Polar Biol (2006) 29: 519–525
DOI 10.1007/s00300-005-0083-4

ORIGINAL PAPER

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# Metabolic energy utilization during development of Antarctic naked dragonfish (*Gymnodraco acuticeps*)

Received: 22 July 2005 / Revised: 20 October 2005 / Accepted: 29 October 2005 / Published online: 29 November 2005


**Abstract** We have capitalised on the availability of eggs and adults of the naked dragonfish *Gymnodraco acuticeps* (Sub-order Notothenioidei, F. Bathydraconidae) near McMurdo Station, Antarctica to examine metabolic energy utilization at different stages of its life cycle. Average egg respiration rates were found to increase from $2.17 \pm 1.02$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ at about 17 h post-fertilization (hpf) to $5.72 \pm 0.56$ nmol $h^{-1}$ $ind^{-1}$ at about 24 hpf, during which time the eggs underwent first cleavage. The respiration rates of embryos from 2–20 days post-fertilization (dpf) averaged $4.11 \pm 1.47$ nmol $O_2$ $h^{-1}$ $ind^{-1}$. About 10 months post-fertilization, oxygen consumption rates of $27.14 \pm 3.92$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ were recorded immediately prior to hatching, with a peak of $112.41 \pm 31.38$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ at the time of hatch. Larvae aged 46–63 days post-hatch had an average respiration rate of $64.4 \pm 15.11$ nmol $O_2$ $h^{-1}$ $ind^{-1}$. Mass-specific respiration rates of hatched larvae (approximately 1–2 months old) were calculated using dry weights (DW) and averaged $16.1 \pm 3.4$ nmol $O_2$ $h^{-1}$ $mg^{-1}$ DW. Adult dragonfish respiration rates (corrected for a 100 g fish and using a 0.8 scaling exponent) averaged $0.91 \pm 0.36$ mmol $O_2$ $kg^{-1}$ $h^{-1}$ after a 48 h acclimatization period, which is not indicative of significant metabolic cold adaptation. The energy contents of dragonfish eggs and larvae were also measured by microbomb calorimetry and used, along with the respiration data, in an initial approach to estimate an energy budget. In order to balance the budget, the bulk of the available post-gastrulation respiratory energy (during 213 days of embryonic incubation) must be consumed at a relatively low average rate (7.1 nmol $O_2$ $h^{-1}$ $ind^{-1}$), which supports the possibility that advanced dragonfish embryos overwinter in a relatively quiescent metabolic state while awaiting a suitable stimulus (such as the return of the sun) to initiate hatching.

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## Introduction

Members of the perciform suborder Notothenioidei dominate the highest latitudes of the Southern Ocean, accounting for about 77% of teleost diversity in these waters (Eastman 2005). The life history strategies of Antarctic notothenioids, however, are generally poorly understood, particularly with respect to their modes of development. Like other marine organisms displaying yolk-dependent development, the survival of notothenioid embryonic and yolk-sac larval stages is critically dependent on a number of factors, including the maternal allocation of energy to the egg, the rate of metabolism during yolk-dependent stages, and the length of the developmental period through to independent feeding (Heming and Buddington 1988; Mommsen and Walsh 1988). Furthermore, suitable food for plankton-dependent larvae in the high-latitude waters of the Southern Ocean may be seasonally restricted (Knox 1994), rendering the timing of hatching critical, and survival dependent on the amount of yolk remaining at the time of larval emergence from the encapsulating chorion.

Eggs of the naked dragonfish *Gymnodraco acuticeps* Boulenger, 1902 (F. Bathydraconidae) are typically

laid in patches on flat rocks at about 15–35 m depth from mid-October to early November in the McMurdo Sound region adjacent to McMurdo Station (77°51.081′S, 166°39.821′E). Development is protracted across a 10–11 month period, with hatching occurring mostly in early September of the following year when sunlight returns to this high-latitude region (Evans et al. 2005). Although both eggs and larvae are negatively buoyant, newly hatched larvae swim immediately for the surface, which at this time of year is covered with a thick layer of sea ice. Diver observations suggest that the larvae remain in the upper reaches of the water column, possibly seeking the protection offered by crevices within the sea ice, while feeding on a variety of planktonic species. Yolk-sac absorption in aquarium-reared fish is typically completed by 2–3 weeks after hatching, and survival beyond this time requires an exogenous energy source (Evans et al. 2005).

