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Egg dormancy in the Australian estuarine-endemic copepods *Gippslandia estuarina* and *Sulcanus conflictus*, with reference to dormancy of other estuarine fauna

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Abstract. The presence of dormant life history stages was investigated for the zooplankton of an annually flooding salt-wedge estuary. Such stages are seen as a potential mechanism for population persistence following environmental adversity. Laboratory incubation experiments were conducted on estuarine sediments. As a result, dormant eggs in Australian estuarine-endemic copepods are reported for the first time. Nauplii of the dominant estuarine-endemic calanoids *Gippslandia estuarina* and *Sulcanus conflictus* commonly hatched from the sediments. Manipulation of the salinity and temperature of experimental media indicated that temperature was the more important hatching trigger for *S. conflictus*, and that both high salinity and high temperature were important for *G. estuarina*. Results of the incubation experiments, including those of 'conversion' experiments (i.e. from freshwater to saline conditions or low temperature to high temperature), help to elucidate the type of dormancy characteristic of each species; it appears that *S. conflictus* may have diapause eggs and *G. estuarina* may have quiescent eggs, although this is yet to be confirmed. Other estuarine fauna developed from the mud during the incubation experiments, most notably the harpacticoids *Onychocamptus chathamensis*, an ectinosomatid and *Schizopera* sp., and the medusa *Australomedusa baylii*. Ecological and evolutionary consequences of dormancy in these estuarine-endemic zooplankton are briefly discussed.

Introduction

A dormant life history stage is often critical to the existence of a species, by ensuring its persistence during periods that are unfavourable for development, reproduction or adult survival. Dormancy represents a spectrum of suppressed development ranging from diapause (arrested development) to quiescence (retarded development). Diapause stages cannot develop, even under optimal conditions, until the passage of a required dormancy period, the refractory phase, the duration of which may be affected by environmental conditions such as temperature (Grice and Marcus 1981; Marcus 1996). Diapause is typically triggered by a stimulus prior to the onset of environmental adversity; it ensures long-term viability and synchronizes the life cycle of the species with the environment (Grice and Marcus 1981; Marcus 1996). In contrast, quiescence is induced by adverse environmental conditions and requires no prior acclimation, and development resumes immediately suitable conditions return (Grice and Marcus 1981).

Stages of dormancy in aquatic organisms may include the encystment of adults or juveniles, or the production of resistant eggs or propagules. Dormancy of adults or juvenile stages is prevalent among freshwater crustaceans (e.g. Morton and Bayly 1977; Elgmork 1980; McKaige 1980; Fryer 1996), but is rarer in marine invertebrates, having been reported in a marine harpacticoid (Coull and Grant 1981) and a few marine calanoids (Grice and Marcus 1981). The most common form of dormancy in aquatic invertebrates is that of dormant eggs or cysts, which are known for both marine and freshwater dinoflagellates, tintinnids, rotifers, nematodes, ostracods, cladocerans, and calanoid, cyclopoid and harpacticoid copepods (Barnes 1974; Wall and Evitt 1975; Coull and Grant 1981; Fryer 1996; Marcus 1996).

Dormant eggs have been found to be particularly prevalent among the neritic marine Calanoida (Marcus 1996). For example, the phenomenon of 'winter disappearance – spring reappearance' is common among temperate neritic calanoid copepods of the Northern Hemisphere and first prompted the discovery of temperature-dependent egg dormancy in this group (e.g. Zillioux and Gonzalez 1972). Subsequently, dormant eggs of at least 38 species of calanoids have now been observed to be in, or to hatch from, marine sediments where they occur seasonally in large numbers (Marcus 1996; Chen and Marcus 1997). The survival of dormant eggs in sediments during periods that are not suitable for the existence of later life-cycle stages in the water-column is seen as critical for the perpetuation of such species (Marcus 1984). Three dormant egg types have been distinguished: quiescent subitaneous, diapause (for 14 species, Marcus 1996) and delayed hatching (for 2 species, Chen and Marcus 1997). Eggs with delayed hatching hatch gradually over an extended period regardless of incubation temperature, and they do not require a change in temperature to hatch (Chen and Marcus 1997).

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The calanoid species known to have dormant eggs are from the families Centropagidae, Temoridae, Pontellidae, Acartiidae and Tortanidae—all of the super-family Diaptomoidea (Andronov 1991) [note: super-family quoted as Centropagoidea in Bowman and Abele (1982) and Lindley (1990,1992)]. Several of these families have euryhaline-marine representatives that occur in estuarine environments, which suggests that the ability to produce dormant eggs may also extend to the estuarine-endemic (i.e. 'true estuarine' species that live and breed exclusively in the estuary) Calanoida. Indeed, such a strategy would seem more useful given the often dynamic and unpredictable nature of estuarine environments compared with the more stable marine environment.

