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MANEJO ALIMENTAR DE JUVENIS DE PEIXE ENXADA (*Chaetodipterus faber*)

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Universidade Federal do Rio Grande
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Dissertação apresentada como parte dos requisitos para a obtenção do grau de mestre em Aquicultura no programa de Pós-Graduação em Aquicultura da Universidade Federal do Rio Grande.

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DEDICATÓRIA

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1. RESUMO GERAL

O peixe enxada *Chaetodipterus faber* é um excelente candidato para o desenvolvimento da aquacultura marinha, devido à algumas características como: fácil adaptação ao confinamento, boas taxas de crescimento e carne de boa qualidade. Como no caso de qualquer espécie aquícola emergente, o sucesso da criação do peixe enxada vai depender, em parte, do atendimento das suas necessidades nutricionais e do correto manejo alimentar. Dentre as diferentes práticas alimentares que podem maximizar a eficiência alimentar e o crescimento estão a taxa de arraçoamento e a frequência alimentar. Nesse estudo foi avaliada a performance de crescimento de juvenis do peixe enxada (3,60 ± 0,03 g, média ± DP) alimentados com 3, 5, ou 7% da biomassa (3% BW, 5% BW, 7% BW), em uma única alimentação (1x) ou divida igualmente entre três refeições (3x). Ganho em peso, taxa de crescimento específico (TCE), conversão alimentar e consumo de alimento foram afetados tanto pela taxa de arraçoamento como pela frequência alimentar. O ganho em peso e a TCE aumentaram com a taxa de arraçoamento, sendo superior nos grupos alimentados 3x ao dia do que os alimentados 1x ao dia. Os peixes alimentados com maiores taxas de arraçoamento acumularam mais ` no corpo em detrimento da umidade, proteína e teor de cinzas, mas a composição da carcaça não foi afetada pela frequência alimentar. O presente estudo sugere que o manejo alimentar do peixe enxada criado em sistema intensivo de recirculação, pode ser otimizado quando a alimentação é ofertada na proporção de 5-7% da biomassa/dia em três alimentações diárias com 7% da biomassa/dia há um crescimento maior, embora menos eficiente.

2. GENERAL ABSTRACT

The Atlantic spadefish *Chaetodipterus faber* is an excellent candidate for aquaculture development, because of its ability to adapt well to confinement, good growth rates and excellent flesh quality and consumer appeal, but success will depend on the identification of proper feeds and feeding regimens for this species. Among the different management practices that may maximize feeding and growth efficiency, feeding rate and feeding frequency have a crucial function in determining overall feed intake, growth, and waste outputs of fish. Accordingly, we evaluated the growth performance of juveniles (3.60 ± 0.03 g, mean ± SE) fed at 3, 5, or 7% of body weight (3% BW, 5% BW, 7% BW), either in a single feeding (1×) or divided equally among three feedings (3×). Weight gain, specific growth rate, feed conversion ratio, and feed intake were significantly affected by both feeding rate and frequency. Weight gain and SGR increased significantly with feeding rate, and growth was generally greater and more efficient in the 3× groups than the 1× groups. Fish fed at higher feeding rates accumulated more lipid within the body at the expense of moisture, protein, and ash content, but carcass composition was unaffected by feeding frequency. We suggest that the growth of juvenile Atlantic spadefish may be optimized when fed at 5-7% BW / day in 3 daily feedings, with 7% BW / day yielding the greatest, albeit less efficient, growth.

3. INTRODUÇÃO GERAL

O setor pesqueiro mundial é movido pela acentuada demanda de organismos aquáticos na tentativa de acompanhar o acrescente aumento da população mundial, que praticamente mais que quadruplicou desde 1900, de 1,6 bilhões no final do século passado para mais de 7 bilhões atualmente (United Nations, 2011). Devido a essa demanda, muitas espécies de relevada importância comercial já atingiram seus limites pesqueiros (Pauly, et al., 2002).

Em meados dos anos 90, o setor pesqueiro tradicional atingiu uma quantidade de captura que parecia ser o máximo possível, com níveis entre 85 e 95 milhões de toneladas anuais, das quais menos de 60 milhões de toneladas eram destinadas ao consumo humano (FAO, 2009). Atualmente, o consumo mundial consiste em 131 milhões de toneladas/ano, ou 18,6 kg por pessoa/ano, e a produção aquícola é responsável por 60 milhões de toneladas/ano, (FAO, 2012). Considerando que até 2050 a população poderá atingir um patamar de 9 bilhões de pessoas (United Nations, 2009), em 2030 a demanda atendida pelo setor aquícola atingirá aproximadamente 160 milhões de toneladas de organismos aquáticos (FAO, 2002).

A maioria dos estoques pesqueiros está completamente explotado, sobreexplotado ou em processo de recuperação (Alder et al., 2008). A produção pesqueira também pode ser afetada por variações climáticas e oceanográficas (por exemplo, eventos de El Niño), principalmente na porção Leste do Oceano Pacífico. Essa região abrange países como Peru e Chile, onde a anchoita é um dos principais recursos comerciais, e é também um dos recursos mais vulneráveis a essas variações (FAO, 2011). No Nordeste, Noroeste e região Centro-Oeste do Oceano Atlântico, a produção pesqueira atingiu picos em 2000, 2001 e 2004, porém nos anos seguintes a captura decresceu 30, 23 e 13%, respectivamente (FAO, 2011). No ano de 2004 a produção pesqueira para as mesmas regiões citadas acima foi de 10, 2,4 e 3,4 milhões de toneladas (FAO, 2005), enquanto que no ano de 2000 foi de 10,9, 2,1 e 3,5 milhões de toneladas (FAO, 2001). O mesmo ocorreu no Mediterrâneo com uma redução de 12% em 2008 em comparação com o ano anterior (FAO, 2011).

