DOI: 10.1111/are.13657

ORIGINAL ARTICLE

WILEY Aquaculture Research

Feeding behaviour and growth of lumpfish (*Cyclopterus lumpus* L.) fed with feed blocks

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Funding information
Norges Forskningsråd, Grant/Award Number: 256199

Abstract

The aim of this project was to investigate if lumpfish can be fed using specially designed feed blocks instead of regular fish feed pellets. Two studies were performed. In Part I different designs of feed blocks were introduced and fish observed with underwater cameras to record feeding behaviour. Results indicate that lumpfish require feed blocks with grooves in order to graze from them and that the acclimation period is relatively short (2–4 hr) before the fish will use them as a feed source. In the second part of the project two duplicate groups of lumpfish with an initial mean (±*SD*) weight of 125.4 ± 45.7 g were individually weighed and randomly distributed into six 3.5 m^3 circular flow-through tanks with 45 fish in each tank. Fish in three tanks were fed using feed blocks with grooves and fish in three tanks were fed using a regular commercially available lumpfish extruded feed. Both groups received a daily feeding rate of 2% body/weight. From day 14 onwards, fish fed with marine pelleted feed had a significantly higher mean weight compared to fish fed with feed blocks. Although not significant, the condition factor was higher in the feed block group during the study period. Results from this study show that lumpfish will readily graze from feed blocks if they are presented in a way that allows them to do. In addition, the acclimation period required before the fish will utilize them appears to be short thus potentially allowing for their use in commercial salmon cages.

KEYWORDS
feed blocks, feeding, growth, lumpfish

1 | INTRODUCTION

The biological control of sea lice in Atlantic salmon farming through the use of "cleaner fish" has recently become a feasible alternative due to the increased occurrence of resistance towards medical treatments in salmon lice, *Lepeophtheirus salmonis* (Igboeli, Fast, Heumann, & Burka, 2012; Torrissen et al., 2013), the reduced public acceptance of chemotherapeutic use in food production, and the urgent need for an effective and sustainable method of parasite control in Atlantic salmon aquaculture (Boxaspen, 2006; Denholm et al., 2002; Treasurer, 2002). As a cold-water cleaner fish, the common lumpfish (*Cyclopterus lumpus*) has been suggested and initial results are very promising with up to 93%–97% less sea lice infection (adult female lice) in sea pens with lumpfish (Imsland, Reynolds, Eliassen, Hangstad, Foss, et al., 2014; Imsland et al., 2014a,b; Imsland et al., 2015a,b). Lumpfish in sea pens are strongly opportunistic and the

*Equal authorship between: Imsland and Reynolds fish do not restrict themselves or rely on a single food source if others are present (Imsland et al., 2015a) and a very high proportion of the lumpfish in salmon sea pens are found with pellets in their stomach (Imsland, Reynolds, Eliassen, Hangstad, Foss, et al., 2014; Imsland et al., 2015a). Thus it is becoming increasingly evident that the supplementary feeding of cleaner fish deployed within commercial salmon pens is necessary (Leclercq, Davie, & Migaud, 2014; Leclercq, Graham, & Migaud, 2015) to maintain the nutritional condition, welfare and efficacy of the biological controls over the Atlantic salmon grow-out cycle typically lasting 18–22 months. Therefore, a feed source adapted to the species grazing feeding habit and to the salmon net-pens rearing environment has first to be developed. Presently, lumpfish stocked in commercial salmon pens are being fed extruded pelleted feed usually delivered from feed automats around the edge of the cages. This method has clear limitations as lumpfish have been shown to be opportunistic feeders and readily exploit available food sources (Imsland, Reynolds, Eliassen, Hangstad, Foss, et al., 2014; Imsland et al., 2014a,b). Such a food source which may be predictable both spatially and temporally may result in most of the lumpfish maintaining position around the periphery of the cages and reduces their potential for grazing sea lice. Therefore, there is a need to develop a feed source adapted to the species grazing feeding habit and to the salmon net-pens rearing environment. Feed blocks have been used in salmon cages stocked with wrasse species (Leclercq et al., 2015) and can be positioned in areas of the cage where the wrasse will be in closer proximity to the salmon thus potentially enhancing grazing potential.