The presence of dormant life history stages, including the occurrence of dormant eggs in copepods, has not previously been investigated for Australian estuarine zooplankton. The Hopkins River estuary is a highly dynamic salt-wedge type which undergoes scouring floods followed by lengthy periods of freshwater inundation, on an almost annual basis (Newton 1994, 1996). Such floods generally result in complete flushing of saline waters from the estuary and the bulk of the zooplankton community is lost by entrainment. In contrast, in years of extreme low rainfall, generally once or twice per decade, the mouth of the estuary may close leading to hypersaline conditions. Despite such environmental variability, the system is dominated by a small but highly abundant assemblage of estuarine-endemic zooplankton species (Newton 1994). The holoplanktonic component of this assemblage is dominated by the calanoid copepods *Gippslandia estuarina*, *Gladioferens pectinatus*, *Sulcanus conflictus* and *Isias uncipes*, an undescribed ectinosomatid harpacticoid copepod, and the medusa *Australomedusa baylii*. The presence of dormant life history stages would be advantageous for members of this assemblage, primarily to overcome the adverse environmental conditions resulting from flooding.

A technique commonly used to test for the presence of viable dormant eggs in marine sediments is that of laboratory incubation of sediments, or 'mud hatching' experiments (e.g. Zillioux and Gonzalez 1972, Johnson 1980; Marcus 1984, 1990; Lindley 1990). Mud hatching experiments were conducted to determine the presence and viability of dormant stages within sediments from the Hopkins River estuary, to assess dormancy status (i.e. diapause or quiescence) of life history stages, and to identify possible environmental triggers of the hatching response of dormant stages.

Materials and methods

Study site

The Hopkins River estuary is a relatively small (9.2 km) truncated salt-wedge type situated on the south-western coast of Victoria, Australia, near Warrnambool (Fig. 1). A detailed description of the geomorphology of the estuary and its annual hydrological cycle are given in Newton (1996). The hydrological cycle has two main periods: High Flow – during late winter to early spring, river discharge peaks and causes high-velocity floods that flush out the ageing and largely deoxygenated salt-wedge, resulting in extended periods of freshwater conditions throughout the estuary (i.e. 1–3 months); and Low Flow – in late spring, as discharge decreases, a new salt-wedge is gradually emplaced along the estuary, and subsequently, the highly stratified salt-wedge stagnates (particularly in deeper waters) over summer and autumn. Sediments of the estuary comprise fine silts and anoxic black mud with a high detrital content.

Sampling

Estuarine sediments were obtained in an Ekman grab, with only the top few centimetres of sediments being retained for the mud incubation experiments. Mud samples were taken from two sites along a perpendicular transect

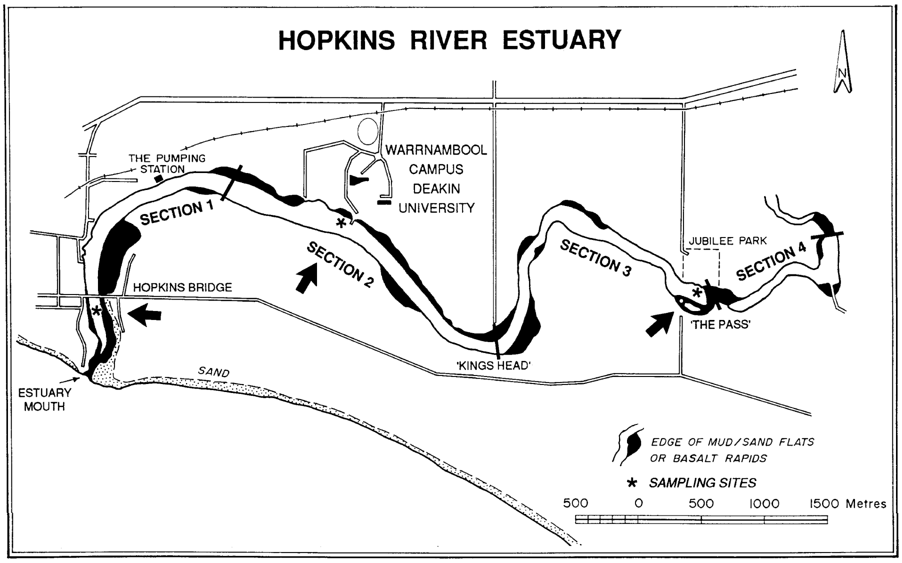

Fig. 1. Map of the Hopkins River Estuary showing the three locations of twin sites sampled for estuarine sediments as used in the mud incubation experiments.

in each of the first three sections of the estuary (Fig. 1). One site was on the border or edge formed where littoral vegetation met open channel waters and the other was situated at approximately mid channel under open waters. Samples were collected on 5 July, 5 August and 16 September 1985, a period that normally coincides with highest river discharge. [Note: although mud sampling coincided with the peak river flow period of 1985 and there were significant decreases in salinity throughout the system (e.g. down to 1.2), discharge did not reach the critical level for complete flushing of saline waters from the estuary (i.e. 4000 ML day^{-1} for several days, Newton 1994)].

