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# The effects of temperature and size on the growth, energy budget and waste outputs of barramundi (*Lates calcarifer*)

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#### ABSTRACT

The effects of temperature and size on the growth, energy budget and waste output were examined in small (21 g) and large (142 g) juvenile barramundi (*Lates calcarifer*) grown for 26 days at one of six temperatures (23 °C, 26 °C, 29 °C, 32 °C, 35 °C and 38 °C). Fish were fed once a day and faeces was collected by stripping at the end of the growth trial to measure the apparent digestibility of dry matter, energy, nitrogen and phosphorous. Survival was not affected by temperature and was high (87-100%) in all treatments. The influence of size (W) on the daily growth rate (DGR, g fish $^{-1}$  d $^{-1}$ ) was modelled with the power function  $DGR = \alpha W^{\gamma}$ , where  $\alpha$  and  $\gamma$  are constants fitted with regressions to incorporate the effect of temperature into the model. Following a parabolic response to temperature, DGR and feed intake were high in both size classes across a temperature range extending from 29 °C to 35 °C. Temperatures for maximum growth and feed intake lay within the thermal zone for optimal feed efficiency from 26 °C to 35 °C. Feed efficiency was higher in small fish than in large fish. Although size had no effect on nutrient digestibility, temperatures influenced the apparent digestibility of dry matter, energy and phosphorous. The partitioning of consumed energy between growth, heat loss, nitrogen excretion and faeces showed increased energy losses through nitrogen excretion at 38 °C. Size and temperature had significant effects on waste outputs in the form of solid, nitrogen and phosphorous waste. In both size classes lowest levels of waste output were seen from 29 °C to 35 °C with levels rising at the cooler temperatures. The energetics and waste output of juvenile barramundi raised at different temperatures are discussed with respect to temperature dependent nutrient digestibility, feed intake and protein turnover.

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#### 1. Introduction

Understanding and maximising the efficiency of aquaculture systems is becoming increasingly critical given the cost of feed inputs and the need to manage farm effluents for the long term sustainability of the waters available for farming. Two primary factors affect feed efficiency and waste output: temperature and size (Glencross, 2008). The effects of temperature have been extensively studied in cultured fish. From a production perspective temperature primarily influences growth potential and therefore feed demand and eventually the waste that needs to be treated or that is released in the environment (Glencross, 2008). Temperature also influences the partitioning of consumed energy between tissue accretion and energy sinks such as heat loss, nitrogen excretion and faeces. The differential effect of temperature on each physiological process plays a dominant role in

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<sup>1</sup> Present address: Food Futures Flagship — CSIRO Marine and Atmospheric Research, PO Box 120, Cleveland, QLD 4163, Australia. Fish somatic growth and nutrient retention efficiency are also highly dependent of size (Imsland et al., 2006; Glencross, 2008). It is generally accepted that in fish metabolic processes such as growth rate increase with size and their relationship can be described as  $\Omega = \alpha W^{\gamma}$ , where  $\Omega$  is the rate of metabolic process, W is live weight,  $\alpha$ is the metabolic level and  $\gamma$  is the weight exponent that standardises  $\Omega$  across sizes. Although the weight exponent is often assumed to be constant for a given species, both  $\alpha$  and  $\gamma$  can be influenced by temperature (Paloheimo and Dickie, 1966; Liu et al., 1998). While declining rates of growth with size are explained by associated declining rates of feeding, increases in size also lead to marked losses in feed efficiency (Imsland et al., 2006). Since both temperature and size affect growth and feed efficiency, the two factors should be



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determining the efficiency of feed utilisation. For instance, the increased feed intake that is generally associated with warmer temperatures will not necessarily result in improved growth rate when it is accompanied by a disproportionally higher increase in nutrients lost through metabolism. This is seen in Atlantic salmon *Salmo salar* L. (Koskela et al., 1997) and wolfish *Anarhichas lupus* L. (McCarthy et al., 1998), where optimal feed efficiency occurs at temperatures below those for maximum feed intake.

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studied together so that the changes in temperature for maximum feed efficiency that can occur through the production cycle (Björnsson et al., 2001) can be reflected in production models (Glencross, 2008).

Previous studies on the effect of temperature on growth, feed intake and nutrient retention in barramundi (Williams and Barlow, 1999; Katersky and Carter, 2005; Williams et al., 2006) have provided useful information on nutrient utilisation and requirements as well as thermal thresholds. However, the scope of those studies was limited because they did not encompass the entire temperature range of production (ca. 20 °C-35 °C) or account for nutritional changes that are known to occur as fish grow during the production cycle (Glencross, 2006; 2008). In the study by Williams and Barlow (1999) a range of sizes was examined from 40 g to 270 g, however, the temperatures studied were restricted to a range of 20 °C to 29 °C, where the barramundi achieved maximum growth and high feed conversion efficiency. The authors did not investigate responses beyond 29 °C or identify an upper thermal limit for production. More recently, Katersky and Carter (2005) examined the growth and nutrient retention of barramundi from 27 °C to 39 °C. This latest study defined the upper thermal limit of juvenile ( $\sim 5 \text{ g}$ ) barramundi. However, it is not known whether the growth response and thermal threshold described in juveniles grown at high temperatures remain constant with size.

The aim of this study was to examine the combined effects of temperature and fish size on growth rate, feed conversion efficiency and nutrient utilisation in barramundi reared between 23 °C and 38 °C. This temperature range was selected in order to establish minimum and maximum thermal thresholds for growth and feed conversion efficiency and determine whether temperature thresholds vary during the production cycle. In addition, the effect of temperature on energy utilisation was studied by partitioning the amount of consumed energy into growth, heat loss, nitrogen excretion or faecal losses in order to identify possible reasons for worsening feed efficiency. From the data collected we were able to compare waste output between sizes and temperature and provide critical information for the management of waste in barramundi culture systems.

