

## Species of the sponge genus *Chondrilla* (Demospongiae: Chondrosida: Chondrillidae) in Australia

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**Abstract** – A new sponge species, *Chondrilla linnaei*, from Australia is described. *Chondrilla australiensis* Carter 1873 and *C. secunda* Lendenfeld 1885 are redescribed with reference to type and new material. The synonymy of *C. corticata* and *C. papillata* with *C. australiensis* is confirmed. Two species previously described from Australia, *C. mixta* and *C. nucula* are discussed, with the conclusion that *C. nucula* is unlikely to occur in Australia. The descriptions of the species incorporate previous molecular results. Information about symbiont relationships and the biogeographical distribution of the species is discussed.

**Keywords:** taxonomy, systematics, sponges, Porifera, *Chondrilla*

### INTRODUCTION

Approximately twenty species of *Chondrilla* Schmidt 1862 appear to be valid worldwide (Boury-Esnault 2002). However, the morphology and skeletal structures of this genus and its sister genus (*Chondrosia*) are remarkably uniform, with few characters useful for differentiation at the species level. Consequently at least two species of *Chondrilla* have been considered to be globally widespread. The type species, *C. nucula* Schmidt 1862 was thought to have a cosmopolitan distribution, and *C. australiensis* Carter 1873 was reported with a widespread Indo-Pacific distribution (Hooper and Wiedenmayer 1994). Recent genetic studies have reduced the known distribution of *C. nucula* to the Mediterranean Sea (Klautau *et al.* 1999).

Six species of the genus *Chondrilla* have been reported from Australian waters. These include *C. australiensis*, *C. secunda* Lendenfeld 1885 and *C. nucula* from mainland Australia, and *C. mixta* Schulze 1877 from Christmas Island, an Australian Territory in the Indian Ocean. Two others, *C. papillata* Lendenfeld 1885 and *C. corticata* Lendenfeld 1885, are regarded as synonyms of *C. australiensis* (Burton 1924). On the basis of DNA sequence analyses of *Chondrilla* specimens from Australian waters, Usher *et al.* (2004a) suggested

the presence of three species: *C. australiensis* and two unidentified species described here. A specimen collected from Australia and previously identified as *C. nucula* was examined and found to be the new species described in this paper. Although fieldwork was undertaken at Christmas Island *C. mixta* was not found. This study reports three valid species of *Chondrilla* from mainland Australia: *C. australiensis*, *C. secunda* and *C. linnaei*. The occurrence of *C. mixta* in Australian waters was not resolved.

Species of *Chondrilla* have been reported from shallow waters (< 50 m depth) in tropical, subtropical and temperate zones, but rarely in deeper waters (Boury-Esnault 2002). Many species are considered to be cryptic, occurring on vertical walls, at cave entrances, and under rocks (Boury-Esnault 2002). In south Western Australia *C. australiensis* is a major space occupant of many temperate limestone reef habitats, and forms large encrustations in shallow depths (1–20 m) in full light and shaded environments (Usher *et al.* 2001). Individuals of this species have been reported to provide refuge for a wide range of invertebrates including brittle stars, molluscs and shrimp (Edgar 1997), and to be a food item for species of the cowrie genus *Zoila* (Wilson and Clarkson 2004). *Chondrilla* aff. *nucula* from Caribbean coral reefs have been found to be a preferred food of

the Hawksbill turtle (Meylan 1988) and some fishes (Randall and Hartman 1968).

Species of *Chondrilla* have been reported to have abundant populations of symbiotic bacteria in their mesohyl (Boury-Esnault 2002), and *C. australiensis* has been found to have both bacteria (Dey *et al.* 2004) and symbiotic cyanobacteria (Usher *et al.* 2001, 2004c). Cyanobacterial symbionts are thought to aid in the rapid growth of larvae at settlement (Wilkinson 1992), and to provide an advantage to adult sponges in competition with algae and other photosynthetic organisms for substrate in high light areas (Wilkinson 1983). In February 1998 a bleaching event was reported for a population of *C. australiensis* at Fremantle (south Western Australia), which coincided with a global hard and soft coral bleaching event (Fromont and Garson 1999).

Recently, *Chondrilla*, *Chondrosia* and two other genera were united in a monophyletic order, Chondrosida, based on molecular data (Boury-Esnault and Lopès 1985), and containing a single valid family, Chondrillidae Gray 1872. Prior to 1985 the family was located in the order Hadromerida but its affinities to this order were not clear.

This study documents species of *Chondrilla* recently collected from Australian waters. Type material of species reported from Australia has been examined and reallocated where necessary. Species currently known to occur in Australia are described and a preliminary assessment of the biogeography of each species is provided. The study also draws on previously published molecular data sets (Usher *et al.* 2004a) and various biological characters such as symbiont relationships, reproductive biology and ecological distributions.

## MATERIALS AND METHODS

Preserved material from various museums (listed at the end of this section) was examined during the course of this study. Collected specimens were preserved in 70% ethanol. Skeletal structure and spicule morphology were examined using light microscopy and scanning electron microscopy (SEM). Spicules were prepared by boiling small pieces of sponge (including the ectosome and choanosome) in concentrated nitric acid, followed by two consecutive washes with both distilled water and absolute alcohol. The resulting spicule extracts were dried on a glass slide and mounted in Shandon EZ-Mount (Thermo Electron Corporation). Spicule dimensions were determined by measurement of 20 randomly selected spicules per specimen using an eyepiece graticule with an Olympus BX50 microscope. Clean spicules were spread on coverslips or

double-sided carbon tape attached to SEM stubs, dried at 70°C and sputter coated with gold prior to examination with a Philips SEM 505 or a Zeiss 1555 SUPRA Variable Pressure SEM operating at 15 kV. Images were recorded electronically.

The skeleton was prepared for examination by cutting a representative section at right angles to the surface of the sponge. The section was dehydrated through an ascending ethanol series, cleared in xylene and infiltrated in paraffin wax (Shandon Histoplast) using an automatic tissue processor on a nine hour cycle. The sponge tissue was further infiltrated with paraffin under a vacuum of 635 mm Hg for 30 min prior to embedding. Blocks were sectioned at 90 µm thickness with a Leitz slide microtome, and section rolling was eliminated by placing filter paper, moistened with distilled water, on top of the paraffin block. Sections were placed on a glass slide smeared with egg albumin for adhesion, dried overnight at 60°C, and dehydrated in two changes of xylene. Sections were mounted in Shandon EZ-Mount and examined using light microscopy. Images were recorded with a Leica DFC420 camera on a Leica DME microscope and saved electronically.

All sequences are available on GenBank (accession numbers, D2 region: AY190190–AY190224, ITS region: AY190225–AY190239). Methods to determine sequence comparisons and phylogenetic trees are detailed in Usher *et al.* (2004a).

Abbreviations used in the text: AM, Australian Museum, Sydney, Australia; BMNH, Natural History Museum, London, United Kingdom; LMJG, Landesmuseum Joanneum, Graz, Austria; NMV, National Museum of Victoria, Melbourne, Australia; NTM, Northern Territory Museum of Arts and Sciences, Darwin, Australia; SAM, South Australian Museum, Adelaide, Australia; WAM, Western Australian Museum, Perth, Australia; ZMB, Museum fuer Naturkunde, Berlin, Germany.

