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ION REPLACEMENT AS A BUOYANCY MECHANISM IN A PELAGIC DEEP-SEA CRUSTACEAN

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Summary

Unlike most pelagic crustaceans, the deep-sea shrimp *Notostomus gibbosus* (A. Milne-Edwards) (Oplophoridae) is positively buoyant, possessing a dorsally enlarged carapace which contains a low-density fluid. This fluid comprises 43 % of the animal's wet mass, has a low pH, and gives a lift of 17·7 mg ml^{-1}; when this fluid is drained, the animal sinks. Low density is achieved by the replacement of less buoyant ions with ions which reduce density *via* two mechanisms: a change in total solute mass by the use of ions of lesser mass, and an ion-specific disruption of the structure of water molecules (resulting in an increase in fluid volume) caused by ions having large, positive partial molal volumes. The presence of large amounts of trimethylamine (Me_3NH^+), a relatively large, heavy ion which, together with NH_4^+, replaces nearly 90 % of the Na^+ in the carapace fluid, results in little change in the total solute mass of the carapace fluid of *N. gibbosus* (33·2 ‰) relative to sea water (approximately 34·1 ‰). Reduced fluid density is primarily a result of the large, positive partial molal volumes of Me_3NH^+ and NH_4^+, rather than a function of reduced solute masses.

Introduction

Pelagic marine invertebrates use a variety of methods to reduce their densities and achieve neutral or positive buoyancies, thereby providing these organisms with energetically conservative means of maintaining their positions in the water column. The more common adaptations found in pelagic invertebrates include reducing overall density *via* an increase in water content, a reduction in exoskeleton calcification, an increase in lipid content and the use of gas-filled spaces. Another approach, found in pelagic squids (Clarke *et al.* 1979), dinoflagellates (Kahn & Swift, 1978), diatoms (Gross & Zeuthen, 1948) and tunicate eggs (Lambert & Lambert, 1978), is the reduction of fluid density by ion replacement. In one group of pelagic cephalopods, the cranchiid squids, NH_4^+ replaces virtually

Key words: buoyancy, pelagic crustacean, ion replacement, ammonia, trimethylamine, *Notostomus gibbosus*, partial molal volume, fluid density.

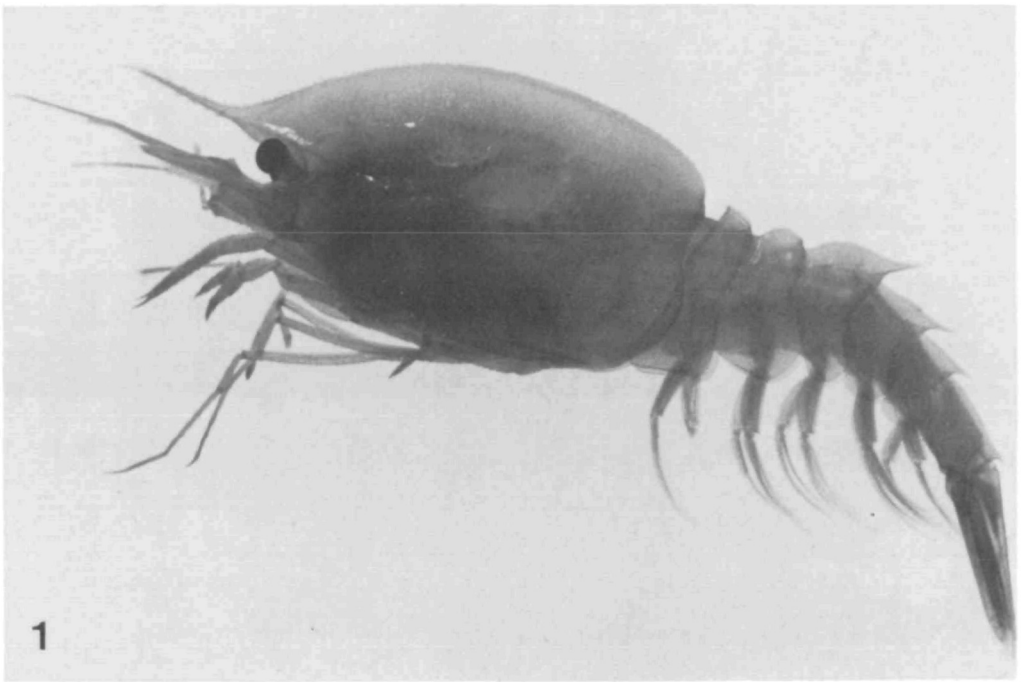

Fig. 1. *Notostomus gibbosus* (45 mm carapace length) showing the dorsally expanded carapace.