Practical feed for lumpfish within salmon net-pens should combine a manufactured base providing a complete and standardized nutrient profile, biosecurity and ease of procurement with high water stability for distribution as a grazing substrate. Furthermore, this methodology has the potential to facilitate lumpfish feeding in sea cages and to allow the monitoring of feed intake to safeguard health, welfare and sea lice grazing activity. As a first step in achieving those goals the objectives of this study was to evaluate different designs of feed blocks for feeding of lumpfish and to compare growth properties using pellets and feed blocks.

2 | MATERIALS AND METHODS

2.1 | Experimental fish and conditions

The lumpfish were produced from fertilized eggs from Senja Akvakultursenter AS, Tromsø. The eggs were incubated at 9–10°C and the juveniles were initially fed with Gemma Micro (150–500 μm, Skretting, Norway). After 30 days, the juveniles were fed with 500–800 μm dry feed pellets (Gemma Wean Diamond, Skretting, Norway). The fish were vaccinated with ALPHA JECT Marin micro 5 (Pharmaq AS, Oslo, Norway) on 7 November 2016. The health status of the fish was assessed immediately prior to transfer to Gifas, Inndyr, Nordland, Norway in early January 2017. Health status was assessed by PCR screening for Vibrio species, atypical furunculosis, pasteurella, moritella, pancreas disease (PD), infectious pancreatic necrosis (IPN), viral hemorrhagic septicemia (VHS), Nodovirus and amoebic gill disease (AGD). From November 2016 to January 2017 the juveniles were fed a high protein low fat marine feed (Skretting, Amber Neptune ST) using Van Gerven 7/L feeding automats (the Netherlands). A 50% mixture of 1.5 mm and 2 mm pellets was used during this period.

All tanks were supplied with full salinity sea water pumped from 70 m depth at a temperature of between 5.4 and 8.5°C and oxygen saturation was maintained above 80% during the whole experimental period. Water temperature and oxygen concentration was recorded in each tank for both studies using a Handy Polaris 2 probe (OxyGuard International A/S).

2.2 | Part I—Feeding behaviour study

One week prior to the start of the trial (16 January 2017), three groups of lumpfish with an initial mean (±*SD*) weight of 15.0 ± 2.0 g were established. The fish were individually weighed and randomly distributed into nine 3.5 m^3 circular flow-through tanks with 100 fish in each tank. At trial start, the pelleted feed (Skretting, Amber Neptune ST) was withdrawn and feed blocks introduced. The chemical composition of the feed blocks was: 50.1% protein; 10.3% lipid; 12.6% carbohydrate; 1.7 fibre % and 20.8% moisture. The energy content of the feed block feed was 17 MJ/kg. The composition of the pelleted feed was: 55% protein; 15% lipid; 11% carbohydrate; 2.5% and 7% moisture. The energy content of the feed pellets was 20.7 MJ/kg.

All feed blocks were weighed prior to placement in the tanks to ensure that the fish received a feeding rate of 2% body weight $(BW)^{-1}$. Once the feed blocks were in place, the fish were observed on a daily basis with underwater cameras to record feeding behaviour. The cameras were activated every 2 hr over a 10 hr period. Different designs and deployment methods were assessed to optimize grazing behaviour. A total of six different designs and deployment methods were assessed during the study period (Figure 1). Each feed block design was tested in three replicate tanks for 5 days starting with design 1–3 followed by design 4–6 after a recovery period of 7 days (fish feed with pellets). Selection of tanks for each design was done randomly. Fish were not fed pelleted feed 2 days prior to testing of each feed block design type. After start of each feed block design trial, the fish were observed for evidence of eating from the feed blocks at 2-hourly intervals over a 10-hour period for 5 days. After the deployment period, the blocks were removed if no feeding activity was evident and the fish were given a marine pelleted feed (Skretting, Amber Neptune ST) using Van Gerven 7/L feeding automats (the Netherlands) to recover. After the 5 day trail for design 1–3 all the fish were individually weighed to record the biomass in each tank and thus regulate the feeding rate prior to designs 4–6 being tested.