Little is known about the metabolic activity of developing Antarctic notothenioid fish. Although a number of studies have examined oxygen consumption rates in adult Antarctic notothenioids (reviewed in Macdonald et al. 1987), none have been made on the eggs and larvae of the naked dragonfish. We have capitalised on the availability of eggs and adults of this species in the McMurdo Sound area to examine metabolic energy utilization at different stages of its life cycle.

## Materials and methods

### Specimens

Dragonfish eggs and adults were collected by divers between August and December in 2003 and 2004 and returned to the aquarium at McMurdo Station where they were kept in flowing local seawater (c. −1.6°C). The average annual water temperature near McMurdo Station recorded from January 2000 to September 2004 was approximately −1.8°C (P. Cziko et al., unpublished observations). Eggs were collected as both fully developed embryos (early September), which hatched soon after collection, and recently fertilized eggs (in mid-October). Larvae were provided with an abundance of local plankton and observations indicated that they began feeding on this within 48 h of hatching, even though the yolk sac was still present at this time. Adults collected at the same location were kept separate from the larvae and were deprived of food for at least 2 weeks prior to experiments to minimise post-prandial effects. As larvae can utilise energy in the yolk sac over at least 15 days (Evans et al. 2005), no attempt was made to control their feeding behaviour. Dry weights (DW) were determined for eggs and larvae using a Sartorius (Goettingen, Germany) Supermicro balance (accuracy ±0.1 μg) after incubation for 2–3 days at 55°C to a constant mass.

### Oxygen consumption

Rates of oxygen consumption of eggs and larvae were measured individually and in groups of up to ten in gas-tight glass vials (0.4–6.6 ml) in micro-filtered (0.2 μm pore size) local seawater (salinity 35‰). Experiments were undertaken under typical laboratory lighting conditions. Late-stage embryos were capable of moving within their chorions and hatched larvae could move within the glass vials used for the respiration rate measurements. No attempt was made to control larval activity. Oxygen levels in egg, embryo and larval experiments were determined using either a Strathkelvin Instruments (Glasgow, Scotland) polarographic oxygen sensor (Model 782 equipped with a 1302 oxygen electrode mounted in a MC-100 cell) as described in Marsh and Manahan (1999) or an Ocean Optics (Boca Raton, FL, USA) oxygen fluorescence-quenching probe (optode) as described in Marsh et al. (2001). Both sensors were calibrated with 0% oxygen and 100% air-saturated standards according to the manufacturers' instructions. In the polarographic method, end-point measurements only were made by injecting a sample from the vial into the measurement cell using a gas-tight syringe. Measurements of oxygen concentrations using the Ocean Optics optode connected to a computer running the manufacturer's software (OOISensor) were performed in real-time. In preliminary tests, both the polarographic and optode methods provided equivalent results (data not shown), and the experimental results were subsequently pooled in the final analyses. The incubation temperature was maintained within the range of −1.5–0°C using an ice bath, and oxygen concentration measurements were taken within a 0.5–12 h period, depending on metabolic activity and the stage being measured. Thirty-minute incubation periods were used for the embryonic measurements. Control vials (seawater only) were loaded with each run to account for any influence on oxygen levels other than that due to the specimens being tested.

Oxygen consumption rates in unsexed adult dragonfish ($N = 6$) were measured using a fully submersible Aanderaa Instruments (Bergen, Norway) Oxygen Optode (model 3835-19). Individual fish were housed in a shaded 6.22 l acrylic plastic closed-box respirometer, which was submerged in a 20 l tank of flowing seawater at ambient local temperature (c. −1.6°C) to act as a thermal buffer. The volume of the respirometer, which included a false bottom and magnetic stir bars to ensure mixing, was corrected for the volume of each fish and all submersible components. Real-time measurements of respiration rates were made at a number of discrete intervals over a maximum of 7 days using proprietary software, and the results were corrected for a 100 g fish using a scaling exponent of 0.8 (Jordan et al. 2001). Fish remained undisturbed in the respirometer between sampling intervals to minimise stress. After a full set of measurements was completed on a single adult dragonfish, the individual was weighed (wet mass; adult fish

were returned live to the sea) and its volume determined by displacement. Temperature and salinity oxygen compensation calculations for oxygen concentration were performed by the proprietary software, with the salinity set manually to 35‰ and the temperature measured simultaneously by the Optode probe. Water temperatures during the measurement periods were generally −1.2°C, but increased to 0°C in the longest measurement periods. Background respiration rates and oxygen leakage into the respirometer were measured and found to be insignificant (< 1% of measured values).

