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EFFECT OF VARIED DAILY FEEDING INTENSITIES ON GROWTH PERFORMANCE AND FEED UTILISATION OF SOUTH AFRICAN DUSKY KOB (ARGYROSOMUS JAPONICUS) JUVENILES REARED IN A RECIRCULATING AQUACULTURE SYSTEM

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ABSTRACT

Several aquaculture studies have concluded that feed constitutes the heaviest cost in aquaculture enterprises. Consequently, researchers continue to investigate strategies that can lower feed costs such as the use of protein rich plant sources and insect meals to partially or completely substitute the expensive fishmeal in aquafeeds. However, this approach has often faced numerous challenges such as anti-nutrients in plant protein sources that limit their use and suboptimal polyunsaturated fatty acids profile in terrestrial protein sources such as insect meal. Precision feeding is a strategy that could also reduce aquafeed feed costs and enhance the economic sustainability of aquaculture. There is discordance regarding the feeding intensity that should be used for fish such as dusky kob. The current study was designed to compare the effect of three feeding intensities (2.8 %, 3.5 % and 4 % of body weight (BW)) on growth performance and feed utilisation of juvenile dusky kob (*Argryrosomus japonicus*). Dusky kob juveniles were randomly and evenly allocated to 18 tanks with a volume of 465 L. Each feeding intensity was randomly allocated to six tanks carrying 66 fish each. The average temperature and dissolved oxygen were 25°C and 5.5-6.0 mg/l, respectively. Commercial dusky kob diet was offered twice daily at the rate of 2.8%, 3.5% and 4% of body weight (BW). A feeding intensity × fish age (weeks) interaction effect was noted on fish feed intake (P<0.05) but not on weight gain, feed conversion ratio (FCR), and specific growth rate (SGR). The overall weight gain after six weeks of feeding was similar across the three feeding intensities (P>0.05). The 2.8% BW feeding rate promoted 1.06%/day SGR as compared to 1.21%/day and 1.48%/day for 3.5% and 4.0% BW feeding rates, respectively. Similar weight gains and FCR recorded across the three feeding intensities may suggest that the lowest feeding intensity (2.8% BW) should be used for dusky kob feeding to reduce feeding costs.

Key words: Aquaculture, Feed, Dusky kob, Rations, Growth performance, FCR, RAS, SGR

INTRODUCTION

Feed availability and management, water quality parameters and feed nutrient quality are important in maintaining a successful aquaculture enterprise [1, 2, 3, 4]. Fish feed has been widely accepted as the single most expensive component in running a viable fish farming business. However, most farmers in Africa tend to source low quality feed to keep their farms running due to high cost at the expense of sustainable fish growth, health, and welfare [5, 6]. Fish reared on low quality diets are more susceptible to diseases because nutrient deficiency compromises their immune system [7]. On the other hand, some farmers overfeed the fish with the intention of maximising fish growth and attain market fish size in a relatively short period [8]. During this period of overfeeding, water quality deteriorates due to large quantities of feed refusals and faeces in the water column. Excess nutrients in the tanks or ponds give rise to toxic ammonia gas that can be detrimental to fish [9]. It is, therefore, important to calculate the correct feeding intensity of the desired fish of interest to ensure an economically and environmentally appropriate feeding strategy. Furthermore, excessive feeding to expedite fish growth may not be a popular strategy as that may lead to more costs associated with labour [10].

A good association between fish growth and feeding frequency has been reported [10, 11]. In addition, it was demonstrated that one might not necessarily feed daily to obtain maximum fish growth [12]. A study by De Silva and Anderson [13] indicated that excessive feeding intensities have no influence on growth and may worsen the feed conversion ratio (FCR). Increase in feed intake may result in poor FCR, as an increased feed ingestion rate may result in reduced gut evacuation time, which will subsequently lower nutrient absorption and ultimately the loss of nutrient in faeces [14].

