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EFEITO DA INOCULAÇÃO DA DIATOMÁCEA *Navicula* sp. NO CULTIVO DE PÓS-LARVAS DE *Litopenaeus vannamei* EM SISTEMA DE BIOFLOCOS

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PRÓ-REITORIA DE PESQUISA E PÓS-GRADUAÇÃO

PROGRAMA DE PÓS-GRADUAÇÃO EM RECURSOS PESQUEIROS E AQUICULTURA

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Resumo

O sistema de bioflocos (BFT) tem sido proposto para a produção intensiva de camarões como uma prática ambientalmente amigável, capaz de reduzir os impactos ambientais, prevenir a introdução de doenças e patógenos através da reduzida ou troca zero de água, aumentando a biossegurança dos cultivos. As microalgas desempenham um papel fundamental na reciclagem dos nutrientes, além de ser as principais produtoras primárias dos ácidos graxos poliinsaturados da família ω-3: EPA e DHA. As PUFAs são consideradas essenciais nas dietas de larvas e pós-larvas de camarões marinhos porque contribuem no crescimento e sobrevivência dos camarões cultivados. O estudo teve como objetivo avaliar o efeito da inoculação da diatomácea *Navicula* sp. no cultivo de pós-larvas de *Litopenaeus vannamei* em sistema de bioflocos. Quatro tratamentos foram realizados: controle (ZWE-B); troca zero de água e adição de ração comercial (ZWE-BF); troca zero de água e adição da diatomácea *Navicula* sp. (ZWE-D) e troca zero de água com adição de ração e *Navicula* sp. (ZWE-FD), todos com três repetições. Os camarões (17,7 ± 0,02 mg) foram estocados a uma densidade de 2,500 PL por m^{-3} e as microalgas inoculadas no 1º, 5º e 15º dia de cultivo, a uma densidade de $5x10^4$ cél mL^{-1}. Os camarões foram alimentados com ração comercial com 42% de proteína bruta, quatro vezes ao dia. Para análise dos dados utilizou-se Cochran, Shapiro-Wilk, ANOVA, Tukey e teste t de Student ($P<0,05$). Os gêneros mais frequentes observados para o fitoplâncton, zooplâncton e cianobactérias foram: *Anabaena, Arcella, Bosmina, Brachionus, Cylindrotheca, Daphnia, Fragilaria, Hemiaulus, Keratella, Orthoseira, Oscillatoria, Phymatodocis, Rhabdonema, Skeletonema, Sckizothrix* e *Ulothrix.* Não foram observadas diferenças significativas ($P>0,05$) para oxigênio dissolvido, temperatura, pH e salinidade. Porém, foram observadas diferenças significativas ($P<0,05$) entre os tratamentos para a TAN, NO_2-N, alcalinidade, peso final, ganho de peso, biomassa final, conversão alimentar, taxa de crescimento específico e sobrevivência. O tratamento com adição de *Navicula* sp. e fornecimento de ração apresentou os melhores parâmetros de produção, indicando os benefícios da inoculação das diatomáceas para as pós-larvas de *L. vannamei*, além de melhorar a qualidade da água e reduzir as densidades de cianobactérias em sistema de bioflocos.

Palavras-chave: Microalgas, Flocos microbianos, Camarão, Troca zero de água.