### Microbomb calorimetry

The heats of combustion of whole naked dragonfish eggs [< 48 h post-fertilization (hpf)], post-gastrulation embryos [89 days post-fertilization (dpf)], egg chorions, 1-day-old hatched larvae and yolk sacs removed from late-stage larvae were determined using a Phillipson oxygen microbomb calorimeter (Gentry Instruments Inc., Aiken, SC, USA). Pelleted samples (5–20 mg) were prepared and combusted as described by Phillipson (1964), with minor modifications as suggested in the user's manual, and correlated to a standard curve (2% accuracy) created by combusting 5–25 mg high-purity benzoic acid (Parr Instrument Company, Moline, IL, USA). Heats of combustion for standards and samples were calculated and corrected according to the manufacturer's instructions. Eggs, embryos and larvae were weighed individually and then combined (3–4 individuals per analysis) to facilitate handling. The reported heat of combustion values are ash-inclusive since ash-free values could not be related directly to the measured dry weights of the samples used for respiration measurements. Calorific values for embryos were calculated by subtracting the mean heat of combustion for individual chorions from the mean heat of combustion for the eggs (egg = embryo + chorion).

Statistical analyses were undertaken using the XLStats macro and Microsoft Excel. Results are presented as the mean ± standard deviation unless specified as standard error of the mean (SE).

## Results

Respiration rates of individual embryos were measured during the first 20 days of development. In an initial set of experiments, changes in egg respiratory activity were measured from 17–24 hpf, during which time the embryo undergoes first cleavage. Average egg respiration rates were found to increase from $2.17 \pm 1.02$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ at about 17 hpf to $5.72 \pm 0.56$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ at about 24 hpf (Fig. 1). In a separate series of experiments, the respiration rates of embryos from 2–20 dpf were measured. There was no apparent trend during this time interval (slope = $0.026 \pm 0.022$ (SE); $P > 0.10$ (slope not greater than zero); $n = 125$), in which the overall average rate of oxygen consumption was $4.11 \pm 1.47$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ (Fig. 2).

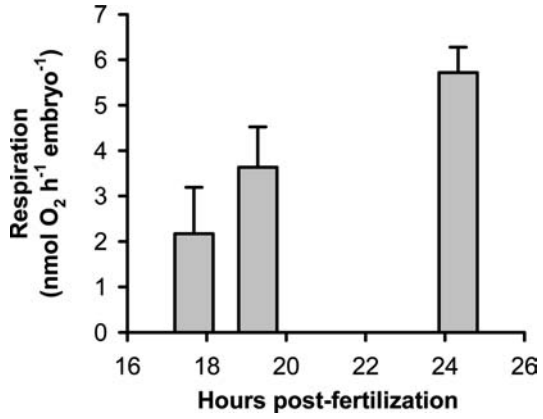


**Fig. 1** Respiration rates increase during the first 24 h of development

Density curves were used to assess the average individual respiration rates of pre-hatch ($y = 27.14x - 6.58$; $r^2 = 0.976$; $n = 6$) and post-hatch ($y = 112.41x - 35.49$; $r^2 = 0.821$; $n = 20$) yolk-sac larvae at an approximate age of 10 months post-fertilization. The slopes of the regression lines yield individual oxygen consumption rates of $27.14 \pm 3.92$ (SE) and $112.41 \pm 31.38$ (SE) nmol $O_2$ $h^{-1}$ $ind^{-1}$, respectively (Fig. 3).

Respiration rates of individual larvae were also measured over the 46–63 day post-hatch period, during which time the overall average respiration rate was $64.4 \pm 15.11$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ (Fig. 4), with no significant trend (slope = $0.498 \pm 0.376$ (SE); $P > 0.09$ (slope not greater than zero); $n = 53$). DW-specific respiration rates of hatched larvae (approximately 1–2 months old) are shown in Fig. 5. The slope of the regression line is $16.1 \pm 3.4$ (SE) nmol $O_2$ $h^{-1}$ $mg^{-1}$ DW ($r^2 = 0.4464$; $n = 30$).