All mud samples were initially held at 1–2°C for several days to kill any organisms present, while at the same time preserving dormant developmental stages (after Marcus 1990). Before use in experiments, the mud samples were sieved through mesh of 1 mm and 70 μm aperture. Microscopic detection of dormant eggs or life stages was not possible because of the high detrital content of the sediments. Techniques that might have overcome this problem, such as gradient separation (e.g. sonication) and/or fluorescence microscopy, were not available.

Incubation experiments

The experiments were qualitative only; no attempt was made to determine the densities of dormant stages in the sediment samples or the proportions hatching. Conditions of incubation (i.e. experimental media) were varied to assess the dormancy status (i.e. diapause or quiescence) and to identify possible environmental factors that may trigger hatching. The incubation experiments were conducted in 750 mL glass jars. The experimental media contained filtered, boiled seawater and distilled water mixed to obtain water of the required salinity. After the jars had been filled with experimental media, a fine layer of sieved mud was introduced. Small amounts of yeast culture were initially supplied to jars as a food source for any organisms that hatched. Jars were gently aerated every few days. To minimize disturbance, jars were inspected visually for the presence of nauplii or other life history stages, and water samples of 10 to 20 mL were pipetted for more detailed microscopic examination.

Salinity is presented according to the Practical Salinity Scale of 1978 (PSS 78). A variety of salinities, from 0 to 25, were used in the experimental jars; this range occurred in the estuary over the high- and low-flow periods of river discharge (Newton 1996). The experimental jars were held in controlled temperatures of 10 ± 3°C or 20 ± 2°C, which were close to estuarine water temperatures before and after a flood, respectively; also some jars were held at 5°C in a refrigerator. [The lowest recorded temperature in the estuary was 7.3°C (Newton 1996)]. Most jars were held under a photoperiod of 14L:10D but the jars at 5°C were held under constant darkness. Conditions for each of the three series of incubation experiments are shown in Tables 1–3. Each series of experiments ran for one to two months. Experiments 1C, 2E and 3D were ‘conversion’ experiments conducted to test for viability of dormant stages and to gain further insight into potential environmental triggers of hatching. After mud had been held at 0 salinity for 25 to 40 days (experiments 1C and 2E), the media was changed to a salinity of 20–25. For experiment 3D, mud was held at 5°C for 21 days, after which the temperature was increased to 20°C.

Table 1. Series 1 incubation experiments (mud collected 5 July 1985)
Fridge time: time mud sample held in refrigerator at 1–2°C

Experiment	No. jars	Fridge time (days)	Temperature (°C)	Salinity
1A	4	4	20 ± 2	15
1A	4	4	20 ± 2	25
1B	1	7	10 ± 3	15
1B	1	7	10 ± 3	25
1C	1	14	10 ± 3	0–20^{A}
1C	1	14	20 ± 2	0–20^{B}

^{A}Salinity converted from 0 to 20 after 32 days. ^{B}Salinity converted from 0 to 20 after 25 days.

Table 2. Series 2 incubation experiments (mud collected 5 August 1985)
Fridge time: time mud sample held in refrigerator at 1–2°C

Experiment	No. jars	Fridge time (days)	Temperature (°C)	Salinity
2A	6	4	20 ± 2	15
2B	6	6	20 ± 2	5
2B	6	6	20 ± 2	10
2C	3	15	10 ± 3	5
2D	1	15	5	5
2E	3	15	20 ± 2	0–25^{A}

^{A}Salinity converted from 0 to 25 after 40 days.

Table 3. Series 3 incubation experiments (mud collected 16 September 1985)
Fridge time: time mud sample held in refrigerator at 1–2°C

Experiment	No. jars	Fridge time (days)	Temperature (°C)	Salinity
3A	4	13	20 ± 2	10
3A	4	13	20 ± 2	20
3B	4	13	10 ± 3	10
3B	4	13	10 ± 3	20
3C	2	13	10 ± 3	15
3C	2	13	10 ± 3	25
3D	2	13	5–20^{A}	25
3D	2	13	5–20^{A}	15
3D	1	13	5–20^{A}	5

^{A}Temperature converted from 5°C to 20°C after 21 days.