## SYSTEMATICS

### Order Chondrosida Boury-Esnault and Lopès 1985

#### Family Chondrillidae Schmidt 1862

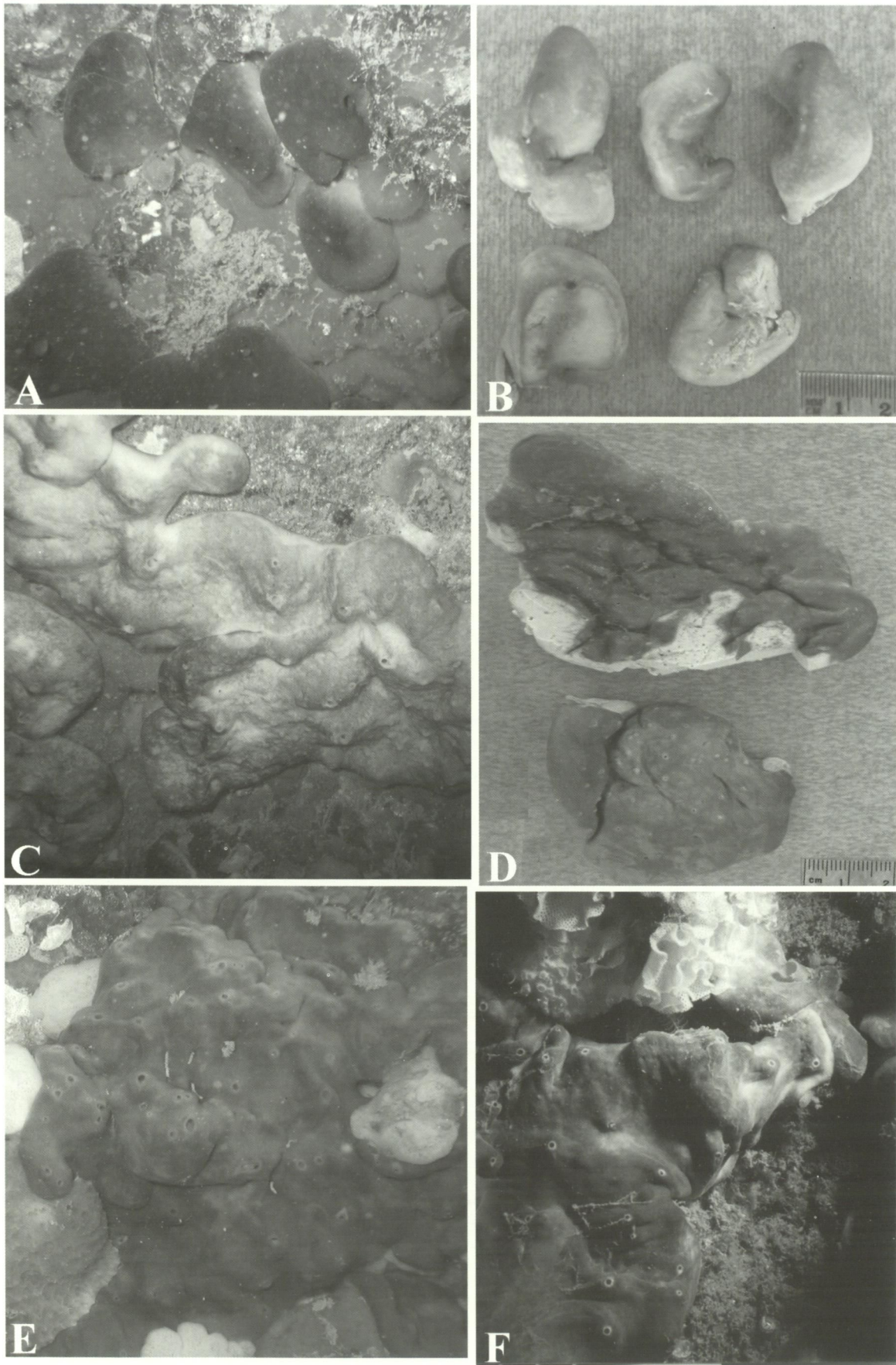
##### *Chondrilla* Schmidt 1862

##### Type species

*Chondrilla nucula* Schmidt, 1862 (subsequent designation by de Laubenfels, 1936).

##### *Chondrilla linmaei* sp. nov.

Figures 1a, 1b, 2–4



**Figure 1** Whole specimen images of Australian *Chondrilla*. A. *C. linnaei* in situ. B. Holotype (WAM Z13267) of *C. linnaei* after preservation in ethanol. C. *C. secunda* in situ. D. *C. secunda* after preservation in ethanol (WAM Z13262). E. *C. australiensis* in situ. F. *C. australiensis* in situ, maroon form.

## Material examined

### Holotype

**Australia: Western Australia:** 5 pieces, in cave, Twilight Cove, Esperance, 33°51'S, 121°55'E, 9.2 m depth, K. Usher, SCUBA, 3 May 2001 (WAM Z13267).

### Paratypes

**Australia: South Australia:** 1 specimen, Western River Cove, Kangaroo Island, 35°40'S, 136°57'E, 3–11 m depth, K. Usher, SCUBA, 8 November 2001 (SAM S1106, ex WAM Z13276). **Western Australia:** 3 pieces, Mistaken Island, Albany, 35°03'S, 117°58'E, 8.0 m depth, K. Usher, SCUBA, 6 May 2001 (WAM Z13256); 1 specimen, under Busselton jetty, 33°30'S, 115°10'E, 8.0 m depth, K. Usher, SCUBA, 21 February 2001 (WAM Z13259); 2 pieces, station JWAM08 transect 1, Essex Rocks Jurien, 30°21.15'S, 114°59.30'E, 7–11 m, J. Fromont, SCUBA, 1 May 2005 (WAM Z31397).

### Other material examined

**Australia: Tasmania:** 1 specimen, Maria Island, ca. 42°38'S, 148°05'E (BMNH 1925.11.1.1331); **South Australia:** 1 specimen, Western River Cove, Kangaroo Island, 35°40'S, 136°57'E, 9.0 m depth, K. Usher, SCUBA, 8 May 2001 (WAM Z13275).

## Diagnosis

*Chondrilla linnaei* is characterised by always forming small, discrete encrusting mounds or lobes, a finely speckled surface in darker shades of brown with a lighter interior, and small oxysphaerasters as the only spicule type. Diameter from ray tip to opposing ray tip varies among specimens (range 15.7–18.4 µm, mean 17.6 µm, n = 140).

## Description

Habitus as in Figure 1a, b. Thickly encrusting with a smooth, shiny surface. Individuals tend to form small thick discrete encrustations or low lobes up to 30 mm high with apical oscules approximately 3 mm wide in preserved sponges. Oscules may have slightly raised rims up to 1 mm in height. Dimensions: The holotype consists of five discrete lobes, the largest of which is 45 × 20 × 18 mm high. Texture: soft alive, but firm, compressible and springy after preservation. Sponges have a dense compact interior with fine internal canals. *Colour:* finely speckled shades of brown to dark brown with a cream, fawn or brown interior. WAM Z13256 has an orange tinge. Some specimens are pigmented throughout the choanosome, with more dense pigmentation towards the surface and around internal canals, although degree of pigmentation varies among specimens.

*General organisation:* (Figure 2a, b). Ectosome: oxysphaerasters form a single layer at the surface or are sparsely distributed throughout this layer. This region is 320–1000 µm thick and is usually apparent macroscopically. Choanosome: oxysphaerasters are distributed throughout the choanosome, but tend to be more numerous around internal canals, where they may form a single boundary layer. The mesohyl of the choanosome is clearly differentiated from the ectosome.

*Spicules:* (Figure 2c–e). Oxysphaerasters usually with fine sharply pointed spines, occasionally with blunt spines and a more ball-like shape. Diameter from ray tip to opposing ray tip varies among specimens (range 15.7–18.4 µm, mean 17.6 µm, n = 140) (Table 1).

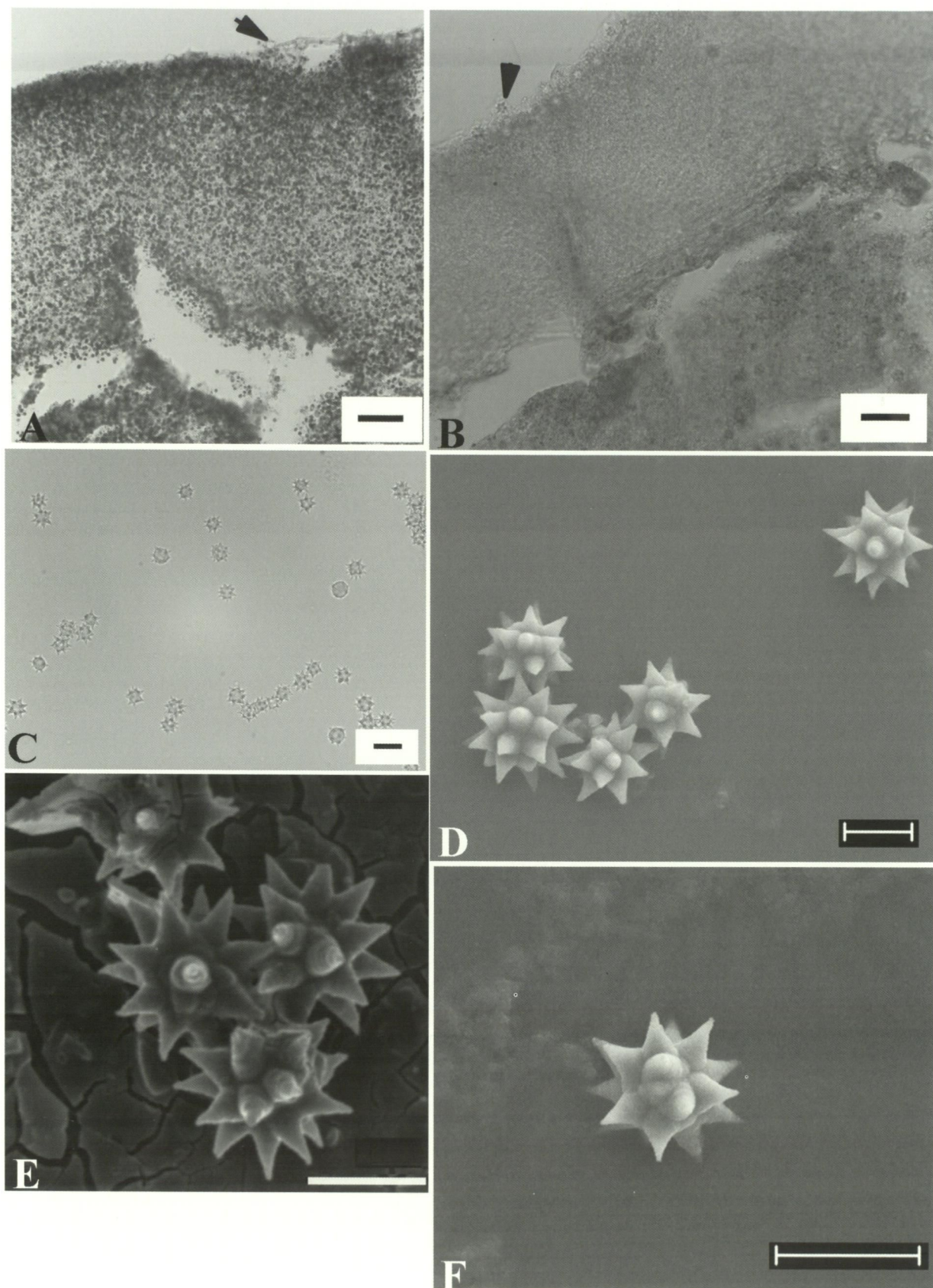
*Cyanobacteria:* sponges were found to contain cyanobacteria in low concentrations with 99.7% partial sequence similarity to *Synechococcus* WH 8103 (Genbank), a species which occurs in the water column (Usher *et al.* 2004c).

