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# Abstract

Specimens of *Daphnia* found in Lake Burley Griffin in December 1994 (austral summer), were taken into laboratory culture where males and ephippial females were generated. All the key morphological features of the females, males and ephippia (resting eggs) fitted those described for *Daphnia obtusa* Kurz, 1874 emend Scourfield, 1942 not yet recorded from Australia. There were major differences between the specimens from Lake Burley Griffin and descriptions of *D. jollyi*, the only other species from the subgenus *Daphnia* in Australia which has a fringe of long setae in the mid-region of the ventral margin of the carapace. In addition, *D. jollyi* is endemic to the south-west of Western Australia. The discovery of *D. obtusa* does not provide evidence for the existence of this taxon in Australia since the breakup of Gondwana in the Cretaceous. Its discovery in an artificial lake opened in 1964, and which has been extensively sampled without detection of this species over the last 10–20 years, suggests a recent introduction from overseas.

# Introduction

Collections of zooplankton from Lake Burley Griffin made during the austral summer of 1994 contained specimens of *Daphnia* that did not correspond to any of the descriptions of taxa previously recorded from Australia (Benzie, 1988). Comparison of the specimens with the world literature showed a close fit with *D. obtusa*, a species originally described from Central Europe and typically found in temporary pools and puddles in lowland and alpine areas. *D. obtusa* has been found in North and South America, Asia, South Africa (Benzie, 1987) and probably New Zealand (Hrbacek, 1987), but never recorded from Australia (Smirnov & Timms, 1983; Benzie, 1987; Benzie, 1988).

This note documents the first record of D. obtusa in Australia, provides descriptions of the key features of the parthenogenetic female, ephippium and the male that identify the specimens as D. obtusa, and discusses the implications of the discovery for daphniid biogeography.

#### **Materials and methods**

Specimens were obtained by plankton tows using a 200  $\mu$ m mesh net at several sites in the east basin of Lake Burley Griffin, the Jerrabomberra Wetlands and Molonglo Reach on 12 December 1994. Environmental parameters of a water sample from 1 m depth in the east basin on the date of collection were as follows: temperature 25.7 °C, O<sub>2</sub> 6.8–7.3 mg 1<sup>-1</sup>, turbidity slight, colour yellow, no surface debris or algal scums, trace concentrations of the blue green alga, *Microcystis aeruginosa* detected. Animals were taken for live culture and identification in the laboratory and all samples were processed within two days of collection. Specimens used in the descriptions were preserved in 70% alcohol. Males and ephippial females were produced

in culture demonstrating the population to be cyclically parthenogenetic, and providing the opportunity to obtain data on male and ephippium characters.

Specimens were dissected in polyvinyl-lactophenol with chlorazol black added to highlight the nonpigmented chitin in the limbs, and left for several days to cure prior to microscopical examination. The specimens used to obtain the descriptions were deposited in the Australian Museum, Sydney (reference numbers P46508-P46511). In the descriptions given below, the mean  $\pm$  the standard error followed by the range (in parentheses) are given for body size measurements (all in mm) taken in lateral aspect of animals not depressed by the weight of cover slips. Body length excludes the tail-spine, body width is taken at the midpoint of the carapace and head width horizontally at the base of the head. Naming of limb parts follows Harnisch (1950).

## Results

### Diagnosis

This species is distinguished from other species in the subgenus *Daphnia* in Australia by the presence of the following combination of characters: the female has a fringe of long setae in the mid-ventral region of the carapace; first and second (proximal) combs of teeth on the postabdominal claw distinctly larger than the third (distal) comb; rounded, not right-angled, fornices; and the first postabdominal process is not distinctly hairy. The male has a lateral seta on the basipodite of the antennule, and a flagellum slightly longer than the basipodite tapering to a setose tip and not a spatulate tip.

Description of female and ephippium (Figures 1a, c, d, f, i, k, m) Body length (BL)  $1.50 \pm 0.09$  (1.4–1.7) Body width (BW)  $0.99 \pm 0.07$  (0.9–1.1) Head width (HW)  $0.55 \pm 0.08$  (0.5–0.7) BW/BL 0.7 HW/BW 0.5 (sample size = 10)

Medium sized (1-2 mm), with short tail-spine (<0.1 body length), ventral and dorsal margins of the carapace only spinose on posterior half (Figure 1a), ventral margin of carapace with long setae at mid-line (Figure 1d). Cephalic shield penetrates dorsal shield as a broad wedge. Well-developed fornices rounded.