Abstract

Biofloc systems (BFT) have been proposed for intensive shrimp production as an environmentally friendly practice that can reduce environmental impacts and prevent the introduction of diseases and pathogens through reduced or zero water exchange, thus increasing the biosecurity of cultivation. Microalgae perform an important role in the recycling of nutrients, in addition to being the main primary producers of polyunsaturated fatty acids of the ω-3 family: EPA and DHA. PUFAs are considered essential in the diets of shrimp larva and post-larva because they contribute to growth and survival of cultivated shrimp. The purpose of this study is to evaluate the effect of inoculation with the diatom *Navicula* sp. in the cultivation of post-larva of *Litopenaeus vannamei* in a biofloc system. Four treatments were conducted: a control (ZWE-B); zero water exchange with the addition of commercial rations (ZWE-BF); zero water exchange and addition of diatom *Navicula* sp. (ZWE-D); and zero water exchange with addition of rations and *Navicula* sp. (ZWE-FD), each with three repetitions. The shrimp (17,7 ± 0,02 mg) were stocked at a density of 2,500 PL per m^{-3} and the microalgae were inoculated on the 1st, 5th and 15th day of cultivation, at a density of $5x10^4$ cells mL^{-1}. The shrimp were fed with commercial rations with 42% raw protein, four times a day. The data was analyzed using Cochran, Shapiro-Wilk, ANOVA, Tukey and the Student t-test ($P<0,05$). The most frequent genres observed for the phytoplankton, zooplankton and cyanobacteria were: *Anabaena, Arcella, Bosmina, Brachionus, Cylindrotheca, Daphnia, Fragilaria, Hemiaulus, Keratella, Orthoseira, Oscillatoria, Phymatodocis, Rhabdonema, Skeletonema, Sckizothrix* and *Ulothrix.* No significant differences were observed ($P>0,05$) for dissolved oxygen, temperature, pH and salinity. However, significant differences were observed ($P<0,05$) between the treatments for TAN, NO_2-N, alkalinity, final weight gain, final biomass, food conversion, specific growth rate and survival. The treatment with the addition of *Navicula* sp. and rations had the best production parameters, indicating the benefits of inoculation of the diatoms for the post-larva of *L. vannamei*, in addition to improving water quality and reducing the density of cyanobacteria in a biofloc system.

Key words: Microalgae, Microbial flocs, Shrimp, Zero water exchange.

Lista de tabelas

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Sumário

1- Introdução

A carcinicultura é considerada uma atividade consolidada no âmbito da produção de alimentos de procedência aquática e o camarão branco do Pacífico, *Litopenaeus vannamei*, se destaca por ser a espécie mais cultivada no mundo. Porém, o crescimento do setor pode ocasionar alguns problemas ambientais, tais como, destruição dos mangues, propagação de doenças no cultivo correlacionadas na maioria das vezes por manejo inadequado e geração de efluentes com alta concentração de nutrientes e matéria orgânica (PRIMAVERA et al., 2006; KRUMMENAUER et al., 2012). Neste sentido, é necessária a busca por técnicas de manejo que melhorem a eficiência da administração de alimentos, da qualidade da água e dos solos, nos quais a renovação de água seja minimizada, mitigando a emissão de efluentes e a transmissão de doenças, no sentido de tornar a atividade sustentável (BRITO et al., 2010). Dentro dessas estratégias pode-se destacar o cultivo de camarões marinhos em sistema de bioflocos, também conhecido como BFT (Biofloc Technology System) ou cultivo de camarões em meio heterotrófico.

O sistema de bioflocos, além de controlar eficientemente a qualidade da água em cultivos com troca zero de água, permite a cultura intensiva e saudável dos camarões (AVNIMELECH, 2012). A força motriz de sistemas BFT é microbiana, ao qual é um agregado orgânico de partículas suspensas com muitas variedades de microrganismos ativos associados com substâncias poliméricas extracelulares (De SCHRYVER et al., 2008; JU et al., 2008; RAY et al., 2010; XU e PAN, 2013). Esse sistema é aludido sobre os efeitos benéficos à carcinicultura, principalmente por remover fontes de nitrogênio tóxicas, como a amônia e o nitrito (RAY et al., 2011; XU et al., 2012); melhor aproveitamento da ração e melhor desempenho do crescimento dos camarões, através do incremento do alimento natural que estimulam a digestão e as atividades enzimáticas (BALLESTER et al., 2010; XU e PAN, 2012; ANAND et al., 2014; KIM et al., 2014), além de aumentar a biossegurança e a saúde dos animais cultivados, exercem possível efeito probiótico (MOSS et al., 2012; HASLUN et al., 2012; XU e PAN, 2013; SOUZA et al., 2014).

