Using modelling approaches to understand the implications of physiological challenges and raw material demands on aquaculture feed designs

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Abstract

Models have been used for sometime in estimating fish production and nutrient/energy demands. One form of these models, the factorial bioenergetic model, is described here and several potential applications from it explored. One such application is the examination of the effects of heat-stress on barramundi (Lates calcarifer) bioenergetics. These were examined using a modelling approach to explore the implications of the key model parameters and subsequently a redefined model was designed. Using the redefined factorial model the optimal iterative feed specifications were then redefined for a range of fish sizes at temperatures of 25°C, 30°C and 35°C. A feed demand model was also developed based on the demand for digestible energy at each of these temperatures. The model outputs suggest that at high temperatures (35°C) that there is an increase in the ratio of digestible protein to digestible energy required and that with increasing size there is a decrease in the digestible protein to digestible energy demand. These model outputs have been independently evaluated in empirical experiments and provide evidence consistent with the model. The use of such a factorial feed demand model also allows for the examination raw material demands over the full production cycle of the fish. By following total feed demand and then overlaying least-cost formulations both with and without fishmeal/oil replacement options it becomes possible to highlight periods of high demand and issues associated with different formulation strategies. The use of such modelling approaches provides a useful tool to identify those strategies that should be followed up with practical empirical research.
Introduction

There has been substantial activity in recent times in the development of nutritional models to examine growth and feed use in fish (reviewed by Dumas et al., 2010). There are many applications for such models, and their use in aquaculture science has seen their adoption as a means of defining fish production potential, estimating feed utilisation demands and even feed specification designs via the use of iterative approaches to diet design (Lupatsch et al., 2001; Glencross, 2008). Those models being developed fall into either empirical or mechanistic categories and although the design of some of these models, particularly the more mechanistic ones, can be quite complex, the added complexity has not necessarily resulted in an improved model for practical purposes, despite that they are certainly more theoretically correct (Dumas et al., 2009). Most empirical models have been based on describing flows of energy and nutrients as governed by fundamental thermodynamic and energetic principles (Ursin, 1967; Cuenco et al., 1985; Cho and Bureau, 1998; reviewed by Dumas et al., 2009).

Those factorial models currently being used (Lupatsch et al., 1998; Glencross, 2008) are an empirical model form, the relative simplicity of which has allowed them to provide a useful platform for compartmentalizing the demands for energy into either somatic or non-somatic components. Those non-somatic components are generally assumed to account for maintenance, heat loss and activity. While the somatic component of energy demand is generally assumed to account for the energy value of the protein and lipid accumulated through growth. Typically the overall energy demand model has been summarised as:

\[
\text{Total Energy Demand (kJ/fish/d)} = M \cdot \text{Liveweight (kg)}^b + G \cdot \text{Energy gain (kJ)}
\]

where M and G are constants describing the efficiency of energy utilisation for maintenance and growth respectively and b is the metabolic weight exponent of the animal (Glencross, 2008).

A growth model for barramundi has been published earlier that was designed based on conservative thermal ranges from 15°C to 35°C for this species (Glencross, 2008). While this is generally adequate for most Australian production conditions, it did pose the question; of what happens at higher temperatures and what particular parameters are conserved and what...
parameters were temperature sensitive. To address these issues a series of studies were undertaken to measure the primary energetic parameters (as used in factorial models) from 20°C to 40°C. These included a better defined growth response curve to temperature (Bermudes et al., 2010), revised estimates of allometric scaling effects with increasing temperature (Glencross and Bermudes, 2010a) and more accurate estimates of maintenance protein and energy demands and partial efficiencies of protein and energy utilisation (Glencross and Bermudes, 2010b). The outcome of these studies combined was a revised model that made allowances for the effects of temperature as it increased above thermally optimal ranges.

Another key element of fish nutrition where modelling has some application is the analysis of raw material use with different feed strategies. The use of nutritional modelling has substantial potential here in providing estimates of the total raw material use throughout the various production phases of fish. Another advantage of this is the potential to use such models to examine different options in raw material use, both in terms of optimising production costs and also resource sustainability.

Accordingly this paper reports on the application of an advanced growth and energetic model for barramundi in examining physiological challenges (heat-stress) and raw material demands (fish meal and oil replacement) on diet specification designs. It is acknowledged that there are numerous other applications of such models and this paper does not seek to examine all those options, but merely highlight some implications of two potential applications.

The findings from these two applications of nutritional modelling are discussed in terms of the implications they have for the barramundi production and feed specifications, and provide an indication of what may be done for many other aquaculture species. However, like all models, the one presented has the potential to be useful, but is still merely an estimation. Therefore caution must be applied when undertaking any practical application of features of the model or applications derived from it, without first verifying their predictions with empirical data.
Methods

**Development of a revised factorial model for barramundi**

**The predictive growth equation**

A functional growth equation is central to an effective factorial energetic model. In this study a combination of commercial farm and our own laboratory data were used to map the growth per day (g/fish/d) with respect to geometric mean weight and mean water temperatures. These data included a range of assessment periods and fish size ranges from numerous fish cohorts, farms and experiments covering temperatures ranging from 18°C to 39°C. In excess of 800 data points were collected. These data were used to define an equation using the regression function of the tools package within Microsoft Excel XP version. The equation takes the form of:

\[
\text{Gain (g/fish/d)} = (K + xT + yT^2 + zT^3) \times (\text{live-weight})^{aT + b}
\]