Head relatively small (HW/BW 0.5); ventral margin of head concave; short, pointed rostrum just exceeds antennule (Figure 1f). Ends of sensory papillae exceed the tip of the rostrum. Eye large, filling antero-ventral quadrant of head, touches ventral margin of head ('forehead'). Ocellus large. Cecae short, breadth 0.3 width (Figure 1a). Colour translucent. Postabdomen long, sub-triangular, finely-tapering with three dorsal processes, length ratios approximately 4:2:1, first naked or slightly spinose, second and third spinose (Figure 1i). Terminal claw slightly curved, three external combs with 11-12, 17-19 and 47-54 spines; first and, particularly, second row of spines distinctly larger than third (Figure 1k). 10-12 sickle-shaped anal spines, length and curvature reducing dorsally. Dorsal margin of postabdomen flat. Serrate seta on last endite of endopodite of second thoracic limb with distinct fine spinules along distal two thirds of seta (Figure 1m).

Ephippium length at mid-line 0.5 mm, width at mid-line 0.4 mm. Tapering posteriorly with dorsal margin gently curved and two small egg chambers with long axes directed at right angles to dorsal margin. No spines on dorsal margin, but small irregularities observed at high magnification. No anterior or posterior spines. Fine irregular polygonal reticulation over whole surface of ephippium. Colour dark-brown to black, no colour differentiation of egg chambers. The only ephippium provided for description was already free from the carapace and the size of the ephippial female is unknown.

Description of male (Figures 1b, e, g, h, j, l) Body length (BL)  $0.87 \pm 0.02$  (0.88-0.93) Body width (BW)  $0.52 \pm 0.02$  (0.50-0.54) Head width (HW)  $0.35 \pm 0.02$  (0.31-0.39) BW/BL 0.6 HW/BW 0.7 (sample size = 5)

Small (<1.0 mm), with short tail-spine (<0.25 body length), ventral and dorsal margins of the carapace only spinose on posterior half (Figure 1b), ventral margin of carapace setose from anterior shoulder to mid-line, setae shortening posteriorly until midline where they lengthen again (Figure 1e). Cephalic shield penetrates dorsal shield as a broad wedge. Well-developed fornices rounded. Ventral margin of head flat or with slight concavity below eye; rostrum reduced to a knob hidden by antennule (Figure 1g). Eye large, filling antero-ventral quadrant of head, touching ventral margin of head ('forehead'). Ocellus large.

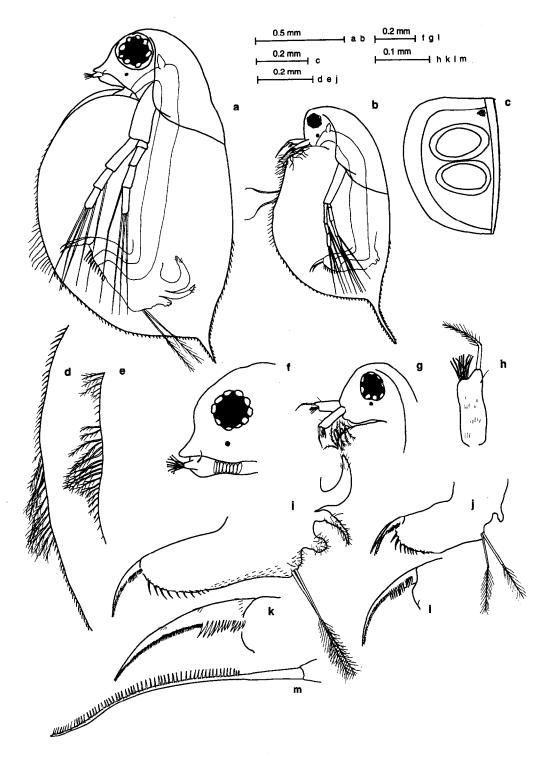


Figure 1. Daphnia obtusa (a) female, left lateral view. (b) male, left lateral view. (c) female, ephippium. (d) female, detail of ventral margin of carapace. (e) male, detail of ventral margin of carapace. (f) female, detail of head and antennule. (g) male, detail of head and antennule. (h) male, antennule. (i) female, postabdomen. (j) male, postabdomen. (k) female, detail of postabdominal claw. (l) male, detail of postabdominal claw. (m) female, serrate seta on last endite of endopodite of second thoracic limb with distinct fine spinules.