Em sistemas de bioflocos, o desenvolvimento de uma comunidade microbiana benéfica deve ser desenvolvida e sustentada (RAY et al., 2010). As algas fornecem um complemento nutricional para camarões, disponibilizam nutrientes para o crescimento de bactérias, além de servir de alimentação básica para o zooplâncton que consecutivamente podem prover de alimentação suplementar para camarões (JU et al., 2008, BALOI et al., 2013). É sabido que nas larviculturas de peneídeos, as exigências nutricionais destes animais são cumpridas com o fornecimento de microalgas marinhas (PIÑA et al., 2006; ZHOU et al., 2009; KHATOON et al., 2009). As diatomáceas são o grupo de microalgas preferidas para a alimentação de larvas e pós-larvas de camarões (YUSOFF et al., 2002; JU et al., 2008; 2009), por contribuir em aminoácidos essenciais e ácidos graxos polinsaturados (PUFAs), principalmente da família ω-3: eicosapentaenóico (EPA) e decosahexanóico (DHA), aos quais são fundamentais para o crescimento e sobrevivência dos camarões cultivados (PIÑA et al., 2006; JU et al., 2009; BELETTINI et al., 2011). Mas, a alta concentração de matéria orgânica em suspensão, que diminui significativamente a luminosidade na coluna d'água reduzindo o processo fotossintético, assim como, a competição por nutrientes com as bactérias heterotróficas, parece impedir o estabelecimento das diatomáceas em sistemas de bioflocos (GODOY et al., 2011), não obstante, a inoculação das diatomáceas em sistemas BFT, poderá favorecer na manutenção destas.

Além disso, o processo fotossintético contribui na ciclagem dos nutrientes, absorve compostos nitrogenados, promovendo a melhoria da qualidade de água, fornecendo oxigênio durante o dia e controlando doenças e patógenos (THOMPSON et al., 2002; BURFORD et al., 2003; LAVÍN e LOURENÇO, 2005; HARGREAVES, 2006; ZHOU et al., 2009; BALLESTER et al., 2010; GODOY et al., 2011; BALOI et al., 2013). Apesar da importância que as diatomáceas tem nas culturas extensivas, semi-intensivas, integradas e intensivas (MARTÍNEZ-CÓRDOVA e PEÑA-MESSINA, 2005; PATIL et al., 2007; ELEZUO, 2011), o entendimento do seu papel em

sistemas de bioflocos permanece pouco depreendido, especialmente no que diz respeito à qualidade de água, crescimento, sobrevivência e desempenho zootécnico dos animais cultivados.

Assim, o objetivo do presente estudo foi avaliar o efeito da inoculação da diatomácea *Navicula* sp. no cultivo de pós-larvas de *Litopenaeus vannamei* em sistema de bioflocos.

2- Revisão de literatura

2.1 Sistema de Bioflocos

Recentemente, vários estudos estão sendo desenvolvidos com o objetivo de limitar ou mesmo zerar a troca de água nos sistemas de cultivo, combinando o tratamento de água com a reciclagem do alimento artificial não consumido, através de uma biota aeróbica e heterotrófica. Dentre outros termos, esse tipo de cultivo é conhecido como, Zero Exchange Aerobic Heterotrophic Culture Systems (ZEAH), Biofloc Technology (BFT), sistema heterotrófico ou simplesmente sistema de cultivo com flocos microbianos (WASIELESKY et al., 2006).

Historicamente, essa tecnologia começou a ser utilizada para o tratamento de efluentes domésticos, mais foi a partir dos anos 80 que ela passou a ter sua história no cultivo de organismos aquáticos. O sistema de bioflocos começou a ser desenvolvido através de pesquisas do grupo AQUACOP, induzindo a formação de flocos microbianos, antigamente chamados de “moulinetes”, como forma de realizar a manutenção de reprodutores de camarões peneídeos na Polinésia Francesa (TACON et al., 2002; CUZÓN et al., 2008). Nos anos 90, em Israel, Avnimelech e colaboradores, realizaram vários experimentos a fim de induzir a formação de uma cadeia composta por bactérias heterotróficas, por meio de mudanças na relação carbono:nitrogênio na água do cultivo. Simultaneamente, Hopkins e um grupo de pesquisadores, iniciaram nos Estados Unidos no Waddell Mariculture Center (WMC), o desenvolvimento de tecnologias ambientalmente amigáveis, com objetivo de diminuir a emissão de efluentes em viveiros revestidos (WASIELESKY et al., 2006a; VENERO et al., 2009). A produtividade nesses sistemas superaram 5000 Kg/ha/safra sendo