In this equation, \( K \), and \( b \) are constants, and \( x \), \( y \), \( z \) and \( a \) are coefficients determined using the regression function. \( T \) is temperature (operational range of 16°C to 39°C) and \( \text{weight} \) is the geometric mean weight of the fish in grams.

\[
\begin{align*}
K &= 2.249522916 \\
x &= -0.327485829 \\
y &= 0.014951694 \\
z &= -0.000203425 \\
b &= 0.72000 \\
a &= -0.00950
\end{align*}
\]

**Revised fish composition modelling**

In addition to the revised growth equation a broader range and greater number of fish samples was collected from the commercial farm and laboratory sourced animals. These fish were analysed and combined with existing data to better define the tissue composition of fish from...
10g to 3000g. Fish were assayed for dry matter (water), crude protein, total lipids, ash and gross energy. All chemical analyses were carried out by NATA (National Association of Testing Authorities) accredited analytical service providers (Chemistry Centre (WA), East Perth, WA, Australia, Animal Health Laboratories, South Perth, WA, Australia and SARDI Pig and Poultry Production Institute, Roseworthy, SA). Dry matter was calculated by gravimetric analysis following oven drying at 105ºC for 24. Protein levels were calculated from the determination of total nitrogen by LECO auto-analyser, based on N x 6.25. Total lipids were measured using the chloroform:methanol method. Gross energy was determined by adiabatic bomb calorimetry of the samples. All data was represented back the animals composition on a live-basis.

**Feed demand modelling**

The amount of feed required to be fed was estimated based on the total energy demand (TED) as derived from a combination of parameters determined in this study (Table 1, Figure 1):

\[
TED (kJ/fish/d) = \text{Maintenance energy demand} + \text{Somatic energy demand} / \text{EUE}
\]

In this equation EUE is the energy utilisation efficiency which is based on \( y = -0.0039T^2 + 0.23T - 2.7779 \) (derived from Glencross and Bermudes, 2010b).

The somatic energy demand (kJ/g/d) was based on \( y = 3.82*(g/fish)^{0.12} \)

The maintenance energy demand (kJ/fish/d) was defined as a three-dimensional factorial equation. This equation has terms for fish live-weight (LW; kg/fish) and water temperature (T; operational range of 16ºC to 39ºC). The coefficient function is derived from Glencross & Bermudes, (2010b) while the exponent is derived from Glencross & Bermudes, (2010a). This combined equation takes the form of:

\[
MED (kJ/fish/d) = (wT^3 + xT^2 + yT + z) * LW \ (aT^4 + bT^3 + cT^2 + dT + e)
\]

\( z = 0.511880952 \)

\( y = -0.084382937 \)

x = 0.004256696
w = -6.03299*10^{-05}

e = 3.198095238
d = -0.443318001
c = 0.029088542
b = -0.000818734
a = 8.43395*10^{-06}

Following the determination of total energy demand (TED), the feed requirement is determined based on the TED divided by the digestible energy density of the feed being fed (Table 1, Figure 2). The digestibility of protein digestibility and energy was also observed to be temperature dependent (derived from Bermudes et al., 2010):

Protein digestibility = -0.0003T^2 + 0.0192T + 0.5416
Energy digestibility = -0.0003T^2 + 0.0226T + 0.4712

Figure 1. Total digestible energy demand with varying temperature and fish weight. Notable is that at higher temperatures an increase in energy demand occurs following the plateau through the optimal growth temperatures. This occurs due to an increasing turnover of protein energy. This effect is also more pronounced in larger fish than smaller fish.

Figure 2. Optimal feed allocation of a 16 MJ/kg digestible energy diet with varying temperature and fish weight. Notable is that at higher temperatures a higher intake is required to offset the increase in maintenance energy demands and a reduced partial efficiency of utilisation. This feed demand model does not consider factors other than gross energy demand in defining feed intake requirements and other factors may impinge on the potential to achieve this intake level at each of the temperatures detailed.

Revision of feed specifications based on iterative modelling

If a given dietary digestible energy density is prescribed, not only can the feed ration required to be fed to achieve the predicted growth be determined, but the concentration of digestible protein required in a diet of that given digestible energy density can also be estimated (Table 2). The amount of digestible protein required in the diet is defined based on:

\[
\text{Digestible Protein (g/kg)} = \frac{(\text{Maintenance Protein Demand} + \text{Somatic Protein Demand} / \text{PUE})}{\text{TED} / \text{Diet Digestible Energy}}
\]
The maintenance protein demand (g/fish/d) was also defined as a three-dimensional factorial equation with terms for fish weight (LW: kg/fish) and water temperature (T: operational range of 16°C to 39°C). The coefficient function for this relationship was derived from Glencross and Bermudes, (2010b) while the exponent was derived from Glencross and Bermudes, (2010a):

\[
\text{MPD (g/fish/d)} = (vT^4 + wT^3 + xT^2 + yT + z) * \text{LW}^{(aT^4 + bT^3 + cT^2 + dT + e)}
\]

\[
z = 14.7156242
\]
\[
y = -2.6703623
\]
\[
x = 0.1811715
\]
\[
w = -0.0053183
\]
\[
v = 0.0000573
\]
\[
e = 0.136428571
\]
\[
d = 0.046585498
\]
\[
c = 0.000260417
\]
\[
b = -0.000101799
\]
\[
a = 2.0715*10^{-06}
\]

The Somatic Protein Demand (protein concentration of the animals weight gain, % live-weight basis) was determined as 17% and was not influenced by fish live-weight (Figure 3).