all the Na^+ present in body fluids, and results in a lift of more than 17 mg ml^{-1} (Denton *et al.* 1969; Clarke *et al.* 1979). Although ion replacement has not been documented for any crustacean, it has been suggested for the pelagic deep-sea shrimp *Notostomus* sp. (Oplophoridae) and the deep-sea ostracod *Gigantocypris* sp. (Herring, 1973, citing Denton, 1971).

Although pelagic crustaceans are generally streamlined and negatively buoyant, the positively buoyant shrimp *Notostomus gibbosus* is a dramatic exception (Fig. 1). *N. gibbosus* possesses a bulky, dorsally expanded carapace which is fluid-filled. Although one species of *Notostomus* has a high water content (91·3 %) and reduced exoskeleton calcification, the lipid content is very low (Childress & Nygaard, 1974), indicating that buoyancy is achieved by some other mechanism. Our initial measurements of the carapace fluid using ammonia electrodes (HNU model ISE-10-10-00) showed high concentrations of NH_4^+ (up to 500 mmol l^{-1}). This, together with the unusual morphology of *N. gibbosus*, led us to examine the possibility that the greatly expanded carapace acts as a buoyancy chamber, with reduced fluid density providing lift to the animal *via* ion replacement.

Although the potential contribution of ion partial molal volume to density has been mentioned by several authors (Denton *et al.* 1969; Denton, 1971; MacDonald, 1975; Kahn & Swift, 1978), it has not generally been discussed in depth.

As a result, textbook discussions of buoyancy *via* ion replacement have centred on decreasing fluid density by the use of 'lighter' ions (i.e. NH_4^+) to replace 'heavier' ions (i.e. Na^+), leaving the assumption that density changes are due primarily to the differences in ion mass (Marshall, 1979; Hainsworth, 1981; Schmidt-Nielsen, 1983). Our analysis of the very low-density carapace fluid of *Notostomus gibbosus*, however, revealed the presence of trimethylamine (Me_3NH^+), a relatively large, heavy ion which replaces more than 25 % of the Na^+ in this fluid. Because of the large mass of Me_3NH^+, there is little change overall in solute mass in the carapace fluid (33·2‰) relative to sea water (34·1‰); rather, the dramatic decrease in fluid density is primarily a result of the large, positive partial molal volumes of Me_3NH^+ and NH_4^+, and not a function of solute mass. Partial molal volume differences result in changes in the volume of a fluid *via* the specific ion effects on the water molecules. Ions having positive partial molal volumes disrupt the structure of the water molecules, thereby increasing fluid volume and reducing density. This is the first report of an animal that uses Me_3NH^+ to reduce fluid density, and the first report of the use of ion replacement for density reduction in a crustacean.

Materials and methods

Notostomus gibbosus and other deep-sea pelagic shrimps (Oplophoridae), as well as the mysid *Gnathophausia ingens* (Dohrn), were collected during July 1987 off the leeward side of the Hawaiian island of Oahu (21°15–37′ N, 158°15–38′ W) from depths of 400–800 m. The deep-sea crustaceans *Gigantocypris agassizii* (Müller) (Ostracoda) and *Cystisoma* sp. (Amphipoda) were collected in January 1987 from San Nicolas Basin off southern California (32°50′–33°10′ N, 119°00–20′ W) from depths of 400–1000 m. These animals were captured with a modified Tucker trawl using a thermally insulated cod end (Childress *et al.* 1978) which protected the animals from surface temperatures. Animals were maintained alive on board ship in individual containers in a refrigerated storage room at 5°C. The mysid *Gnathophausia longispina* (Sars) was captured with an Isaacs–Kidd midwater trawl at Southeast Hancock Seamount (29°50′ N, 179°10′E) from depths of 100–150 m, and live specimens were bled immediately after capture. Only samples from live, undamaged animals were used in these experiments.