principalmente atribuída à diminuição da renovação de água, indução e estabilização da cadeia microbiana (AVNIMELECH, 1993; HOPKINS et al., 1993; AVNIMELECH et al., 1994; CHAMBERLAIN e HOPKINS, 1994). Posteriormente este sistema foi adaptado para fazendas em Belize. Onde a Belize Aquaculture Ltda foi a primeira fazenda comercial a utilizar este sistema de cultivo com sucesso. Na primeira tentativa a produção foi de 13,5 toneladas/camarões/ha, posteriormente, alcançando uma média de produção de 20 toneladas/camarão/ha (BURFORD et al., 2003).

O processo de formação dos bioflocos envolve interações físicas, químicas e biológicas. O início da agregação das partículas ocorre pela adição de uma fonte de carbono orgânico diretamente na água e/ou pelo uso de alimentos com especial taxa C/N (BALLESTER et al., 2010; CRAB et al., 2012). Com o balanceamento e a manutenção da relação carbono:nitrogênio (C/N) próximos a 12-20, bactérias heterotróficas imobilizam o íon amônio, diminuindo-o no sistema, para a produção de proteína microbiana (AVNIMELECH, 1999; SCHENEIDER et al., 2005). A partir daí, microorganismos começam a se desenvolver e diversificar cada vez mais, assim, além de bactérias heterotróficas e autotróficas, observa-se protozoários ciliados e flagelados, nematoides, microalgas, rotíferos, copépodos, dentre outros. Resumidamente, este processo de formação dos flocos ocorre com aumento da carga orgânica e o tempo, seguindo os passos: água clara, bloom de algas, grande quantidade de espumas na superfície, acúmulo de material orgânico dissolvido, mudança na coloração da água para marrom, desaparecimento das espumas e finalmente o surgimento dos flocos (AVNIMELECH, 2009).

Estes sistemas permitem que o nitrogênio gerado pelos alimentos não consumidos e excretos dos organismos seja convertido em biomassa proteica, voltando a ser disponibilizado no cultivo e consumido por esses mesmos indivíduos. Com isto, se torna possível minimizar a troca de água sem que esta perca sua qualidade e, por conseguinte a quantidade de nutrientes descarregados em água adjacentes é diminuída, aumentando a biossegurança (LEZAMA-CERVANTES e PANIAGUA-

MICHEL, 2010). Comparando os sistemas tradicionais de cultivo com o sistema de bioflocos, para produzir 1 Kg de camarão em práticas tradicionais de produção são necessários de 20 a 64 m^3 de água (HOPKINS et al., 1993; TIMMONS e LOSORDO, 1994; MOSS et al., 2001). Em contraste, Samocha et al. (2010) utilizaram apenas 98 litros de água para produzir os mesmos 1 Kg de camarão *L. vannamei* em sistemas de bioflocos com troca zero de água. Além disso, a mesma água pode ser utilizada por vários ciclos de produção sem influenciar no desempenho do camarão cultivado (KRUMMENAUER et al., 2014).