Collect [15] designed a study to develop a rearing protocol for South African juvenile dusky kob. The study concluded that fish fed a restricted ration produced a better FCR (42%) than those fish under ad libitum feeding and recommended that feeding rate for juvenile dusky kob should not exceed 3.85% of body weight (BW) per day. Considering the feed cost and maintaining a good water quality, earlier studies [16, 17, 18, 19, 20] capped the feeding intensity at 2.8% BW per day with satisfactory dusky kob growth performance. However, there are still questions regarding this restricted feeding strategy, as other studies recommended the use of higher feeding intensities such as 3.8 % BW or feeding to satiation [21]. The current study provides a comparative assessment of the effect of three feeding intensities (2.8, 3.5, and 4 % of BW) on feed intake, feed utilisation, and growth performance of dusky kob juveniles. These feeding intensities were mainly

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selected to test the hypothesis that feeding at more than 2.8% does not significantly improve growth performance of dusky kob juveniles with reference to feed costs.

MATERIALS AND METHODS

Study site

The feeding trial was performed at the Aquaculture Research Station of the Department of Forestry, Fisheries and the Environment (DFFE) in Cape Town, South Africa. A recirculating aquaculture system (RAS) that was previously described by Madibana *et al.* [20] was used. The system consisted of 18 tanks, which were connected to multiple filtration components that included the biological filter, sand filter, foam fractionator/protein skimmer and the de-gassing chamber. The RAS was connected to heat pumps (25 °C), UV lights (55 W) and the air-lines that provided dissolved oxygen (6 mg/l) to the system.

Experimental fish

The South African Animals Protection Act, 1962 (Act 71 of 1962) was adhered to when handling the live fish. Ethical clearance to conduct the feeding trial was granted by the Cape Peninsula University of Technology's Animal Ethics Committee (CPUT/AEC 2018/01). A total of 1188 juvenile dusky kob (25.72 ± 0.85 g average) were sourced from a dusky kob hatchery in Eastern Cape Province, off the east coast of South Africa. The transportation of the fish from the hatchery to the Research Aquarium was conducted as described by Madibana and Mlambo [18]. The fish were randomly distributed to 18 replicate tanks (66 fish in each) and allowed to acclimatise for a period of two weeks.

Feeding and sampling strategies

A commercial dusky kob feed (Western Cape, South Africa) (Table 1) was used in the current study to evaluate three feeding intensities (2.8% (control), 3.5%, and 4.0% BW per day). The 2.8% feeding intensity was successfully used in previous studies [18, 20] and produced satisfactory results and the two additional feeding intensities (3.5 and 4%) are new, chosen to test if there is a need to increase the feeding rate for dusky kob. After the acclimatisation period, fish were not offered feed for 24 hours for complete gastric evacuation. Each feeding intensity was randomly allocated to six tanks and offered to fish for a period of six weeks. Due to the dusky kob photophobia tendency, no artificial light was used on the RAS, but fish were exposed to minimal sunlight coming into the Aquarium during the day (0800h – 1800h). From each of the 18 tanks, 40 fish were randomly sampled weekly and the body weight was determined (Viper SW 15, Mettler Toledo, South Africa).

Calculations

Equation 1:

Specific growth rate (SGR) (%/day) was calculated using the formula below [22]:

$$\text{SGR (\%)} = \left[\frac{(\text{Logn Final fish weight.} - \text{Logn Initial fish weight.})}{\text{Time interval (days)}}\right] \times 100$$

Where, Log_n Final fish weight = natural logarithm of the final weight and Log_n Initial fish weight = natural logarithm of the initial weight.

Equation 2:

Feed utilisation (expressed by feed conversion ratio (FCR) was calculated using the formula below [22]:

$$\text{FCR} = \frac{\textbf{Feed consumed (g)}}{\textbf{Weight gained (g)}}$$

Statistical analysis

Data was tested for normality using the NORMAL option in the Procedure Univariate statement before being subjected to analysis of variance. Measurements from each tank were averaged before analysis to obtain one value representing the experimental unit (tank). The growth performance (SGR and fish weight gain) data were analysed using repeated measures analysis [23] according to the following model:

$$Y_{ijk} = \mu + D_i + W_j + DW_{ij} + E_{ijk} \, ,$$

Where, Y_{ijk} = dependant variable, μ = population mean, D_i = effect of diet, W_j = effect of week, DW_{ij} = interaction effect of diet and week, and E_{ijk} = random error associated with observation ijk, assumed to be normally and independently distributed.