The PUE is the utilisation efficiency of dietary protein as determined from the regression of digestible protein intake against crude protein gain (derived from Glencross and Bermudes, 2010b). The PUE was also observed to be temperature affected. The relationship between PUE and temperature (T) is defined as:

\[
\text{PUE} = -0.0039T^2 + 0.2185T - 2.5585
\]

On a similar basis, by specifying a fixed dietary digestible protein density, the iterative process can also be used to define the digestible energy density to optimise the use a fixed level of dietary digestible protein.
Table 1. Derived energy and protein demands in growing barramundi at different temperatures based on the growth, energy and protein utilisation models

<table>
<thead>
<tr>
<th>Temperature</th>
<th>25°C</th>
<th>30°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fish weight (g/fish)</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>500</td>
<td>1000</td>
<td>2000</td>
</tr>
<tr>
<td>Expected growth (g/d)</td>
<td>2.11</td>
<td>2.88</td>
<td>2.27</td>
<td>4.24</td>
<td>5.55</td>
<td>7.27</td>
</tr>
</tbody>
</table>

**Energy requirement**

<table>
<thead>
<tr>
<th></th>
<th>25°C</th>
<th>30°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metabolic BW Exponent</td>
<td>0.797</td>
<td>0.804</td>
<td>0.868</td>
<td>0.868</td>
<td>0.868</td>
<td>0.868</td>
</tr>
<tr>
<td>Metabolic BW (kg)</td>
<td>0.159</td>
<td>0.157</td>
<td>0.135</td>
<td>0.548</td>
<td>1.000</td>
<td>1.826</td>
</tr>
<tr>
<td>DEmaint (kJ/fish/d)</td>
<td>4.72</td>
<td>7.40</td>
<td>10.16</td>
<td>41.11</td>
<td>75.05</td>
<td>137.02</td>
</tr>
<tr>
<td>Energy gain (kJ/fish/d)</td>
<td>14.00</td>
<td>19.14</td>
<td>15.09</td>
<td>34.14</td>
<td>48.54</td>
<td>69.00</td>
</tr>
<tr>
<td>Energy utilisation efficiency (%)</td>
<td>53.5</td>
<td>61.2</td>
<td>49.5</td>
<td>49.5</td>
<td>49.5</td>
<td>49.5</td>
</tr>
<tr>
<td>DEgrowth (kJ/fish/d)</td>
<td>26.20</td>
<td>31.27</td>
<td>30.50</td>
<td>69.03</td>
<td>98.13</td>
<td>139.50</td>
</tr>
<tr>
<td>DEtotal (kJ/fish/d)</td>
<td>30.92</td>
<td>38.67</td>
<td>40.66</td>
<td>110.14</td>
<td>173.18</td>
<td>276.52</td>
</tr>
</tbody>
</table>

**Protein requirement**

<table>
<thead>
<tr>
<th></th>
<th>25°C</th>
<th>30°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein BW Exponent</td>
<td>0.682</td>
<td>0.698</td>
<td>0.830</td>
<td>0.830</td>
<td>0.830</td>
<td>0.830</td>
</tr>
<tr>
<td>Protein BW (kg)</td>
<td>0.208</td>
<td>0.201</td>
<td>0.148</td>
<td>0.563</td>
<td>1.000</td>
<td>1.777</td>
</tr>
<tr>
<td>DProt-maint (g/fish/d)</td>
<td>0.098</td>
<td>0.096</td>
<td>0.17</td>
<td>0.648</td>
<td>1.152</td>
<td>2.047</td>
</tr>
<tr>
<td>Protein gain (g/fish/d)</td>
<td>0.359</td>
<td>0.49</td>
<td>0.386</td>
<td>0.721</td>
<td>0.943</td>
<td>1.233</td>
</tr>
<tr>
<td>Protein utilisation efficiency (%)</td>
<td>46.7</td>
<td>48.7</td>
<td>31.2</td>
<td>31.2</td>
<td>31.2</td>
<td>31.2</td>
</tr>
<tr>
<td>DProt-growth (g/fish/d)</td>
<td>0.769</td>
<td>1.008</td>
<td>1.240</td>
<td>2.314</td>
<td>3.027</td>
<td>3.959</td>
</tr>
<tr>
<td>DProt-total (g/fish/d)</td>
<td>0.867</td>
<td>1.104</td>
<td>1.411</td>
<td>2.962</td>
<td>4.179</td>
<td>6.007</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>25°C</th>
<th>30°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
<th>35°C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Optimal DProt:DE (g/MJ)</td>
<td>28.0</td>
<td>28.5</td>
<td>34.7</td>
<td>26.9</td>
<td>24.1</td>
<td>21.7</td>
</tr>
</tbody>
</table>

a Growth rate defined using growth function.
b Defined based on the exponent value of MED.
c Metabolic body weight based on live-weight *Metabolic body weight exponent.d MED based on MED function.e Energy gain is based on y = 3.82*(g/fish)^0.12; Where g/fish is the expected growth.f Energy utilisation efficiency = (-0.0039T^2 + 0.23T - 2.7779)*100.g DEgrowth = Energy gain / Energy utilisation efficiency.h DETotal = DEgrowth + DEmaint.i Defined based on the exponent value of MPD.j Protein body weight based on live-weight *Protein body weight exponent.k MPD based on function.l Protein gain is based on y = 17.0%*(g/fish); Where g/fish is the expected growth.m Protein utilisation efficiency = (-0.0039T^2 + 0.2185T - 2.5585).n DProt-growth = Protein gain / Protein utilisation efficiency.o DProt-total = DProt-growth + DProt-maint.p Based on exponential functions.