Na produção de camarões em BFT alguns requisitos mínimos em termos de qualidade de água devem ser considerados. Para temperatura, oxigênio dissolvido, salinidade, são semelhantes com os sistemas tradicionais de cultivo. Contudo, para os parâmetros de pH, CO_2, fósforo, amônia, nitrito, nitrato, alcalinidade e sólidos suspensos, se diferenciam. Segundo Furtado et al. (2011), em BFT o pH e a alcalinidade podem diminuir em função do aumento do dióxido de carbono dissolvido e dos sólidos suspensos totais. Essa redução do pH e da alcalinidade, ocorre devido a ação das bactérias nitrificantes e heterotróficas que formam o bioflocos. Outro fator que também deve ser levado em consideração é que como esse sistema oferece a possibilidade de utilizar elevadas densidades de estocagem (KRUMMENAUER et al., 2011; SILVA et al., 2013; FRÓES et al., 2013), pode ocorrer o acúmulo de nitrogenados proveniente da excreção dos animais e da matéria orgânica em decomposição, as concentrações de amônia podem alcançar níveis elevados. Assim, a fertilização orgânica de carbono deve ser realizada para estimular o rápido crescimento bacteriano 5-7 semanas, metabolizando a amônia em nitrito e posteriormente a nitrato (SAMOCHA et al., 2011), visto que esses compostos podem atingir concentrações tóxicas aos animais cultivados. Silva et al. (2013), observaram que mais de 16% do fósforo orgânico e inorgânico dissolvido podem ficar acumulados em cultivos com bioflocos. A acumulação de fósforo nestes sistemas é devido à ração não consumida (lixiviadas) na água, favorecendo a eutrofização (PEÑAFLORIDA, 1999). Apesar

deste composto não afetar diretamente o camarão, pode favorecer no crescimento de cianobactérias, que podem obstruir as brânquias do camarão e produzir toxinas (WASIELESKY et al., 2006b).

Partículas de bioflocos em geral podem contribuir substancialmente para as necessidades nutricionais dos camarões agindo como uma fonte de suplementação alimentar, promovendo uma maior taxa de crescimento, aumento do peso final e redução no fator de conversão alimentar (COHEN et al., 2005; VENERO et al., 2009; RAY et al., 2010; BALLESTER et al., 2010; CRAB et al., 2012; FRÓES et al., 2012; KRUMMENAUER et al., 2014). Além disso, é possível diminuir os níveis de proteína das rações para camarões, reduzindo significativamente os custos com alimentação exógena (AVNIMELECH, 2000; BALLESTER et al., 2010). Bauer et al. (2012) substituindo a farinha de peixe, por flocos microbianos e proteína de soja em dietas para *L. vannamei*, observaram que a farinha de peixe pode ser substituída por flocos microbianos e proteína de soja, sem representar efeitos adversos no desempenho do camarão cultivado. Anand et al. (2014), estudaram o efeito da suplementação dietética de bioflocos no crescimento e atividade de enzimas digestivas em juvenis de *Penaeus monodon*, elucidaram que o biofloco pode ser utilizado como um suplemento dietético, onde a nível de 4% na ração aumentou o crescimento e a atividade de enzimas digestivas (>57%) em juvenis do camarão tigre *P. monodon*. Recentemente Kim et al. (2014) descobriram que os bioflocos além de favorecer o crescimento das pós-larvas de *L. vannamei*, aumentou a resposta imune destes crustáceos.

2.2 Microalgas na aquicultura

Microalgas são seres microscópicos, eucarióticos, fotossintetizantes, pertencentes ao Reino Protista (RICHMOND, 2004). Reproduzem-se utilizando a energia luminosa em energia química (biomassa) através da fotossíntese, completando todo um ciclo de crescimento em poucos dias. Além disso, podem crescer praticamente em qualquer lugar, exigindo unicamente a luz solar e de alguns nutrientes, não obstante, as suas taxas de crescimento, composição bioquímica, pode ser

otimizada pela adição de específicos nutrientes, aeração, temperatura, intensidade luminosa, pH, entre outros (ASLAN et al., 2006).

As microalgas são os principais componentes do primeiro nível trófico de uma cadeia alimentar, ao converter autotroficamente a energia luminosa em energia assimilável pelos seres vivos (ALONSO et al., 2012). Sua biomassa é conhecida como uma fonte natural e ilimitada de compostos biologicamente ativos, tais como carotenoides, ficobilinas, vitaminas, aminoácidos, proteínas, onde atualmente, diferentes espécies são produzidas em escala comercial, favorecendo o seu desenvolvimento em diversos campos como na aquicultura e nas indústrias químicas, farmacêuticas e nutracêuticas (PULZ e GROSS, 2004; GOUVEIA et al., 2007; GRIFFITHS et al., 2012; DRAAISMA et al., 2013).