Results

Calanoida

Nauplii of *Gippslandia estuarina* and *Sulcanus conflictus* commonly appeared in the experimental jars; although rarer, copepodites of both species were also observed, including several adult (CVI) stages of each species after 30–35 days. No nauplii or copepodites of the other two calanoids common in the estuary, *Gladioferens pectinatus* and *Isias uncipes*, were observed. From a total of 62 experimental jars, *G. estuarina* hatchings occurred in 56.5% (25 jars with channel mud and 10 jars with vegetation-edge mud) and *S. conflictus* hatchings occurred in 31% (10 jars with channel mud and 9 with vegetation-edge mud) (Table 4).

Gippslandia estuarina hatched from mud collected in all three months, with similar hatching times and responses observed from mud collected in each month. No hatching was observed at a salinity of 5 or at a temperature of 5°C (Table 4, Fig. 2). Hatching time ranged from 2 to 15 days for the other salinities and temperatures tested, with a shorter duration recorded for the higher salinities (20 and 25) and temperature (20°C) (Fig. 2). Hatching was delayed by either

Table 4. Hatching success of *Gippslandia estuarina* (GE) and *Sulcanus conflictus* (SC) in mud incubation experiments

Channel, Vegetation: source of mud. 1, 2, 3: estuary section. +, nauplii hatched from mud; –, no nauplii hatched from mud; #, nauplii hatched after conversion of salinity or temperature in conversion experiment; x, no nauplii hatched in conversion experiments; blank space, media combination not attempted. Numbers given with symbols for experiments at 20°C indicate that multiples occurred for given experimental conditions (i.e. jars) and thus results

Salinity		Temperature 5°C					Temperature 10°C					Temperature 20°C					
		Channel			Vegetation^A		Channel			Vegetation^A		Channel			Vegetation^A		
		1	2	3	2	3	1	2	3	2	3	1	2	3	1	2	3
5	GE	–	–				–	–	–			–	–3x	–	–	–	–
	SC	+	–				–	+	–			+	+3#	–	–	–	–
10	GE							+	+	–	–	+	2+	+–	+	+–	2–
	SC							+	–	–	–	–	2–	+–	–	+–	2+
15	GE			–		–	–	+	+			2+	2+	+3x	+–	+–	+3#
	SC			–		–	–	–	–			+–	2–	–3x	2–	2–	+3x
20	GE							+ x	+	–	–		+#	–		+	+
	SC							– x	–	–	–		–x	+		+	+
25	GE		–		–		+	+	+			+2#	+3#	2x	+	+3x	
	SC		–		–		–	–	+			–2x	–3x	2x	–	–3#	

[A] No mud samples taken from the Vegetation site in Section 1 were held at 5° or 10°C.

lower salinity (10–15) or lower temperature (10°C), or by a combination of both, which also appeared to result in delayed development and poor survival of the nauplii. Hatching times were more variable at 10°C than at 20°C (Fig. 2). For the duration of the experiments, no copepodites of *G. estuarina* were observed at 10°C.

In contrast, hatching times of *S. conflictus* nauplii varied between mud samples from the three months of collection. None hatched from mud collected in July. Hatching duration was ~12–14 days from the mud collected in August, at temperatures of 5°, 10° and 20°C, and at salinities of 5, 10 and 15. Hatching time of nauplii was much shorter, generally ~4–5 days, from the mud collected in September at a temperature of 20°C and at salinities of 10 and 20; however, hatching times were longer at 10°C (18–24 days) and at a salinity of 25 (24 days). Hatching of *S. conflictus* nauplii was most successful at the higher temperature and at lower salinities (Fig. 2). The combination of high salinity (25) and low temperature (5°C) delayed hatching, and the combination of high salinity (25) and high temperature (20°C) was generally unsuccessful in stimulating hatching for this species (Table 4, Fig. 2).

The three 'conversion' experiments confirmed the dormancy status of eggs of *G. estuarina* and *S. conflictus*, and indicated that dormant stages occurred in ostracods, rotifers and an ectinosomatid harpacticoid copepod. The latter is an undescribed species which probably represents a new genus for the family Ectinosomatidae (R. Hamond, personal communication); it was the most abundant harpacticoid of the Hopkins River estuary zooplankton community, indicating an increased pelagic lifestyle for this normally meiobenthic group. After extended periods in the original experimental media, *G. estuarina* nauplii hatched within 1 or 2 days of the change from fresh water to high salinity, and from low temperature to high temperature. *Sulcanus conflictus* nauplii hatched within 2 days of the change from low to high temperature; however, hatching was not observed when the change was from fresh to saline water. The influence of temperature on *S. conflictus* eggs was clear in the temperature conversion experiments conducted using September mud. Hatching was not observed for muds held in a salinity of 5, 15 or 25 for 21 days at 5°C, but when temperature was then changed to 20°C *S. conflictus* hatched after 2 days in a salinity of 5 or 25. In contrast, *S. conflictus* nauplii hatched from August mud after 12 days at salinity 5 and a temperature of 20°C. This suggests either that low temperature maintains the diapause condition of eggs, i.e. temperature may affect the length of the refractory phase (Grice and Marcus 1981), or that high temperature triggers the hatching response.