During the testing of the different designs feeding response was scored using a frequency distribution table (Table 1). Response was scored from a scale of 0 to 7. Zero equals no evidence of feeding and 7 that more than 50% of the fish in the tank were observed grazing from the blocks. Fresh feed blocks were placed in the tanks every day for each design.

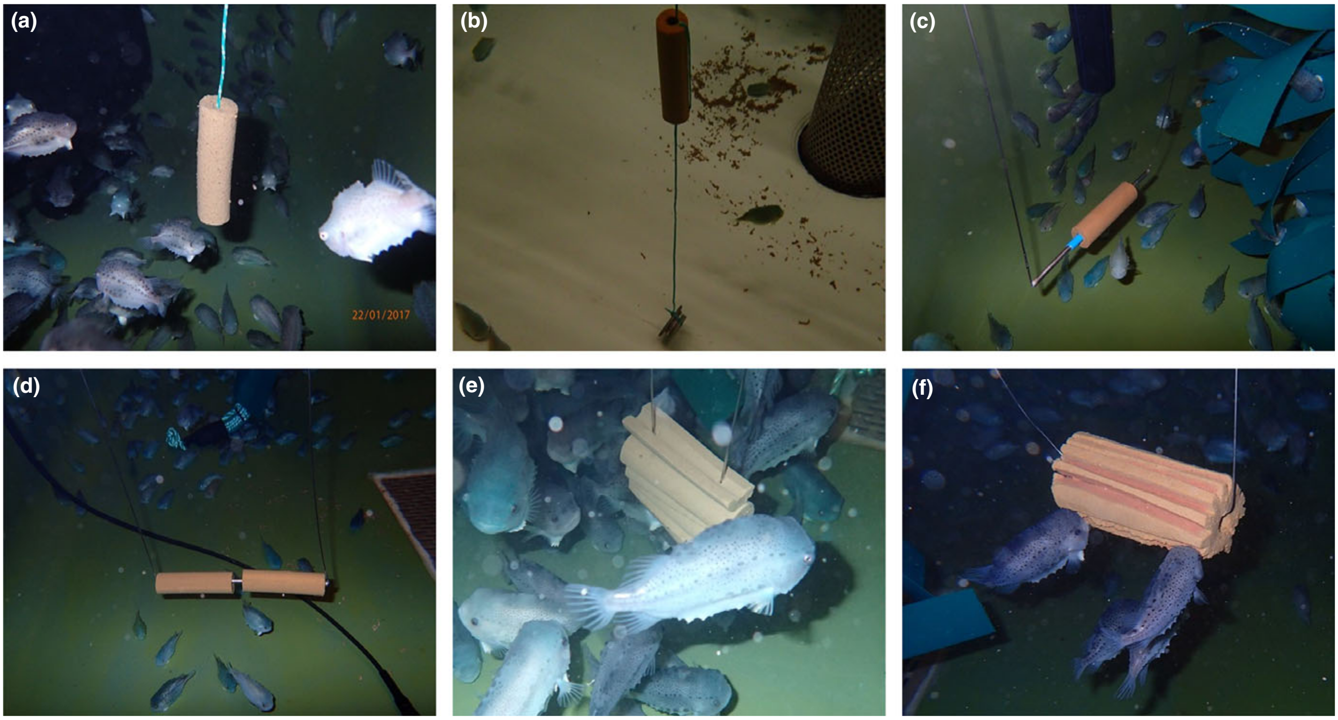

FIGURE 1 Different designs and deployment methods used during Part I of the study. (a) Feed block with smooth surface deployed suspended in the water column; (b) feed block suspended on a rope and weighed at the bottom of the tank; (c) feed block suspended with 3 mm metal wire; (d) two feed blocks suspended with 3 mm metal wire; (e) feed blocks cut in half and 3 mm wire passed through each one to form a stack and (f) feed block with grooves cut longitudinally suspended with 3 mm metal wire [Colour figure can be viewed at wileyonlinelibrary.com]

TABLE 1 Frequency distribution of recorded feeding response behaviour used during the study period in Part I. There were 100 fish in each experimental tank