Apesar dos valores da produção pesqueira não diminuírem drasticamente com o passar do tempo, os estoques estão em declínio. Segundo valores da FAO (2009), entre 2002 a 2006 a produção pesqueira manteve-se no patamar médio de 93 milhões de toneladas. Nos anos entre 2007 a 2009 essa mesma produção média consistiu em 90 milhões de toneladas (FAO, 2011), mesmo patamar que o último relatório da FAO (2012), três milhões de toneladas a menos do que nos anos anteriores.

O declínio dos estoques pesqueiros marinhos impulsionou o rápido crescimento da aquacultura (Naylor et al., 2000). Entre os anos 1987 e 1997, a produção global de organismos aquáticos mais que duplicou em termos de peso e valor, contribuindo para o fornecimento mundial de proteína animal de alta qualidade (FAO, 1999). De 2002 a 2006, a produção aquícola passou de 40,4 para 51,7 milhões de toneladas (FAO, 2009), e continuou crescendo, atingindo 55,1 milhões de toneladas em 2009 (FAO, 2011). Um terço desta produção está restrito a criações que utilizam farinha e óleo de peixe nas dietas, como por exemplo, peixes carnívoros marinhos (Tacon e Metian, 2009).

Esse setor da aquacultura, também denominado de piscicultura marinha de peixes carnívoros é uma prática controversa, devido ao fato de utilizar como matéria-prima de seus insumos a farinha e/ou o óleo de peixe, e também o uso de peixes inteiros como itens alimentares (Welch et al., 2010). Uma das principais críticas consiste no fato de que uma grande biomassa de peixe é utilizada apenas com a finalidade de produzir poucos organismos de topo de cadeia; um padrão que é considerado por muitos como insustentável para uma escala industrial e comercial (Allsopp et al., 2008; Pauly, 2009). Contudo, os níveis de farinha e óleo de peixe utilizados em dietas aquícolas diminuíram na última década, e o consumo global destas matérias-primas pelas indústrias do ramo da aquacultura também decresceu, em dietas para salmão, por exemplo, no ano 2000 o uso de farinha e óleo de peixe correspondia respectivamente a 33 e 23% dos ingredientes contidos na dieta. Já no ano de 2008 essa porcentagem caiu para 24 e 15%, respectivamente (Jackson, 2010). Isso ocorreu em resposta simultânea dos altos preços que estas matérias-primas atingiram e também ao melhoramento tecnológico e científico voltado para a fabricação de dietas com fontes de proteína de origem vegetal mais eficientes (Naylor et al., 2009).

Neste contexto global, a piscicultura marinha no Brasil, por mais que seja incipiente, deve ser fomentada de forma sustentável, buscando a preservação da biodiversidade e dos recursos marinhos da costa Brasileira, contemplando o ecodesenvolvimento da atividade por meio da pesquisa e a busca por novas espécies com potenciais para a aquacultura. Apesar de não haver uma produção brasileira oficial, o potencial para essa atividade é enorme devido à grande área de costa com distintos aspectos geomorfológicos e oceanográficos, possibilitando criações próximas e afastadas da costa; boas condições naturais e climáticas; vasta gama de espécies com potenciais aquícolas, como vermelhos (*Lutjanus* sp) (Watanabe, 2001), garoupas (*Epinephelus* sp) (Sanchez et al., 2007), robalos (*Centropomus* sp) (Souza-Filho e Cerqueira, 2003;

Cerqueira, 2005), tainhas (*Mugil* sp) (Okamoto et al., 2006; Miranda-Filho et al., 2010), corvinas da família Scianidae (Burkert, 1999), linguados (*Paralichthys* sp) (Sampaio e Bianchini, 2002; Sampaio et al., 2010), pampos da família Carangidae (Costa et al., 2008) e bijupirá (*Rachycentron canadum*) (Sampaio et al., 2010; Sampaio et al., 2011).

A atividade encontra-se em um momento oportuno, pois há grande interesse tanto do setor público e privado, quanto de universidades, de fomentar a produção de peixes marinhos por meio da produção de juvenis em laboratório, como é o caso do bijupirá. Essa espécie já possui um pacote tecnológico de reprodução, larvicultura e engorda, que apenas necessita ser adaptado e melhorado para cada região brasileira. Atualmente, universidades desenvolvem pesquisas visando à consolidação da produção dessa espécie, e há alguns empreendimentos pioneiros e experimentais. Dessa forma, ao curto ou médio prazo a criação de bijupirá estará estabelecida, e será responsável pelo desenvolvimento da produção de peixes marinhos do Brasil.

No entanto, é importante a diversificação de espécies criadas tanto para atender o mercado como também para evitar a disseminação de doenças que possam causar mortalidade total, resultando em grande perda econômica; além de degradar o ambiente (Buschmann et al., 2006; 2009). A criação de salmão no Chile é um exemplo negativo, que enfatiza a importância do planejamento inicial que inclui: a diversificação de espécies criadas, a determinação da capacidade suporte de cada região, o controle sobre o uso de antibióticos e a introdução de espécies exóticas (FAO, 2007).

Sendo assim, deve-se continuar o fomento ao desenvolvimento de pacotes tecnológicos para espécies com potencial, a fim de impulsionar o desenvolvimento e a produção da piscicultura marinha brasileira. Nesse aspecto, uma espécie aquícola emergente que possui aceitação de mercado e apresenta boas características de crescimento é o peixe enxada *Chaetodipterus faber*.