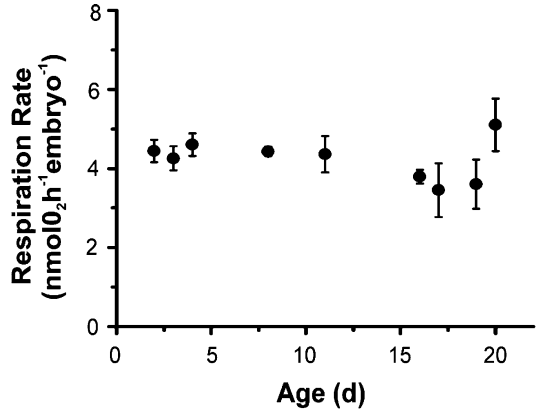


**Fig. 2** Respiration rates of individual embryos during the first 2–20 days of development. The average respiration rate is $4.11 \pm 1.47$ nmol $O_2$ $h^{-1}$ $ind^{-1}$ ($n = 125$)

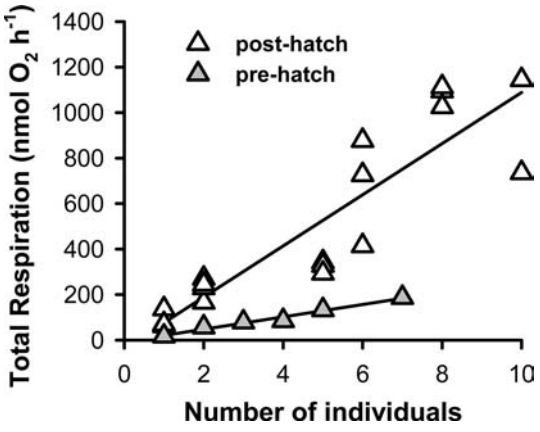


**Fig. 3** Respiration rates of pre- and post-hatch larvae at approximately 10 months post-fertilization. The slopes of the regression lines yield individual oxygen consumption rates of 27.14 ± 3.92 (SE; $n = 6$) and 112.41 ± 31.38 (SE; $n = 20$) nmol $O_2$ $h^{-1}$ $ind^{-1}$, respectively

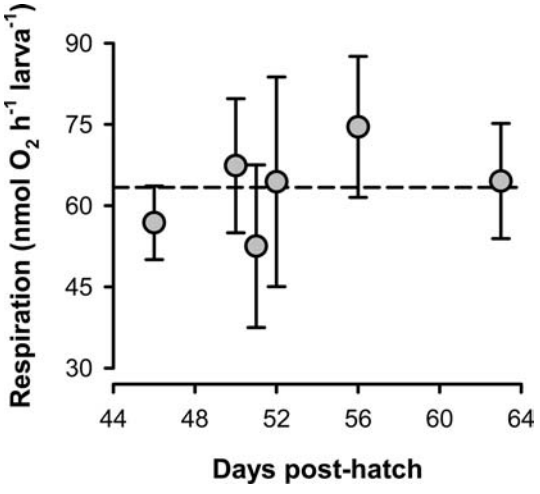


**Fig. 4** Respiration rates of individual larvae measured at 46–63 dph. The average respiration rate during this period (*dashed line*) was 64.4 ± 15.11 nmol $O_2$ $h^{-1}$ $ind^{-1}$ ($n = 53$)

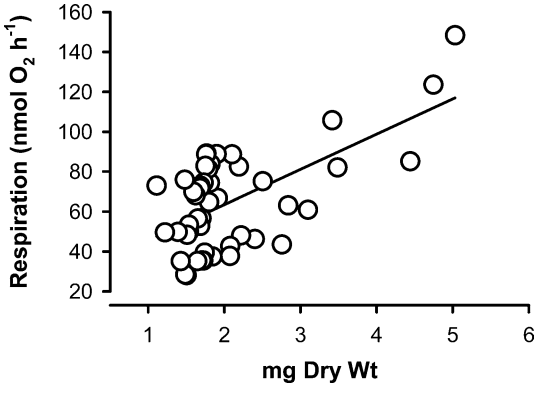


**Fig 5** Mass-specific respiration rates of post-hatch larvae (approx. 6–7 weeks). The slope of the regression line is 16.1 ± 3.4 (SE) nmol $O_2$ $h^{-1}$ $mg^{-1}$ DW ($r^2 = 0.4464$; $n = 30$)