For parameters where week × diet interaction effects were not significant, a one-way ANOVA of overall values was done using the general linear models (GLM) procedures in SAS [21] according to the following linear model:

$$Y_{ij} = \mu + D_i + E_{ij} \, ,$$

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where, Y_{ij} = dependant variable, μ = population mean, D_i = effect of diet, and E_{ij} = random error associated with observation ij, assumed to be normally and independently distributed. For all statistical tests, significance was declared at P<0.05. Least squares means were compared using the probability of difference (pdiff) option in the lsmeans statement of SAS.

RESULTS AND DISCUSSION

Feed intake, growth performance and feed utilisation

The performed repeated measures analysis showed that the interaction between fish age (weeks) and feeding intensity was significant for fish feed intake (P<0.05) suggesting that as the fish grew the ranked effect of three feeding intensities on feed intake changed. The ranking (2.8% BW > 3.5% BW> 4.0 BW) of the three feeding intensities in terms of their effect on feed intake was similar in weeks 1, 2, 5 and 6 (Table 2). In these weeks, 2.8% BW promoted lower feed intake compared to 3.5% BW and 4.0% BW, which promoted similar feed intake (P>0.05). In week 3 of the trial, 2.8% BW promoted lower feed intake while 4.0% BW promoted higher feed intake. During the same week, 3.5% BW promoted similar feed intake as both 2.8% BW and 4.0 BW (P>0.05). In week 4, 3.5% BW promoted the highest feed intake, while 2.8% BW promoted the lowest feed intake. In week 4, 4.0% BW promoted similar feed intake as 2.8% BW and 3.5% (P>0.05). It was expected that feed intake of the group fed lower ration would be lower as compared to the groups fed higher rations, hence the 2.8% BW fed group exhibited lower feed intake throughout the trial. However, since most fish are ectotherms, a slight change in optimum temperature for dusky kob would affect fish growth performance [16]. This is because a decrease or increase due to changes in the ambient temperature is well known in the aquaculture industry to play a key role on fish metabolic rate (appetite) [24] that could ultimately affect feed intake.

Based on the repeated measures analysis, there was no interaction effect of fish age (weeks) and feeding intensity on weight gain (P>0.05). In weeks 1, 2, 4, and 6, fish across the three feeding intensities had similar weight gains (P>0.05). However, in weeks 3 and 5, weight gain differed significantly (P<0.05) across the feeding intensities. The group fed 4.0% BW had the least weight gain average of 0.04 g in week 3 as compared to the two other feeding intensities that resulted in fish weight gain average of over 4 g. In week 5, fish fed 3.5% BW gained the least weight average of 0.92 g, which was inferior to the groups fed 2.8% BW and 4.0 BW that gained average of 3.94 g and 4.78 g, respectively. The overall weight gain after six weeks of feeding was similar across the three feeding intensities (2.8%

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BW = 16.18g, 3.5% BW = 17.70g, 4.0% BW = 18.67g) (P>0.05). Overall standard length gain was significantly different from one feeding ration to the other (P>0.05) (Table 3).

In weeks 1, 2, 3, 4, and 6 the different feeding intensities promoted similar weight derived (P>0.05) SGR (Table 4). In week 5, fish fed 3.5% BW and 4.0% BW had significantly different weight derived SGR, however both feeding intensities produced similar SGR (P>0.05) to the fish fed 2.8% BW. Table 5 shows similar length derived SGR from initial feeding week to the last. Water parameters have a greater influence on the feeding behaviour of fish, which in turn would translate into growth performance [24]. Different SGR between fish fed 3.5% BW and fish fed 4.0% BW in week 5 may be due to water quality changes as indicated earlier or simply due to other groups showing superior feed utilizing capability as the fish entered week 5 [25]. No interaction effect between fish age (weeks) and the feeding intensity on FCR (P>0.05) (Table 6) was noted on weekly basis. A similar (P>0.05) weekly and overall FCR was recorded for the three feeding intensities.