A Família Ephippidae é composta por 20 espécies de peixes onívoros, comprimidos lateralmente, marinhos e de água salobra (Randall, 1967), incluindo o peixe enxada (*Chaetodipterus faber*), único membro da Família presente no Atlântico Oeste (Robins e Ray, 1986). O ciclo de vida e a distribuição dessa espécie ainda não são completamente conhecidos (Ditty et al., 1994), mas é sabido que habitam águas costeiras desde o litoral norte americano até o sudeste do Brasil, incluindo o Golfo do México (Johnson, 1978) e Caribe (Burgess, 1978).

O peixe enxada utiliza uma grande variedade de ecossistemas dependendo da etapa do ciclo de vida, incluindo mangues e marismas, próximos a águas costeiras e recifes (Robins e Ray, 1986). Os juvenis são geralmente encontrados em estuários e águas abrigadas, enquanto os adultos têm preferência por águas costeiras e oceânicas (Johnson, 1978); frequentemente se reúnem em grandes cardumes nas águas costeiras, tornando um atrativo para a pesca esportiva (Bohlke e Chaplin, 1993; Robbins e Ray, 1986). Na natureza, podem atingir 91 cm de comprimento e 9 kg (Robbins e Ray, 1986).

O peixe enxada é um excelente candidato para o desenvolvimento da aquacultura (Tucker e Jory, 1991) devido à sua robustez física e resistência às doenças, a capacidade de boa adaptação ao confinamento, boas taxas de crescimento (Gaspar e Larez, 1984) e a tolerância à baixas temperaturas e salinidades. A ausência de comportamento agressivo ou canibal tanto durante a larvicultura como na engorda e sua carne de boa qualidade completam suas características de espécie emergente (Bohlke e Chaplin, 1993). Além do seu potencial aquícola como fonte de proteína, sua forma jovem possui importância ornamental (Gaspar, 1995) devido à sua morfologia e à sua impressionante coloração branca e preta.

Como no caso de qualquer espécie aquícola emergente, o sucesso da criação do peixe enxada vai depender, em parte, da correta identificação dos alimentos e manejos alimentares. Entre as diferentes práticas de manejo que podem maximizar a eficiência de alimentação e o crescimento, estão a taxa de arraçoamento e a frequência alimentar. Elas possuem uma função crucial para determinar o consumo total de ração, o crescimento e a eliminação de resíduos (Silva et al., 2007). Por exemplo, a alimentação esporádica e baixas taxas de arraçoamento podem resultar em redução do crescimento, bem como o aumento da fome, a agressão intra-específica e o aumento das taxas de canibalismo (Folkvord e Ottera, 1993). Como alternativa, a alimentação frequente e altas taxas de arraçoamento podem levar ao desperdício de alimento, deterioração da qualidade da água, reduzir a produção e aumentar os custos de produção (Booth et al., 2008). A otimização da frequência alimentar e da taxa de arraçoamento podem minimizar o desperdício de alimento, levando à melhoria da qualidade de água e maior homogeneidade nas classes de tamanho (Dwyer et al., 2002; Tucker et al., 2006), e também pode influenciar o uso de nutrientes nas dietas e na alimentação (Mihelakakis et al., 2002).

Estratégias alimentares e necessidades nutricionais são espécie-específicos, ou seja, podem variar conforme a espécie em questão, tamanho do peixe, fase do ciclo de vida, sistema de criação (Cho et al., 2003) e do tipo do alimento empregado (Chua e Teng, 1978, 1982). Estudos são realizados com o intuito de determinar o ótimo regime alimentar avaliando o crescimento, sobrevivência, consumo de alimento e composição corporal para diversas espécies, principalmente nas fases larval e juvenil (Dwyer et al., 2002; Cho et al., 2003; Tucker et al., 2006; Wang et al., 2009). Biswas et al. (2010) sugerem que a melhor frequência alimentar para larvas de “sea bass” (*Lates calcarifer*) é de 3 vezes ao dia, quando criadas em tanques-rede em ambiente salobro. Entretanto, larvas de “ayu” (*Plecoglossus altivelis*) possuem maiores taxas de crescimento quando alimentadas 6 vezes ao dia, ao invés de uma única alimentação diária (Cho et al., 2003). Estudos realizados com juvenis de “Australian snapper” (*Pagrus auratus*) criados em tanques em laboratório em um sistema de reuso parcial de água concluíram que o melhor regime alimentar consiste na alimentação até a saciedade e ofertada de 2 vezes ao dia (Booth et al., 2008). Para juvenis de peixe de água doce como, por exemplo, o tambaqui (*Colossoma macroponum*) quando criado em tanques-rede a otimização do regime alimentar é alcançada através da oferta de 10% da biomassa dividida em 3 partes iguais ao dia (Silva et al., 2007).

Um importante aspecto a respeito dessas práticas de manejo alimentar que deve ser ressaltado é a interação entre elas, pois quando se alimenta uma criação com uma frequência adequada e altas taxas de arraçoamento há um desempenho produtivo maior, porém se elevarmos demais a oferta de ração o crescimento se tornará menos eficiente. Isso ocorre devido ao fato dos organismos utilizarem o excesso de nutrientes disponível não para o crescimento em termos protéico, que é o desejado na aquacultura e sim em termos de acúmulo de gordura (Silva et al., 2007). E se o inverso ocorrer, o desempenho produtivo também será afetado (Booth et al., 2008). Desse forma a interação entre a taxa de arraçoamento e frequência alimentar é um fator de relavada importância que deve ser analisado e otimizado afim de obter um bom desempenho de produção.