Score	Response
0	No response to the feed blocks. Fish are distributed and no fish near the blocks.
1	Fish swimming towards feed blocks or hovering around them. No evidence of grazing.
2	Periodic grazing by less than 10 fish
3	Regular grazing by 10–19 fish
4	Regular grazing by 20–29 fish
5	Regular grazing by 30–39 fish
6	Regular grazing by 40–49 fish
7	Regular grazing by over 50 fish

2.3 | Part II—Growth study

One week prior to the start of the trial (May 2017), two triplicate groups of lumpfish with an initial mean (±*SD*) weight of 125.4 ± 45.7 g were established from the original population. The fish were individually weighed and randomly distributed into six 3.5 m^3 circular flow-through tanks with 45 fish in each tank ($N = 270$). On 8 May 2017, feed blocks were introduced to three tanks and pelleted feed was stopped 3 days prior to feed block deployment. The design deployed was based on observations from Part 1 of the study. Design number 6 (longitudinal blocks with grooves, Figure 1f) was used during the study period. Feed blocks were weighed prior to placement to ensure sufficient feed was available to maintain a daily feeding rate of 2%/BW. The three other tanks received the same daily feeding rate using a commercially available lumpfish extruded feed (Biomar lumpus 2.2 mm) using Van Gerven 7/L feeding automats. The fish in all tanks were individually weighed every 2 weeks for 41 days.

2.4 | Feed block design and placement in tanks

Each individual feed block was 26 × 100 mm with a 10 mm hole through the centre. The surface structure as stated was either smooth or grooves cut in them. The grooves were 3–5 mm wide. The blocks are extruded under cool temperature and were relatively dense although they have a small amount of softness.

In order to increase potential access the placement of the feed blocks was either at:

- A minimum of 50 cm from then side of the tank and 40 cm from the bottom of the tank
- A minimum of 50 cm from then side of the tank and 70 cm from the bottom of the tank

The blocks were also placed randomly around the tank allowing for the greatest distance between them. Each block was weighed to calculate the number required based on the biomass; so for each tank, 4 blocks were placed at 2 different depths at the start increasing to 5/6 blocks towards the end of the study.

2.5 | Growth

All fish in Part II were individually weighted and their total length measured every second week for 42 days. Specific growth rate (SGR) of individual lumpfish was calculated according to the formula of Houde and Schekter (1981):

$$\mathrm{SGR} = (e^{g} - 1) \times 100 \tag{1}$$

where g = (ln (W_2)-ln (W_1)/(t_2-t_1) and W_2 and W_1 are weights on days t_2 and t_1 respectively.

Condition factor (K) of individual lumpfish in Part II (calculated at each weighing interval) was defined as:

$$\mathrm{K} = 100 * W = L^3 \tag{2}$$

where W is the weight (g) of the fish and L the corresponding total length (cm).

2.6 | Statistics

All statistical analyses were conducted using Statistica™ 12.0 software. A Kolmogorov–Smirnov test (Zar, 1984) was used to assess for normality of distributions. The homogeneity of variances was tested using the Levene's F test (Zar, 1984). Possible differences in feeding behaviour (Part I), mean weights, condition factor and growth rates (Part II) between the experimental groups were tested with two-way nested analysis of variance (ANOVA), where replicates are nested within feeding types. Significant differences revealed in ANOVA were followed by Student–Newman–Keuls (SNK) post-hoc test to determine differences among experimental groups. A significance level (α) of 0.05 was used if not stated otherwise. In cases with non-significant statistical tests, power (1–β) analysis was performed in Statistica using $\alpha = .05$.

3 | RESULTS

3.1 | Feeding behaviour (Part I)

Designs 1–4 (Figure 1a–d) elicited only weak responses from the fish with no direct evidence of grazing with only occasional bite marks noted on the feed blocks (Table 2) with average score between 0.24 and 0.4 (no evidence of grazing). Design 5 (Figure 1e) elicited a stronger response with an average response (±SE) of 2.68 ± 0.37 (between 10%–20% of the fish grazing). Design 6 (Figure 1f) elicited the strongest (two-way nested ANOVA, $F_{5,25} = 9.12$, $p < .001$) and most frequent feeding response of all six designs with an average response per replicate tank of 4.56 ± 0.07 (between 20%–30% of the fish grazing of feed blocks).