Adult dragonfish respiration rates were measured at discrete intervals over a maximum of 7-day period. After initial acclimation to the respirometer, adult dragonfish seldom moved, even when slightly disturbed at the start of a measurement period or when a small window in the cover was opened for observation. Pectoral fin movements and postural changes were infrequent, or non-existent, except in two fish which displayed continual pectoral fin movements and occasionally changed position within the respirometer. The starting respiration rate was 4.19 ± 2.03 mmol $O_2$ $kg^{-1}$ $h^{-1}$ (134.02 ± 64.90 mg $O_2$ $kg^{-1}$ $h^{-1}$). The average lowest recorded respiration rate (corrected for a 100 g fish) was 0.91 ± 0.36 mmol $O_2$ $kg^{-1}$ $h^{-1}$ (29.06 ± 11.56 mg $O_2$ $kg^{-1}$ $h^{-1}$). All adult fish in the study started with an initially high respiration rate which dropped to a lower steady state (taken as the routine metabolic rate), typically over a 48 h period. The average decrease in respiration rate during acclimation to the respirometer (as a percentage of the starting value of each adult dragonfish) was 70.51 ± 23.35% (range 24.36–88.62%). A typical plot of the rate of change in respiration in a single adult dragonfish is illustrated in Fig. 6.

The energy content of dragonfish embryos without their chorions (< 48 hpf) was found to be 81,603 ± 2,473 mJ $ind^{-1}$ using microbomb calorimetry ($N = 8$, using a total mass equivalent to 24 eggs at 4.57 mg $egg^{-1}$ DW). The average energy of embryos without their chorions at 89 dpf was 54,976 ± 1,769 mJ $yolk^{-1}$ ($N = 3$, using a total mass equivalent to 12 embryos at 3.3 mg $ind^{-1}$ DW) and that of immediate pre-hatch yolks was 12,288 ± 732 mJ $yolk^{-1}$ ($N = 4$, using a mass equivalent to 59 yolks at 0.5 mg $yolk^{-1}$ DW). The average energy content of a chorion was 12,106 ± 190 mJ ($N = 2$, using a mass equivalent to 27 chorions) and that of a newly hatched larva (< 48 h) was 39,290 ± 2,377 mJ $ind^{-1}$ ($N = 2$, using a mass equivalent to eight larvae).

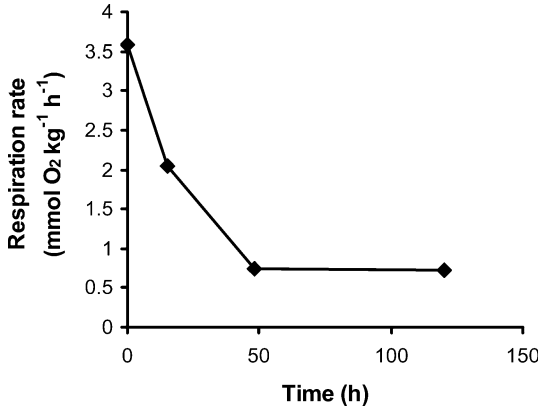


**Fig. 6** Representative adult dragonfish respiration rate time-course after transfer to the respirometer; at least 48 h is required for acclimatization. The respiration rate during the final measuring period (after 5 days) was 0.729 mmol $O_2$ $kg^{-1}$ $h^{-1}$ (23.32 mg $O_2$ $kg^{-1}$ $h^{-1}$)

## Discussion

Eggs of the naked dragonfish are laid in McMurdo Sound early in the Austral spring. Significant development during the embryonic phase takes place over the summer months, but hatching is delayed until the end of the winter (Evans et al. 2005). Aquarium observations suggest that the eggs may be ready to hatch well in advance of this date, and it is tempting to speculate that the hatching process is not initiated until an appropriate stimulus (such as the return of the sun) is received. Coincidentally, the delay in hatching may enable the larvae to optimise feeding opportunities associated with the appearance of the spring plankton bloom.

The protracted embryonic phase raises questions with respect to the metabolic status of the developing embryo since the yolk remains its only energy source during this time (10–11 months). Remarkably, considerable yolk (approximately 18% initial DW) remains at the time of hatch, and the yolk-sac larval stage can extend over about another 2–3 weeks under standard aquarium conditions.