Poucos estudos de alimentação foram realizados com o peixe enxada, embora haja experimentos de engorda em tanques-rede na Venezuela. Gaspar e Larez (1984) realizaram experimentos de engorda com juvenis de peixe enxada em tanques-rede e em um ano de criação os peixes atingiram o peso comercial de 300 – 400 g. Outros estudos foram realizados em tanques de concreto com fluxo contínuo com o intuito de testar diferentes dietas para engorda de juvenis. Em um trabalho o melhor resultado encontrado foi com dieta a base de camarão, com um ganho de peso de 390% (Robaina e Salaya, 1993). Um segundo trabalho mostrou o potencial de crescimento do peixe enxada durante um ano de criação, quando os peixes atingiram 300 g ao serem alimentados com sardinha fresca (Gaspar, 2002). Ambos os estudos sugerem o grande potencial aquícola da espécie e a necessidade de mais trabalhos sobre engorda e nutrição na fase juvenil. Dado o interesse no desenvolvimento da criação intensiva do peixe enxada, este será o primeiro estudo a avaliar a influência da taxa de arraçoamento e da frequência alimentar sobre o desempenho de crescimento de juvenis criados em sistema de recirculação.

4. OBJETIVOS

4.1. Geral

Avaliar o efeito da taxa de arraçoamento e da frequência alimentar na produção do peixe enxada.

4.2. Específicos

Determinar a melhor taxa de arraçoamento e frequência alimentar para o peixe enxada.

Avaliar o efeito do manejo alimentar sobre a sobrevivência do peixe enxada.

Analisar a composição de carcaça para a determinação de proteína, cinzas e lipídeos.

Avaliar se existe interação entre a taxa de arraçoamento e frequência alimentar.

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ARTIGO ANEXO

MANEJO ALIMENTAR DE JUVENIS DE PEIXE ENXADA (*Chaetodipterus faber*), ESPÉCIE AQUÍCOLA EMERGENTE

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OPTIMAL FEEDING OF JUVENILE ATLANTIC SPADEFISH *Chaetodipterus faber*, AN EMERGING AQUACULTURE SPECIES

Abstract

The Atlantic spadefish *Chaetodipterus faber* is an excellent candidate for aquaculture development, but success will depend on the identification of proper feeds and feeding regimens for this species. Accordingly, we evaluated the growth performance of juveniles (3.60 ± 0.03 g, mean ± SE) fed at 3, 5, or 7% of body weight (3% BW, 5% BW, 7% BW), either in a single feeding (1×) or divided equally among three feedings (3×). Weight gain, specific growth rate, feed conversion ratio, and feed intake were significantly affected by both feeding rate and frequency. Weight gain and SGR increased significantly with feeding rate, and growth was generally greater and more efficient in the 3× groups than the 1× groups. Fish fed at higher feeding rates accumulated more lipid within the body at the expense of moisture, protein, and ash content, but carcass composition was unaffected by feeding frequency. We suggest that the growth of juvenile Atlantic spadefish may be optimized when fed at 5-7% BW / day in 3 daily feedings, with 7% BW / day yielding the greatest, albeit slightly less efficient, growth.

Key Words: *Chaetodipterus*, Atlantic spadefish, marine aquaculture, feeding rates, feeding frequency

1. Introduction

The family Ephippidae is comprised of 20 species of omnivorous, deep-bodied, laterally compressed, marine and brackish fishes (Froese and Pauly 2010), including the Atlantic spadefish, *Chaetodipterus faber*, the only Ephippid present in Western Atlantic waters (Robins and Ray 1986; Walker 1991). The life history and distribution of this species is not completely understood (Ditty et al. 1994), but it is known to inhabit coastal waters from the United States to southern Brazil, including the Gulf of Mexico (Johnson 1978) and Caribbean (Burgess 1978). Atlantic spadefish utilize a wide variety of ecosystems depending on life stage, including mangroves and salt marshes, to near-shore waters and offshore reefs (Robins and Ray 1986). Juveniles are generally found in estuaries and protected waters, whereas adults predominate near shore and offshore waters (Johnson 1978). Adult Atlantic spadefish often congregate in large schools in coastal waters, where in the U.S. they support a popular recreational fishery (Bohlke and Chaplin, 1993; Robins and Ray 1986). In the wild, specimens can reach 91 cm in total length (TL) and 9 kg in weight in tropical waters (Robins and Ray 1986).

The Atlantic spadefish is an excellent candidate for aquaculture development (Tucker and Jory 1991) because of its physical hardiness and disease resistance, ability to adapt well to confinement, good growth rates (Gaspar and Larez, 1984), tolerance of low temperatures and salinities, lack of aggressive or cannibalistic behavior during both larviculture and growout, and excellent flesh quality and consumer appeal (Randall 1966; Bohlke and Chaplin 1993). In addition to food fish culture, young spadefish may have ornamental importance (Gaspar 1995) owing to their morphology and striking black and white coloration.

As is the case for any emerging aquaculture species, successful culture of Atlantic spadefish will depend, in part, on the identification of proper feeds and feeding regimens for this species. Among the different management practices that may maximize feeding and growth efficiency, feeding rate and feeding frequency have a crucial function in determining overall feed intake, growth, and waste outputs of fish (Silva et al., 2007). For example, sporadic feeding and low feeding rates may contribute to reduced growth as well as increased hunger, intra-specific aggression and increased rate of cannibalism (Folkvord and Ottera, 1993). Alternatively, frequent feeding and high feeding rates may lead to feed wastage, deteriorating water quality, reduced fish production, and ultimately greater production costs (Booth et al., 2008). Optimizing feeding frequency and feeding rate may minimize feed wastage, leading to improvement in water quality and greater size class homogeneity (Dwyer et al., 2002; Tucker et al., 2006). Few feeding studies have been conducted with Atlantic spadefish; though cage culture has been investigated in Venezuela (Gaspar and Larez 1984; Robaina and Salaya 1993; Gaspar 2002). However, proper feeds and feeding strategies can vary with fish size, rearing system (Cho et al., 2003) and feed type (Chua and Teng 1978, 1982). Given the interest in developing intensive aquaculture of Atlantic spadefish, we assessed the influence of feeding rate and feeding frequency on the growth performance of juveniles reared in land-based recirculation systems.