3.2 | Growth (Part II)

No fish died during the experimental period. The overall initial mean weight (SD) was 125.4 (45.7) g and did not differ (three-way ANOVA, P [Power (1–β)] > 0.7, Figure 2a) between the two feed type groups. From day 14 onwards, fish fed with marine pelleted feed had a significantly higher mean weight compared to fish fed with feed blocks (SNK post-hoc test, $p < .05$). The condition factor (K) in both groups varied between 2.6 and 4.2 (Figure 2b). The mean K tended to be higher in the feed block group throughout the trial period being significantly higher at the termination of the trial (SNK test, $p < .05$, Figure 2b). With the exception of the first period the mean specific growth rate (SGR, Figure 2c) was higher in the pellet group (SNK test, $p < .05$).

4 | DISCUSSION

4.1 | Feeding behaviour

In this study six designs of feed blocks were assessed with design 5 (feed blocks cut in half and stacked together) and particularly design

TABLE 2 Results from behavioural observations recorded for six of the feed block designs and deployment methods. The scoring was based on a constructed frequency distribution (Table 1). Numbers are given as sum for both replicate tanks for each design tested

Day																									
	1					2					3					4					5				
	Observations within day																								
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Design	Response																								
1	0	0	0	0	0	0	0	0	0	2	0	2	0	0	0	0	2	2	0	2	0	0	0	0	2
2	2	2	0	0	0	2	0	0	0	2	0	0	0	2	2	0	0	2	2	2	0	0	0	2	0
3	0	0	0	0	0	0	0	2	2	0	0	0	0	0	0	2	2	0	0	0	2	0	0	0	2
4	2	2	2	2	0	0	0	0	2	0	0	2	0	0	0	2	0	0	0	2	0	0	2	2	0
5	2	4	2	4	4	2	6	4	6	6	8	8	10	4	6	8	10	4	2	0	6	12	8	6	2
6	0	10	12	12	10	8	14	10	8	6	14	10	8	6	10	14	14	4	2	10	12	10	14	8	2

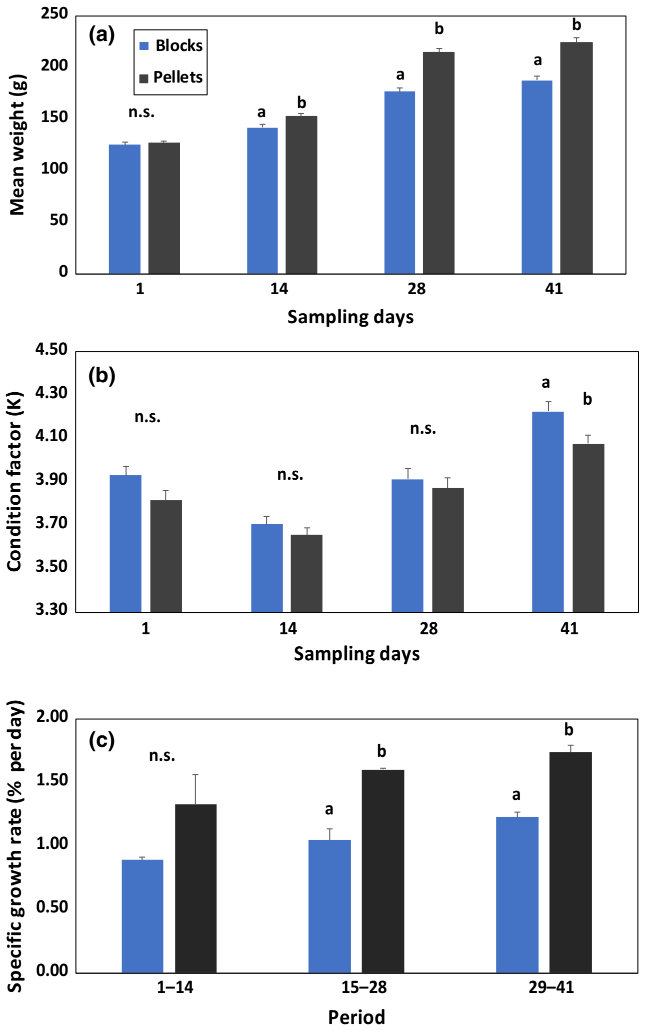

FIGURE 2 (a) Mean weight (*g*); (b) Condition factor (K) and (c) Specific growth rates (SGR) of lumpfish fed either feed blocks or extruded pelleted feed. Values represent means ± *SE*. Different letters indicate significant differences (SNK test, $p < .05$); n.s., not significant [Colour figure can be viewed at wileyonlinelibrary.com]