Other taxa

Other organisms commonly observed in the experimental jars were harpacticoid copepods, ostracods, nematodes and the medusa *Australomedusa baylii* (Table 5). It is uncertain what life history stage these organisms had developed from, although it is possible that the medusoid form of *A. baylii* developed from resistant bodies produced by the hydroid stage (as yet undescribed, see Bayly 1971). Bayly (1971) suggested that the possession of a resistant stage would be advantageous for *A. baylii*, which also occurs in saline lakes that have salinity fluctuations likely to be greater than its range of tolerance. However, harpacticoid nauplii were often abundant and this suggests that dormant eggs were present for this group. Adults of the true estuarine harpacticoids *Onychocamptus chathamensis*, ectinosomatid sp. and *Schizopera* sp. were recorded, some of which produced egg-sacs. Occasionally, some organisms were observed soon after the experimental

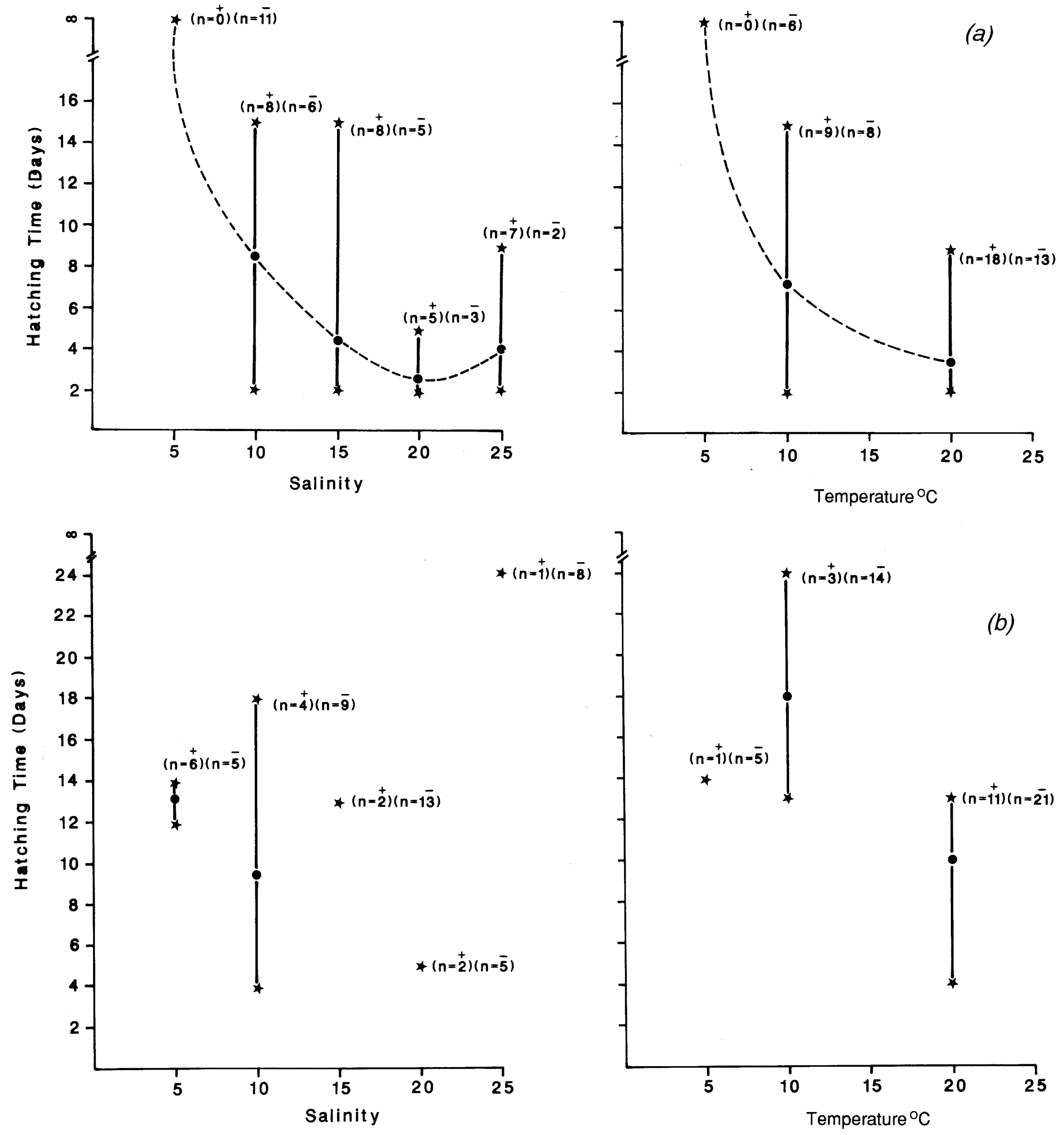

Fig. 2. Hatching time (mean and range, days) for *(a) Gippslandia estuarina* nauplii and *(b) Sulcanus conflictus* nauplii, in mud incubated at a range of salinities and temperatures. No range shown where mean and range are equal. Numbers of successful (+) and failed (–) hatching responses for each salinity or temperature tested in experimental jars are indicated in parentheses. [Note: dashed line in (a) is indicative only of apparent trends.]