2. Methods and Materials

Juvenile spadefish (3.60 ± 0.03 g, mean ± SE; 7 fish/tank), sourced from the Virginia Seafood Agricultural Research and Extension Center's (VSAREC) aquaculture program, were stocked into a recirculation system at the VSAREC consisting of 24, 10-L tanks, a fluidized KMT biofilter, 50 µm particle filter and 50 watt UV unit (Aquatic Habitats, Apopka, Florida, USA). Fish were cultured under optimal conditions and fed a commercially-available feed (Otohime S2, Reed Mariculture, California, USA; guaranteed analysis: crude protein = 50% minimum, crude fat = 10% minimum, crude fiber = 3% maximum, crude ash = 16% maximum; average pellet size = 1.4 mm; floating feed) at 3, 5, or 7% of body weight (3% BW, 5% BW, 7% BW), either in a single feeding (1×) or divided equally among three feedings (3×). Each feeding rate/feeding frequency treatment combination was randomly assigned to 4 replicate tanks (N = 4) Feeding rates were adjusted for growth every 10 days after group weighing the fish by tank. Fish in the 1× treatments were fed at 1300 hours, whereas fish in the 3× treatments were fed at 0700, 1300 and 1900 hours.

Water temperature and dissolved oxygen (DO) were monitored daily using a YSI-85 Series dissolved oxygen meter (YSI Inc., Yellow Springs, Ohio, USA). Total ammonia-, nitrite-, and nitrate-nitrogen (spectrophotometric analysis, Hach Inc., Loveland, Colorado, USA), pH (YSI-pH100; YSI Inc., Yellow Springs, Ohio, USA), and alkalinity (bromocresol green-methyl red titration method, Hach Inc., Loveland, Colorado, USA) were also quantified once daily. Throughout the experiment, photoperiod was kept at a 12:12 light/dark cycle, tank inflow rates were maintained at 0.6 L/min, and water quality parameters were maintained as follows: temperature = 28.9 ±

0.9 °C, salinity = 27.1 ± 0.2 ppt, dissolved oxygen = 6.1 ± 0.6 mg/L, total ammonia nitrogen = 0.34 ± 0.08 mg/L, nitrite nitrogen = 0.35 ± 0.13 mg/L, nitrate nitrogen = 27.0 ± 3.6 mg/L, alkalinity = 140 ± 6 mg/L, and pH = 7.51 ±0.02 (mean ± SD).

After 39 days of culture, the study was terminated and production performance was assessed. Standard metrics of production performance were calculated as follows:

$$\text{Weight Gain (\%)} = 100 \times \frac{(\text{average final weight} - \text{average initial weight})}{\text{average initial weight}}$$

$$\text{Feed Conversion Ratio (FCR)} = \frac{\text{average individual dry matter feed intake}}{\text{average individual weight gain}}$$

$$\text{Specific Growth Rate (SGR, \% body weight/day)} = 100 \times \frac{\ln \text{average final weight} - \ln \text{average initial weight}}{\text{days of feeding}}$$

$$\text{Feed Intake (\% body weight/day)} = 100 \times \frac{\text{average individual dry matter feed intake}}{\frac{(\text{initial individual weight} \times \text{final individual weight})^{0.5}}{\text{days of feeding}}}$$

Three fish/tank were randomly selected, euthanized via overdose of tricaine methanesulfonate (MS-222; Finquel®; Argent Chemical Laboratories, Inc., Redmond, Washington, USA; immersion in >200 mg/L bath until cessation of opercular movement), and frozen (-80˚C).

Frozen carcass samples were packed in ice and transported (~36 hours in transit) to the Fisheries and Illinois Aquaculture Center (FIAC) in Carbondale, Illinois (USA) where the samples were refrozen (-80˚C) for storage prior to compositional analysis. Carcass samples were sectioned (~1 cm thickness) and lyophilized (Freezone 6, Labconco, Kansas City, Missouri, USA) to determine moisture content. Lyophilized samples were pulverized and pooled by tank before determination of protein, ash, and lipid. Total lipid was determined gravimetrically following chloroform/methanol extraction modified from Folch et al. (1957). Ash content was determined gravimetrically after incineration in a muffle furnace for 4 hours at 650°C. A LECO protein analyzer (FP-528, LECO Corporation, St. Joseph, Michigan) was used to determine protein content.

Although multiple fish were sampled from each tank, replicate tanks served as the experimental units for all statistical analyses (n = 3). As multiple weight measurements were taken from each experimental unit (i.e., tank) through time, it was determined that a repeated measures statistical analysis would be most appropriate for the fish weight data in order to reduce error variability arising from within-subject ('within-tank') effects. These data were analyzed by one-way, repeated measures two-way analysis of variance (ANOVA) within the Mixed Model framework of the Statistical Analysis System, version 9.1 (SAS Institute, Cary, North Carolina, USA) to determine significance of differences among treatment means at each time point as well as whether the stress treatments differed in their overall effect on the response parameters. All other data were analyzed by two-way ANOVA within the Mixed Model framework to determine the significance of feeding rate and feeding frequency as main effects, as well as to test for a significant interaction effect. When two-way ANOVA revealed significant main or interactive effect(s), Tukey's HSD tests were used for post-hoc pair-wise comparisons. In all cases, differences were considered significant at $P < 0.05$.