We first measured changes in egg respiratory activity following fertilisation, including events leading up to the first cleavage. Average egg respiration rates were relatively low ($2.17 \pm 1.02$ nmol $O_2$ $h^{-1}$ $ind^{-1}$) when first measured at about 17 hpf (Fig. 1). Two hours later the respiration rate had increased 1.7× and it had continued to increase when measured at 24 hpf (2.6×). This small respiratory burst presumably reflects post-fertilization activities leading up to the first cleavage at about 24 hpf. In a separate series of experiments we measured egg respiration rates following first cleavage through to 20 dpf (Fig. 2). The rate of oxygen respiration during this period (2–20 dpf) averaged nearly twice the 17 hpf level. Observations of egg respiration rates ceased prior to the beginning of epiboly, which was underway by 25 dpf.

Respiration rates were also measured in late-stage embryos from the previous season around the time of hatching. Fully developed, pre-hatch embryos displayed 12.5× higher respiration rates relative to eggs measured at 17 hpf. A significant respiratory burst (nearly 4× that found immediately pre-hatch and 50× that found in 17 hpf embryos) was detected immediately post-hatch. The larval respiration rate measured at 46–63 dph, after the yolk has been completely resorbed, averaged 30× the rate shown by 17 hpf embryos. There is no statistically significant change in respiration rate during this post-hatch interval ($P = 0.0953$; slope not greater than zero), although the larvae increase in mass from hatch under aquarium conditions (Ln wet mass $= 0.012$ (days) $+$ 1.813; $r^2 = 0.51$).

Data on notothenioid egg and larval respiration rates are notably absent from the literature. Nevertheless, studies across a broad range of teleost species suggest oxygen consumption is generally low shortly after fertilization, with a fluctuating increase during epiboly to a peak near blastopore closure. This is followed by a decline during organogenesis and then another rise towards hatch, after which the rate of oxygen consumption changes in a variable manner, depending on the species and their feeding state (reviewed in Rombough 1988). Although direct comparisons across phylogenetically and/or ecologically diverse groups may not be particularly rewarding, Finn et al. (1995) found that oxygen consumption in developing yolk-sac larvae of the Atlantic halibut *Hippoglossus hippoglossus*, increased slowly from hatching (about 10 nmol $O_2$ $h^{-1}$ $ind^{-1}$) up to about 20 dph (15 nmol $O_2$ $h^{-1}$ $ind^{-1}$), after which there was a rapid rise to a peak (52 nmol $O_2$ $h^{-1}$ $ind^{-1}$) around 33 dph and then a general decline to the completion of yolk-sac resorption (28 nmol $O_2$ $h^{-1}$ $ind^{-1}$), which is around 45 dph for this species. The eggs of the Atlantic halibut may be of similar diameter (3–3.5 mm) to those of *G. acuticeps* ($3.42 \pm 0.19$ mm), but they are incubated in significantly warmer water (6–8°C). Furthermore, although the newly hatched larvae of the two species are of similar mass, the Atlantic halibut contains significantly more yolk, which presumably contributes to the lengthy yolk-sac larval stage and may influence metabolic dynamics. Early larvae (0.04 mg DW) of the Atlantic cod *Gadus morhua* have a respiration rate of about 5 nmol $O_2$ $h^{-1}$ $ind^{-1}$ measured at 7°C (Finn et al. 2002), which is considerably lower than that shown by *G. acuticeps* larvae ($64.4 \pm 5$ nmol $O_2$ $h^{-1}$ $ind^{-1}$). However, there are marked differences in size (*G. acuticeps* early larval DW is $1.34 \pm 0.15$ mg) and measured incubation temperature (between −1.5°C and 0°C for *G. acuticeps*), which again will influence metabolic dynamics.

The increase in respiration rate seen in many teleost species prior to hatching has been attributed to weakening of the chorion (removing a restriction to oxygen supply) and to changes in activity. Observations of developing *G. acuticeps* embryos show that they are capable of turning within the egg soon after the pigmented eye stage, and that this activity becomes increasingly vigorous as the larvae attempt to break free of the enclosing chorion at the time of hatch. Respiration rates measured in early eggs prior to the embryos developing the capacity for free movement represent a basal (or standard) state for this stage of development. Since older larvae and adults were able to move in the experimental chambers, the values determined for these represent the routine (or resting) metabolic condition associated with spontaneous activity. As the level of activity has a profound effect on the rate of oxygen consumption, some variation in the data seems inevitable.