The group fed on the low ration (2.8 % BW) had a lower feed intake as compared to the other two groups which were fed on higher feeding intensities. This observation was consistent with the observation by Saether and Jobling [26], who reported that feeding lower restricted rations of 0.25 and 0.38 % BW per day as compared to the control feeding of 1 % BW per day in turbot (Scophthalmus maximus L.), resulted in significantly lower feed intake. These observations suggest that, the lower the feeding ration, the more the likelihood of recording lower fish feed intake and vice versa.

The current study showed that feeding juvenile dusky kob restricted rations of 2.8 %, 3.5 % and 4 % of body weight daily, resulted in similar overall weight gain after 42 days of feeding. Collett [15] also reported no weight gain difference when feeding juvenile dusky kob 3.41 and 3.6 % BW restricted rations and satiation feeding in a 56-day feeding trial. Different feeding regimes of 3.4, 3.8, 4.2, 4.6, and 5 % BW per day and feeding to satiation at various frequencies per day also produced similar juvenile dusky kob weight gain after 42 days [27]. Feeding 2-year-old (1.7 kg) mulloway (dusky kob) at three feeding intensities of 0.6 %, 0.8 % and 1.0 % BW per day resulted in similar weight gains after 30 days of feeding [28]. All these studies suggest that feeding juvenile dusky kob at lower rates (2.8 % in the current study) or as high as 5 % BW per day resulted in similar fish weight gain. Study with Asian seabass (Lates calcarifer) fry also revealed similar final weight between groups fed once, twice, thrice, and four times a day over five weeks [29]. If indeed dusky kob weight gain does not depend on feeding frequency, this

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suggests that if protein content is maintained at an optimal level as suggested by Daniel [30], the lower 2.8 % BW per day feeding should be further encouraged. Even though, feeding frequency does not seem to make a difference in dusky kob weight gain, a study by Richie *et al.* [10] suggested that there is a positive relation between growth and feeding frequency in Nile tilapia.

The highest feeding intensity (4 % BW per day) in the current study, promoted similar SGR as to the other two lower daily feeding intensities (2.8 % BW and 3.5 % BW) in five weeks, but one, which agrees with Collett [15] who observed similar SGR of dusky kob groups fed restricted diets (3.41 and 3.6 % BW per day) and the group fed to satiation. It has been reported previously that dietary energy ingested between maintenance ration, when no growth occurs, and maximum ration, when maximum growth occurs, is the scope for growth [31]. Hepher [31] further indicated that, the higher the ration the higher the growth rate. However, based on the SGR results from the current study, the aforementioned relationship between food ration and growth rate was not observed as the current different feeding rations exhibited mostly similar fish SGR during the feeding trial period. This suggests that lower and higher feeding rates provide similar maintenance energy and growth was independent of the feeding rate in dusky kob.

The FCR from the current study was not affected by feeding intensity throughout the six weeks. These observations are consistent with Collett [15] who reported that feeding intensities of 3.8 % BW and below, produced FCR that was 42% better than when fish were fed to satiation. Kaiser *et al.* [21] also concluded that feeding juvenile dusky kob restricted daily rations (3.85 % BW and lower) achieve lower FCR, than feeding fish to satiation. This suggests that for sustainable dusky kob farming in South Africa, fish should be fed restricted as opposed to satiation feeding, as there are no added benefits associated with satiation feeding in terms of growth and FCR. Jobling [14] suggested that when feed is readily available, fish eat more than they necessarily require and use the diet less efficiently. An increase in feed intake may lead to a reduction in FCR because gut evacuation time will be reduced, limiting the utilisation of essential nutrients while more nutrients are lost in faeces. In addition, feeding causes a 2 to 3-fold post-prandial increase in metabolic rate compared to the pre-prandial level. This suggests that large rations associated with satiation feeding, extend the time that fish metabolic rate remains elevated above the pre-prandial level leading to a high-energy requirement and reduced FCR. Indeed, European sea bass (*Dicentrarchus labrax*) fed at 4.5 % body weight per day had significantly poor FCR compared to those fed at 3.1 % body weight per day [32]. The current study corroborates the position that lower feeding intensities, such as the 2.8 % employed in this study and in all other previous studies [16,17,18,19,20], promotes similar or better FCR than higher intensities.