jars had been set up: nematodes after 3 days, ostracods after 2 days, and a harpacticoid, *Onychocamptus chathamensis*, bearing an egg-sac, after 6 days (experiment 2A). This may imply that some of these smaller organisms (e.g. nematodes) were present in the estuarine mud, but were robust enough to survive the period of refrigeration and anoxic conditions within the mud. It may also imply that resting juvenile or adult stages appear rapidly in the water column given appropriate conditions. However, most organisms appeared in the experimental jars after a greater length of time, on average 18–30 days, and these were likely to have originated from resting stages such as eggs or cysts. Ostracods, the ectinosomatid harpacticoid and rotifers developed in the salinity ‘conversion’ experiments, confirming the presence of dormant life history stages for these taxa. Temperature appeared to be the more important environmental stimulus for non-calanoid taxa, because most organisms were found at high temperatures but across a wide range of salinities (excluding fresh water).

Table 5. Non-calanoid taxa (adult to near–adult forms) observed in jars of incubated mud

*, harpacticoid with an egg-sac; #, included ectinosomatid sp.; +, included *Onychocamptus cathamensis*; ++, included *Schizopera* sp.; ##, ostracod and ectinosomatid sp. developed at salinity 25 following 25 days in freshwater medium

Taxon	Vegetation mud	Channel mud	Salinity	Temp. (°C)	Days to first observation Range	Days to first observation Mean	No. jars
A. baylii	2	2	15	20	1–42	35	3
			10	20	33	33	1
Nematodes	4	4	10	20	11	11	1
			15	20	3–42	23	5
			25	20	29	29	1
			25	10	59	59	1
Ostracods	6	7	5	20	29–33	31	2
			10	20	11	11	1
			15	20	11–42	28	6
			20	20	20	20	1##
			25	20	10–16	13	2
			25	10	2	2	1
Harpacticoids	8	3	5	20	22–33*	28	2#,+
			10	20	22–33*	28	2
			15	20	6*–42	24	3+
			20	20	27	27	1##
			25	20	29–31*	30	2++
			10	10	18	18	1

Discussion

Dormant eggs of estuarine copepods

Nauplii were the most common form to develop in the experimental jars, which suggested that eggs were the main dormant stage present, as also noted by Marcus (1990) for neritic copepods. The production of dormant eggs by *G. estuarina* and *S. conflictus* is reported for the first time by the present study, which is apparently also the first record of dormant eggs for an Australian estuarine-endemic copepod although nauplii of an unknown copepod have been observed to hatch from what were previously thought to be microfossil dinoflagellate cysts taken from estuarine sediments of Tuggerah Lake, NSW (McMinn *et al*. 1992). Dormancy was confirmed in the present study by results obtained from the 'conversion' experiments. The appearance first of nauplii and younger copepodid stages during salt-wedge emplacement following flooding of the Hopkins River estuary further supports the existence of dormant eggs for these species (Newton 1994). *Gippslandia estuarina* (family Centropagidae) and *S. conflictus* (family Sulcanidae) are both members of the super-family Diaptomoidea (Andronov 1991), as are all marine calanoids with dormant eggs (Lindley 1992; Marcus 1996). The two other dominant calanoid species in the Hopkins River estuary, *G. pectinatus* and *I. uncipes*, both also centropagids, were not observed to hatch in any of the experiments. However, the bulk of the *Isias* population in the estuary occurred in Section 4 (see Fig. 1), which was not sampled for mud.

Harpacticoid nauplii hatched in the mud incubation experiments, and adults of at least three species of true estuarine forms were recorded. These results represent the first report of dormancy among estuarine-endemic Harpacticoida. Although the observation that *O. chathamensis* developed to egg-bearing female stage in six days at 20°C may suggest that dormancy occurred at an advanced copepodid stage rather than the egg (see Cole 1953), it is not unprecedented. For example, the calanoid *Acartia tonsa* has been reared from egg to egg-bearing female in eight days at 21°C (Zillioux and Gonzalez 1972) and generation times for harpacticoid copepods at 20°C have been reported to be as short as four days, although they are generally in the order of 10–30 days (see Hicks and Coull 1983).