## Remarks

This species is comparable in spicule type to *Chondrilla nucula* Schmidt 1862 having a single size category of oxysphaeraster. The specimen from Tasmania examined in this study (BMNH 1925.11.1.1331) was previously identified as *C. nucula* by Shaw. We examined that specimen and have assigned it to the new species *C. linnaei*. *Chondrilla nucula* has also been reported from the Great Barrier Reef (Burton 1934). This material requires checking but it is unlikely that *C. nucula* is present in Australia, and more likely that early records are of a different species, possibly the new species described here.

We compared our specimens of *Chondrilla linnaei* to the type material of *C. nucula*, type locality Adriatic Sea (holotype LMJG 15108/0 and paratypes LMJG 15687/0 and BMNH 1867.7.26.1). The average size of the oxysphaerasters of these specimens was 27, 26 and 31 µm, respectively (n = 20) (Table 1, Figure 2f). We also obtained recently collected fresh material of *C. nucula* from Marseille and Portofino in the Mediterranean (WAM Z13268 and Z13261, respectively) and found similar average spicule sizes (26 and 25 µm, respectively; n = 20) to the type material. Sequences of these *C. nucula* specimens showed 100% similarity to each other and 89.1% similarity to the *C. linnaei* holotype Z13267 (Figure 3). The two species are clearly differentiated by spicule morphology and size, molecular dissimilarity, and geographic locality.

We also examined a specimen of *Chondrilla* from Bermuda (BMNH 1948.8.6.55) and found oxysphaerasters with an average size of 23 µm

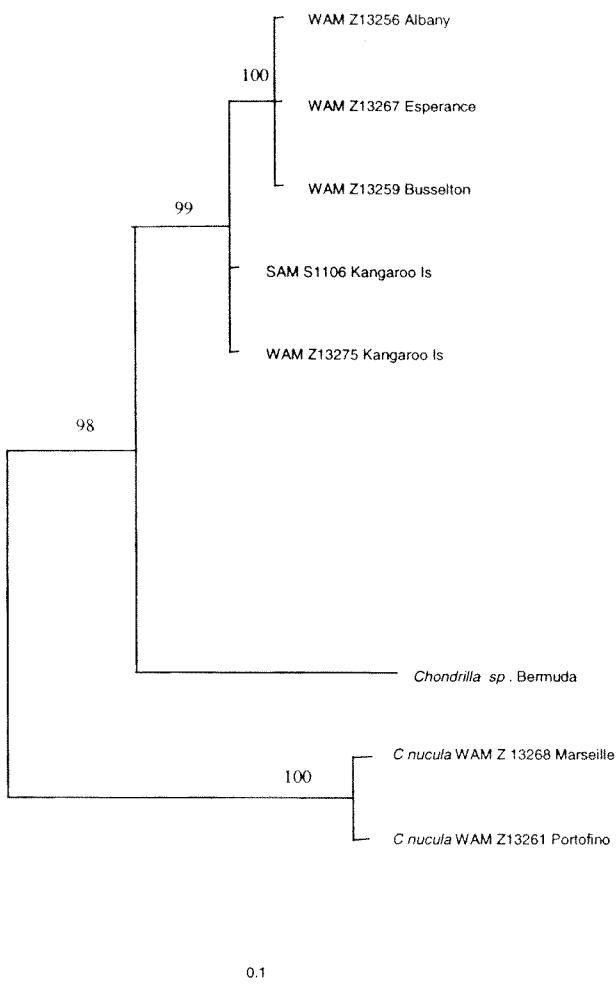


**Figure 2** Internal organisation and spicules of *Chondrilla linnaei*. A. Internal organisation of the holotype (WAM Z13267), scale bar = 50  $\mu$ m, spicules indicated by arrow. B. Internal organisation of a paratype (WAM Z13276) scale bar = 50  $\mu$ m, spicules indicated by arrow. C. Spicules of the holotype (WAM Z13267), scale bar = 20  $\mu$ m. D. SEM image of spicules of the holotype (WAM Z13267), scale bar = 10  $\mu$ m. E. SEM image of spicules of a paratype (WAM Z13275), scale bar = 10  $\mu$ m. F. SEM image of spicules of *C. nucula*, (LMJG 15108/0), scale bar = 20  $\mu$ m.

**Table 1** Spicule dimensions of *Chondrilla linnaei*, *Chondrilla nucula* and *Chondrilla* sp.<sup>1</sup>

<i>Chondrilla linnaei</i>							<i>Chondrilla nucula</i>					<i>Chondrilla</i> sp.
BMNH 1925.11.1.1331	SAM S1106	WAM Z13275	WAM Z13267	WAM Z13256	WAM Z13259	WAM Z31397	BMNH 1867.7.26.1	LMJG 15687/0	LMJG 15108/0	WAM Z13261	WAM Z13268	BMNH 1948.8.6.55
20	15	18	20	18	18	18	30	25	28	25	25	20
18	20	18	18	15	13	18	35	25	25	28	25	28
18	20	18	15	18	15	18	33	25	28	28	28	25
18	15	20	15	15	15	18	25	30	28	25	25	20
15	20	20	18	15	13	15	30	25	28	25	13	25
20	18	18	18	18	18	18	35	25	23	25	23	20
20	20	18	18	20	15	15	33	23	30	25	23	23
18	18	18	18	18	18	15	35	25	25	25	28	23
18	20	20	18	18	15	18	28	25	25	23	30	20
18	18	20	15	18	13	15	30	25	30	28	30	20
18	15	20	20	18	15	18	35	25	25	25	25	23
20	15	15	18	15	13	18	35	28	28	25	28	20
18	18	18	18	18	15	18	30	28	25	28	30	25
18	18	18	18	20	15	18	28	30	23	25	28	23
20	18	20	18	18	18	18	33	28	28	25	28	23
20	20	18	15	18	18	20	30	25	28	25	25	23
18	18	18	18	18	15	20	33	25	30	20	28	25
18	15	20	18	18	15	20	28	25	30	23	30	30
18	18	18	15	18	18	15	33	25	28	25	25	25
15	18	15	20	18	18	20	25	25	25	25	25	20
Mean	18.3	17.9	18.4	17.6	17.6	15.7	31.2	25.9	27.0	25.2	26.1	23.1
SD	1.5	1.9	1.5	1.7	1.5	1.9	3.3	1.9	2.3	1.9	3.9	2.9
Range	15–20	15–20	15–20	15–20	15–20	13–18	25–35	23–30	23–30	20–28	13–30	20–30

<sup>1</sup> n = 20



**Figure 3** Phylogenetic tree of *Chondrilla linnacii* specimens produced with MrBayes, using the C2D2 region of 28S rDNA. Confidence levels are given at nodes. Scale bar = 0.1 substitutions per site.

(n = 20) (Table 1). Molecular results from this specimen showed it to be distinct from *C. nucula* (91.1% similarity) and *C. linnacii* (91.5% similarity to the *C. linnacii* holotype), and we suggest it is another new species of *Chondrilla* awaiting formal description. *Chondrilla nucula*, *C. linnacii* and *Chondrilla* sp. from Bermuda all formed a cluster in the C2D2 and ITS phylogenetic analyses (although the Bermuda sample was not included in the ITS tree, Usher *et al.* 2004a) and all have the same spicule complement, although spicule size is distinctive and geographic separation is very large (Figure 3).

Kumar (1925) described *Chondrilla kilakaria* from south India with oxysphaerasters as the only spicule type. This is a very thin encrusting species from tropical coral reefs. *Chondrilla kilakaria* differs from *C. linnacii* in having a thin encrusting growth form, a tropical habit and larger oxysphaerasters (20–24  $\mu\text{m}$  compared to the mean size of 17.6

$\mu\text{m}$  for *C. linnacii*). This is the only other species of *Chondrilla* with oxysphaerasters as the only spicule type to have been described from the Indian Ocean.

The rDNA sequences of Western Australian specimens of *C. linnacii* were 100% similar to each other. The two South Australian specimens had 97.7% similarity to the Western Australian specimens. However, as 5 of the 8 “mismatches” in these sequences included uncertain base pairs, the true sequence similarity may be higher. Alternatively, Kangaroo Island (South Australia) and Esperance (Western Australia) are separated by an approximate distance of 1600 km around the coastline of the Great Australian Bight (Figure 4) and it is possible that speciation is occurring between these populations.

### Distribution and habitat

*Chondrilla linnacii* is found in Tasmania and South Australia, and in south Western Australia as far north as Jurien Bay (Figure 4). It is a temperate species occurring on heavily shaded rock faces, under jetties and in caves at depths less than 15 m (Usher *et al.* 2004a). This species is rare.

### Etymology

Named in honour of Carolus Linnaeus and the 250<sup>th</sup> anniversary of the publication of *Systema Naturae*.

### *Chondrilla secunda* Lendenfeld 1885

Figures 1c–d, 4–5

*Chondrilla secunda* Lendenfeld 1885: 15, plate 4, figures 10–12; Hooper and Wiedenmayer 1994: 125.

### Material examined

#### Lectotype

**Australia: Victoria:** piece of a specimen and four slides, Port Phillip Bay ca. 38°09'S, 144°52'E (ZMB Por 1131).

#### Paralectotypes

**Australia: Victoria:** three slides, Port Phillip Bay, ca. 38°09'S, 144°52'E (BMNH 86.6.7.95, BMNH 86.6.7.96, BMNH 1954.2.10.15).