3. Results

Juvenile Atlantic spadefish weight gain (feeding rate $P < 0.001$, feeding frequency $P < 0.001$, interaction $P = 0.002$), SGR (feeding rate $P < 0.001$, feeding frequency $P < 0.001$, interaction $P = 0.072$), FCR (feeding rate $P = 0.004$, feeding frequency $P < 0.001$, interaction $P = 0.288$), and feed intake (feeding rate $P < 0.001$, feeding frequency $P = 0.024$, interaction $P = 0.036$) were significantly affected by both feeding rate and frequency. Fish weight increased significantly over the course of the 39-day experiment, with treatment groups becoming statistically distinct from one another by 20 days (Figure 1). Weight gain and SGR increased significantly with feeding rate (Figure 2). Regardless of feeding rate, growth was generally greater and more efficient in the 3× groups than the 1× groups. The growth enhancing effect of greater feeding frequency was particularly evident within the 7% BW treatment. Feeds were offered at specific levels, and feed intake varied expectedly with feeding rate (Figure 2). Although feeding rates were constant, regardless of feeding frequency, feed intake expressed as a percent of body weight was elevated among fish in the 3× fed at 7% BW.

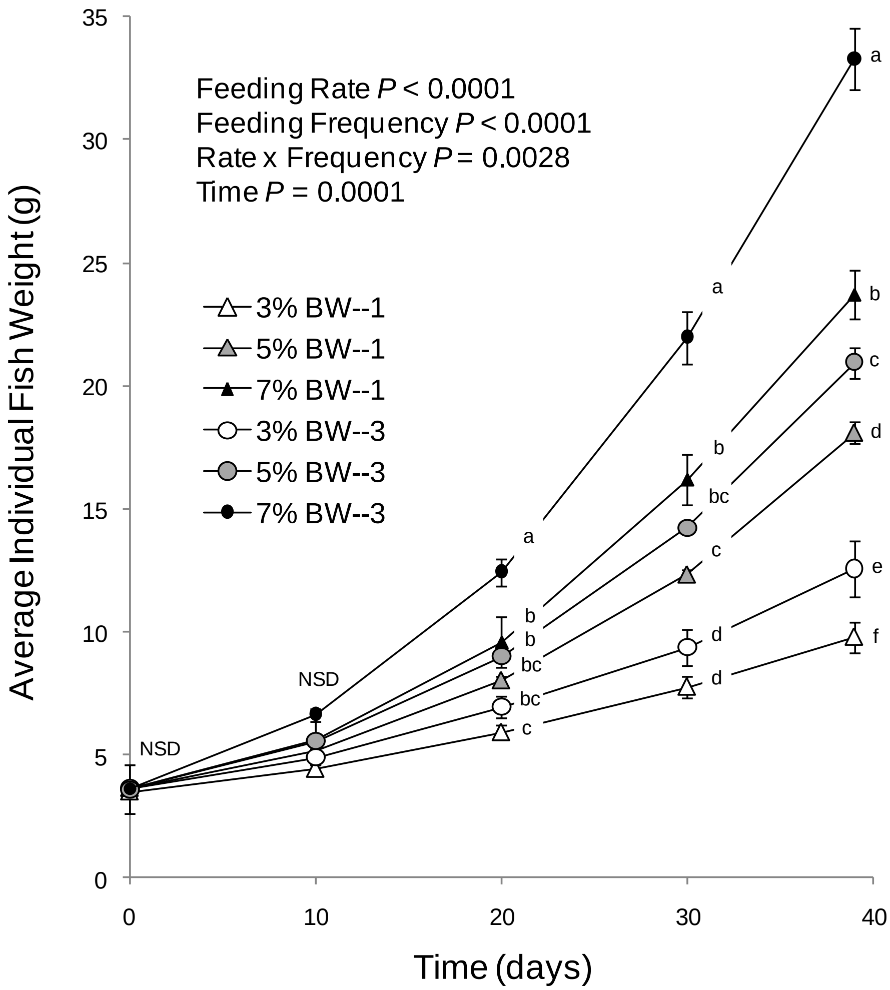

Figure 1. Average individual of juvenile Atlantic spadefish fed at 3, 5, or 7% of body weight (BW) per day in single (1×) or multiple (3×) feedings. Each data point represents the treatment combination mean ± standard error of the mean. Data points at a given time point with common letters are not significantly different ($P > 0.05$). *P*-values generated by a two-way repeated measures ANOVA assessing feeding rate and frequency as main effects are also provided; "NSD" indicates no significant difference between means at a time point.