To examine the possibility of density reduction *via* ion replacement, we first estimated the fraction of body mass present as carapace fluid. Live *Notostomus gibbosus* were weighed at sea on a precision shipboard balance system (Childress & Mickel, 1980), the carapace was then cut at the dorsal margin to drain the fluid, and the animal was reweighed.

Relative buoyancy represents the weight which 1 g wet mass of animal would have to support to maintain its position in the water column (Childress & Nygaard, 1974). Negative values for relative buoyancy indicate that an animal will float. To determine the relative buoyancies (mg wet mass in sea water/g wet mass in air) of intact *Notostomus gibbosus* at 1 atmosphere (101·3 kPa), three live animals were anaesthetized with N_2 gas, weighed in sea water at 5°C (34·1‰, 1 atmosphere),

then weighed in air. After draining the carapace fluid, the animal was reweighed in sea water and in air.

To determine the pH of carapace fluid and blood of *Notostomus gibbosus*, samples were removed from the animals with a syringe and immediately introduced into a water-jacketed chamber containing a Markson electrode (model GM 989) calibrated with precision buffers (Radiometer S1500 and S1510). The electrode was stabilized with carapace fluid or blood for several minutes before each pH measurement was made. Carapace fluid was sampled by inserting a syringe into the carapace chamber at the dorsal margin; blood samples were obtained from the ventral sinus for all shrimps. Discrete samples of carapace fluid and blood of *N. gibbosus*, *N. elegans* (A. Milne-Edwards) and *Meningodora mollis* (Smith) (a representative of the genus most closely related to *Notostomus*), and blood samples from the following shrimps were frozen for 1–3 months at −70°C for later analysis: *Acanthephyra curtirostris* (Wood-Mason), *A. acutifrons* (Bate), *A. smithi* (Kemp), *Oplophorus gracilirostris* (A. Milne-Edwards) and *Systellaspis debilis* (A. Milne-Edwards) (all oplophorids), and the mysids *Gnathophausia ingens* and *G. longispina*. Body fluids from the pelagic deep-sea ostracod *Gigantocypris agassizii* and the amphipod *Cystisoma* sp. were sampled *via* syringe and stored in a similar fashion.

Ion concentrations were analysed with single-column ion chromatography (Wescan cation and anion columns) in an HPLC system with conductimetric detection for the following ions: Na^+, K^+, NH_4^+, Me_3NH^+, Ca^{2+}, Mg^{2+}, SO_4^{2-} and Cl^-. Samples from individual animals were deproteinated with methanol (1:1), and then centrifuged; a 1:10 dilution of the resultant supernatant was analysed for ion composition. Na^+, NH_4^+, K^+ and Me_3NH^+ were separated on a cation column using a 2·3 mmol l^{-1} nitric acid eluent (Ultrex HNO_3, Baker Chemical Co.) pumped at 2·0 ml min^{-1}; Ca^{2+} and Mg^{2+} were measured on the same column with an ethylene diamine eluent (0·033 ml l^{-1}) at pH 6·1, pumped at 2 ml min^{-1}. Cl^- and SO_4^{2-} were determined using an anion column with a 0·004 mol l^{-1} potassium hydrogen phthalate eluent at pH 4·7, pumped at 2 ml min^{-1}.

The presence of high levels of Me_3NH^+ in chromatograms from carapace fluid and blood of several species was unexpected, and the identity of this peak was verified by mass spectrometry. A deproteinated, diluted sample of *Notostomus gibbosus* carapace fluid was analysed for Me_3NH^+ using a Vacuum Generator Instruments model VG70-250 HS double-focusing mass spectrometer.

Carapace fluid density was calculated for *Notostomus gibbosus* using the mean ion concentration from nine individually measured samples (Table 1), together with the mass and partial molal volume of each ion (Table 2). Additionally, a pooled sample of carapace fluid taken from three *N. gibbosus* was weighed on an electronic balance to measure directly fluid density relative to sea water. The ion concentration of this pooled sample was measured by single-column ion chromatography and the fluid density then calculated from these data for comparison with the density of the mean of nine individual samples.