CONCLUSION

Restricted feeding at lower 2.8% BW per day, as employed in this study, showed that it has potential to lower production costs by reducing the quantity of feed used while maintaining satisfactory growth rates similar to feeding 3.5% BW and 4.0% BW per day. This feeding approach has the potential to improve economic and environmental sustainability of dusky kob aquaculture and thus attract more players into this industry.

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DECLARATION OF COMPETING INTEREST

The authors declare that they have no competing interests.

INSTITUTIONAL REVIEW BOARD STATEMENT

The study was conducted according to the guidelines of the Declaration of Helsinki and approved by the Animal Research Ethics Committee of the University of Mpumalanga (FANS17; 21 May 2020).

DATA AVAILABILITY STATEMENT

The data presented in this study are available on request from the corresponding author. The data are not publicly available because the diet formulae used in this study are proprietary brands.

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Table 1: Gross and chemical composition of the commercial dusky kob diet used in the current study

[1]Ingredients (%)	
Fishmeal 66	30.00
Maize	18.71
Soya oilcake	10.00
Full fat soya 58	8.50
Blood meal 90	6.00
Wheat gluten	10.91
Pork meal 28	8.17
Fish and poultry oil	5.36
Vit/Min Premix	2.35
Proximate composition (calculated values)	
Dry matter (DM) (%)	86.80
Ash (% DM)	0.62
Moisture (% DM)	13.20
Crude protein (% DM)	46.58
Crude fat (% DM)	12.15
Digestible energy (MJ/kg DM)	17.38
Metabolisable energy (MJ/kg DM)	15.75
Chitin (% DM)	0.50
Essential amino acids (g/100 g DM)	
Arginine	3.54
Threonine	1.66
Methionine	0.71
Valine	2.47
Phenylalanine	1.93
Isoleucine	1.98
Leucine	3.42
Histidine	1.34
Lysine	2.92
Tryptophan	5.40
Non-essential amino acids (g/100 g)	
Alanine	2.45
Tyrosine	1.22
Proline	2.45
Serine	1.85
Aspartic acid	3.36
Glutamic acid	6.17
Glycine	2.68

[1]ingredients, Fishmeal, wheat gluten, pork meal, maize, fish and poultry oil, and vit/min premix were all sourced from Johannesburg, South Africa. Blood meal was sourced from Germany. Soya oilcake and full-fat soya were sourced from Argentina. Poultry meal was sourced from Netherlands

Table 2: Effect of different feeding intensities on feed intake of juvenile dusky kob (g/fish/day)

	Intensity[1]				
	Ration28	**Ration35**	**Ration40**	**SEM[2]**	**Significance[3]**
Week 1	5.66[a]	6.52[b]	6.07[b]	0.14	*
Week 2	6.55[a]	7.45[b]	7.71[b]	0.19	*
Week 3	6.81[a]	7.63[a,b]	8.16[b]	0.20	*
Week 4	7.59[a]	8.80[b]	8.15[a,b]	0.18	*
Week 5	7.81[a]	9.92[b]	9.13[b]	0.26	*
Week 6	8.58[a]	10.14[b]	10.47[b]	0.25	*

[1]Intensity: Ration28 = 2.8% body weight; Ration35 = 3.5% body weight; Ration40 = 4.0% body weight
[a,b]Means along the same row with different superscripts denote significant differences ($p < 0.05$).
[2]SEM: standard error of the mean
[3]Significance: * = $p < 0.05$