#### Other material examined

**Australia: Western Australia:** 2 pieces, Cape Le Grande, Esperance, 34°01'S, 122°07'E, 6.4 m depth, K. Usher, SCUBA, 2 May 2001 (WAM Z13264); 2 pieces, Two People's Bay, Albany, 34°57'S, 118°11'E, 11.2 m depth, K. Usher, SCUBA, 8 May 2001 (WAM Z13262); 2 pieces, in cave, Mistaken Island, Albany, 35°03'S, 117°58'E, 6.2 m depth, K. Usher, SCUBA,

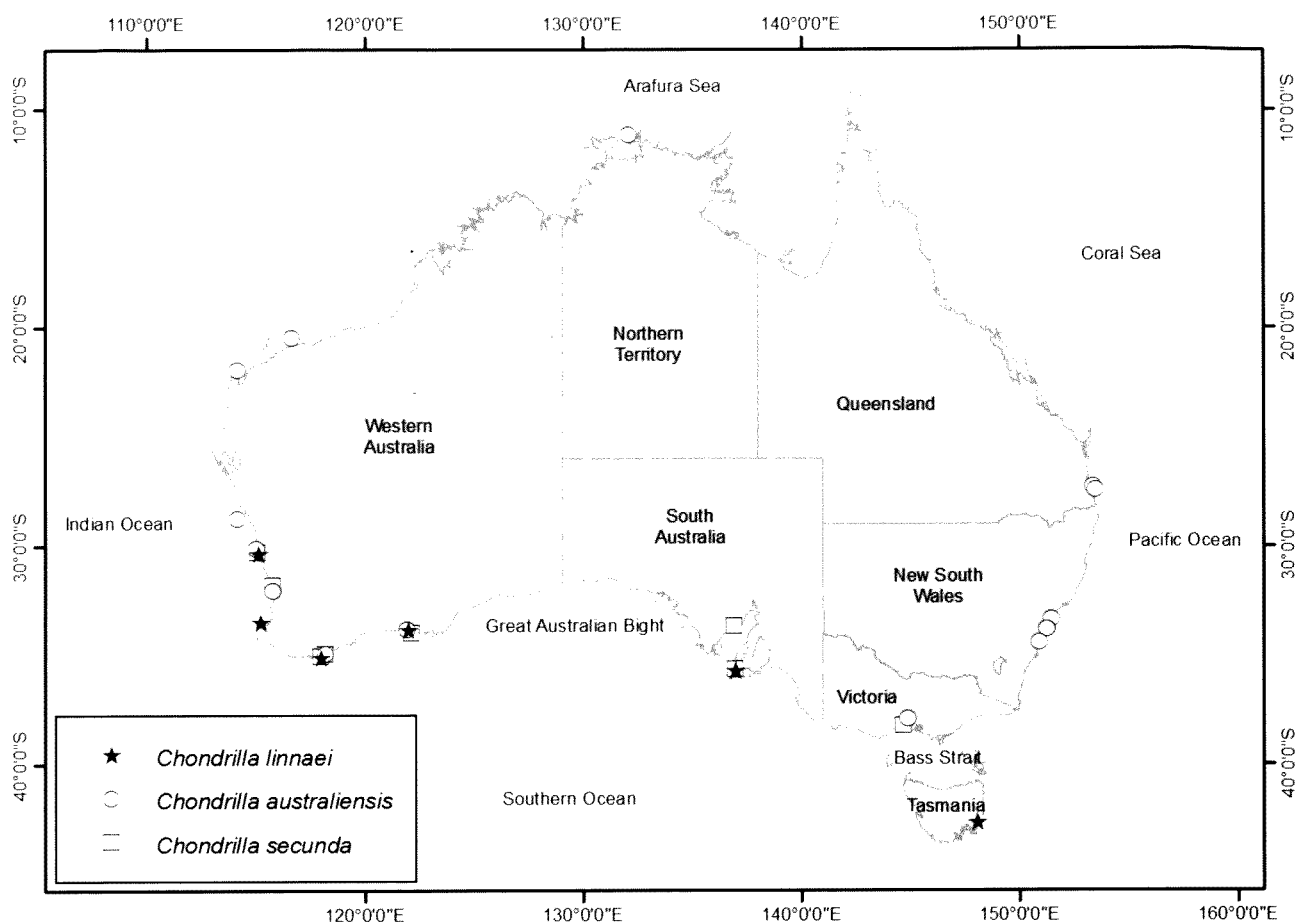


Figure 4 Map of distributions of Australian species of *Chondrilla*.

6 May 2001 (WAM Z13270); 1 specimen, Mistaken Island, Albany, 35°03'S, 117°58'E, 2.3 m depth, K. Usher, SCUBA, 6 May 2001 (AM Z.6973 ex WAM Z13271); 1 specimen, in cave, Two People's Bay, Albany, 34°57'S, 118°11'E, 20.5 m depth, K. Usher, SCUBA, 5 May 2001 (BMNH 2008.9.15.1 ex WAM Z13273); 1 specimen, station SC18, Marmion Lagoon, 31°50'S, 115°45'E, 4–10 m depth, collector L. McQuillan, SCUBA, 31 October 1999 (WAM Z12501); 2 pieces, station JWAM05, transect 3, Booka Rocks, Jurien, 30°17.85'S, 115°01.17'E, 7 m depth, J. Fromont, SCUBA, 28 April 2005 (WAM Z31396). **Victoria:** 2 pieces, Cottage by the Sea, Queenscliff, 38°16'S, 144°40'E, 5.4 m depth, collector K. Usher, SCUBA, 15 November 2001 (NMV F157468, exWAM Z13260). **South Australia:** 1 specimen, Western River Cove, Kangaroo Island, 35°40'S, 136°57'E, 3–11 m depth, K. Usher, SCUBA, 8 November 2001 (SAM S1107, exWAM Z13274); 1 specimen, prawn trawl out of Cowell, Spencer Gulf, ca. 33°41'S, 136°55'E 40 m depth, 16 April 1982, B. Mills (NTM Z1619).

#### Diagnosis

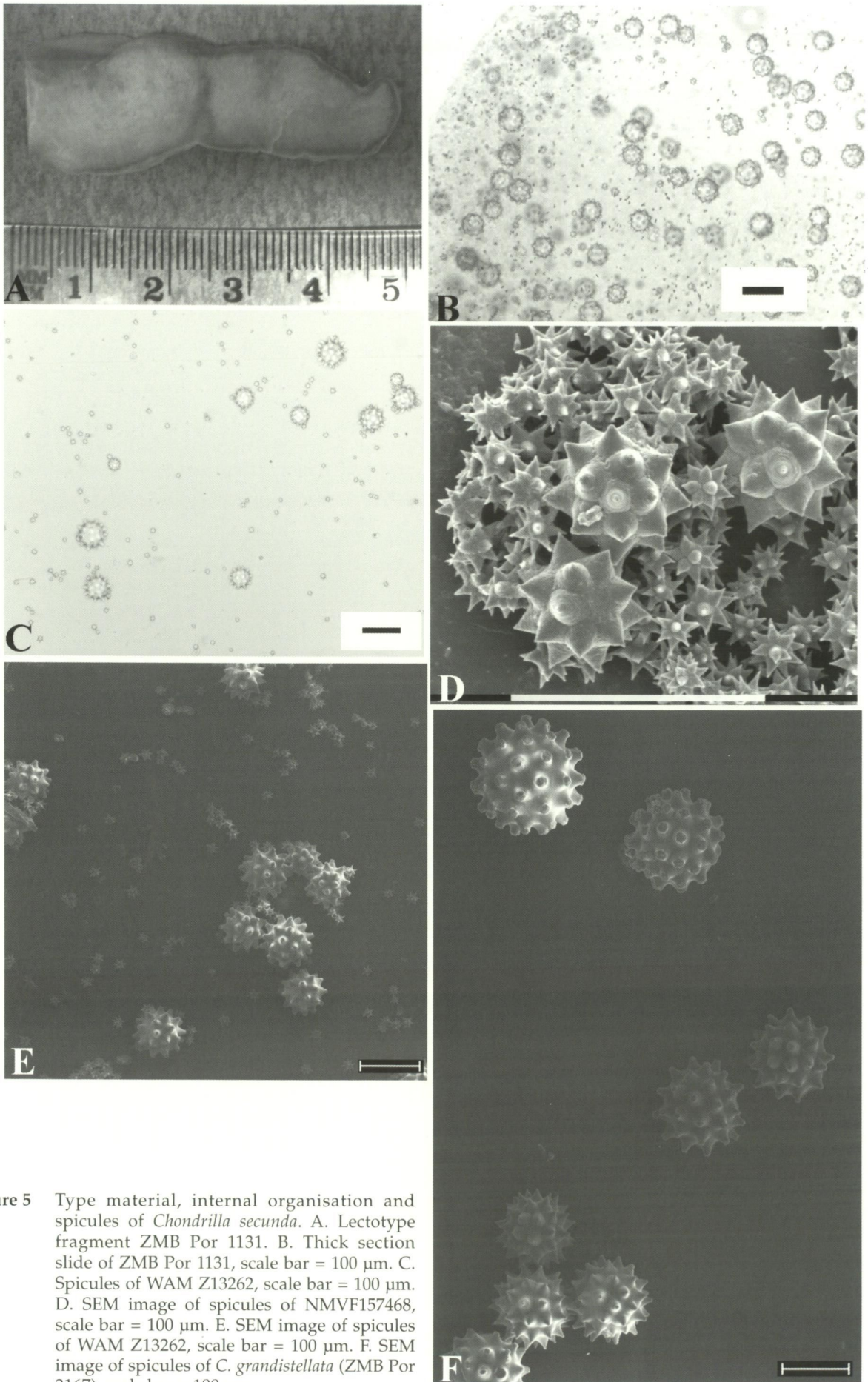
*Chondrilla secunda* is characterised by a thick spreading, encrusting growth form, an irregular

undulating surface coarsely mottled in various shades of brown with a lighter interior, and 2 size classes of oxysphaerasters as the only spicule type. Diameter of large oxysphaerasters ranges from 30–100 µm (mean 65.4 µm,  $n = 280$ ), and the small oxysphaerasters range from 15–30 µm in diameter (mean 23.3 µm,  $n = 280$ ). Density of spicules and specimen colour varies among specimens.