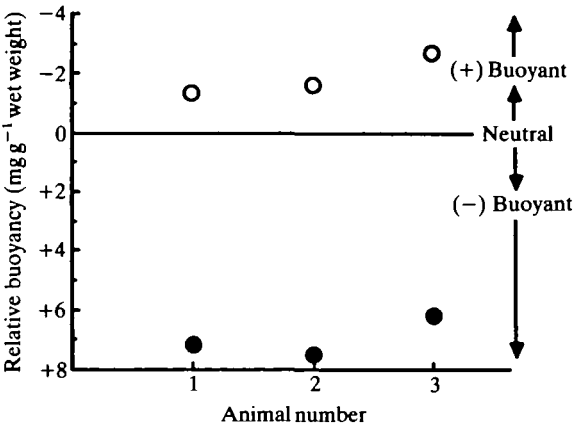

Fig. 2. Relative buoyancies of three *Notostomus gibbosus*. Negative values for intact animals (open circles) indicate that the animal floats; positive values for relative buoyancy (animal with carapace fluid drained) indicate that the animal sinks (closed circles).

Results

The fluid within the expanded carapace of *Notostomus gibbosus* represented 42·9 ± 4·6 % (S.D., $N = 9$) of the animal's wet mass, and the range was 39·6–52·9 %. There was no apparent relationship between animal size and the percentage of body mass as carapace fluid. At 5 °C, the pH of the carapace fluid was very low (6·59 ± 0·08, $N = 8$) and the blood pH (7·52 ± 0·06, $N = 8$) was also unusually low for crustaceans (Mangum, 1980). At these low pH levels, virtually all the ammonia would be present in the ionized NH_4^+ form.

The relative buoyancies of three *Notostomus gibbosus* are shown in Fig. 2. These animals were all slightly buoyant when intact (relative buoyancies = −1·36, −1·59 and −2·65 mg g^{-1}), and sank when their carapace fluid was removed (relative buoyancies = 7·15, 7·50 and 6·24 mg g^{-1}). The difference in relative buoyancy between intact and drained animals represents the amount of lift which the fluid must provide to account for the observed buoyancy. Values for drained animals, however, represent a conservative estimate because of the difficulty of removing all the trapped air bubbles once the carapace is drained.

The ion compositions of fluids are presented in Table 1. Although the carapace fluid and blood of *Notostomus gibbosus* were both isosmotic with sea water, their ion compositions differed dramatically. Reduced concentrations of Na^+ and high concentrations of NH_4^+ and Me_3NH^+ were present in both fluids but were most evident in the carapace fluid. In addition, the levels of all divalent ions were greatly reduced. Ion compositions of the carapace fluids and bloods of *N. elegans* and *Meningodora mollis* were also very different from sea water, and demonstrated ion replacements similar to those in *N. gibbosus*. The remaining oplophorid shrimps and the mysids had bloods similar in ionic composition to sea

Table 1. *Ion composition of sea water and invertebrate body fluids*

Source	Sample	*N*	Ion composition ($mmol\,l^{-1}$)							
			Na^+	NH_4^+	Me_3NH^+	K^+	Mg^{2+}	Ca^{2+}	SO_4^{2-}	Cl^-
Sea water	NA	NA	470·2	0	0	9·97	53·35	10·32	28·20	549·0
Notostomus gibbosus (three pooled)	Fluid	1	106	223	144	10·0	6·8	0	7·9	478
Notostomus gibbosus	Fluid	9	62 ± 23	296 ± 51	128 ± 20	10·8 ± 1·0	3·5 ± 1·4	0	1·2 ± 1·0	511 ± 41
N. gibbosus	Blood	9	222 ± 63	217 ± 54	113 ± 19	13·5 ± 5·0	9·9 ± 4·0	1·1 ± 1·7	5·2 ± 1·5	537 ± 32
N. elegans	Fluid	2	74	115	142	47·8	1·7	0	1·3	541·2
N. elegans	Blood	2	318	131	137	18·4	14·3	9·0	9·3	599·3
Meningodora mollis	Fluid	2	145	133	79	14·5	9·2	6·6	2·7	557·3
M. mollis	Blood	2	296	129	15	79·0	8·9	4·3	7·3	547·7
Acanthephyra curtirostris	Blood	2	451	12	Trace	11·8	11·5	8·9	9·4	467
A. acutifrons	Blood	2	458	18	Trace	10·6	26·3	5·2	17·7	549
A. smithi	Blood	2	446	Trace	Trace	13·6	17·4	6·6	17·8	481
Oplophorus gracilirostris	Blood	2	480	Trace	Trace	14·0	7·9	3·9	15·5	545
Systellaspis debilis	Blood	2	493	23	Trace	15·6	25·0	6·7	17·8	538
Gnathophausia ingens	Blood	2	519	0	0	21·8	15·0	6·6	4·5	523
G. longispina (approx. 100 pooled)	Blood	NA	482	29	64	20·8	16·4	1·3	20·0	566
Gigantocypris agassizii	Fluid	2	345	0	0	8·1	30·3	5·4	6·6	434
Cystisoma sp.	Fluid	2	491	0	0	13·5	17·6	8·2	10·4	512

