Carnarvon Station Reserve, which are within entirely separate water catchments, and the distinct groundwater habitats involved, it is likely that the two species collected at Carnarvon Station Reserve are new species, separate to *Chillagoe thea*, albeit with more work needed to establish if they should be placed in the same family.

The aims of this paper are to comprehensively examine the two new stygobiont amphipod species discovered at Carnarvon Station Reserve using morphological and molecular data to: 1) produce robust species hypotheses; 2) attempt to place the new taxa within the current classification hierarchy; and 3) provide new species descriptions.

## Materials and methods

#### **Specimens**

Twenty amphipod individuals were collected at one groundwater habitat site within the Carnarvon Station Reserve: "Lady Spring", a spring-fed upwelling of groundwater within a gravel creek bed. Samples were collected using a Bou-Rouch pump and *ca.* 50 litres of water were pumped onto a 0.5 mm sieve from *ca.* 70 cm depth in the gravel bank. Specimens were immediately preserved in 95% EtOH and kept refrigerated for optimum preservation of tissue for molecular analyses.

An initial morphological examination of samples was carried out and provisional identification to "paramelitidlike" was made based on the morphology of the uropods (particularly the third uropod, but also the absence of robust basofacial setae on the peduncle of the first uropod). Morphological analyses involved micro-dissection, light microscopy and illustration using a Nikon Eclipse 80i (Nikon, Tokyo, Japan) compound microscope with a camera lucida attachment. Type material was dissected primarily along the left-hand side (as per traditional taxonomic practice) and mounted on temporary slides in glycerol for examination. Type material is deposited at the Queensland Museum, Brisbane (QM) and the South Australian Museum, Adelaide (SAMA); sequence data is available through GenBank.

#### DNA extraction, PCR, and sequencing

DNA was extracted from eight specimens: two or three percopods or pleopods removed from the right side of the animal whenever possible. DNA was extracted using a modified version of the Canadian Centre for DNA Barcoding Glass Fiber Plate DNA Extraction Protocol, with Insect Lysis Solution and an Acroprep ADVANCE 96-Well Filter Plate (1 ml 3.0  $\mu$ m Glass Fiber/ 0.2  $\mu$ m Supor; bio-strategy; Tullamarine, VIC, Australia) (Ivanova *et al.*, 2006). Partial sequences of *cytochrome c oxidase subunit I (COI)* were Polymerase Chain Reaction (PCR)-amplified using the degenerate Folmer primers COIF (TCWACNAAYCAYAARGAYATTGG) and COIR (ACYTCNGGRTGNCCRAARARYCA) (Folmer *et al.*, 1994).

PCR-amplification was carried out in 25  $\mu$ L reaction volumes containing 1 × Immolase PCR buffer (Meridian Bioscience; Cincinnati, Ohio, USA), 1.5 mM MgCl<sub>2</sub>, 0.8 mM dNTP mix, 0.05 mg/ml BSA, 0.24  $\mu$ M primer, 0.5 units Immolase DNA polymerase and approximately 1 ng of extracted DNA. Thermal cycling conditions for *COI* involved an initial hold at 94°C for 10 min followed by 38 cycles of denaturation at 94°C for 45 s, annealing at 48°C for 45 s and extension at 72°C for 60 s. Following the 38 cycles, a final extension step at 72°C for 6 min completed the reaction. PCR products were then verified by agarose gel. Cleanup of the PCR reaction and Big Dye Terminator sequencing in both directions was carried out at The Australian Genome Research Facility (AGRF).

Uncorrected pairwise sequence divergences were determined using PAUP\* version 4.0b8 (Swofford 2001).

#### Results

Detailed examination of the Carnarvon Station Reserve material based on morphological and molecular data indicated that two undescribed species were present. Gnathopod, coxal and uropodal morphology indicated that they should be placed within separate genera within the family Chillagoeidae. Type specimens of *Chillagoe thea* as well as additional material were borrowed from the Australian Museum (AM) (holotype AM P44066, paratypes AM P44071, and other cited material AM P44072) for comparative analyses of all three taxa to now be included within that family, and an updated diagnosis of the family with additional characters is presented herein.