#### Description

Habitus as in Figure 1c, d. Thickly encrusting with a smooth, shiny surface. Individuals tend to form thick spreading encrustations up to 19 mm high with small and generally apical oscules less than 1 mm wide in preserved sponges. Oscules may have slightly raised rims up to 0.5 mm in height. Holotype specimen is a thick section of an individual 50 mm long by 12 mm high and 3 mm thick. Texture: Stiff alive, and firm and slightly compressible after preservation. Sponges have a dense compact interior with fine internal canals. *Colour:* Coarsely mottled shades of fawn, brown to dark brown with a cream or fawn interior. Specimens vary from little differentiation in pigmentation between the choanosome and ectosome (WAM Z31396) to dense pigmentation





**Figure 5** Type material, internal organisation and spicules of *Chondrilla secunda*. A. Lectotype fragment ZMB Por 1131. B. Thick section slide of ZMB Por 1131, scale bar = 100 µm. C. Spicules of WAM Z13262, scale bar = 100 µm. D. SEM image of spicules of NMVF157468, scale bar = 100 µm. E. SEM image of spicules of WAM Z13262, scale bar = 100 µm. F. SEM image of spicules of *C. grandistellata* (ZMB Por 3167), scale bar = 100 µm.

towards the surface and around internal canals. Degree of pigmentation varies among specimens.

*General organisation:* (Figure 5a, b). Ectosome: large oxysphaerasters common throughout. This region varies in thickness from 140–800  $\mu\text{m}$ , frequently with a narrow outer strongly pigmented layer 50–150  $\mu\text{m}$  thick. In the type material ZMB Por 1131 the two sizes of oxysphaerasters are equally abundant in the ectosome. In recently collected specimens the ectosome may contain more small oxysphaerasters than large (e.g. WAM Z13260, WAM Z13270). Occasionally the ectosome is barely differentiated from the choanosome, and has no additional pigmentation to differentiate it (WAM Z13273). Choanosome: The interior is dense and compact with fine canals, but less so than the ectosome. Both types of oxysphaeraster are more dense around internal canals. WAM Z13264 has a basal layer of both sizes of oxysphaerasters.

*Spicules:* (Figure 5c–e). Two types of oxysphaeraster. Oxysphaerasters: large, either conical with faintly mammillate ray tips or with flattened, faintly spined 'mesa-topped' rays. These spicules are very variable in size, with the diameter from ray tip to opposing ray tip ranging from 30–100  $\mu\text{m}$  (mean 65.4  $\mu\text{m}$ ,  $n = 280$ ). Oxysphaerasters: small, consistently tapered, conical rays extending from a large central disc. These spicules have a size range of 15–30  $\mu\text{m}$  in diameter (mean 23.3  $\mu\text{m}$ ,  $n = 280$ ) (Table 2).

*Cyanobacteria:* sponges were found to contain cyanobacteria in low concentrations with 99.8% partial sequence similarity to *Synechococcus* WH 8103 (Genbank), a species which occurs in the water column (Usher *et al.* 2004c).

### Remarks

*Chondrilla secunda* has not been reported since its first description in Lendenfeld, 1885. Our collection of this species has extended its geographical range from the type locality in Port Phillip Bay, Victoria to the mid west coast of Western Australia. It has also enabled a thorough redescription of the species including field characters.

We discovered that the only extant type specimen of this species is a piece three mm thick with four associated microscope slides all labeled ZMB Por 1131, and lodged in the Museum für Naturkunde, Berlin. We have designated this syntype material to be the lectotype of the species. All the syntype specimens of *Chondrilla secunda* Lendenfeld 1885 from the Natural History Museum and the Australian Museum (AM G9057, BMNH 1886.6.7.92, BMNH 1886.6.7.93–94) were examined and found to be specimens of *C. australiensis* or other non-related species (BMNH 1886.6.7.92 is a lithistid). The only

type material remaining of this species in the Natural History Museum are three historic slides (BMNH 1886.6.7.95–96 and BMNH 1954.2.10.15) we have designated paralectotypes of this species. We examined slides (ZMB 650) previously thought to be syntype material (Hooper and Wiedenmayer 1994) and they are not *C. secunda*. We have distributed recently collected specimens of this species to the Museum of Victoria, South Australian Museum, Australian Museum, Natural History Museum and Western Australian Museum to assist future studies on this species.

The density of spicules, and the relative proportions of the large and small oxysphaerasters in the choanosome and ectosome of *C. secunda* varies among specimens. The cortical region of this species is less pronounced than in *C. linnaei* but more pronounced than in *C. australiensis*. This species is consistently thicker than *C. australiensis* and with a more irregular undulating surface, and a more mottled, less finely speckled appearance than *C. linnaei*. In some specimens (e.g. SAM S1107) the two size categories of oxysphaeraster grade into each other (Table 2). However, the morphology of the oxysphaerasters differs, with the small size category having consistently long, thin conical rays, and the large size category having short, thick conical, mammillate or mesa-topped rays.

Three species of *Chondrilla* have been described with large oxysphaerasters: *Chondrilla secunda* from southern Australia, *C. sacciformis* Carter 1879 from Mauritius and *C. grandistellata* Thiele 1900 from Indonesia (Ternate).

*Chondrilla grandistellata* was synonymised with *C. sacciformis* by Dendy (1916). The type specimen of *C. sacciformis* from the Natural History Museum, London (BMNH 95.8.9.2) had oxeas as well as sphaerasters. Carter mentioned both spicule categories in the type description (Carter 1879, p. 299), so this is likely to be the true type of the species, but oxeas are not a spicule type ever reported in the genus *Chondrilla*. The sphaerasters in the specimen BMNH 95.8.9.2 are more like sterrasters in form, and this species may need to be reassigned to the family Geodiidae. Dendy (1916, p. 245) examined teased fragments mounted in balsam and a spicule preparation (presumably made by Carter), and in the absence of oxeas concluded that *C. sacciformis* was a good species of *Chondrilla*. We suggest that the latter preparations are compared to the type specimen and a final conclusion made as to whether *C. sacciformis* is a valid species of *Chondrilla*.

The type specimen of *Chondrilla grandistellata* (ZMB Por 3167) and a second specimen (ZMB Por 3007) have large mesa-shaped oxysphaerasters with a mean diameter of 140  $\mu\text{m}$  ( $n = 20$ )

Table 2 Spicule dimensions of *Chondrilla secutata*.<sup>1,2</sup>

ZMB		BMNH 86.6.7.95		BMNH 86.6.7.96		BMNH 1954.2.10.15		NMV F157468		SAM S1107		NTM Z1619		WAM Z13264		WAM Z13273		WAM Z13262		WAM Z13270		WAM Z13271		WAM Z12501		WAM Z31396	
L	S	L	S	L	S	L	S	L	S	L	S	L	S	L	S	L	S	L	S	L	S	L	S	L	S	L	S
70	25	70	25	60	25	60	23	55	27	45	22	55	25	50	25	95	25	90	25	80	20	65	20	95	25	95	25
60	25	50	25	65	25	65	25	55	20	35	20	56	20	35	25	80	25	90	21	76	23	50	20	80	25	90	23
75	20	65	25	60	20	60	20	55	20	35	25	60	22	45	28	80	20	75	22	85	25	55	22	75	26	85	23
35	25	65	25	70	25	65	25	55	22	35	25	60	22	50	24	85	25	85	25	70	25	55	25	65	25	60	25
75	25	60	23	70	20	75	25	42	20	36	25	42	20	45	22	75	20	45	24	70	25	85	25	70	23	60	25
65	25	65	20	65	28	65	28	60	23	30	20	60	22	46	21	90	25	85	20	92	22	75	25	70	27	70	25
70	25	60	23	55	20	68	22	60	27	35	21	60	24	55	20	90	25	80	24	70	25	65	24	75	23	90	25
70	18	60	15	70	25	55	25	55	15	35	20	60	21	30	21	70	20	45	20	85	25	70	25	72	24	90	25
60	25	65	25	70	25	75	23	60	20	40	16	62	20	50	25	90	20	80	25	75	23	65	25	50	25	85	25
70	20	65	30	70	25	70	25	56	23	35	21	62	24	55	25	100	27	75	20	85	20	65	20	60	21	95	20
70	23	65	25	65	22	65	30	40	22	36	24	60	22	50	25	92	24	85	25	65	25	70	20	60	25	80	25
70	20	70	23	65	20	70	25	35	25	45	24	45	25	45	26	95	24	40	20	75	15	75	20	52	26	85	25
75	25	60	25	65	25	58	25	60	23	40	20	45	22	50	25	80	20	95	25	90	20	90	25	46	25	80	25
65	24	65	25	65	25	60	25	60	22	42	21	45	25	55	25	65	20	85	27	85	20	80	25	65	24	90	25
60	25	65	20	70	20	68	25	60	25	30	25	40	20	50	22	90	25	85	22	85	25	85	26	60	25	75	25
60	30	70	20	65	25	60	20	55	20	38	20	45	20	60	24	80	25	80	25	82	23	90	25	60	24	60	27
55	25	70	27	70	25	55	25	65	21	35	25	42	22	55	25	90	26	85	22	70	24	75	25	80	22	80	23
55	25	65	20	75	25	65	25	60	20	45	20	45	22	50	20	75	25	45	22	70	25	80	22	75	20	95	23
75	25	70	27	70	25	60	25	62	25	42	22	55	22	60	25	80	25	40	25	78	20	90	20	80	20	85	23
65	20	70	24	70	25	65	20	60	20	40	20	55	20	55	20	80	28	75	25	72	25	70	25	80	20	60	24

n = 20.

L = large oxyphaerasters; S = small oxyphaerasters.