NA, not applicable.
Mean values ± S.D.

Table 2. *Contributions of partial molal volume (V°) at 5°C and molecular weight (MW) to density in sea water and* Notostomus gibbosus *carapace fluid*

Ion	MW ($g\,mol^{-1}$)	V° ($cm^3\,mol^{-1}$)	Sea water ($mmol\,l^{-1}$)	Sea water ($g\,l^{-1}$)	Sea water ($ml\,l^{-1}$)	Carapace fluid ($mmol\,l^{-1}$)	Carapace fluid ($g\,l^{-1}$)	Carapace fluid ($ml\,l^{-1}$)
Na^+	23·0	−2·90	470·20	10·800	−1·370	62·1	1·428	−0·180
NH_4^+	18·0	+17·49	0·0	0·000	0·000	296·0	5·328	+5·176
K^+	39·1	+7·59	9·96	0·389	+0·076	10·8	0·422	+0·082
Me_3NH^+	60·0	+71·51	0·00	0·000	0·000	127·6	7·656	+9·284
Mg^{2+}	24·3	−21·66	53·57	1·302	−1·160	3·5	0·085	−0·076
Ca^{2+}	40·1	−19·26	10·23	0·410	−0·197	0·0	0·000	0·000
Cl^-	35·3	+16·65	548·30	19·465	+9·124	511·7	18·165	+8·520
SO_4^{2-}	96·1	+11·70	28·25	2·715	+0·331	1·2	0·115	+0·014
Totals				35·081	+6·804		33·199	+22·659

Density was calculated as $g\,ml^{-1}$ for sea water ($1035{\cdot}081\,g\,l^{-1}/1006{\cdot}804\,ml\,l^{-1} = 1{\cdot}028\,g\,ml^{-1}$) and carapace fluid ($1033{\cdot}199\,g\,l^{-1}/1022{\cdot}659\,ml\,l^{-1} = 1{\cdot}0103\,g\,ml^{-1}$), resulting in $17{\cdot}7\,mg\,ml^{-1}$ lift for carapace fluid in sea water.

water, although very low levels of NH_4^+ and Me_3NH^+ were present in some (Table 1).

Using the mean ion composition for *Notostomus gibbosus* carapace fluid from nine individual samples (Table 1), the density of this fluid ($1{\cdot}0103\,g\,ml^{-1}$) was calculated (Table 2). The high concentrations of NH_4^+ provided the greatest contribution to lift ($9{\cdot}1\,mg\,ml^{-1}$ fluid relative to sea water having a density of $1{\cdot}028\,g\,ml^{-1}$), and Me_3NH^+ gives an additional lift of $1{\cdot}8\,mg\,ml^{-1}$ carapace fluid. The reduction in divalent ions provided $5{\cdot}4\,mg\,ml^{-1}$ lift, and the total lift contribution calculated for carapace fluid was $17{\cdot}7\,mg\,ml^{-1}$. The density of a pooled sample of carapace fluid ($1{\cdot}011\,g\,ml^{-1}$) was also measured directly by weighing a known volume and represented a lift of $17{\cdot}0\,mg\,ml^{-1}$ relative to sea water. For comparison, the density calculated from the ion composition of this pooled sample was $1{\cdot}0112\,g\,ml^{-1}$ (a lift of $16{\cdot}8\,mg\,ml^{-1}$), compared with $1{\cdot}0103\,g\,ml^{-1}$ calculated from the mean carapace fluid given above. These three separate analyses of carapace fluid are in excellent agreement, varying by only $0{\cdot}9\,mg\,ml^{-1}$. The ion composition of the blood of *N. gibbosus* results in a lift of $12\,mg\,ml^{-1}$ when calculated from the mean ion composition given in Table 1.