Uncorrected pairwise sequence divergence of *COI* data indicate that *Warregoensis lowryi* sp. nov. (GenBank *COI*: OP596266.1, OP596267.1, OP596268.1, OP596269.1, OP596270.1) and *Carnarvonis katjae* sp. nov. (GenBank *COI*: OP596271.1, OP596272.1, OP596273.1) are at least 15% divergent from each other and 22.7–23.2% divergent from *Chillagoe thea* sequences (Barcode of Life database (BOLD: MSAPB2517–19.COI-5P, MSAPB2518–19. COI-5P, https://www.boldsystems.org/#). Amphipod species and genus divergences have been recorded as between 5–20% for *COI* (Havermans *et al.*, 2013; King *et al.*, 2022) and so these divergence values correlate well with both species and genus level determinations.

Amphipoda

Crangonyctoidea Bousfield, 1973

# Chillagoeidae Lowry & Myers, 2012

Type genus. Chillagoe Barnard & Williams, 1995

Diagnosis. (After Lowry & Myers, 2012) Body laterally compressed. Eyes absent. Antennae 1-2 calceoli crangonyctoid (type 9). Antenna 1 longer than antenna 2; peduncular article 1 subequal to, or longer than article 2; article 2 longer than article 3; article 3 shorter than article 1; peduncular articles 1-2 not geniculate; accessory flagellum short. Mandible molar triturative; palp symmetrical. Maxilla 1 basal endite setose along medial margin or apically setose; palps symmetrical. Maxilla 2 basal endite with oblique setal row of 2-3 setae. Labium inner lobes vestigial or absent. Coxal gills on percopods 2-6 or 2-7, stalked (without proximal restriction); sternal gills present, simple, paired; sternal blisters absent; oostegites fringing setae simple. Coxa 1 distinctly smaller than coxa 2, as long as broad, with a robust seta at the posterodistal corner and additional simple seat along distal margin; Coxae 2-3 similar, longer than broad with a robust seta at the posterodistal corner and additional simple seat along distal margin; coxa 4 similar length to coxae 2–3, as long as broad, with small but distinct excavated corner on the posterior margin, with a robust seta at the posterodistal corner and additional simple setae along distal margin. Coxae 5-6 similar, approximately half length of coxae 2-4, anteroventral lobe distinct, posteroventral lobe indistinct, with setae along posterior margin. Coxa 7 similar length to coxae 5-6, rounded, with setae along posterior margin. Gnathopod 1-2 subchelate, similar in males and females (not sexually dimorphic); gnathopod 1 smaller (or weaker) than or similar in size to gnathopod 2; gnathopods 1-2 propodus lateral face with distinct excavation above and parallel to palm margin, palm without robust setae along palm margin, with rows of 1-5 robust setae either side of palm corner (where dactyl fits to the propodus). Pereopods 3–4 not sexually dimorphic. Pereopod 4 basis without distinct posteroventral lobe. Pereopod 5 shorter than percopod 6. Percopod 7 subequal in length to, or longer than percopod 6. Pleonites 1-3 without dorsal carinae. Urosomites 1-3 free; with slender or robust dorsal setae. Urosomite 1 with distoventral robust seta. Urosomites 1-2 with paired dorsolateral robust setae. Uropod 1 with or without basofacial robust setae on lateral margin of peduncle, distal margin of peduncle with group of 1 large and 2 small robust setae laterally and 1 large robust seta medially. Uropod 2 distal margin of peduncle with group of 2 small robust setae laterally and 1 small robust seta medially. Uropod 3 not sexually dimorphic; uniramous, without plumose setae. Telson cleft; apical robust setae present.