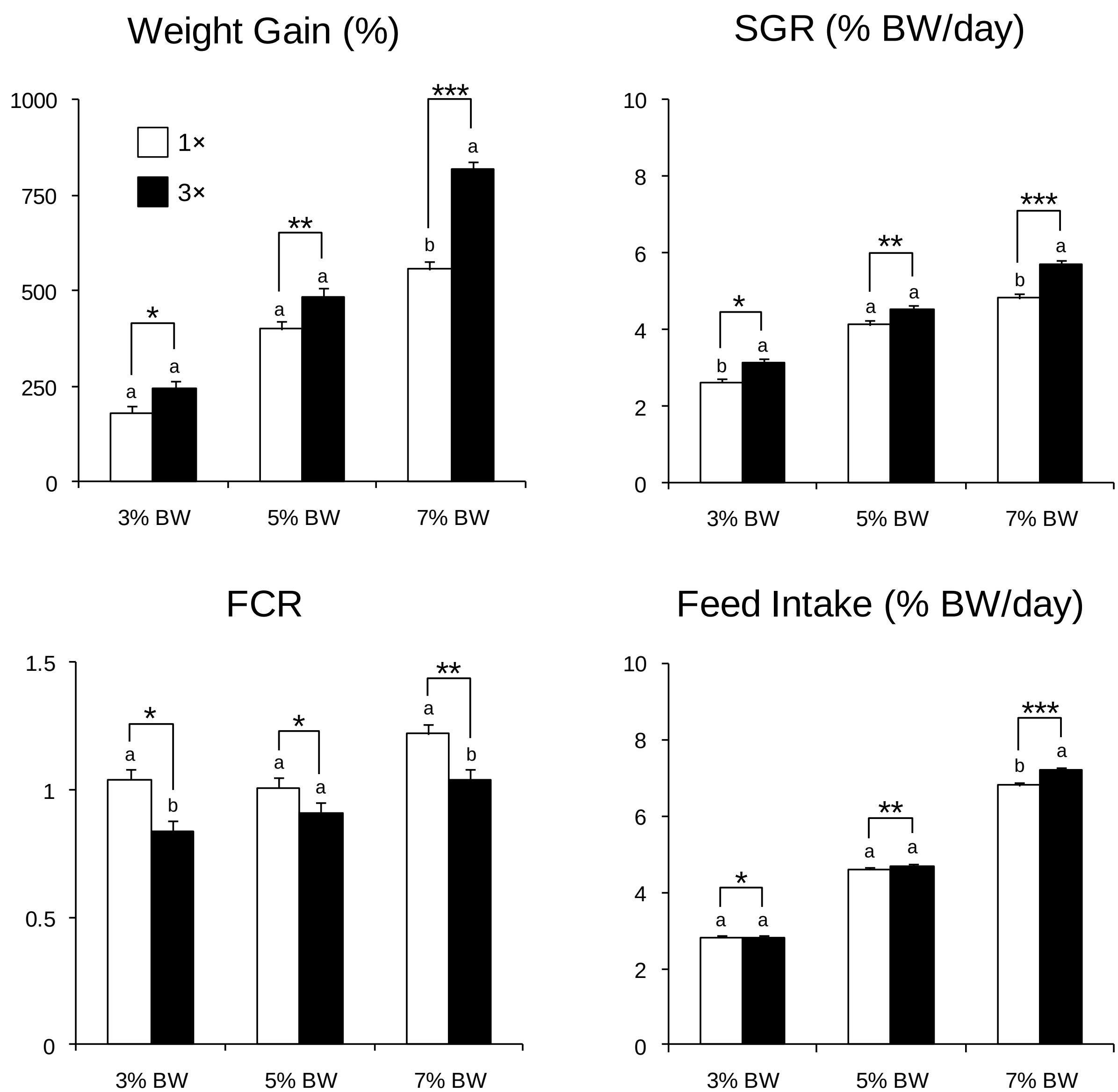

Figure 2. Growth performance of juvenile Atlantic spadefish fed at 3, 5, or 7% of body weight (BW) per day in single (1×) or multiple (3×) feedings. Bars represent least-square means; error bars represent pooled standard error. Significant differences between feeding frequencies within a feeding rate are indicated by different letters; significant differences among feeding rates are indicated by different numbers of asterisks ($P < 0.05$).

Carcass proximate composition was affected by feeding rate, but not by feeding frequency (Table 1). Spadefish fed at higher feeding rates accumulated more lipid within the body at the expense of moisture, protein, and ash content.

Table 1. Carcass proximate composition of juvenile spadefish at the end of the feeding trial. Except moisture, all parameters are reported as percent of dry matter basis (%). Least-square means ± SE are shown for each treatment factor combination in normal text; means across diet and stress exposure replacement factors are shown in italics. *P*-values for each response parameter and their interaction are also provided. For response parameters exhibiting significant factor effects, means without common superscripts are significantly different ($P < 0.05$).

	Feeding Frequency	Feeding rate: %BW / day				*P*-values		
Parameter	(times / day)	3	5	7	*Means*	Rate	Frequency	Interaction
Moisture	1×	76.4 ± 0.9	73.2 ± 0.9	70.5 ± 0.9	*73.4*	0.001	0.104	0.222
	3×	73.9 ± 0.9	73.6 ± 0.9	68.9 ± 0.9	*72.2*			
	Means	*75.1 a*	*73.4 a*	*69.7 b*				
Lipid	1×	10.6 ± 1.1	19.9 ± 1.1	25.7 ± 1.1	*18.7*	<0.001	0.066	0.061
	3×	12.9 ± 1.1	18.6 ± 1.1	29.2 ± 1.1	*20.2*			
	Means	*11.7 c*	*19.3 b*	*27.4 a*				
Protein	1×	69.0 ± 1.1	63.2 ± 1.1	56.6 ± 1.1	*62.9*	<0.001	0.074	0.385
	3×	65.7 ± 1.1	63.1 ± 1.1	54.7 ± 1.1	*63.1*			
	Means	*67.3 a*	*63.1 b*	*55.7 c*				
Ash	1×	17.8 ± 0.5	14.0 ± 0.5	12.0 ± 0.5	*14.6*	<0.001	0.076	0.217
	3×	16.7 ± 0.5	14.3 ± 0.5	10.5 ± 0.5	*13.8*			
	Means	*17.2 a*	*14.1 b*	*11.2 c*				