The significant respiratory burst detected in *G. acuticeps* larvae immediately after hatch presumably reflects the energy expenditure associated with rupturing and escaping from the protective chorion, and arguably an element of stress as the larvae leave the protection afforded by their egg case. The negatively buoyant larvae swim continuously from hatch, displaying a subca-

rangiform locomotory mode with undulations traversing about half the body. The pectoral fins beat regularly, presumably providing a means of auxiliary propulsion in addition to influencing manoeuvrability and stabilisation. Larvae must devote considerable aerobic energy towards continuous swimming, as shown by a relatively high average post-hatch respiration rate (Fig. 3), in order to maintain their position in the water column and to seek out prey. Periods of burst activity are brief and confined to occasional darting and vigorous wriggling.

The average adult naked dragonfish metabolic rate ($29.06 \pm 11.56$ mg $O_2$ $kg^{-1}$ $h^{-1}$ corrected for a 100 g fish using a 0.8 scaling exponent) determined in the current study is significantly lower than that reported by Wells for the same species (Macdonald et al. 1987). In that study, an average metabolic rate of 45.3 mg $O_2$ $kg^{-1}$ $h^{-1}$ was determined at −1.5 °C, using a 0.9 scaling exponent. Wells (1987) also reported uncorrected oxygen uptake data for adult *G. acuticeps* (unfed for 10–14 days), with an average rate of $46.7 \pm 12.3$ mg $O_2$ $kg^{-1}$ $h^{-1}$ for fish weighing $74.4 \pm 12.8$ g ($N = 5$) after a 12 h acclimation period. The relatively high oxygen consumption rates for Antarctic fish reported in these and other studies (reviewed in Clarke and Johnston 1999) have been considered by some to provide evidence of metabolic cold adaptation (Krogh 1914, 1916). However, they are almost certainly overestimates as a consequence of handling stress and other confounding factors (Holeton 1974; Steffensen et al. 1994; Steffensen 2002). Significantly, we found that even the simple transfer of adult dragonfish from aquaria (where they had been acclimatized over at least 2 weeks without feeding) to a respirometer (in which they could still move and turn) led to artificially high oxygen consumption rates, which took at least 48 h to reach low steady-state levels (Fig. 6).

As an initial approach to deriving an energy budget for the early life history of the naked dragonfish, we have estimated energy consumption during development (Table 1; Fig. 7) using measured values of $81{,}603 \pm 2{,}096$ mJ $ind^{-1}$ for chorion-subtracted naked dragonfish eggs (< 48 hpf), $54{,}976 \pm 1{,}049$ mJ $ind^{-1}$ for chorion-subtracted embryos at 89 dpf, and $12{,}288 \pm 732$ mJ $yolk^{-1}$ for immediate pre-hatch yolks. An average oxyenthalpic equivalent of 484 kJ $mole^{-1}$ $O_2$ (Gnaiger 1983) was used to convert respiration rates

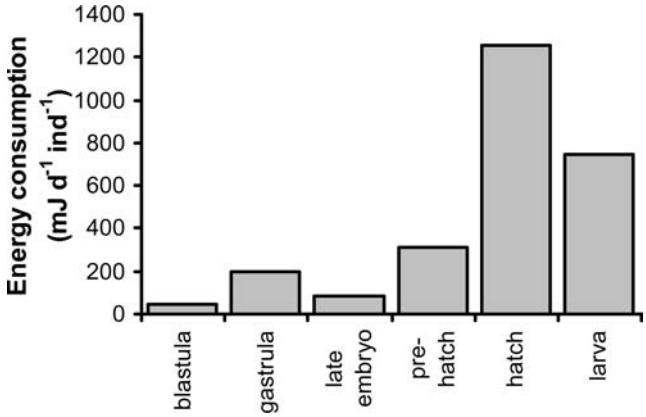


**Fig. 7** Rates of energy consumption during development as estimated in the respiration energy budget. The late-stage embryo (spanning approximately 213 days and including the overwintering period) has a relatively low rate of respiration consuming energy at a rate of about 82 mJ $day^{-1}$ $ind^{-1}$