Antennule long, mobile, with one lateral seta on dorsal margin on basipodite, width:length ratio of basipodite 0.3 (Figure 1h). Terminal seta 1.1 times length of basipodite, setose towards tip. No spatula or thickening of terminal seta towards tip. Cecae short, broad 0.3-0.5 length (Figure 1b). Colour translucent. Postabdomen sub-triangular, finely-tapering with one spinose dorsal process poorly developed and slightly spinose (Figure 1i). Terminal claw long, slightly curved, with three external combs with 8-10, 17-23 and 38-43 spines, first two rows of spines markedly larger than the third (only clear at high magnifications). 8-9 sickle-shaped anal spines, length and curvature reducing dorsally. Dorsal margin of postabdomen flat. Serrate seta on last endite of endopodite of second thoracic limb with distinct spinules, more robust than those of the female, along distal half of seta.

## Discussion

The fact that the cephalic shield penetrates dorsal shield in a broad curve showed the species was a member of the subgenus *Daphnia*. In the subgenus *Ctenodaphnia*, the only other subgenus in the genus *Daphnia*, the dorsal shield has an anterior extension which penetrates the cephalic shield (Brooks, 1959).

The possession of a fringe of long setae in the midventral region of the carapace in the female restricts the identity of the specimens to three described taxa in the subgenus Daphnia: Daphnia obtusa, D. peruviana and D. jollvi. Only D. jollvi has been recorded in Australia, where it is endemic to the south-west of Western Australia (Petkovski, 1973; Benzie & Bayly, 1996). However, female D. jollyi differ from the Lake Burley Griffin specimens in that they have a distinctly setose first abdominal process, a shorter, less pointed rostrum, teeth in the first two combs of the post-abdominal claw which are similar in size to each other and to the teeth of the third comb, and right angled, not rounded, fornices. In addition, the ephippium of D. jollyi is sub-rectangular in shape and the orientation of the relatively large egg chambers is at an angle to the dorsal margin of the carapace. Males of D. jollyi do not have a lateral seta on the basipodite of the antennule, but have a seta situated terminally on the basipodite near to the terminal flagellum.

D. peruviana has been recorded only from the high Andes near Lake Titicaca in Peru (Harding, 1955), and is distinguished from D. obtusa by having a truncated rostrum, and an ephippium where the anterior margin makes more than a right angle with the dorsal margin of the carapace. Harding (1955) even expressed some doubt about separating *D. peruviana* from *D. obtusa*. The Lake Burley Griffin specimens do not have the truncated rostrum characteristic of *D. peruviana*, and the anterior margin of the ephippium makes a sharp right angle with the dorsal margin of the carapace. Finally, *D. peruviana* (and *D. jollyi*) are deeply pigmented while *D. obtusa* is more variable in colour and can be pigmented to some degree like specimens Harding (1955) obtained from the high Andes, or, more commonly, translucent, like the specimens from Lake Burley Griffin.

The females from Lake Burley Griffin show the key characteristics of *D. obtusa*: a fringe of long setae in the mid-region of the ventral margin on the carapace, a short but pointed rostrum and larger teeth on the first and second combs, but particularly the second comb, on the postabdominal claw. The ephippium of the Lake Burley Griffin specimens tapers posteriorly and has two relatively small egg chambers whose long axis is oriented perpendicular to the dorsal margin of the carapace. This is typical of the subgenus (Brooks, 1959) and is similar to that described for *D. obtusa* by Scourfield (1942) except for the complete lack of a posterior spine. Negrea (1983), however, illustrated the ephippium of European *D. obtusa* without a posterior spine, identical to the Australian material.

Comparison of the Australian material with illustrations in the original description by Kurz (1874) suggest the Australian females have a shorter rostrum, more rounded carapace and larger ocellus than European specimens. Males from Lake Burley Griffin also appeared to have a larger carapace and ocellus than those described by Kurz (1874). However, the general body shape of Australian females more closely resemble illustrations of European material given by Negrea (1983) and Scourfield (1942), although the specimens illustrated by Scourfield have a more deeply concave ventral margin on the head, and, perhaps as a result, a longer more distinctive rostrum. The males from Europe appear to have a larger head width (Negrea, 1983). Otherwise the descriptions of Australian males are identical to descriptions of D. obtusa males (Kurz, 1874; Scourfield, 1942; Negrea, 1983). They have a lateral seta on the basipodite, a flagellum slightly longer than the basipodite and which tapers to a setose tip. Hudec (1990) illustrates the serrate spine on the distal endite of the epipodite of the second thoracic limb as having large robust spinules. The spinules in Australian females were distinct, long, relatively fine

and more numerous than those illustrated by Hudec, but if crossing the plane of focus, can appear to have a broader base and more robust appearance. It is not clear whether the differences between Hudec's illustrations and the Australian material reflect differences in the animals or the field of focus used in the illustrations. In any case, no differences from the descriptions of *D. obtusa* were observed that might suggest the specimens represent a species new to science.