**Remarks**. Examination of type material for the family and comparison of this to material of the two new taxa described herein has led to a revision of diagnostic characters of the Chillagoeidae. New characters to the diagnosis are highlighted in bold text. Significant changes include the morphology of the maxilla 2 and the setation of the uropod

1 peduncle. Firstly, the original description of *Chillagoe thea* indicated that maxilla 2 basal endite was missing an oblique row of setae, which was included as a family-level character by Lowry & Myers (2012). However, subsequent examination of the holotype and paratype material showed that while there was variation between individuals, there was an oblique setal row of at least 2–3 setae that is also present in the new genera described herein. Secondly, in the original diagnosis of Chillagoeidae the presence of a robust basofacial seta on the uropod 1 peduncle was included as an important family-level character (Lowry & Myers, 2012) but we conclude that this character is variable among genera.

More importantly, we find that the distinct gnathopod, coxal and uropodal morphology of all three genera now included within the Chillagoeidae should be raised to family-level diagnoses. This includes the sculpturing of gnathopods 1–2 propodus lateral face (with distinct excavation above and parallel to palm margin) as well as setation of the palm corner (with rows of 1–5 robust setae either side of where the dactyl fits to the propodus; coxal morphology (coxae 1 short, coxae 2–4 similar length, coxae 5–7 half length of coxae 1–4); and the setation of uropods 1–2 peduncle distal margin (uropod 1 with group of 1 large and 2 small robust setae laterally and 1 large robust seta medially, uropod 2 with group of 2 small robust setae laterally and 1 small robust seta medially). Together, a suite of characters is formed that clearly defines the family.

Chillagoeidae taxa are most easily separated from the albeit poorly defined Paramelitidae by the uniramous uropod 3 as well as urosomites 1-2 with paired dorsal robust setae. Two other Australian groundwater-associated families are also characterized in-part by a uniramous uropod 3: Uronyctidae (erected for the monotypic Uronyctus longicaudus Stock & Iliffe, 1990 from sinkholes in Mt Gambier, South Australia) and Giniphargidae (erected for the monotypic Giniphargus pulchellus (Sayce, 1899) from Gippsland, Victoria). Both of these species show significantly more "vermiform" slender bodies typically associated with true stygobiotic lifestyle, compared to Chillagoeidae taxa. In addition, U. longicaudus further differs from Chillagoeidae taxa in that it has sexually dimorphic gnathopods with distinctive propodus morphology, coxal gills on pereopods 2-7, coxa 4 without a distinct excavated corner on the posterior margin, percopod 5 much shorter than percopods 6-7 and an extremely elongate uropod 3 outer ramus. G. pulchellus has reduced coxae 1-7 of similar lengths, a robust antenna 2 peduncle, gnathopods with distinctive propodus morphology, coxa 4 without a distinct excavated corner on the posterior margin, and an elongate uropod 3 outer ramus of two equally sized articles.

### Carnarvonis gen. nov.

urn:lsid:zoobank.org:act:C6E7BFF2-9532-4F36-8A53-63F5CBB104F5

Type species. Carnarvonis katjae sp. nov.

**Diagnosis**. Body laterally compressed, not vermiform. Eyes absent. Antenna 1 longer than antenna 2, accessory flagellum of two articles. Antenna 2 flagellum shorter than peduncle, calceoli absent. Mandibular palp of 3 articles, right and left palp with slightly different setation. Labium (lower lip) lacking inner lobes. Coxae 1–4 distinctly longer than coxae 5–7; coxa 1 distinctly shorter than coxae 2–3; coxa 4 with

small but distinct excavated corner on the posterior margin. Gnathopods 1–2 not distinctly large (propodus smaller than head length), not sexually dimorphic; gnathopod 2 propodus slightly longer than in gnathopod 1; gnathopods 1–2 propodus lateral face with distinct excavation above and parallel to palm margin, palm margins smooth, without robust setae along palm margin, with 1 mesial (inner) and 1–2 lateral (outer) robust setae either side of palm corner (where dactyl fits to the propodus). Pereopod 6 and 7 of similar lengths, bases broad (less than 1.5 times as long as wide). Coxal gills on pereopods 2–6, sternal gills on thoracic segments 2–6, slender, not greater than ½ length of coxal gills. Uropod 3 outer ramus of 1 article, inner ramus absent. Telson shallowly cleft, with apical setae.