#### 2. Material and methods

#### 2.1. Experimental system and design

A flow-through system of 24×150 L cylindrical tanks with conical bottoms was designed to operate with six different temperatures, in blocks of four tanks, by mixing ambient and heated seawater. Water flow rate in each tank was adjusted to  $3 L \min^{-1}$  to maintain dissolved oxygen above 80% saturation. Two sizes of fish were grown in the system so that the experimental design was 6 temperatures  $\times 2$  sizes  $\times 2$  replicates. Temperature was measured every 20 min in each tank using data loggers (TCS, Thermocron) and the six temperature treatments measured as means  $(\pm SD)$  of daily averages between tanks (n = 4 tanks  $\times 26$  days) were: 23.3  $\pm$  0.2,  $26.0 \pm 0.0, 29.0 \pm 0.0, 32.0 \pm 0.0, 35.0 \pm 0.0, 37.7 \pm 0.8$  °C. The system was run on a 12 L:12D photoperiod. Minimum and maximum dissolved oxygen measurements were obtained daily in each tank, one and seven hours following feeding, respectively, using a Handy Gamma Oxygard oxygen electrode. Salinity was measured daily with a conductivity meter (WTW, Cond 315i).

#### 2.2. Fish acclimation

Barramundi were grown from eggs sourced from the Darwin Aquaculture Centre. Two groups of fish were grown to ~20 g (small) and ~140 g (large) at the Department of Fisheries Western Australia (Fremantle and Hillarys, Australia) at temperatures of 19–24 °C. Fish were gradually acclimated to their experimental temperature over a period of three weeks during which the temperature was raised at a maximum rate of ~1 °C per day. Acclimation was staggered so that each treatment group reached its experimental temperature simultaneously. Once at their experimental temperature fish were acclimated for a further two days before the start of the experiment. Fish were also acclimated to the experimental feed for 7 days prior to the start of the experiment to avoid interference from any previous feeding regime (Dickson and Kramer, 1971; Kaushik, 1980).

#### 2.3. Initial stocking and sampling procedures

Following acclimation fish were weighed on day 0 (Table 2) and the number of fish in each tank adjusted to 15 and 24 in large and small fish tanks, respectively. At the same time (day 0), three fish from each tank were euthanized, pooled and minced to determine initial proximate composition and gross energy content. Following a 26-day growing period all fish were weighed and a final sample of three fish per tank taken for chemical analysis. At the end of the growth study and to determine nutrient digestibility, faecal samples containing vttrium as inert marker (see diet composition, Table 1) were collected from all tanks, each day for a period of two weeks. Faeces were sampled between four and six hours following the last meal to account for differences in gut transit between warm and cool water grown fish. Faeces were collected from the hind gut region by applying gentle pressure to the abdominal area (Austreng, 1978; Glencross, 2008). Care was taken to ensure that the faeces were not contaminated by urine and mucous. Fish and faecal samples were freeze dried prior to analyses. Fish were sedated with AQUI-S® (AQUI-S New-Zealand Ltd) for weighing and faecal sample collection.

#### 2.4. Feeding

Throughout the experiment, every morning and over a period of 2 h, fish were fed a 3 mm floating extruded diet (Table 1) to slight excess. The diet was designed to meet the energy and protein requirements of barramundi (Glencross, 2006). Uneaten feed was recovered at the surface of each tank 2 h following the end of feeding. Daily feed intake was calculated as the difference between fed and uneaten feed amounts. Amounts of uneaten feed were oven dried at 105 °C for 24 h, weighed and multiplied by 1.16, the correction factor determined to convert uneaten dry feed back to fresh feed equivalent taking into account the loss of dry matter that occurred through leaching when soaking in the tank. Feed intake was expressed on a dry matter basis.

#### 2.5. Chemical analysis

Diet and faecal samples were analysed for dry matter, yttrium, ash, phosphorus, nitrogen and gross energy content. Dry matter was

#### Table 1

Ingredient composition and proximate analysis of the experimental diet.

Ingredients (g kg <sup>-1</sup> )	
Fish meal	700.0
Wheat flour	144.0
Fish oil	150.0
Vitamin and mineral premix <sup>a</sup>	5.0
Yttrium oxide	1.0
Chemical composition (g kg $DM^{-1}$ )	
Dry matter (g kg <sup>-1</sup> )	894.7
Crude protein	530.9
Total lipid	178.8
Phosphorus	17.0
Ash	126.4
Energy (MJ kg $DM^{-1}$ )	23.0

<sup>a</sup> Vitamin and mineral premix includes (IU/kg or g/kg of premix): Vitamin A, 2.5MIU; Vitamin D3, 0.25 MIU; Vitamin E, 16.7 g; Vitamin K, 3, 1.7 g; Vitamin B1, 2.5 g; Vitamin B2, 4.2 g; Vitamin B3, 25 g; Vitamin B5, 8.3; Vitamin B6, 2.0 g; Vitamin B9, 0.8; Vitamin B12, 0.005 g; Biotin, 0.17 g; Vitamin C, 75 g; Choline, 166.7 g; Inositol, 58.3 g; Ethoxyquin, 20.8 g; Copper, 2.5 g; Ferrous iron, 10.0 g; Magnesium, 16.6 g; Manganese, 15.0 g; Zinc, 25.0 g.

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