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Table 3: Effect of different feeding intensities on overall fish standard length (cm) and weight gain (g) of the juvenile dusky kob during a six-week feeding period

Intensity[1]	Length gain	Weight gain
Ration28	1.66[a]	16.18[a]
Ration35	2.05[a,b]	17.70[a]
Ration40	2.30[b]	18.67[a]
SEM[2]	0.11	0.92
Significance[3]	*	NS

[1]Intensity: Ration28 = 2.8% body weight; Ration35 = 3.5% body weight; Ration40 = 4.0% body weight
[a,b]Means along the same row with different superscripts denote significant differences ($p < 0.05$)
[2]SEM: standard error of the mean
[3]Significance: NS = $p > 0.05$; * = $p < 0.05$

Table 4: Effect of different feeding intensities on weekly weight derived specific growth rate (%/day)

	Intensity[1]				
	Ration28	**Ration35**	**Ration40**	**SEM[2]**	**Significance[3]**
Week 1	0.32 [a]	0.32[a]	0.57[a]	0.06	NS
Week 2	0.12 [a]	0.06[a]	0.14[a]	0.03	NS
Week 3	0.26[a]	0.34[a]	0.01[a]	0.06	NS
Week 4	0.07[a]	0.29[a]	0.26[a]	0.05	NS
Week 5	0.22[a,b]	0.05[a]	0.33[b]	0.05	*
Week 6	0.07[a]	0.16[a]	0.18[a]	0.07	NS
Overall	1.06[a]	1.21[a,b]	1.48[b]	0.07	*

[1]Intensity: Ration28 = 2.8% body weight; Ration35 = 3.5% body weight; Ration40 = 4.0% body weight
[a,b]Means along the same row with different superscripts denote significant differences ($p < 0.05$).
[2]SEM: standard error of the mean
[3]Significance: NS = $p > 0.05$. * = $p < 0.05$

Table 5: Effect of different feeding intensities on weekly length derived specific growth rate (%)

	Intensity[1]				
	Ration28	Ration35	Ration40	SEM[2]	Significance[3]
Week 1	0.08[a]	0.14[a]	0.19[a]	0.02	NS
Week 2	0.03[a]	0.02[a]	0.03[a]	0.01	NS
Week 3	0.08[a]	0.11[a]	0.00[a]	0.02	NS
Week 4	0.11[a]	0.10[a]	0.07[a]	0.03	NS
Week 5	0.00[a]	0.01[a]	0.14[a]	0.03	NS
Week 6	0.01[a]	0.06[a]	0.04[a]	0.02	NS
Overall	0.31[a]	0.39[a,b]	1.48[b]	0.02	*

[1]Intensity: Ration28 = 2.8% body weight; Ration35 = 3.5% body weight; Ration40 = 4.0% body weight
[a,b]Means along the same row with different superscripts denote significant differences ($p < 0.05$).
[2]SEM: standard error of the mean
[3]Significance: NS = $p > 0.05$. * = $p < 0.05$

Table 6: Effect of different feeding intensities on weekly feed conversion ratio (FCR)

	Intensity[1]				
	Ration28	Ration35	Ration40	SEM[2]	Significance[3]
Week 1	2.31[a]	5.19[a]	1.90[a]	1.23	NS
Week 2	2.13[a]	10.64[a]	22.87[a]	5.58	NS
Week 3	2.46[a]	0.69[a]	0.14[a]	2.32	NS
Week 4	1.70[a]	0.79[a]	4.09[a]	2.66	NS
Week 5	2.6[a]	8.54[a]	3.65[a]	1.55	NS
Week 6	10.57[a]	1.96[a]	1.86[a]	3.57	NS
Overall	2.88[a]	2.92[a]	2.77[a]	0.16	NS

[1]Intensity: Ration28 = 2.8% body weight; Ration35 = 3.5% body weight; Ration40 = 4.0% body weight
[a]Means along the same row with different superscripts denote significant differences ($p < 0.05$).
[2]SEM: standard error of the mean
[3]Significance: NS = $p > 0.05$

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