Mean 65.0 23.8 64.8 23.6 66.8 24.0 64.2 24.3 56.2 22 37.7 21.8 52.7 22 50.6 23.7 84.1 23.7 73.3 23.2 78 22.8 72.8 23.2 68.5 23.8 80.5 24.3  
 SD 9.6 2.8 5.0 3.3 4.7 2.3 5.8 2.5 7.1 2.9 4.5 2.5 8.0 1.8 8.1 2.3 9.1 2.6 18.7 2.2 7.8 2.8 12.0 2.4 12.2 2.1 12.3 1.5  
 Range 35-75 18-30 50-70 15-27 55-75 20-28 55-75 20-30 35-65 15-27 30-45 16-25 40-62 20-25 30-65 20-28 65-100 20-28 40-95 20-25 65-92 15-25 50-90 20-26 46-95 20-27 60-95 20-27

and occasional smaller (35–45 µm diameter) identical forms, possibly developmental in nature (Figure 5f, Table 2). This species has larger oxysphaerasters than *C. secunda* (mean diameter 65.4 µm), and does not have the second smaller and morphologically distinct oxysphaeraster seen in *C. secunda*. Sequence data for these specimens could not be obtained, but a large geographical disjunction occurs between the tropical *C. grandistellata* (Indonesia) and the temperate *C. secunda* (south coast of Australia), as well as the differences in spicule sizes and morphologies, therefore we consider *C. secunda* to be a valid temperate species.

Sequencing of rDNA of nine specimens of *Chondrilla secunda* showed close molecular similarity between all specimens (Usher *et al.* 2004a), with up to 99.5% sequence identity.

### Distribution and habitat

*Chondrilla secunda* is found in Victoria, South Australia, and south Western Australia as far north as Jurien Bay (Figure 4). It is a temperate species occurring on heavily shaded rock faces at depths less than 15 m (Usher *et al.* 2004a). This species is uncommon. Specimen NMV F157468 from Queenscliff, Victoria collected on the 15<sup>th</sup> November 2001 at 5.4 m depth has synchronously developing spermatocysts 33 µm wide.

### *Chondrilla australiensis* Carter 1873

Figures 1, 4, 6

*Chondrilla australiensis* Carter 1873: 23, plate I, figures 10–14, 16; Lendenfeld 1885: 15.

*Chondrilla corticata* Lendenfeld 1885: 18, plate 4, figures 18–20, plate 5, figure 17; Hooper and Wiedenmayer 1994: 123.

*Chondrilla papillata* Lendenfeld 1885: 17, plate 5, figures 13–16; Hooper and Wiedenmayer 1994: 123.

### Material examined

*Holotype* of *Chondrilla australiensis*

**Australia: New South Wales:** Port Jackson, ca. 33°51'S, 151°16'E (BMNH 1895.8.9.1).

*Syntype* of *C. corticata*

**Australia: New South Wales:** Port Jackson, ca. 33°51'S, 151°16'E (AM G9050).

*Syntype* of *C. papillata*

**Australia: New South Wales:** Port Jackson, ca. 33°51'S, 151°16'E (AM G9051)

### Other material examined

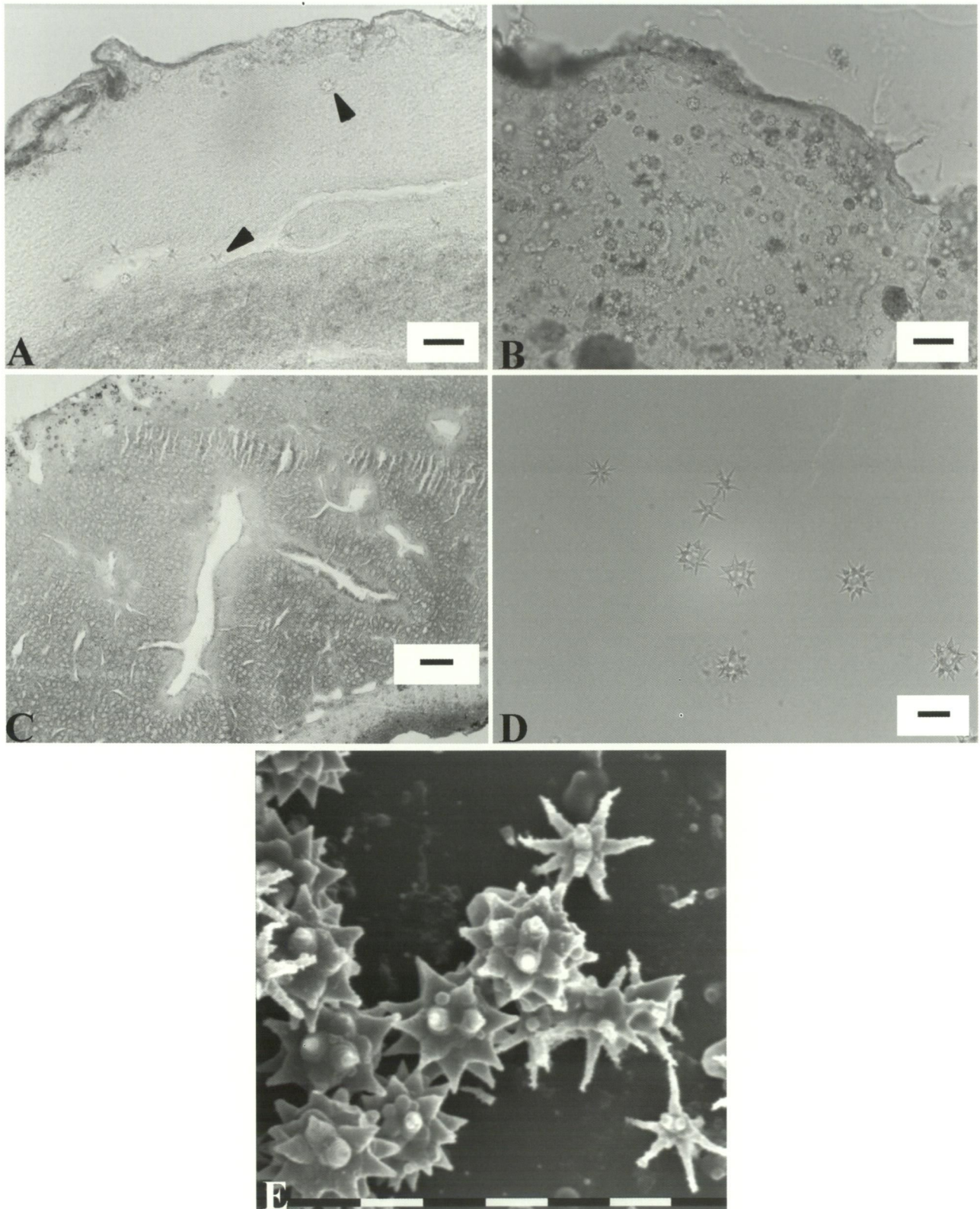
**Australia: Northern Territory:** 1 specimen, Coral Bay, Point Essington, 11°11'S, 132°03'E, < 1 m depth, collector J.N.A. Hooper and A.J. Bruce, snorkel, 19 July 1981 (NTM Z377). **Queensland:** 2 pieces, Roby Bay, Moreton Bay, 27°18'S, 153°22'E, < 1 m depth, collector S. Cook, 7 June 2001 (WAM Z13269); 2 pieces, Point Lookout, North Stradbroke Island, 27°28'S, 153°28'E, < 1 m depth, collector S. Cook, 2 June 2001 (WAM Z13263). **New South Wales:** 3 pieces, Bateau Bay, Central Coast, 33°23'S, 151°29'E, intertidal, collector J. Fromont & D. Sutton, 5 January 2001 (WAM Z13254); 2 pieces, Flinders Island, Wollongong, 34°26'S, 150°53'E, 11 m depth, collector A. Davis, SCUBA, 1 March 2001 (WAM Z13265). **Victoria:** 1 specimen, Port Phillip Bay, 37°58'S, 144°54'E (BMNH 1886.6.7.87–89). **Western Australia:** 2 pieces, Esperance jetty no.1, 33°51'S, 121°55'E, 10.7 m depth, collector K. Usher, SCUBA, 1 May 2001 (WAM Z13266); 4 pieces, Two People's Bay, Albany, 34°57'S, 118°11'E, 5.3 m depth, collector K. Usher, SCUBA, 8 March 2001 (WAM Z13272); 1 specimen, South Mole, Fremantle, 32°03'S, 115°45'E, 4–10 m depth, collector K. Usher SCUBA, 31 October 1999 (WAM Z13257); 1 specimen, South Mole, Fremantle, 32°03'S, 115°45'E, 4–10 m depth, collector K. Usher SCUBA, 31 October 1999 (WAM Z13255); 1 specimen, station JWAM13, transect 1, Julia Rocks, Jurien, 30°09.36'S, 114°59.72'E, 2.5–4.7 m depth, collector J. Fromont SCUBA, 3 May 2005 (WAM Z31393); 1 specimen, Mid-reef, Houtman Abrolhos, 28°46'S, 114°08'E, 24.8 m depth, collector K. Usher, SCUBA, 7 May 2000 (WAM Z13257); 1 fragment, Outer reef, Exmouth, 21°57'S, 114°07'E, 9.8 m depth, collector K. Usher, SCUBA, 15 May 2001 (WAM Z13278); 1 specimen, station DA3/99/42, Georgeff Reef, Dampier Archipelago, 20°29.34'S, 116°36.80'E, intertidal, collector J. Fromont, 28 August 1999 (WAM Z5419).

### Diagnosis

Characterised by forming thin encrusting sheets that vary greatly in size. Colour varies from maroon to ochre exterior with a cream interior. Spicule complement of oxysphaerasters (diameter range 16–38 µm, mean 25.9 µm, n = 360) and oxyasters (diameter range 15–35 µm, mean 23.8 µm, n = 360).