**Etymology**. Named for the Carnarvon Station Reserve where the holotype material was collected. Gender: feminine.

**Remarks**. *Carnarvonis* gen. nov. and *Warregoensis* gen. nov. share some distinct traits from *Chillagoe*. They do not have a basofacial seta on uropod 1 and the uropod 3 outer ramus is composed of a single article (vs. 1 basofacial seta present on uropod 1 and uropod 3 outer ramus of two articles in *Chillagoe*). *Carnarvonis* gen. nov. can be clearly identified from the smooth palm margin of gnathopods 1–2, a shallowly cleft telson and pereopods 5–7 bases broad (less than 1.5 times as long as wide).

# Carnarvonis katjae sp. nov.

urn:lsid:zoobank.org:act:797AACEF-2E9E-46C4-BF7E-73A03D5FEA66

### Figs 1-3

**Holotype**: QM W29606 (RL 2296.6, Genbank *CO1*: OP596273.1), female, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014. **Paratypes**: QM W29607, male, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014; SAMA C14815 (RL 2296.5, Genbank *CO1*: OP596271.1, OP596272.1), males and juveniles, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014; SAMA C14815 (RL 2296.5, Genbank *CO1*: OP596271.1, OP596272.1), males and juveniles, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014.

Description. Holotype, female, 4.5 mm. Head. Without rostrum, eyes absent. Antenna 1 length less than half length of animal; peduncle with three articles progressively shorter; accessory flagellum of two articles, distal article small; flagellum of 12 articles. Antenna 2 approximately 1/2 length of antenna 1; peduncle longer than flagellum, articles 4-5 of peduncle of subequal length; flagellum of 4 articles. Labrum (upper lip) evenly rounded, setose apically. Labium (lower lip) without inner lobes. Mandible incisor with 5 teeth, lacinia mobilis with 4 teeth, setal row of 5 plumose setae present, molar narrow and denticulate; palp of three articles, article 3 shorter than article 2, right palp with short row of plumose setae ("D-setae"; after Lowry & Stoddart, 1993). Maxilla 1 outer plate with nine apical denticulate robust setae; inner plate with two apical plumose setae; palp of two articles, distal article longest, with apical setae. Maxilla 2 outer plate with two rows of simple and plumose setae apically and simple setae laterally; inner plate with simple setae apically and an oblique row of medial plumose setae. Maxilliped palp

slender (no articles enlarged), of four articles plus spine; outer plate with robust and simple setae along apical and lateral margin; inner plate with robust and plumose setae apically and laterally.

Pereon. Gills include coxal gills on percopods 2-6, sternal gills on thoracic segments 2–6, slender, not greater than  $\frac{1}{2}$ length of coxal gills. Gnathopods 1-2 not distinctly large (propodus smaller than head length). Gnathopod 1 coxa short (only slightly longer than wide), with robust seta at posterior corner and few simple setae scattered along margin; propodus length 1.5 times width, lateral face with distinct excavation above and parallel to palm margin, palm margins smooth (no distinct crenulations), without robust setae along palm margin, with 1 mesial (inner) and 1 lateral (outer) robust seta either side of palm corner (where dactyl fits to the propodus), posterior margin of propodus with 1 oblique row of long simple setae. Gnathopod 2 coxa longer than coxa 1, distinctly longer than wide, with robust seta at posterior corner and few simple setae scattered along margin; propodus length 1.8 times width, lateral face with distinct excavation above and parallel to palm margin, palm margins smooth (no distinct crenulations), without robust setae along palm margin, with 1 mesial (inner) and 1 lateral (outer) robust setae either side of palm corner (where dactyl fits to the propodus), posterior margin of propodus with 2 oblique rows of long simple setae. Pereopod 3 coxa similar length to coxa 2, longer than wide, with robust seta at posterior corner and few simple setae scattered along margin; dactylus with single seta. Pereopod 4 coxa similar length to coxa 2–3, longer than wide, small but distinct excavated corner on the posterior margin, with robust seta at posterior corner and few simple setae scattered along margin. Pereopod 5 coxa with rounded anteroventral lobe with seta, posteroventral lobe indistinct, with 1 seta on posterior margin. Pereopod 6 similar length to pereopod 7; coxa with rounded anteroventral lobe, posteroventral lobe indistinct, with 1 seta on posterior margin; basis 1.5 times as long as wide; propodus longer than carpus; dactylus with single seta. Pereopod 7 coxa rounded, with 1 seta on posterior margin; basis 1.6 times as long as wide; propodus longer than carpus; dactylus with single seta.