Hatching response

The results of the conversion experiments suggest that both temperature and salinity are important triggers for the hatching response of dormant *G. estuarina* eggs. The failure of *G. estuarina* to hatch at a temperature of 5°C or a salinity of 5 suggests that hatching would not occur in the estuary under flood conditions. Shorter hatching durations were recorded under high salinities (15–25) and/or high temperature (20°C), conditions that would more closely resemble post-flood conditions in the estuary when the new salt-wedge is in place (Newton 1996).

Sulcanus conflictus hatched at salinities of 5–25 and at all temperatures tested, although hatching was more successful and rapid at the highest temperature. In addition, *S. conflictus* hatched in response to temperature conversion but not to salinity conversion. Embryonic and post embryonic development of *S. conflictus* has been shown to be strongly temperature dependent (Ough and Bayly 1989). The above factors suggest that temperature is likely to be a more important trigger of hatching than salinity for this species.

Previous experiments with dormant eggs of marine calanoids have found that hatching is controlled by factors such as temperature, oxygen and photoperiod (Kasahara *et al.* 1975; Landry 1975; Uye and Fleminger 1976; Johnson 1980; Ban 1992; Lutz *et al.* 1992). Salinity is likely to be an important factor in the estuarine environment, as exemplified by *G. estuarina* in the present study. Dormant eggs of marine calanoid species do not hatch in anoxic waters or while buried in sediments (Kasahara *et al.* 1975; Uye and Fleminger 1976; Uye *et al.* 1979). Although it could be expected that estuarine plankton would be more tolerant of low dissolved oxygen conditions than their marine counterparts, it is possible that the reoxygenation of estuarine waters (which stagnate because of stratification over the low flow period) during the emplacement of a new salt-wedge after a flood (Newton 1996), may influence hatching of the dormant stages of some planktonic organisms in the Hopkins River estuary.

Type of dormancy

Diapause eggs have been confirmed for only 14 species of calanoid copepods (Marcus 1996), including *Acartia tonsa* and *A. californiensis*, which are euryhaline-marine forms common in Northern Hemisphere estuaries (Johnson 1980; Grice and Marcus 1981; Uye 1985). However, both diapause and quiescent eggs may respond in similar ways to hatching tests if the diapause eggs have completed the refractory phase, thus making distinction difficult. With regard to the unpredictable flooding of an estuary, both egg types could be an important mechanism of population re-establishment after flooding, particularly if saline waters return within one to two months, as is generally the case for the Hopkins River estuary (Newton 1994, 1996). Periods of freshwater inundation longer than a few months could render quiescent eggs unviable, because the period of viability is considerably shorter for quiescent eggs than for diapause eggs (e.g. months rather than years, see Marcus 1996).

Evidence from the present study suggests that the dormant eggs of *G. estuarina* are the quiescent type and those of *S. conflictus* are the diapause type, but further testing is required. While the *G. estuarina* population in the estuary declined over winter, the *S. conflictus* population virtually disappeared, particularly in upstream sections (Newton 1994). The hatching of viable eggs from sediments long after the disappearance of adults from the plankton strongly indicates a diapause condition (Grice and Marcus 1981). The *S. conflictus* nauplii that developed from mud taken in September would have hatched from eggs laid at least 3 months before, because by July the *S. conflictus* population had already virtually disappeared from the plankton (Newton 1994). The failure of *S. conflictus* to hatch from July mud samples may indicate that the refractory phase of the diapause eggs had not yet been completed. The longer hatching time of *S. conflictus* nauplii from August mud than from September mud, too, may reflect the stage of progression of the refractory phase. In contrast, a reduced population of *G. estuarina* remained in the estuary over the sampling period and the duration to hatching of *G. estuarina* nauplii was similar from muds taken in all three months.

Ecological adaptations

A variety of holoplanktonic and facultative zooplankters were observed in the estuarine mud incubation experiments. Dormant stages buried in sediments are part of the pool of potential recruits to the plankton and may contribute to the dynamics of both the pelagic and benthic communities (Marcus 1984; Lindley 1990). Dormant eggs of calanoids can remain viable for years (Lindley 1990; Ban and Minoda 1992; Marcus *et al.* 1994). It is likely that zooplankton populations could be regenerated after several unfavourable years if diapause stages are present, and this would be an important mechanism of population persistence in dynamic, flood-prone estuaries such as the Hopkins River. Experimentation is required to determine if dormant stages are continually produced by estuarine species, or if their production is triggered by specific temporal environmental cues. Some neritic copepod species switch between direct-developing subitaneous eggs and dormant eggs under different environmental conditions (Grice and Marcus 1981; Madhupratap *et al.* 1996; Marcus 1996), and recent work suggests that intermediate states of egg dormancy can also occur (e.g. delayed-hatching, Chen and Marcus 1997). In the Australian fresh- water cladoceran *Daphnia carinata*, a proportion of the population is ephippial (with resting eggs) throughout the population cycle and this is apparently unrelated to environmental conditions (Mitchell and Williams 1982).