6 (feed blocks with grooves cut longitudinally) eliciting the strongest feed response. Fish started responding to the presence of the feed blocks after only 2 hr and readily grazed from them throughout the assessment period. It was expected that the fish would readily graze from the feed blocks given their strong opportunistic feeding behaviour as seen in previous studies (Imsland, Reynolds, Eliassen, Hangstad, Foss, et al., 2014; Imsland et al., 2014a,b;Imsland et al., 2015a) and their need to attach themselves to suitable substrates (Imsland et al., 2015b) to conserve energy. Current data imply that the design of the feed block is vital if it to be used as a food source for lumpfish. Almost no feeding response was observed with several types of smooth surface feed blocks suspended in the water column. In contrast the expected feeding response was observed once the block was compromised of edged or grooved surface. The fish exploited the presence of these edges to graze and at closer examination reveals that their mouths are rounded with no teeth that jut outwards (Figure 3c). This may result in them being unable to graze from a smooth surface, whereas wrasse species have protractile mouths, usually with separate jaw teeth that jut outwards (Wainwright, Alfaro, Bolnick, & Hulse, 2005; Figure 3a–b) allowing them to graze easily from smooth surfaced feed blocks. The smooth feed blocks used in the present study have been used in salmon cages stocked with wrasse species in Scotland to good effect (James A. Mackie, pers. comm), whereas current data imply that smooth surface feed blocks are not suitable for lumpfish due to the species anatomical structure of their mouth (non-protractile).

Initially there was a concern that the reason why the fish were showing very little response to the first designs (no. 1–4) was that there may have been a palatability issue for the fish. However, given that lumpfish grazed frequently on subsequent designs highlighted that palatability was not the reason for the poor responses from the first attempted designs and deployment methods but rather an inability to graze from the blocks largely due to their smooth surface.

FIGURE 3 Mouth of (a) Goldsinny wrasse (*Ctenolabrus rupestris*) (Modified from: http://light.rockfishing.co.uk); (b) Ballan wrasse (*Labrus bergylta*) (photo, Camilla Utgård, Institute of Marine Research, Bergen, Norway) and (c) Lumpfish (photo, Patrick Reynolds)

The overall aim is to deploy feed blocks in commercial salmon cages. To achieve this goal, the lumpfish must be able to access them and readily graze from them. One advantage of using such a feed type is that they can be deployed in areas in the cage where they would be in close proximity to the salmon and thus enhance their lice grazing potential. Presently, most commercial farms using lumpfish fed them with pelleted feed (Imsland et al., 2015a; Powell et al., in press) which usually is delivered from the edge of the cage using automatic feeders. This limits their ability to deliver feed away from the edges of the cage and thus encourage lumpfish to colonize these areas due to feed availability. By using feed blocks, lumpfish can be encouraged to occupy areas of the cage where the salmon are predominantly found, thus increasing the interaction between salmon and lumpfish.

A prerequisite for successful use of lumpfish is that they need attachment areas to rest when not actively grazing or looking for food. This is particularly important when the fish are first introduced into cages. In the wild, juvenile lumpfish are typically found among algae, both attached and free floating during their first year of life (Ingólfsson, 2000; Ingólfsson & Kristjánsson, 2002), but are also found attached to substrates (Moring, 1989). In general, members of the family Cyclopteridae use their ventral adhesive disc to adhere to rocks, vegetation and other available substrates (Brown, 1986; Moring, 1989). Small juvenile lumpfish (c. 15–20 g) are routinely deployed in salmon cages and previous studies have shown that they require areas of attachment to rest when not foraging for food (Imsland, Reynolds, Eliassen, Hangstad, Foss, et al., 2014; Imsland et al., 2015b). The lack of suitable attachment sites are likely to result in increased stress thus increasing the probability of disease particularly bacterial agents. Ongoing research in our research group is focussed on using feed blocks in combination with artificial substrates thus providing stand-alone units which can be deployed in any area inside commercial cages. These "lumpfish stations" provide suitable habitats for the fish which in turn may enhance their lice grazing efficacy.