Estimation of nutrient and raw material demand

Table 2. Iteratively determined diet specifications at varying temperatures based on the redefined growth and utilisation parameters.

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>25</th>
<th>25</th>
<th>25</th>
<th>30</th>
<th>30</th>
<th>30</th>
<th>35</th>
<th>35</th>
<th>35</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fish weight (g/fish)</td>
<td>100</td>
<td>500</td>
<td>1000</td>
<td>100</td>
<td>500</td>
<td>1000</td>
<td>100</td>
<td>500</td>
<td>1000</td>
</tr>
<tr>
<td>Expected growth (g/d)</td>
<td>2.11</td>
<td>4.59</td>
<td>6.41</td>
<td>2.88</td>
<td>5.81</td>
<td>7.85</td>
<td>2.27</td>
<td>4.24</td>
<td>5.55</td>
</tr>
<tr>
<td>DE-total (kJ/fish/d)</td>
<td>30.9</td>
<td>86.1</td>
<td>134.5</td>
<td>38.7</td>
<td>103.4</td>
<td>159.4</td>
<td>40.7</td>
<td>110.1</td>
<td>173.2</td>
</tr>
<tr>
<td>DProt-total (g/fish/d)</td>
<td>0.87</td>
<td>1.97</td>
<td>2.81</td>
<td>1.10</td>
<td>2.32</td>
<td>3.22</td>
<td>1.41</td>
<td>2.96</td>
<td>4.18</td>
</tr>
<tr>
<td>Optimal DP:DE (g/MJ)</td>
<td>28.0</td>
<td>22.8</td>
<td>20.9</td>
<td>28.5</td>
<td>22.5</td>
<td>20.2</td>
<td>34.7</td>
<td>26.9</td>
<td>24.1</td>
</tr>
</tbody>
</table>

Diet specification based on predetermined digestible energy density

<table>
<thead>
<tr>
<th>Digestible Energy (MJ/kg)</th>
<th>16</th>
<th>18</th>
<th>20</th>
<th>16</th>
<th>18</th>
<th>20</th>
<th>16</th>
<th>18</th>
<th>18</th>
</tr>
</thead>
<tbody>
<tr>
<td>Digestible Protein (g/kg)</td>
<td>449</td>
<td>411</td>
<td>417</td>
<td>456</td>
<td>405</td>
<td>405</td>
<td>555</td>
<td>484</td>
<td>434</td>
</tr>
<tr>
<td>Crude Protein (g/kg)</td>
<td>541</td>
<td>495</td>
<td>503</td>
<td>537</td>
<td>476</td>
<td>476</td>
<td>653</td>
<td>569</td>
<td>511</td>
</tr>
<tr>
<td>Crude Fat (g/kg)</td>
<td>142</td>
<td>230</td>
<td>285</td>
<td>130</td>
<td>225</td>
<td>285</td>
<td>60</td>
<td>160</td>
<td>192</td>
</tr>
<tr>
<td>Starch (g/kg)</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
</tr>
<tr>
<td>Required Feed Intake (g/fish/d)</td>
<td>1.93</td>
<td>4.78</td>
<td>6.72</td>
<td>2.42</td>
<td>5.74</td>
<td>7.97</td>
<td>2.54</td>
<td>6.12</td>
<td>9.62</td>
</tr>
<tr>
<td>Expected FCR</td>
<td>0.92</td>
<td>1.04</td>
<td>1.05</td>
<td>0.84</td>
<td>0.99</td>
<td>1.01</td>
<td>1.12</td>
<td>1.44</td>
<td>1.73</td>
</tr>
</tbody>
</table>

Diet specification based on predetermined digestible protein density

<table>
<thead>
<tr>
<th>Digestible Protein (g/kg)</th>
<th>500</th>
<th>450</th>
<th>400</th>
<th>500</th>
<th>450</th>
<th>400</th>
<th>550</th>
<th>500</th>
<th>450</th>
</tr>
</thead>
<tbody>
<tr>
<td>Digestible Energy (MJ/kg)</td>
<td>17.8</td>
<td>19.7</td>
<td>19.2</td>
<td>17.5</td>
<td>20.0</td>
<td>19.8</td>
<td>15.9</td>
<td>18.6</td>
<td>18.6</td>
</tr>
<tr>
<td>Crude Protein (g/kg)</td>
<td>602</td>
<td>542</td>
<td>482</td>
<td>588</td>
<td>529</td>
<td>471</td>
<td>647</td>
<td>588</td>
<td>529</td>
</tr>
<tr>
<td>Crude Fat (g/kg)</td>
<td>116</td>
<td>195</td>
<td>212</td>
<td>108</td>
<td>205</td>
<td>227</td>
<td>47</td>
<td>150</td>
<td>182</td>
</tr>
<tr>
<td>Starch (g/kg)</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
<td>90</td>
</tr>
<tr>
<td>Required Feed Intake (g/fish/d)</td>
<td>1.74</td>
<td>4.37</td>
<td>7.02</td>
<td>2.21</td>
<td>5.16</td>
<td>8.06</td>
<td>2.56</td>
<td>5.92</td>
<td>9.29</td>
</tr>
<tr>
<td>Expected FCR</td>
<td>0.82</td>
<td>0.95</td>
<td>1.10</td>
<td>0.77</td>
<td>0.89</td>
<td>1.03</td>
<td>1.13</td>
<td>1.40</td>
<td>1.67</td>
</tr>
</tbody>
</table>

a Growth rate defined using function.
b Defined based on the GED.
c Defined based on the MPD+DProt-growth/PUE.
d Based on exponential functions.
e Energy density arbitrarily assigned within practical regimes.
f Defined based on function using optimal DP:DE.
g Assumes protein digestibility varies with temperature.
h Varied to satisfy DE demands. Assumes lipid digestibility is the counterpart of protein digestibility in the energy digestibility function.
i Fixed at a value of 90 g/kg for diet processing reasons. Assumes starch digestibility is 50%.
j Required feed intake = DE-total / Diet Digestible Energy.
k Expected FCR = Required feed intake / Expected growth.
l Digestible protein density arbitrarily assigned within practical regimes.
m Defined using optimal DP:DE.