Lindley (1992) suggested that dormant eggs pre-adapted species to colonize inland waters. However, dormant eggs have mainly been shown for either stenohaline or euryhaline neritic marine copepod species, several of which also inhabit estuaries (Marcus 1996; George and Lindley 1997; Lindley *et al.* 1998). Little has been written on the occurrence of such stages in the life cycles of estuarine-endemic organisms, even though estuaries are also dynamic unpredictable environments and dormancy is known to be common in both the adjacent neritic and freshwater environments. If diapause eggs are confirmed for *S. conflictus*, it would be useful to ascertain whether this egg type were produced only in local environments which undergo flooding. For example, after severe flooding of the Fitzroy River in North Queensland, the return of adult *S. conflictus* was preceded by earlier developmental stages, which led Kennedy (1978) to speculate that this species may have resistant eggs. This feature has now been confirmed by the present study. Such investigations may also provide further information on the triggers that induce the production of dormant eggs in this species, because floods tend to occur more during summer in the tropics. In temperate estuaries such as the Hopkins River, floods tend to occur in late winter or early spring, thus making it more difficult to differentiate between the production of dormant eggs as an 'over-wintering' or 'flood' adaptation. There is also some suggestion that *S. conflictus* may produce resting eggs under conditions of deoxygenation, as was caused by a bloom of toxic algae in a Queensland lagoon (Thomson and Dunstan 1968).

Distribution and evolutionary consequences

The occurrence of *G. estuarina* in the Hopkins River estuary represents the most westerly extension of its known distribution range. The monotypic *G. estuarina* is a primitive centropagid first described from the Gippsland Lakes in eastern Victoria (Bayly and Arnott 1969). It has been recorded in fewer than ten estuaries since, all in south-eastern Australia, with records from Tasmania, Victoria and the extreme south of New South Wales (Neale and Bayly 1974; Bayly 1975; Jarvis 1979; Arnott 1983; Arnott and McKinnon 1983; Tafe and Griffiths 1983; Newton 1994). In contrast, the monotypic *S. conflictus* is generally common in Australian estuaries, and occurs from 30°S in Western Australia around southern Australia to 23°S in northern Queensland (Bayly 1975; Ough and Bayly 1989).

The differential distribution patterns of the two species may be influenced by their ability to produce different types of dormant eggs. It is generally accepted that diapause has played a key role in the adaptive radiation and ecological success of the freshwater Copepoda (Fryer 1996). Diapause eggs offer longer-term viability, and therefore the ability to disperse over greater distances. The ability of diapause eggs to survive many years in sediments means that where the accumulation of eggs occurs, a pool of genetic information would be sustained (i.e. an 'egg bank', Marcus *et al.* 1994) that may slow down the rate of evolutionary change (Hairston and De Stasio 1988). Maly and Bayly (1991) produced indirect evidence that rates of evolution in centropagid calanoids may indeed be slow, and that some species have remained morphologically unaltered over considerable spans of geological time – a feature that may be contributed to in part by the possession of dormancy. If confirmed, the ability of *S. conflictus* to produce diapause eggs may help explain its widespread distribution with little morphological (and presumably genetic) variation (Nicholls 1945; Bayly 1963), in both eastern and western Australian estuaries. It may also be a contributing factor to the ability of the species to occupy a wide range of estuarine habitats.

Whereas diapause confers the capacity for long-term survival, quiescence enables individuals to survive shorter and less predictable periods of adverse conditions (Marcus 1996). Therefore, it could be expected that the production of quiescent eggs may restrict the dispersal ability of a species and possibly the range of habitat types suitable for colonization. If it is shown that *G. estuarina* can only produce quiescent subitaneous eggs, this may, in part, explain the more limited distribution of this species and the fact that it has only sporadic and limited abundance over most of its range. It is only known to be dominant in the Hopkins River estuary and the Gippsland Lakes, both of which are stratified and flood-prone estuaries (Arnott 1983; Newton 1994). However, in an extensive survey of lakes and ponds (freshwater and saline) in Western Australia, Maly *et al.* (1997) found that of the 23 centropagid copepods recorded (some of which are known to have dormant eggs), 16 species had fewer than 10 known populations. They concluded that distribution patterns are moulded primarily by historical events rather than dispersal ability. It is likely that *G. estuarina* is currently mainly represented by 'refuge populations' in favourable habitats such as stratified estuaries or regions of estuaries with more limited tidal exchange (Newton 1994).

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