into energy utilization rates, assuming a 50:50 balance of protein and lipid consumption in fuelling metabolism (Marsh et al. 1999). After hatching, there is sufficient energy (12,288 mJ) remaining in a larva as unused yolk to fuel metabolism (without feeding) for a maximum of 16–17-day period using the determined average larval respiration rate of 64.4 nmol $O_2$ $h^{-1}$ $ind^{-1}$ measured 46–63 dph. For most organisms that reproduce with yolky eggs, there is a general trend for ~ 60% of the initial egg mass to be utilized for producing organic tissues to produce a free-feeding larva or juvenile stage at hatch. The other 40% is required to provide the metabolic energy necessary to sustain that anabolic growth (Needham 1931; Calow 1977). Thus, we can generally assume for developmental energy budgets that at most, only 50% of the egg's initial energy content is available to fuel metabolism (Marsh et al. 1999). By applying this estimate to post-hatch yolk-derived energy utilization, a naked dragonfish larva may be able to survive 8–9 days at the determined larval respiration rate without feeding (Table 1). This result suggests that a reasonable tolerance window is available for naked dragonfish larvae to hatch and begin feeding in the water column before impacting organic mass. In a related study we have determined a yolk absorption time of about 15 days for

**Table 1** Estimated energy budget during the developmental period

| Stage | Respiration rate (nmol $O_2$ $h^{-1}$) | Metabolic rate (mJ $h^{-1}$) | Time (days) | Energy consumed (mJ) | Energy remaining (mJ) | Energy remaining (%) |
|---|---|---|---|---|---|---|
| Egg | | | | | 81,603 | 100 |
| Blastula | 4.1 | 1.98 | 29 | 1,381 | 80,222 | 98 |
| Gastrula | 17.2 | 8.32 | 60 | 11,988 | 68,234 | 84 |
| Late embryo | 7.1 | 3.44 | 213 | 17,567 | 50,667 | 62 |
| Pre-hatch | 27.1 | 13.12 | 7 | 2,204 | 48,464 | 59 |
| Hatch* | 112.4 | 54.4 | 1 | 1,306 | 47,158 | 58 |
| Larvae | 64.4 | 31.17 | 8.6 | 6,433 | 40,725 | 50 |

*At the time of hatch there is about $12{,}288 \pm 732$ mJ energy available in the remaining yolk (18% whole larval DW) which, if we assume only 50% is available to fuel metabolism, will provide enough energy for a larva to survive 8–9 days without feeding and without impacting significantly on its organic mass

naked dragonfish larvae (Evans et al. 2005). Other published yolk absorption times for notothenioids are in the range 21–35 days (Koch and Kellermann 1991), but both data sets are likely to be overestimates since they do not fully account for the possibility that the rate of yolk utilization is confounded by the availability of exogenous food.

To balance the energy equation, we have utilised a respiration rate of 4.1 nmol $O_2$ $h^{-1}$ $ind^{-1}$ (measured 2–20 dpf) for the blastula period of about 29 days. Since we know that 89 dpf embryos contain about 54,976 mJ $ind^{-1}$, then assuming 50% energy is available to fuel metabolism, we can calculate a residual 68,234 mJ at the end of the estimated gastrula period (89 dpf) with an energy utilization rate of about 11,988 mJ per 60 days post-blastula (Table 1). To achieve this rate of energy utilisation, the respiration rate must rise to an average of about 17.2 nmol $O_2$ $h^{-1}$ $ind^{-1}$ during the 60-day period which includes gastrulation. If we now assume that the pre-hatch respiratory rate represents a short-term ramping-up of energy expenditure over a nominal 7-day period prior to hatching, and that the respiratory burst at the time of hatching is of relatively short duration (1 day) to enable the larva to break free of the encompassing chorion, then we can estimate that the bulk of post-gastrulation respiratory energy expenditure (during 213 days of incubation) must be at a relatively low average rate (7.1 nmol $O_2$ $h^{-1}$ $ind^{-1}$) for the budget to balance.

The pattern of energy expenditure reflected in the naked dragonfish energy budget is broadly in line with trends shown by a range of teleosts (reviewed in Rombough 1988). Interestingly, the inferred low average respiration rate during the post-gastrulation period prior to hatching (7.1 nmol $O_2$ $h^{-1}$ $ind^{-1}$) supports the possibility that embryos of this species overwinter in a relatively quiescent metabolic state awaiting a suitable stimulus (such as the return of the sun) to initiate hatching.

**Acknowledgements** We thank Kevin Hoefling, Ben Hunt and Luke Hunt for their efforts in collecting eggs and adult fish by SCUBA at McMurdo Station, Antarctica, and Paul Ulrich and Allison Green for their assistance with some of the respiration equipment. This work was supported by: the National Science Foundation, Office of Polar Programs (#02-31006 to ALD and C-HCC; #02-38281 to AGM). CWE is grateful to the University of Auckland Research Committee for their continuing support.

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