D. obtusa is widespread in Eurasia, North and South America and is found in South Africa (Benzie, 1987). The discovery of D. obtusa in Lake Burley Griffin might suggest that this widespread taxon has also existed in Australia since its evolution, presumably prior to the breakup of Gondwana. However, Lake Burley Griffin is an artificial lake opened in 1964, and has been extensively sampled without detection of this species, over the last 10-20 years (Benzie, 1988). Extensive collecting throughout the continent (Benzie, 1988; Smirnov and Timms, 1983; Bayly, 1982; Bayly, 1992) has not revealed the species before, even in refugia such as the south-west of Western Australia that have produced relict species such as D. occidentalis (Benzie, 1986a) and D. jollyi (Petkovski, 1973, Benzie & Bayly, 1996). The latter species represent discrete clades within the subgenus (Benzie, 1986b, 1987; Benzie & Bayly, 1996). Given the absence of D. obtusa in previous collections from throughout Australia, the detection of D. obtusa in only one location, and one which has a recent origin, suggests the species is a recent introduction to Australia, though the mechanism of introduction remains unknown.

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# References

- Bayly, I. A. E., 1982. Invertebrate fauna and ecology of temporary pools on granite outcrops in southern Western Australia. Aust. J. mar. Freshwat. Res. 33: 599–606.
- Bayly, I. A. E., 1992. The microcrustacea and physicochemical features of temporary ponds near Northcliffe, Western Australia. J. R. Soc. Western Australia 75: 99–106.
- Benzie, J. A. H., 1986a. Phenetic and cladistic analyses of the phylogenetic relationships within the genus *Daphnia* worldwide. Hydrobiologia 140: 105–124.
- Benzie, J. A. H., 1986b. Daphnia occidentalis, new species (Cladocera: Daphniidae) from western Australia: new evidence on the evolution of the north american D. ambigua D. middendorffiana group. J. Crust. Biol. 6: 232–245.
- Benzie, J. A. H., 1987. The biogeography of Australian Daphnia: Clues to an ancient (>70 m.y.) origin from the genus. Hydrobiologia 145: 51–65.
- Benzie, J. A. H., 1988. The systematics of Australian Daphnia (Cladocera: Daphniidae). Species descriptions and keys. Hydrobiologia 166: 95-161.
- Benzie, J. A. H. & I. A. E. Bayly, 1996. Male and ephippial female Daphnia jollyi Petkovski 1973, discovered in Western Australia and the parthenogenetic female redescribed. Hydrobiologia (in press).
- Brooks, J. L., 1959. Cladocera. In W. T. Edmodson (ed.), Freshwater Biology. Wiley and Sons, New York: 587–656.
- Harding, J. P., 1955. The Percy Sladen Trust Expedition to Lake Titicaca in 1937. Crustacea; Cladocera. Trans. linn. Soc. Lond. (Zoology) 1: 329–354.
- Harnisch, O., 1950. Vergleichende Beobachtungen zum Nahrungserwerb von Daphnia und Ceriodaphnia. Zool. Jhrb. Abt. Syst. 78: 173–192.
- Hudec, I., 1990. Vyskyt a biologia druhov rodu Daphnia, podrodu Daphnia (Caldocera, Daphniidae) na slovensku. 1. Cast: D. obtusa, D. pulicaria, D. parvula. Biologia (Bratislava) 45: 491–499.
- Hrbacek, J., 1987. Systematics and biogeography of *Daphnia* species in the northern temperate region. Mem. Ist. ital. Idrobiol. 45: 37– 76.
- Kurz, W., 1874. Dodekas neuer Cladoceren nebst einer kurzen Ubersicht der Cladocerenfauna Bohmens. Osterreichische Akademie der wissenschaften Mathematisch-naturwissenschaftliche Klasse. Sitzungsberichte. 70: 1–88.
- Negrea, S., 1983. Crustacea, Cladocera. Fauna Republicii Socialiste Romania. Vol IV, Fascicula 12. Academiei Republicii Socialiste Romania, Bucuresti, 380 pp.
- Petkovski, T. K., 1973. Zur Cladoceren-fauna Australiens. I. Daphniidae und Chydoridae. Acta Mus. maced. Sci. nat. 13: 133–157.
- Scourfield, 1942. The 'pulex' forms of *Daphnia* and their separation into two distinct series, represented by *D. pulex* (De Geer) and *D. obtusa* Kurz. Ann mag. nat. Hist. 9: 202–219.
- Smirnov, N. N. & B. V. Timms, 1983. A revision of the Australian Cladocera (Crustacea). Rec. Aust. Mus. Suppl. 1: 1–132.