### Description

Habitus as in Figure 1e, f. Encrusting sponge of variable thickness 0.2 to 3.0 cm at thickest dimension. Individuals may form small encrusting patches to extensive spreading mats up to 1 m across. Oscules occur on the upper surface and are closed or very small (300 µm wide) in the preserved state. They can occur on low raised lobes 5 mm in height with slightly raised rims up



**Figure 6** Internal organisation and spicules of *Chondrilla australiensis*. A. Internal organisation of the holotype (BMNH 1895.8.9.1), scale bar = 50  $\mu$ m, spicules indicated by arrows. B. Internal organisation of the holotype (AM G9051) of *C. papillata*, scale bar = 50  $\mu$ m. C. Internal organisation of WAM Z13266, scale bar = 200  $\mu$ m. D. Spicules of WAM Z13266, scale bar = 20  $\mu$ m. E. SEM image of spicules of WAM Z13254, scale bar = 10  $\mu$ m.

to 1 mm high. The surface is shiny and slippery. Texture: Collagenous, compressible but not elastic. The interior is dense and compact with fine vertical canals throughout. *Colour*: Variable from chocolate brown to ochre or maroon. Surface colour is more homogenous in *C. australiensis* than in the other *Chondrilla* species occurring in Australia. Specimens in high light tend to be ochre and those in shade are maroon. Sides of sponges can be cream.

*General organisation*: (Figure 6a–c). Ectosome: Thin superficial layer 5–15 µm wide, usually more densely pigmented than the interior. Less pigmented layer of ectosome beneath is 110–270 µm thick. Spicules variable in this region, with some specimens including the holotype having sparse spicules. In the syntypes of *C. corticata* and *C. papillata* oxysphaerasters are dense in this region. Choanosome: The interior is differentiated from the ectosome in colour and density of the mesohyl. Spicules can be sparse in this region but are more dense around the edges of canals, or oxysphaerasters are dispersed evenly, or both oxysphaerasters and oxyasters are dense. In most specimens oxysphaerasters are the dominant spicule but in some the oxyasters are common or dominate, particularly lining the edges of canals (e.g. WAM Z13254 and Z13265). Some specimens have a basal layer of oxysphaerasters 150–400 µm thick (WAM Z13266, Z13258).

*Spicules*: (Figure 6d, e). Small oxysphaerasters with short thick rays tapering abruptly to points, or occasionally mammillate (diameter range 16–38 µm, mean 25.9 µm, n = 360). Small oxyasters with tapering microspined rays frequently bi- or multi-rayed and irregularly bent (diameter range 15–35 µm, mean 23.8 µm, n = 360) (Table 3).

*Cyanobacteria*: sponges were found to contain high concentrations of the unicellular cyanobacterium "*Candidatus* Synechococcus spongiarum" (Genbank) in surface tissues. "*Candidatus* S. spongiarum" was also found in samples of *Chondrilla nucula* from the Ligurian Sea, with the two symbionts having 100% 16S rDNA sequence similarity. This cyanobacterial species has not to date been found free-living in seawater (Usher *et al.* 2004c).

#### Remarks

Given the enormous variability in live colour, the variable distribution and abundance of the two spicule types within the skeleton, and the large geographical distribution of *Chondrilla australiensis* in Australia, it was extremely helpful in this study to have undertaken molecular analyses to complement the morphological analyses of this species (Usher *et al.* 2004a). We had previously successfully sequenced the

type material (BMNH 1895.8.9.1) and 17 other specimens of *C. australiensis*. Sequence similarities between the specimens ranged from 99.1% (4 base pair differences) to 100%, with most sequences only being 1 or 2 base pairs different from each other. However, the sample from Dampier typically had only 97.5% sequence similarity to other samples of *C. australiensis*. This species is now known from almost the entire coastline of Australia (Figure 4).

Numerous species of *Chondrilla* with oxysphaerasters and oxyasters as the spicule complement have been described including *C. mixta* Schulze 1877 from the Red Sea, *C. distincta* Schulze 1877 from the Caroline Islands, *C. nuda* Lendenfeld 1897 from Zanzibar, *C. media* Hentschel 1912 from Indonesia, and *C. agglutinans* Dendy 1916 from India (the latter four are all accepted as synonyms of *C. mixta*). *Chondrilla mixta* was reported from Christmas Island in the Indian Ocean by Kirkpatrick (1900). We did not examine this material and cannot determine if this species assignment is valid, or whether Kirkpatrick had found *C. australiensis*. The size of the spicules he described (oxysphaerasters 25–30 µm, oxyasters 24–28 µm) are within the size ranges we determined for *C. australiensis*. We attempted to collect *Chondrilla* from a limited number of locations at Christmas Island but were unsuccessful in finding any specimens.

*Chondrilla globulifera* Keller 1891 from the Red Sea and *C. ternatensis* Thiele 1900 from Indonesia (Ternate) are considered synonyms of *C. australiensis* (Burton, 1924). *Chondrilla jinensis* Hentschel 1912 from Indonesia is one of the few species with a spicule complement of oxysphaerasters and oxyasters to have been retained as a valid species, and it has larger spicules than *C. mixta* and *C. australiensis* (oxysphaerasters ca. 50 µm and oxyasters ≤ 80 µm).

#### Distribution and habitat

*Chondrilla australiensis* is found from the Northern Territory along the east coast of Australia to Victoria, and from south to north Western Australia. We did not find this species in South Australia, where sampling was minimal, and no collecting occurred in Tasmania. This is a widespread distribution, with specimens occurring in shallow tropical and temperate habitats from the intertidal to 30 m depth in both shaded and full light environments. The species is common, and is always encrusting with a highly variable extent of cover.

#### Key to Australian species of *Chondrilla*\*

1. Oxyasters present..... *Chondrilla australiensis*

Table 3 Spicule dimensions of *Chondrilla australiensis*<sup>1,2</sup>.

	NTM Z377		WAM Z13269		WAM Z13263		WAM Z13254		BMNH 1895.8.9.1		AM G9050		AM G9051		WAM Z13265		BMNH 1886.6.7.87 -89		WAM Z13266		WAM Z13272		WAM Z13257		WAM Z13255		WAM Z31393		WAM Z13258		WAM Z13278		WAM Z5419			
	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa	Os	Oa
23	25	25	20	25	20	25	25	25	20	20	23	18	20	20	20	20	30	23	20	25	25	23	20	20	20	20	22	28	25	23	18	35	25			
20	25	20	20	25	20	25	21	23	23	20	18	25	23	22	20	20	28	23	25	20	28	20	23	20	23	26	28	25	38	25	33	30				
20	25	20	20	25	20	25	25	28	23	18	18	25	20	20	22	20	20	28	28	28	20	25	20	28	20	25	26	28	23	35	25	33	25			
30	28	24	20	26	22	28	20	25	20	23	18	23	23	22	17	20	20	28	25	28	23	23	18	25	18	25	20	30	28	30	28	35	28			
33	28	24	20	22	25	26	20	25	25	20	18	18	23	23	20	20	30	23	28	20	28	23	28	20	26	25	28	28	23	23	25	25				
35	30	22	15	22	23	26	21	23	25	20	18	20	18	20	21	20	20	30	25	28	18	23	23	25	18	26	20	33	25	30	23	30	30			
35	25	24	20	25	23	22	22	28	20	20	18	20	20	23	18	18	20	33	25	28	20	28	23	23	20	25	25	33	25	18	25	28	28			
25	28	25	20	25	25	25	17	25	23	20	18	25	18	21	20	20	20	33	23	28	20	25	23	20	18	22	25	28	23	33	25	30	23			
33	28	25	20	25	25	25	23	23	23	20	28	23	22	20	23	23	30	28	20	18	25	23	25	25	20	20	33	28	35	25	30	23				
28	30	22	20	20	25	27	23	23	20	20	18	25	23	18	18	18	20	28	23	30	23	25	20	23	20	25	20	30	23	38	25	33	25			
28	28	25	20	26	22	27	17	23	25	23	20	23	20	16	18	23	18	28	25	28	28	28	20	25	20	23	22	28	25	35	25	33	28			
33	25	24	22	28	25	27	20	23	23	20	20	23	20	18	20	20	23	30	25	30	23	28	20	20	23	25	22	30	23	35	25	33	25			
30	33	24	20	26	20	25	23	25	20	20	20	25	23	20	22	18	23	30	25	30	25	28	20	20	23	27	20	33	25	38	25	33	25			
33	25	25	20	25	25	27	20	30	23	23	18	25	23	16	20	20	18	28	25	30	20	30	20	25	23	20	20	30	25	33	23	30	28			
35	25	23	25	20	20	20	20	25	20	23	18	20	23	20	18	20	23	33	23	28	23	28	20	20	20	25	25	30	25	20	23	30	25			
30	25	24	20	30	22	25	22	33	23	23	18	25	20	16	20	23	20	33	23	28	23	28	23	20	20	20	20	30	23	25	20	28	28			
28	28	25	20	30	30	26	20	23	20	25	20	25	23	20	20	20	18	30	20	28	23	30	23	28	23	26	22	30	25	38	25	35	28			
35	28	21	20	30	26	27	25	20	25	23	20	20	20	20	20	20	18	30	23	28	18	28	23	25	20	25	25	30	25	33	28	33	28			
35	25	26	22	25	25	25	25	28	20	20	20	20	20	20	15	18	18	33	25	28	25	25	23	25	20	22	25	28	25	40	25	28	28			
35	28	25	20	25	25	26	16	28	25	23	20	25	20	20	20	20	18	33	25	33	20	28	23	25	23	25	20	30	25	35	23	33	35			
Mean	30.2	27.1	23.7	20.2	25.3	23.4	25.5	21.3	25.3	22.3	21.35	18.9	23.2	21.05	19.9	19.5	20.1	20.0	30.3	24.3	27.7	21.8	26.8	21.6	23.7	20.7	23.8	22.5	29.9	25.0	31.8	24.2	31.4	27		
SD	5.0	2.3	1.8	1.7	2.8	2.7	1.8	2.7	3.0	2.1	1.8	1.0	2.6	1.9	2.1	1.7	1.5	1.8	2.0	1.8	3.0	2.8	2.1	1.7	2.8	1.9	2.3	2.4	1.8	1.6	6.5	2.3	2.8	2.8		
Range	20	25	20	15	20	20	22	16	20	20	18	18	18	16	16	18	18	28	20	20	18	23	18	20	18	20	20	28	23	18	20	25	23			
	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to	to		
	35	33	25	25	30	30	28	25	30	25	25	20	25	23	23	22	23	23	33	28	33	28	30	23	28	23	27	26	33	28	40	28	35	35		

<sup>1</sup> n = 20.