Pleon. Epimera 1-3 posteroventrally subquadrate, posteroventral corner with robust seta and posterior margins with several small robust setae along length; epimera 2-3 with robust setae along the ventral margin. Urosomite 1 with distinct distoventral robust seta, with paired dorsolateral setae on distal margin. Urosomite 2 with paired dorsolateral setae on distal margin. Uropod 1 peduncle without basofacial robust seta, distal margin of peduncle with group of 1 medium-large and 2 small robust setae laterally and 1 large robust seta medially. Uropod 2 distal margin of peduncle with group of 2 robust setae laterally and 1 small robust seta medially. Uropod 3 distal margin of peduncle with robust setae laterally and medially; outer ramus of 1 article, approximately 3 times length of peduncle, with clusters of 2–3 robust setae along length; inner ramus absent. Telson cleft shallowly, with robust and simple setae apically.

**Etymology**. Named for Dr Katja Hogendoorn, scientist and partner of Remko Leijs.

**Remarks**. *Carnarvonis katjae* sp. nov. is clearly a distinct genus and species with 15% CO1 divergence from *Warregoensis lowryi* sp. nov. and 22% CO1 divergence from *Chillagoe thea*.



**Figure 1**. *Carnarvonis katjae* sp. nov., holotype female, TL 4.5 mm, Carnarvon Station Reserve, Queensland, QM W29606. (*A*) whole animal in lateral view; (*B*) maxilla 1; (*C*) maxilla 2; (*D*) maxilliped; (*E*) mandible; (*F*) upper lip; (*G*) lower lip. Scale (*A*): 0.1 mm.



Figure 2. *Carnarvonis katjae* sp. nov., holotype female, TL 4.5 mm, Carnarvon Station Reserve, Queensland, QM W29606. (*A*) gnathopod 1; (*B*) gnathopod 2.

## Warregoensis gen. nov.

urn:lsid:zoobank.org:act:5B239C3B-3E9D-4CB2-AB23-9D139C0AE8B2

#### Type species. Warregoensis lowryi sp. nov.

Diagnosis. Body laterally compressed, not vermiform. Eyes absent. Antenna 1 longer than antenna 2, accessory flagellum of two articles. Antenna 2 flagellum shorter than peduncle, calceoli absent. Mandibular palp of 3 articles. Labium (lower lip) lacking inner lobes. Coxae 1-4 distinctly longer than coxae 5-7; coxa 1 distinctly shorter than coxae 2-3; coxa 4 with small but distinct excavated corner on the posterior margin. Gnathopods 1–2 large (propodus as large or larger than head length), not sexually dimorphic; gnathopod 2 propodus slightly longer than in gnathopod 1; gnathopods 1-2 propodus lateral face with distinct excavation above and parallel to palm margin, palm margins crenulate and with 2 excavations, without robust setae along palm margin, with rows of 3 mesial (inner) and 2-3 lateral (outer) robust setae either side of palm corner (where dactyl fits to the propodus). Pereopod 6 and 7 similar lengths, bases narrow (close to 2 times as long as wide). Coxal gills on percopods 2-7, sternal gills on thoracic segments 2–6, slender, around  $\frac{1}{2}$  length of coxal gills. Uropod 3 outer ramus of 1 article, inner ramus absent. Telson cleft to 1/3 depth, with apical setae.

**Etymology**. Named for the Warrego River Basin where the type specimens of *Warregoensis lowryi* sp. nov. were collected. Gender: masculine.