4.2 | Growth

The growth rates observed during this study were similar to growth rates from previous studies (Imsland et al., 2015a,b). However, growth rates were significantly higher for fish fed with pelleted feed compared to fish fed with feed blocks even though both feed types were offered a daily feeding rate of 2% /BW based on biomass gain. This difference in growth performance may be attributed to the lumpfish not eating all of the offered feed blocks. It was also observed that as the fish grazed on them, small pieces would break off and sink to the bottom of the tank. Some of these fragments would be eaten by fish near the bottom whilst some were lost through the flow-through system thus reducing the feed available to the fish. Alternatively the higher growth in the group fed pelleted feed could be linked to higher energy and lipid content of pellets (17 vs. 21 MJ/kg). However, it should be noted that high growth is not an aim for lumpfish used as cleaner fish. Imsland et al. (2016) found that small lumpfish (initial size approx. 20 g) have a higher overall preference for natural food items, including sea lice, compared to larger conspecifics. This makes slow to moderate and uniform growth of lumpfish more desirable than fast growth for its optimal use as cleaner fish in salmon aquaculture.

There were variations in K throughout the study period with fish fed feed blocks having higher K values at the termination of the trial. Lumpfish exhibit a high degree of opportunistic feeding behaviour as seen in previous studies (Imsland, Reynolds, Eliassen, Hangstad, Foss, et al., 2014; Imsland et al., 2014a,b, 2015a,b) and individual fish exhibit different food selection choices within populations. These selection differences can greatly affect body condition due to differences in the nutritional quality of different food sources. The variation in K values observed in this study may in part be attributed to food choice selection between individuals within both experimental groups. In addition, feeding hierarchies (Imsland, Folkvord, & Nilsen, 1998; Imsland, Jenssen, Jonassen, & Stefansson, 2009) may have been established within both groups of fish resulting in some fish being less able to compete for food when available as this behaviour was observed on several occasions throughout the study, particularly with fish fed feed blocks. The presence of dominant fish is not such a surprise with fish fed feed blocks as this food source is available in single persistent locations within the tanks whilst pelleted feed is delivered by automatic feeders and is more spread throughout the tank thus resulting in less chance of dominant hierarchies forming. To prevent dominant fish controlling areas where feed blocks are deployed in commercial cages it will be necessary to establish several feeding stations within the cage.

It is important that lumpfish used as cleaner fish in salmon cages have access to a regular food source particularly in winter time when naturally occurring food items become scarce. This food source is vital to maintain healthy and robust populations. Pelleted feed is normally used to feed these fish in commercial cages, however, its availability is generally limited to the periphery of the cage and lumpfish which have regular access to pelleted feed would also compete for salmon feed once they have grown larger (Imsland et al., 2015a). Feed blocks offer the advantage that they can be deployed anywhere in the cage and used as a maintenance rather than a self-sustaining food source.

5 | CONCLUSION

Results from this study show that lumpfish will readily graze from feed blocks if they are presented in a way that allows them to. In addition, the acclimation period required before the fish will utilize them appears to relatively short, thus potentially allowing for their use in commercial salmon cages.

ACKNOWLEDGMENTS

The authors would like to thank the technical staff at GIFAS aquaculture research station at Inndyr for valuable assistance prior to and during the experimental period and Meløy Videregående Skole for providing access to their marine hall. Financial support was given by the Research Council of Norway (LUSINFER 256199). There is no conflict of interest in relation to this study.

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How to cite this article: Imsland AK, Reynolds P, Hangstad TA, et al. Feeding behaviour and growth of lumpfish (*Cyclopterus lumpus* L.) fed with feed blocks. *Aquac Res*. 2018;49:2006–2012. https://doi.org/10.1111/are.13657