**Defining feed and nutrient demands**

Using the energy demand based elements of the factorial model it becomes possible to estimate the volumes of any theoretical feed formulation applied to that model. Based on the digestible energy density of any theoretical feed, a ration will be allocated to satisfy the TED. This will then equate to a unit of feed (g/fish/d) that is required to be fed. As was also detailed in the previous section the application of an appropriate protein content of that feed can also be estimated for any fish size and any feed energy density. Therefore the process of defining key feed specifications can be satisfied on an iterative basis (Table 1 and 2).

To examine the implications of different feed design strategies a range of different feed specifications were designed based on either a single diet approach to production or a successional specification plan (Glencross, 2006). This later option being the application of changing dietary protein to energy specifications with increasing fish size:

1. The first option was based on a diet of 450 g/kg digestible protein and 15.0 MJ/kg of digestible energy.

2. The second option (which is that typically used by progressive barramundi farmers in Australia) is based on a series of three or four diets varying in digestible protein from 450 g/kg to 360 g/kg and diet digestible energy densities from 15.0 MJ/kg to 21.5 MJ/kg (Glencross, 2008).

Each of these specifications was altered based on nominal fish weight ranges of 10-200g, 200-1000g, 1000-2500g and greater than 2500g. The optimal protein to energy ratio was also maintained within appropriate limits within these size ranges. The different diet specifications are given in Table 3.

By applying these different feed strategies across the full production range for barramundi (10g to 3000g) it becomes possible to use the model to examine the total feed use and also total protein and lipid use with any given strategy.
Table 3. Feed formulations and composition as applied to the raw material demand modelling. Diets in the A column are formulated based on only fishmeal, wheat and fish oil being options. Diets in the B column are formulated with all raw materials as options.

<table>
<thead>
<tr>
<th>FEED Formulations</th>
<th>A</th>
<th>B</th>
<th>A</th>
<th>B</th>
<th>A</th>
<th>B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fish meal ($1500/tonne)</td>
<td>69.1%</td>
<td>28.8%</td>
<td>62.4%</td>
<td>32.0%</td>
<td>55.3%</td>
<td>32.0%</td>
</tr>
<tr>
<td>Fish oil ($1500/tonne)</td>
<td>3.3%</td>
<td>0.4%</td>
<td>14.2%</td>
<td>6.1%</td>
<td>23.1%</td>
<td>10.8%</td>
</tr>
<tr>
<td>Wheat ($300/tonne)</td>
<td>27.6%</td>
<td>13.7%</td>
<td>23.4%</td>
<td>14.1%</td>
<td>21.6%</td>
<td>14.1%</td>
</tr>
<tr>
<td>Poultry Offal Meal($1000/tonne)</td>
<td>32.6%</td>
<td>25.9%</td>
<td>19.4%</td>
<td>19.1%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lupin meal ($500/tonne)</td>
<td>23.7%</td>
<td>15.8%</td>
<td>12.9%</td>
<td>7.9%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Canola oil ($1500/tonne)</td>
<td>0.4%</td>
<td>6.1%</td>
<td>10.8%</td>
<td>11.8%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Formulation Cost ($/tonne)</td>
<td>$1,168</td>
<td>$964</td>
<td>$1,219</td>
<td>$1,069</td>
<td>$1,240</td>
<td>$1,124</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>FISH SIZE RANGES</th>
<th>10-200g</th>
<th>200-1000g</th>
<th>1000-2500g</th>
<th>&gt;2500g</th>
</tr>
</thead>
<tbody>
<tr>
<td>FISH SPECIFICATIONS</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dry matter (g/kg)</td>
<td>900</td>
<td>900</td>
<td>900</td>
<td>900</td>
</tr>
<tr>
<td>Protein (g/kg)</td>
<td>500</td>
<td>450</td>
<td>400</td>
<td>400</td>
</tr>
<tr>
<td>Lipid (g/kg)</td>
<td>100</td>
<td>200</td>
<td>280</td>
<td>300</td>
</tr>
<tr>
<td>Gross Energy (MJ/kg)</td>
<td>18.0</td>
<td>20.0</td>
<td>23.0</td>
<td>23.0</td>
</tr>
<tr>
<td>Digestible Protein (g/kg)</td>
<td>450</td>
<td>400</td>
<td>360</td>
<td>360</td>
</tr>
<tr>
<td>Digestible Energy (MJ/kg)</td>
<td>15.0</td>
<td>18.0</td>
<td>21.0</td>
<td>21.5</td>
</tr>
</tbody>
</table>

**Defining raw material demands**

Because of this process of satisfying the energy and protein demands, this modelling approach also allows for co-application of estimation of raw material demands. Based on that the volume of a specific feed used can be estimated, different formulations can be applied and the net utilisation of individual raw materials examined on both a discrete and cumulative basis.
To examine one application of this, for each of these different diet specifications two different feed formulations were designed based linear least-cost formulations based on either of a series of raw material constraints/options:

1. Only resources available: fishmeal, fish oil, wheat and a vitamin and mineral premix.
2. A range of raw materials available, including: fishmeal, fish oil, wheat, lupin kernel meal, canola meal, poultry offal meal, canola oil and a vitamin and mineral premix (Glencross, 2010).