**Remarks**. *Warregoensis* gen. nov. can be clearly identified from the large overall size of the adult animals (6.2 mm vs 4.5 mm for *Carnarvonis katjae* sp. nov.), the large gnathopods

1–2, and crenulations of the palm margin in gnathopods 1–2, telson cleft to  $\frac{1}{3}$  depth, and percopod 5–7 bases narrow (close to 2 times as long as wide).

## Warregoensis lowryi sp. nov.

urn:lsid:zoobank.org:act:D631937A-D112-46F2-8D55-E713B7E92030

### Figs 4–6

Holotype: QM W29604 (RL 2296.4, Genbank *CO1*: OP596266.1), male, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014. Paratypes: QM W29605 (RL 2296.3, Genbank *CO1*: OP596269.1), female, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014; SAMA C14814 (RL 2296.1, RL2296.2, Genbank *CO1*: OP596268.1, OP596270.1), female and male, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014; SAMA C14814 (RL 2296.1, RL2296.2, Genbank *CO1*: OP596268.1, OP596270.1), female and male, Carnarvon Station Reserve, Lady Spring, 24.80255°S 147.8265°E, Bou-Rouch pump in gravel bank, coll. R. Leijs, 15 October 2014.

**Description**. Holotype male, 6.2 mm. *Head*. Without rostrum, eyes absent. Antenna 1 length greater than half length of animal; peduncle with three articles progressively shorter; accessory flagellum of two articles, distal article small; flagellum of 18 articles. Antenna 2 approximately ½ length of antenna 1; peduncle longer than flagellum, articles 4–5 of peduncle of subequal length; flagellum of 7 articles. Labrum (upper lip) evenly rounded, setose apically. Labium (lower lip) without inner lobes. Mandible incisor with 5 teeth, lacinia mobilis with 4 teeth, setal row of 5 plumose setae



Figure 3. *Carnarvonis katjae* sp. nov., holotype female, TL 4.5 mm, Carnarvon Station Reserve, Queensland, QM W29606. (*A*) pereopod 3; (*B*) pereopod 6; (*C*) pereopod 7; (*D*) uropod 1; (*E*) uropod 2; (*F*) telson; (*G*) uropod 3.

present, molar narrow and denticulate; palp of three articles, article 3 shorter than article 2, with short row of plumose setae ("D-setae"; after Lowry & Stoddart, 1993). Maxilla 1 outer plate with seven apical denticulate robust setae; inner plate with two apical plumose setae; palp of two articles, distal article longest, with apical setae. Maxilla 2 outer plate with two rows of simple and plumose setae apically and simple setae laterally; inner plate with simple setae apically and an oblique row of medial plumose setae. Maxilliped palp slender (no articles enlarged), of four articles plus spine; outer plate with robust and simple setae along apical and lateral margin; inner plate with robust and plumose setae apically and laterally.

*Pereon*. Gills include coxal gills on pereopods 2–7; sternal gills on thoracic segments 2–6, slender, around ½ length of coxal gills. Gnathopods 1–2 large (propodus similar length to

head). Gnathopod 1 coxa short (wider than long), with robust seta at posterior corner and few simple setae scattered along margin; propodus length 1.8 times width, lateral face with distinct excavation above and parallel to palm margin, palm margins crenulate and with 2 excavations, without robust setae along palm margin, with rows of 3 mesial (inner) and 3 lateral (outer) robust setae either side of palm corner (where dactyl fits to the propodus), posterior margin of propodus with 4 oblique rows of long simple setae. Gnathopod 2 coxa longer than coxa 1, distinctly longer than wide, with robust seta at posterior corner and few simple setae scattered along margin; propodus length 2 times width, lateral face with distinct excavation above and parallel to palm margin, palm margins crenulate and with 2 excavations, without robust setae along palm margin, with rows of 3 mesial (inner) and 2 lateral (outer) robust setae either side of palm corner (where



**Figure 4**. *Warregoensis lowryi* sp. nov., holotype male, TL 6.2 mm, Carnarvon Station Reserve, Queensland, QM W29604. (*A*) whole animal in lateral view; (*B*) maxilla 1; (*C*) maxilla 2; (*D*) mandible; (*E*) maxilliped; (*F*) lower lip; (*G*) upper lip. Scale (*A*): 0.1 mm.