4. Discussion

Naturally, feeding rate affects the growth of fishes: increasing feeding rate increases the availability of resources (amino acids, structural lipids, energy, etc.) for growth, and weight gain is typically greater among fish at higher feeding rates. However, if any of the required elements for somatic growth become limiting (e.g., certain amino acids or phospholipids) or if nutrient intake simply exceeds that which can be used according to the maximum intrinsic growth rate of the fish, surplus resources will be lost via increased fecal losses or stored as glycogen or neutral lipid deposits within the hepatic, muscular, and adipose tissues (Jobling 1994). In turn, carcass levels of energy storage products and FCR will increase over time. Our data illustrates this pattern quite clearly: regardless of feeding frequency, Atlantic spadefish that are fed at higher rates gain more weight, but growth efficiency is reduced at 7% BW / day and the composition of the gain is increasingly skewed towards greater adiposity. Similarly, cuneate drum *Nibea miichthioides* fed 1-6% BW / day grew more at the highest feeding rates, but grew less efficiently, had a reduced nitrogen retention efficiency, and accumulated higher carcass lipid levels (Wang et al. 2007). Comparable results have also been reported for gilthead seabream *Sparus aurata* (Mihelakakis et al. 2002), Chinese sucker *Myxocyprinus asiaticus* (Yuan et al. 2010), rainbow trout *Oncorhynchus mykiss* (Reinitz 1983; Storebakken et al. 1991), white sturgeon *Acipenser transmontanus* (Hung et al. 1993), grass carp *Ctenopharyngodon idella* (Du et al. 2006), and flounder *Paralichthys olivaceus* (Kim et al. 2007). Given the wide range of taxa for which this phenomenon is reported, perhaps this pattern is exhibited by all fishes including Atlantic spadefish. Focusing on the effect of feeding rate exclusively, our data suggest Atlantic spadefish should be fed at 5-7% BW / day to maximize growth, but lean growth may be more efficient at the lower end of this range.

Feeding rate has a strong effect on the growth performance of fishes, but feeding frequency has been shown to independently and interactively influence the growth and growth efficiency as well. When fish are fed to apparent satiety, increasing the frequency of feeding events tends to increase total feed intake up to a threshold determined, perhaps in part, by gastrointestinal evacuation rate (Jobling 1994). As a result, feeding rate is effectively increased and growth is enhanced, though as mentioned above, growth efficiency may be somewhat reduced at very high feed intake rates. This phenomenon has been demonstrated in yellowtail flounder (Dwyer et al. 2002), cuneate drum (Wang et al. 2007), channel catfish *Ictalurus punctatus* (Peterson and Small 2006), black sea trout *Salmo trutta labrax* (Başçınar et al. 2007), Korean rockfish *Sebastes schlegeli* (Lee et al. 2000), great sturgeon *Huso huso* (Mohseni et al. 2006), hybrid sunfish (female green sunfish *Lepomis cyanellus* × male bluegill *L. macrochirus*; Wang et al. 1998), cuneate drum (Wang et al. 2007), and pikeperch *Sander lucioperca* (Wang et al. 2009). When fish are fed at a fixed feeding rate, increasing feeding frequency still tends to improve growth to a point, but the magnitude of the effect is diminished and is likely the result of moderate improvements in conversion efficiency associated with gastrointestinal adaptation to more consistent, but lower instantaneous 'gut fill' (Jobling 1982; Peterson and Small 2006). For example, Asian seabass *Lates calcarifer* gained more weight and grew more efficiently when fed the same ration in 3 daily feedings compared to 1 or 2 feedings, but feeding 4× daily failed to increase growth further and resulted in an elevated FCR (Biswas et al. 2010); largely consistent results were also reported for this species by Salawa (2008). Similar responses to increasing feeding frequencies have been reported for ayu *Plecoglossus altivelis* (Cho et al. 2003), Australian snapper *Pagrus auratus*

(Tucker et al. 2006), and red-spotted grouper *Epinephelus akaara* (Kayano et al. 1993). Our results are broadly consistent with literature: although differences were not significant in all cases, regardless of feeding rate, Atlantic spadefish fed 3× daily grew more and more efficiently than those fed 1× daily. Although a significant difference in feed intake was observed within the 7% BW treatment, the magnitude of the difference was relatively small, and is likely an artifact arising from the differences in body weight observed for these two treatments which grew more marked over time. Based on our data, it would seem that a 3× daily feeding rate would be advantageous for Atlantic spadefish, regardless of feeding rate.

We observed significant interaction between feeding rate and frequency in terms of weight gain which suggests that the performance enhancing effects of higher feeding frequencies are more pronounced among fish fed higher rates (a significant interaction effect was also observed for feed intake, but as mentioned above, this is likely an artifact of the differences in weight gain). Fewer studies have simultaneously evaluated the effects of different feeding rates and frequencies, and the literature is somewhat divided among species. Similar to what we observed in Atlantic spadefish, tambaqui *Colossoma macropomum* performed better when fed more frequently at 10% BW / day, but these effects were not observed when feeding 5% BW / day (Silva et al. 2007). Conversely, increasing feeding frequency in ayu beyond 1× daily did not improve performance of fish fed 3% or 6% BW / day (Cho et al. 2003). Despite these conflicting results, given the negative effect of excessively high feed intake on growth efficiency via greater fecal losses (in the absence of adequate gastrointestinal adaptation to compensate for larger meal sizes), it is logical to hypothesize that dividing high intake into multiple feeding events would improve efficiency and yield greater weight gain. Increasing feeding frequency among fish fed at high feeding rates may also be beneficial in the sense that it would the duration of each feeding event, and thus the possibility of feeding inefficiencies due to pellets sinking or being flushed from the system before they could be consumed. Certainly, behavioral interactions such as social dominance play a role in feeding efficiency and feed intake in fishes (Jobling 1994), but for species such as Atlantic spadefish that do not appear to exhibit strong social hierarchies and competitive feeding behaviors in intensive culture, increasing feeding frequencies may be beneficial in terms of digestive/absorptive efficiencies and growth performance, particularly at high feeding rates. Taking this and the rest of our results into consideration, we suggest that the growth performance of Atlantic spadefish may be optimized when fed at 5-7% BW / day in 3 daily feedings, with 7% BW / day yielding the greatest, albeit slightly less efficient, growth.

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6. CONCLUSÕES

Os resultados do presente estudo mostram que o manejo alimentar do peixe enxada *Chaetodipterus faber* criado em sistema intensivo de recirculação, pode ser otimizado quando a alimentação é ofertada com 5-7% da biomassa/dia em 3 alimentações diárias, com 7% da biomassa/dia há um crescimento maior, embora menos eficiente.