Comparisons of fishmeal use were made based on the application of the different diet specifications and their constraints on diet formulations. This was done with both the conservative raw materials range (option 1) and with known viable alternatives in Australia (option 2). Options both with and without fish oil replacements were also examined based on the replacement of 50% of the fish oil with canola oil (Turchini et al., 2009). Similarly the application of a “finisher-diet” was also examined for fish >2500g and comparisons made on the effect that such a strategy would have on the net savings in fish oil use and the theoretical fatty acid composition of the fish based on the principle of fatty acid dilution (Glencross & Turchini, 2010).

Results

Iterative feed design modelling

The iterative feed design model, as used in this study demonstrated that as the energy density of the diet increased, the response of the model was to reduce the required ration. But despite the reduced ration there was still and need to maintain the protein demand. Therefore as diet energy density increased, the required protein concentration in the diet also increased. For example, the optimal predicted protein concentration for each diet, within a specific fish size, increased with increasing energy density of the diet (Table 1 and 2).

With increasing fish size there was also a declining demand for digestible protein (DP) in terms of the g/MJ consumed (Figure 3). This relationship is independent of dietary digestible
energy (DE) density. However, there were also different temperature based responses for each relationship and these can be described by the following equations:

\[ y_{25} = 50.37 \times \text{live-weight}^{0.1275}, \quad R^2 = 1.0000 \]
\[ y_{30} = 55.691 \times \text{live-weight}^{-0.1466}, \quad R^2 = 0.9996 \]
\[ y_{35} = 70.515 \times \text{live-weight}^{-0.1549}, \quad R^2 = 0.9999 \]

The relationships between digestible protein: digestible energy demand (g/MJ) observed at 25°C and 30°C were very similar. At 35°C there was a substantially higher DP:DE demand at all fish sizes. Based on these relationships a 100 g fish will require 28.5 g DP / MJ DE at 30°C, but the same fish will require 34.7 g DP MJ\(^{-1}\) DE at 35°C (Table 1). By contrast a 1000 g fish will require 20.2 g DP MJ\(^{-1}\) DE at 30°C, but the same fish will require 24.1 g DP MJ\(^{-1}\) DE at 35°C (Table 2).

Figure 3. Optimal digestible protein to digestible energy ratio with varying temperature and fish weight. At higher temperatures a higher protein to energy ratio is clearly noted and this is required to off-set the increase in maintenance protein requirements and a reduced partial efficiency of utilisation that occurs at these higher temperatures.

Nutrient and raw material demand modelling

The feed demand modelling underpinning the raw material demands was based on the predicted energy demands for barramundi grown to 3000g at a uniform temperature of 30°C.
Therefore the amount of feed that needs to be fed varies with the digestible energy density of the feed being used. As feed digestible energy density increased, the amount required to be fed to the fish declines. With the successional feed (SF) strategy there were a series of increases in diet digestible energy and as such a series of steps in reduction in feed ration allocation were observed relative to the standard 50-10 strategy (Figure 4). Using the 50-10 strategy a theoretical FCR of 1 : 1.37 is possible in growing the fish from 10g to 3000g. Using the SF strategy this FCR over the same size range is 1 : 1.03.

Figure 4. Total feed demand depending on feed plan (50-10, constant feed specification) or successional plan during a full production cycle for barramundi.

With the use of the SF strategy a significant decline in protein use through the production cycle is achieved (Figure 5). This reduction is equivalent to a total protein intake of 2051 g/fish for the 50-10 strategy and 1304 g/fish for the SF strategy, a difference of 747 g/fish over the production cycle. However a significant increase in lipid use occurs with the SF strategy. Using the SF strategy an equivalent lipid intake of 772 g/fish is needed. In comparison with the 50-10 strategy only 410 g/fish of lipid is required. Therefore the proportional increase in lipid use with the SF strategy is far greater than its proportional decrease in protein use, with the protein ratio (50-10:SF) being 157% and the lipid ratio (SF:50-10) being 188% (i.e. 100% being maintained as the same).
Figure 5. Total protein and lipid demands depending on feed plan (50-10, constant feed specification) or successional plan during a full production cycle for barramundi.

With the raw material use in the absence of alternatives, the SF strategy resulted in a significant decline in fishmeal use through the production cycle (Figure 6). This reduction was equivalent to a total fishmeal use of 2834 g/fish for the 50-10 strategy and 1858 g/fish for the SF strategy, a difference of 976 g/fish over the production cycle. However a significant increase in fish oil use occurs with the SF strategy. When using the SF strategy an equivalent fish oil use of 627 g/fish is needed. In comparison with the 50-10 strategy only 135 g/fish of fish oil is required, a difference of 492 g/fish. Therefore the proportional increase in fish oil use with the SF strategy is far greater than its proportional decrease in protein use, with the fish meal ratio (50-10:SF) being 153% and the fish oil ratio (SF: 50-10) being 463% (i.e. 100% being maintained as the same).

Figure 6. Total fish meal and fish oil demands depending on feed plan (50-10, constant feed specification) or successional plan during a full production cycle for barramundi. Diets assume no raw material substitution.