Figure 5. *Warregoensis lowryi* sp. nov., holotype male, TL 6.2 mm, Carnarvon Station Reserve, Queensland, QM W29604. (*A*) gnathopod 1 with enlargement to show palm detail; (*B*) gnathopod 2 with enlargement to show palm detail.

dactyl fits to the propodus), posterior margin of propodus with 5 oblique rows of long simple setae. Pereopod 3 coxa similar length to coxa 2, longer than wide, with robust seta at posterior corner and few simple setae scattered along margin. Pereopod 4 coxa similar length to coxa 2-3, longer than wide, small but distinct excavated corner on the posterior margin, with robust seta at posterior corner and few simple setae scattered along margin; dactylus with single seta. Percopod 5 shorter than percopods 6–7: coxa with rounded anteroventral lobe with seta, posteroventral lobe indistinct, with two setae on posterior margin; basis 1.7 times as long as wide; propodus longer than carpus; dactylus with single seta. Pereopod 6 similar length to pereopod 7; coxa with rounded anteroventral lobe, posteroventral lobe indistinct, with two setae on posterior margin; basis 1.9 times as long as wide; propodus longer than carpus; dactylus with single seta. Pereopod 7 coxa rounded, with two setae on posterior margin; basis 1.9 times as long as wide; propodus longer than carpus; dactylus with single seta.

*Pleon*. Epimera 1–3 posteroventrally subquadrate, posteroventral corner with robust seta and posterior margins

with several small robust setae along length; epimera 2–3 with robust setae along the ventral margin. Urosomite 1 with distinct distoventral robust seta, with paired dorsolateral setae on distal margin. Urosomite 2 with paired dorsolateral setae on distal margin. Uropod 1 peduncle without basofacial robust seta, distal margin of peduncle with group of 1 large and 2 small robust setae laterally and 1 large robust seta medially. Uropod 2 distal margin of peduncle with group of 2 small robust setae laterally and 1 small robust seta medially. Uropod 3 distal margin of peduncle with robust setae laterally and medially; outer ramus of 1 article, approximately 3 times length of peduncle, with clusters of 2–3 robust setae along length; inner ramus absent. Telson cleft to 1/3 depth, with robust and simple setae apically.

**Etymology**. Named in honour of Dr Jim Lowry, a friend and colleague.

**Remarks**. *Warregoensis lowryi* sp. nov. is clearly a distinct species with 15% CO1 divergence from Carnarvonis katjae sp. nov. and 22% CO1 divergence from Chillagoe thea.



**Figure 6**. *Warregoensis lowryi* sp. nov., holotype male, TL 6.2 mm, Carnarvon Station Reserve, Queensland, QM W29604. (*A*) pereopod 4; (*B*) pereopod 5; (*C*) pereopod 6; (*D*) pereopod 7; (*E*) uropod 1; (*F*) uropod 2; (*G*) uropod 3; (*H*) telson.

# Conclusion

Herein, we present two species hypotheses, largely based on morphological analyses, but with corroborating *CO1* divergence data. We describe two distinct new genera and species, *Carnarvonis katjae* sp. nov and *Warregoensis lowryi* sp. nov. within the family Chillagoeidae, existing within a single spring at Carnarvon Station Reserve in north-eastern Australia. We have revised the family Chillagoeidae and present new diagnostic characters that unite the three monotypic genera within the family. There are now three species of stygobiotic amphipods described from two discrete groundwater systems in north-eastern Australia, and it is highly likely that additional and potentially diverse stygobiont taxa will be discovered as groundwater habitats across the region are further explored. ACKNOWLEDGEMENTS. We thank the editors Shane Ahyong, Alan Myers, Lucia Fanini and Penny Berents for their invitation to contribute to this memorial volume for Jim Lowry. Over the course of Rachael King's career, Jim has been a mentor, colleague, professional champion, and living encyclopaedia of all things Amphipoda and he will be missed.

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