<sup>2</sup> Os = oxysphaeraster, Oa = oxyaster.

- Oxyasters absent..... 2
2. Oxysphaerasters present 1 small size.....  
 .....*Chondrilla linnaei*
- Oxysphaerasters present 2 sizes .....  
 .....*Chondrilla secunda*

\*we have not included *Chondrilla mixta* in this key as it was not found in this study and its presence in Australia remains to be determined.

## DISCUSSION

In this study we found three species of *Chondrilla* with distinctive spicule complements. *Chondrilla australiensis* has two spicule types; oxysphaerasters and oxyasters of similar small size. *Chondrilla linnaei* only has oxysphaerasters of a single small size category, while *C. secunda* has two types of oxysphaerasters, one a mammillate oxysphaeraster with a large size range and a second smaller oxysphaeraster with pointed, conical rays and a small size range. The distribution and abundance of spicules throughout the sponge body is highly variable for all three species. In most specimens spicules are abundant in the ectosome, or in the apical surface layer of the sponge and/or around internal canals, and more dispersed in the choanosome. Some specimens had a basal layer of spicules. In other specimens spicules were more evenly distributed throughout the sponge body. The most reliable morphological characters of these three species are their distinctive spicule complements and their growth forms. *Chondrilla linnaei* forms small discrete encrusting lobes, *C. secunda* comprises larger thick encrusting individuals and *C. australiensis* forms extensive thin encrusting mats. Neither *C. linnaei* nor *C. secunda* ever form the extensive mats common to *C. australiensis*. However, even a trained collector will have difficulty accurately differentiating these species in the field, and a spicule check will be essential.

Cavalcanti *et al.* (2007) recently reported very high variability in some of the common characters used in sponge taxonomy, including skeletal organization, surface morphological features and spicule sizes, and they could not distinguish cryptic species of *Chondrilla* on the basis of these characters. We are fortunate that the Australian species described here have distinctive spicule complements.

This morphological study of Australian *Chondrilla* species supports molecular data that clearly distinguished the three species using two different gene regions (Usher *et al.* 2004a). For the species described here, the greatest genetic distance occurred between *C. secunda* and *C. australiensis* (85.5% and 86.1% sequence similarity for the C2D2 and ITS regions, respectively). The

C2D2 sequence similarity between the holotypes of *C. australiensis* and *C. linnaei* was 89.7% and between *C. secunda* and *C. linnaei* 88.1%. These results confirm the existence of three species. Intraspecific genetic similarity was consistently high within all three species, and confirmed the south and west coast distributions of *C. secunda* and *C. linnaei*, and the widespread, almost circum-Australian, distribution of *C. australiensis*.

Klautau *et al.* (1999) identified five distinct genetic forms within *Chondrilla nucula* using allozyme techniques, and found that variation in spicule size did not correlate with the boundaries defined genetically. Some of these genetic forms have recently been studied by Vilanova *et al.* (2007) who found that the sulphated polysaccharide content distinguished cryptic species. This technique can be used on formalin fixed, frozen and dried specimens as well as those preserved in ethanol (Vilanova *et al.* 2007). These results suggest that there are a number of species with almost identical spicule complements and sizes that have been found in the Caribbean Sea and south western Atlantic that would previously have been called *C. nucula*, thus erroneously contributing to the cosmopolitan distribution of this species. Instead these are new species awaiting formal description, and the distribution of *C. nucula* is thought to be restricted to the Adriatic and Mediterranean seas (Klautau *et al.* 1999). The incorporation of novel character sets is essential for the determination of species within *Chondrilla* and many other sponge genera with few distinguishing morphological characters.

The three Australian species occurred in sympatry along the south and west coasts of Australia. *Chondrilla australiensis* has the most widespread distribution including both tropical and temperate regions of Australia, while *C. secunda* and *C. linnaei* appear restricted to temperate south and west Australia. It may be that the species have temporal separation of reproductive activity. We found spermatocysts in the specimen of *C. secunda* collected from Victoria in mid November, while Usher *et al.* (2004b) reported sperm development occurring over two weeks in February/March in *C. australiensis* from Fremantle, Western Australia. If *C. secunda* also has short periods of sperm development (it has now been found that most sponge species do), then these species are reproducing at different times of the year. Studies on the reproduction of *C. secunda* and *C. linnaei* would progress this hypothesis.

In a recent study Usher and Ereskovsky (2005) noted that coeloblastulae larvae of *Chondrilla australiensis* are short lived and begin to settle after a free-swimming period of 24–36 h. These short



dispersion abilities are now commonly reported for sponges and may account for the short range endemism of many species. The almost circum-Australian distribution of *C. australiensis* could be explained partly by longshore currents along continuous coastline gradually dispersing larvae or asexual products. Fromont (1999) first suggested that asexual fragmentation may occur in *C. australiensis*, and this has been supported by Zilberberg *et al.* (2006) who found similar asexual products of *Chondrilla* species from the Caribbean and Brazilian coastline. Their study found low clonality (7%) in a heterogeneous environment with strong upwelling, and higher clonality (39%) in a more homogeneous and temporally stable environment. A similar study of the population genetics of Australian *Chondrilla* species would increase understanding of their dispersal abilities and thus their differing biogeographic distributions.

Individuals of *Chondrilla australiensis* have been found to contain the unicellular cyanobacterial symbiont "*Candidatus Synechococcus spongiarum*" (Usher *et al.* 2004c). These symbionts are apparently transferred to the young by vertical transmission via developing eggs and occasionally sperm (Usher *et al.* 2005). This symbiont was not found in *C. secunda* or *C. linnaei*, which contain symbionts with 99.8% and 99.7% partial sequence similarity, respectively, to *Synechococcus* WH 8103 (GenBank), a cyanobacterium from the water column (Usher *et al.* 2004c). Although the presence of "*Candidatus Synechococcus spongiarum*" distinguished *C. australiensis* from *C. secunda* and *C. linnaei*, some sponge symbionts occur in more than one species and over vast geographic distances. For example, Usher *et al.* 2004c found "*Candidatus Synechococcus spongiarum*" in samples of *C. nucula* from the Ligurian Sea had 100% 16S rDNA sequence similarity to those sequenced from *C. australiensis*.

This is the third recent publication describing new species of *Chondrilla*. Desqueyroux-Faundez and Van Soest (1997) described a new species from the Galápagos Islands, and two new species have been described from Mexico (Carballo *et al.* 2003). The recent awareness of the cryptic nature of *Chondrilla* species (Klautau *et al.* 1999, Usher *et al.* 2004a, Vilanova *et al.* 2007) suggests that many more species are likely to be found.

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# Guide to Authors

## Subject Matter

Original research, reviews and observations in all branches of natural science and human studies will be considered for publication. However, emphasis is placed on studies pertaining to Western Australia and neighboring regions. Longer papers will be considered for publication as Supplements to the *Records of the Western Australian Museum*. Such publications may attract charges to the authors to offset the costs of printing – authors should consult the editors before submitting large manuscripts. Short communications should not normally exceed three typed pages and this category of paper is intended to accommodate observations, results or new records of *significance*. All material must be original and not have been published elsewhere.

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An abstract must be given in full length papers but not short communications, summarizing the scope of the work and principal findings. It should normally not exceed 2% of the paper and be suitable for reprinting in reference periodicals. At the end of the abstract, provide several keywords not already included in the title.

The International System of units should be used. Spelling should follow the *Concise Oxford Dictionary*. Numbers should be spelled out from one to nine in descriptive text; figures used for 10 or more. For associated groups, figures should be used consistently (e.g., “5 to 10”, not “five to 10”).

Systematic papers must conform with the International Codes of Botanical and Zoological Nomenclature and, as far as possible, with their recommendations.

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## References

In the body of the text, references should be cited as follows:

McKenzie and colleagues (McKenzie 1999, 2000; McKenzie *et al.* 2000) found that bat frequencies were highest on full moons, contra previous workers (Smith and Jones 1982; Berman 1988; Zucker *et al.* 1992).

For citing taxonomic groups and the author, a comma occurs between them:

The family Carphodactylidae consists of *Carphodactylus* Smith, 1999, *Nephrurus* Jones, 1999, *Orroya* Couper, Covacevich and Hoskin, 2001, *Phyllurus* Sprong, 1888 and *Saltuarius* Hammond, 1901.

All references must be cited in the text by author and date and all must be listed alphabetically at the end of the paper. The names of journals are to be given in full. Consult a recent edition of the *Records* for style. For taxonomic papers, include full references for all taxonomic groups mentioned in the text. In manuscripts dealing with historical subjects references may be cited as footnotes.

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## Editors

Manuscripts can be submitted to either Paul Doughty (paul.doughty@museum.wa.gov.au); human studies [anthropology, archaeology or history] and vertebrate animals) or Mark Harvey (mark.harvey@museum.wa.gov.au); invertebrate animals).

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