Discussion

Marine crustacean bloods are isosmotic with sea water and are generally similar to sea water in ionic composition, although levels of divalent ions are often reduced (Mangum, 1983; Bridges *et al.* 1984; Sanders *et al.* 1988). In *Notostomus gibbosus*, body fluids are indeed isosmotic with sea water, but ion compositions differ dramatically (Table 1). For example, the Na^+ concentration in the carapace fluid of *N. gibbosus* is nearly 90 % lower than in sea water, and very high levels of

NH_4^+ and Me_3NH^+ are present (Table 1). Similarly, our methods detected no Ca^{2+} in this fluid, and very low levels of Mg^{2+} and SO_4^{2-} (Table 1). These changes in ion composition influence density in two ways: first, a change in density can result from the differences in solute masses alone. Second, and most significant here, is the change in solution volume and resultant change in fluid density due to the differences in the partial molal volumes of the replacement ions (partial molal volume, $V° = cm^3\,mol^{-1}$; Millero, 1972). When ions with a negative partial molal volume are added to a solution, the total volume of the solution is less than that of the separate ions and solvent. The addition of ions with a positive partial molal volume results in a solution having a greater volume than the sum of its components. This volume increase occurs because ions with positive partial molal volumes ('structure breakers') disrupt the structure of water in their vicinity, whereas small ions with negative partial molal volumes ('structure makers'), especially those with multiple charges, cause a constriction in the water structure around them (Horne, 1969). For example, if only solute masses are examined in relation to carapace fluid density, the fluid (33·2‰) would be nearly as dense as sea water (34·1‰) due to the relatively large mass of Me_3NH^+ (Table 2). The partial molal volumes at 5°C of Me_3NH^+ and NH_4^+, however, are large and positive, and in the carapace fluid result in a lift of $10{\cdot}9\,mg\,ml^{-1}$, with an additional $5{\cdot}4\,mg\,ml^{-1}$ lift from a reduction in the concentrations of Ca^{2+}, Mg^{2+} and SO_4^{2-} (Table 1). Even in an organism that completely replaces Na^+ with NH_4^+, and where no Me_3NH^+ is present, such as in the cranchiid cephalopods (Denton *et al.* 1969), only $2{\cdot}4\,mg\,ml^{-1}$ lift would be explained by solute mass differences alone. (It is possible that Me_3NH^+ was present in cephalopod fluids, but the methods previously employed to measure NH_4^+ did not discriminate between the amines present, Denton *et al.* 1969; Clarke *et al.* 1979.) In *N. gibbosus* carapace fluid, the total lift from solute mass and partial molal volume (Table 1) is $17{\cdot}7\,mg\,ml^{-1}$ (Table 2). Similarly, *N. gibbosus* blood has a reduced level of Na^+, high levels of NH_4^+ and Me_3NH^+, and a reduction in the levels of divalent ions relative to sea water. The calculated lift contribution of the blood in $12\,mg\,ml^{-1}$.

The mean relative buoyancy (Fig. 2) of *Notostomus gibbosus* ($-1{\cdot}87\,mg\,g^{-1}$) is lower than that observed in 14 other midwater crustaceans (relative buoyancies of $-0{\cdot}7$ to $+40{\cdot}0\,mg\,g^{-1}$) (Childress & Nygaard, 1974), and a negative relative buoyancy indicates that the animal is positively buoyant. The total lift required to account for the observed relative buoyancy in *N. gibbosus* can be calculated using the mean relative buoyancies for intact ($-1{\cdot}87\,mg\,g^{-1}$) and drained ($6{\cdot}96\,mg\,g^{-1}$) animals, and assuming that sea water has a density of $1{\cdot}028\,g\,ml^{-1}$. A 10-g animal having 42·9 % of its wet mass as carapace fluid would weigh 5·71 g drained, with 4·29 g carapace fluid. The volume of 4·29 g of carapace fluid ($1{\cdot}0103\,g\,ml^{-1}$, Table 2) is 4·25 ml. The overall change in mean relative buoyancy ($8{\cdot}83\,mg\,g^{-1}$) multiplied by the animal's mass without carapace fluid (5·71 g) is the total lift (50·4 mg) that must be accounted for in a 10-g animal. Therefore, the carapace fluid (4·25 ml) must be less dense than sea water ($1{\cdot}028\,g\,ml^{-1}$) by $12\,mg\,ml^{-1}$ to provide 50·4 mg of lift. This requires that the density of the fluid be $1{\cdot}016\,g\,ml^{-1}$