Figure 7. Total fish meal and fish oil demands depending whether diets consider some raw material substitution or not. In both cases the successional feed strategy is used during a full production cycle for barramundi.

With raw material use with the use of alternatives, the SF strategy a significant decline in fishmeal use through the production cycle 1329 g/fish vs 1858 g/fish (Figure 7). With the 50-10 strategy a total fishmeal use of 2834 g/fish is required without use of alternatives and with the use of alternatives a total fish meal use of 1181 g/fish is required over the production cycle. However a significant increase in fish oil use occurs with the SF strategy. Using the SF strategy an equivalent fish oil use of 627 g/fish is needed, while with a 50% fish oil replacement strategy using this feed strategy a total of 314 g/fish are used (Figure 8). In comparison the 50-10 strategy only uses 135 g/fish of fish oil. A 50% fish oil replacement option with this feed strategy would use 68 g/fish, a difference of 68 g/fish. With the use of a fish oil replacement strategy with use with the SF strategy, followed by a finisher-diet the total fish oil use would 381 g/fish, which is about 61% that used in the SF strategy without fish oil replacement, but is still 282% that used in the 50-10 strategy without the use of fish oil replacement.

Based on the principle of growing a barramundi to 2100 g/fish using a 50% dilution of canola and fish oil it was estimated that a fatty acid composition of the fish at that stage based on a fish of 9% lipids equivalent to 189 g lipid/fish would be a fatty acid content of 30% saturates (SFA), 40% monounsaturated fatty acids (MUFA), 15% polyunsaturated fatty acids (PUFA) and 15% long-chain polyunsaturated fatty acids (lcPUFA). If the fish are then fed a diet rich

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in fish oil (30% SFA, 40% MUFA and 30% lcPUFA) then the subsequent gain in biomass to 3000g bodyweight of fish at 10% lipid equivalent to 285 g/fish would constitute a gain of 96 g of lipid/fish and the final diluted fatty acid composition would be 30% saturates (SFA), 40% monounsaturated fatty acids (MUFA), 10% polyunsaturated fatty acids (PUFA) and 20% long-chain polyunsaturated fatty acids (lcPUFA). This then shows that such a “finisher-diet” strategy would increase the lcPUFA content of a 100g fillet of fish from 1500 mg/100 g of fillet to 2000 mg/100 g of fillet.

![Graph showing total demands for fish oil with varying feed strategy.](image)

Figure 8. Total demands for fish oil with varying feed strategy. Considered are the successional feed strategy (with no fish oil replacement), a successional feed strategy (with 50% fish oil replacement) ending with a finisher-diet and the 50-10 strategy (with no fish oil replacement) during a full production cycle for barramundi.
Discussion

This paper aimed to examine the application of an advanced growth and energetic model for barramundi in considering the effect of physiological challenges (heat-stress) on diet specification designs and also the implications to variations in raw material demands (fish meal and oil replacement) throughout the animals production cycle. As such this paper examines just a small sample of the potential applications of such models and proposes their application as a means of exploring notional concepts prior to empirical experiments.

Implications of high water temperatures on modelling

The assembly of a revised factorial model for barramundi that considers not only a broader temperature range, but more notably those effects of upper temperature limits on total energy and protein demands is a novel step forward in nutritional modelling. Earlier models had assumed that energy demands largely mimicked those of growth demands (Lupatsch et al., 1998; Glencross, 2008; Dumas et al., 2009). This was based on the observation that the somatic energy demand associated with weight gain was a disproportionate part of total energy demand. However, the recent empirical evidence underpinning the present advanced model demonstrates that above thermal optimal ranges there is a decoupling of the animals regulation of energy and protein demands (Bermudes et al., 2010; Glencross & Bermudes, 2010a;b). This is underpinned by observations that the allometric relationships with energy/protein maintenance demands and protein/energy utilisation efficiencies alter as a consequence of these high temperatures. It is hypothesised that this is due to a dramatic increase in protein losses associated with the animals mucosal and gut epithelial linings (the animals primary contact points with its environment) as temperature stress increases. As a consequence this decoupling dramatically magnifies the maintenance energy demands at high temperatures and reduces the ability of the animal to effectively use dietary energy to supply its maintenance and growth needs. The net result is an increase in total energy demand at higher temperatures once the animal begins to undergo heat-stress. Most of the change in demand was also observed to be through a disproportionate increase in protein losses and reduction in protein utilisation efficiencies (Glencross & Bermudes, 2010a; b).
By applying these changes to the iterative feed design elements of the model it was possible to examine the implications of these physiological challenges on total protein and energy demands and how these might best be met through a modified dietary strategy. In doing so, in the present study a series of iterative diets were developed using the model based on varying fish size and temperatures. Through this approach it was demonstrated that within a temperature and fish size class, as the energy density of the diet increased, the response of the model was to reduce the required ration, but despite the reduced ration there was still and need to maintain the protein demand. Therefore as diet energy density increased, the required protein concentration in the diet also increased and these effects are consistent with other similar models (Lupatsch et al., 1998; Glencross, 2008). With increasing fish size though a declining demand for digestible protein (DP) in terms of the g/MJ consumed was observed and this can be seen as a clear inverse power-function relationship (Figure 5). It was also noted that this relationship was independent of dietary digestible energy (DE) density, being manifested as an ideal ratio being between DP and DE only. However, different temperatures induced different optimal DP : DE ratios. The relationships between digestible protein : digestible energy demand (g/MJ) observed at 25°C and 30°C were very similar. At 35°C though there was a substantially higher DP : DE demand at all fish sizes. Based on these relationships it is suggested that a 100 g fish will require 28.5 g DP / MJ DE at 30°C, but the same fish will require 34.7 g DP / MJ DE at 35°C (Table 1). By contrast a 1000 g fish will require 20.2 g DP / MJ DE at 30°C, but the same fish will require 24.1 g DP / MJ DE at 35°C (Table 2). So in essence what is observed at elevated temperatures is a clear increase in the demand for digestible protein within a certain digestible energy density. This observation is consistent with independent empirical data from a study examining an increase in DP : DE in diets fed to juvenile barramundi at either 30C or 37C. In that study significant benefits were observed from the elevated DP : DE ratio, not only at the higher water temperature but also at the lower one (Glencross and Rutherford, 2010). Both observations are in retrospect easily explainable by the outcomes of the model.