The observed carapace fluid density ($1{\cdot}0103\,g\,ml^{-1}$, Table 2), however, gives a lift of $17{\cdot}7\,mg\,ml^{-1}$, and provides the 10-g animal with 75·2 mg of lift. Using these numbers, the lift contribution of the carapace fluid alone is more than sufficient to explain the observed differences in the relative buoyancies of intact and drained *N. gibbosus*. This discrepancy, however, may be due to a conservative mass estimate for drained animals. When drained animals are resubmerged to measure their mass in sea water, air bubbles can remain trapped in the carapace making precise mass estimates difficult. High levels of NH_4^+ and Me_3NH^+ were also found in the bloods and carapace fluids of the congener *Notostomus elegans* as well as in *Meningodora mollis*. No evidence of ion replacement was found in *Gigantocypris agassizii* or *Cystisoma* sp. (Table 2), and both these animals have a very high water content (Childress & Nygaard, 1974). *Cystisoma* sp. fluid, however, is apparently slighly hyposmotic relative to sea water (Table 1). The small amounts of NH_4^+ and Me_3NH^+ detected in the bloods of the other oplophorid shrimps may indicate a predisposition of this family for ion replacement as a buoyancy mechanism. The other oplophorid species examined in this study, however, are all negatively buoyant and do not have enlarged carapaces.

The high NH_4^+ and Me_3NH^+ concentrations in *Notostomus gibbosus* might present toxicity problems if the fluids were not confined to specific body compartments which prevented direct contact with other tissues. Evidence for isolation of the carapace fluid in a discrete chamber comes from direct observations in preserved specimens of a membrane which contains the fluid in the bulky, dorsally expanded region of the carapace. The pH values were low for both fluids (blood pH = 7·52, carapace fluid pH = 6·59) and were sufficiently acidic to maintain virtually all the ammonium ions in the NH_4^+ ionized form. Additionally, although both the carapace fluid and blood have low pH values, these values are significantly different from one another (99 % confidence interval, paired Student's *t*-test); the ionic compositions of the carapace fluid and blood also differ significantly for the monovalent cations, Mg^{2+} and SO_4^{2-} (95 % confidence interval, paired Students's *t*-test), and these differences support the separate nature of these two fluids.

Although other extracellular body fluids were not measured, both the carapace fluid and the blood of *Notostomus gibbosus* have unusual ion compositions. High levels of ammonia were not found in the blood of cranchiid squids and other cephalopods (Denton *et al.* 1969; Denton, 1971; Clarke *et al.* 1979), and ammonia concentrations as high as those found in coelomic fluids and other body compartments would be sufficient to impair nervous function in these animals if these fluids were not isolated in separate chambers. The ammonia levels found in the blood of *N. gibbosus* (Table 1) would interfere with the ability of nerve fibres to conduct nerve impulses if found in a squid (Denton, 1971); we do not know how this shrimp handles the potential toxicity of the high ammonia concentrations found in the blood.

Animals which are negatively buoyant must either swim or sink; a positively buoyant animal such as *Notostomus gibbosus* must swim to stay at depth. This

raises the question of the adaptive significance of positive buoyancy in a midwater crustacean. It may be a predator avoidance strategy in that most deep-sea crustaceans respond to a predator by rapidly swimming to escape, or by ceasing swimming and sinking. In maintenance conditions, when startled, *N. gibbosus* generally stops swimming immediately and slowly floats upwards. This novel behaviour may well enable the shrimp to escape predators which would 'expect' disturbed prey to sink or swim away.

The use of the ammonium ion to provide buoyancy for *Notostomus gibbosus* is the first report of a crustacean that uses ion replacement to reduce fluid density. Whereas high concentrations of NH_4^+ (a relatively small ion) have been found to contribute to positive buoyancy in other pelagic organisms (Kahn & Swift, 1978; Lambert & Lambert, 1978; Clarke *et al.* 1979), the use of a large ion with a large positive partial molal volume (Me_3NH^+) to replace Na^+ is unique to this group.

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