Examining the application of raw material demands

Using the model to consider total feed demands and by association changes in nutrient and raw material demands, is another potential application for such models. In this paper we have explored the implications of the use of a successional feed strategy, with the use of increasing
energy density of feeds throughout the production cycle, against that of using a single high-protein diet fed throughout the entire production cycle. This successional feed strategy is a strategy similar to that being currently promoted by the feed industry and accepted by most modern fish production industries, principally because of its ability to minimise FCR and also introduce efficiencies in feed costs (Glencross, 2006).

The modelled feed use of a successional feed strategy has a significant impact on reducing total feed demand throughout the production cycle. It was shown that a reduction in FCR from the 50-10 single diet strategy of 1.37 : 1 to 1.03 : 1 should be theoretically possible. Evidence with large fish (>1000g) fed such diets certainly supports this notion as possible (Glencross et al., 2008). The successional feed strategy also minimises the protein use in producing the animal and indeed this has been one of the main drivers for pursing this strategy. Especially as cost sensitivities with aquaculture feeds remain tightly linked to fish meal prices. Therefore one of the key roles of the successional feed strategy is to minimises the amount of fish meal required in such formulations due the ability of these lower-protein diets to accommodate cheaper low-protein alternative raw materials. But the strategy also demands a much higher inclusion of fish oil (or other oils) and as such the feeds tend to be more expensive on a formulated cost basis relative to a higher-protein low-oil diet (Table 3). However, the benefits gained with a reduction in FCR using the successional feed strategy more than offset this cost and it becomes a more cost-effective strategy and certainly on a protein basis, a more sustainable strategy.

In contrast though this successional feed strategy is shown to be very lipid “expensive” and causes a dramatic increase in the demand for oil resources. As such it increases the pressure being placed on fish oil resources. Indeed recent commentary has rightly pointed out that it is the fish oil resource sustainability issue that is a more pressing one that the fish meal issue (Naylor et al., 2009). One strategy to minimise fish oil use has been the blending of fish oils with plant or terrestrial animal oils (Turchini et al., 2009). However, even with a 50% replacement of fish oil with a plant oil (e.g. canola oil) the successional strategy is still a greater consumer of lipids than that achieved using the higher-protein diet throughout the full production cycle. The use of an alterative oil source in replacement of lcPUFA rich fish oil also results in a significant shift in fatty acid profile of the animal, which has prompted the consideration of the use of “finisher-diets” (Glencross et al., 2003).
The use of the model however shows that the benefits of using finisher-diet on satisfying product quality demands are nominal at best. Without the use of a finisher-diet a fillet of barramundi would have 1500 mg lcPUFA/100 g fillet. With the use of a finisher diet strategy this would increase to only 2000 mg lcPUFA/100 g fillet (only a 25% increase at best). With recommended intakes of lcPUFA being ~500 mg/d (NHMRC, 2006) then to achieve a weekly intake of lcPUFA of 3500 mg a person would have to consume either 230 g of fillet with the fish-oil-substitution option or 180 g of fillet with the fish oil finisher diet option. A fish fed a diet (either the successional plan or the 50-10 plan) where only fish oil was used would probably result in a fillet lcPUFA content of 3000 mg/100g of fillet. This would then require consumption of about 120 g of fillet. If one assumes that the standard meal would be around 200 g of fish meat, then this constitutes only a few meals of fish what ever the feed lipid strategy (and in some cases less than one meal). And this point needs to be considered in view that with this species, this flesh lipid content is relatively lean. A fish such as an Atlantic salmon has a lipid content almost twice that of barramundi (Glencross et al., 2008).

So despite that it can be modelled that the impact of fish oil replacement in aquaculture feeds has limited impact in terms of total lcPUFA contribution to the human diet, this practice has only “bought-us-time” in the resolution of total lcPUFA resource allocation to the aquaculture feeds sector. At best we have doubled the volume of effective oil source we can use via the practice of dilution (assuming we only dilute fish oils 50%). With the present growth rate of the aquaculture feed sector and current use of lcPUFA rich oils by this sector we will still need to identify and produce new lcPUFA sources in the near-term future (Naylor et al., 2009).

**Conclusion**

This paper provides several examples that show that the key value in nutritional models lies in their ability to explore ideas and assist in the refinement of experimental designs for further empirical study. However, like all models, the studies presented here have the potential to be useful, but are still only an estimation of the potential that might be achieved. Therefore caution must be applied when applying such features of the model or applications derived from it, without first verifying their predictions with empirical data. And ideally such models

should be used as a “screening” method to explore ideas and concepts prior to their validation using empirical experiments.
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