Além das aplicações biotecnológicas, a grande demanda da produção de microalgas concentra-se na aquicultura, onde são utilizadas como fonte alimentar, principalmente pelo seu conteúdo proteico e de ácidos graxos poliinsaturados, para moluscos bivalves (PETTERSSEN et al., 2010; PERNET et al., 2012), rotíferos (COSTA et al., 2008; ROMERO e YUFERA, 2012; YIN et al., 2013), artêmias (MAKRIDIS et al., 2006; DEHGHAN et al., 2011; INTERAMINENSE et al., 2014), copépodos (MARTÍNEZ-CÓRDOVA et al., 2012) e de outros invertebrados marinhos, como os camarões (THOMPSON et al., 2002; SOARES et al., 2006; KENT et al., 2011; VIAU et al., 2013; KHATOON et al., 2013). Além de servir de corantes para organismos intensamente cultivados, melhorando o preço desses no mercado (CHIEN et al., 2003; NIU et al., 2009). Rações incrementadas com 5-20% de *Arthrospira* (rica em carotenos) aumentam os padrões de vermelho e amarelo em carpas e intensificam o brilho das partes brancas. Essa definição de cor aumenta o valor da venda. Outro exemplo é a tradicional técnica francesa de “esverdeamento de ostras”, que consiste na indução de cor verde-azulada nas brânquias e nos palpos labiais de ostras utilizando a diatomácea *Haslea ostrearia*, aumentando o valor do produto em 40% (SPOLAORE et al., 2006; GAGNEUX-MOUREAUX et al., 2007; HEMAISWARYA et al., 2011).

Como as microalgas são primordiais nas cadeias produtivas da aquicultura, representam custo relativamente alto na produção de animais. O valor dos custos envolvidos na produção dependerá da espécie do animal cultivado e da duração do período em que as microalgas serão ofertadas (fase exponencial e/ou fase estacionária), podendo representar 40% do custo total para a produção de sementes de ostras ou 30% da produção de pós-larvas de peneídeos (LAING e HELM, 1981; KUBAN et al., 1983; BENEMAM et al., 1992; BOROWITZKA, 1997; CAÑAVATE e FERNÁNDES-DÍAZ, 2001). Por conta disso, para ser viável em aquicultura, a microalga deve apresentar altas taxas de crescimento, ser de fácil cultivo, ser resistente às condições de cultivo, atóxica, apresentar tamanho adequado e alta qualidade nutricional para ser ofertada ao animal de interesse, como, apresentar parede celular digerível (ou ausente) para facilitar o acesso aos nutrientes contidos nas células (HEMAISWARYA et al., 2011).

As espécies utilizadas na alimentação de organismos aquáticos pertencem a vários grupos Cryptophyceae (*Rhodomonas* spp. Karsten), Chrysophyceae (*Monochrysis* spp. Skuja), Haptophyceae (*Isochrysis* spp. Parke), Prasinophyceae (*Tetraselmis* spp. Stein), Cyanophyceae (*Arthrospira* spp. e *Spirulina* spp.) e Chlorophyceae (*Chlorella* spp., *Dunaliella* spp. e *Scenedesmus* spp. Bourrely). Dentre outras classes e diversas espécies, as mais comumente utilizadas em larviculturas de peneídeos são as diatomáceas Bacillariophyceae centrales (*Chaetoceros* spp. Ehrenberg, *Thalassiosira* spp. Cleve, Skeletonema spp. Greville) e as penales (*Phaeodactylum tricornutum* Bohlin, *Nitzchia* spp. Hustedt, *Amphora* spp. Kützing) (MULLER-FEUGA, 2004).

As diatomáceas são organismos unicelulares, estão dentro da classe Bacillariophyceae, possuindo uma característica peculiar às outras microalgas, porque possui parede celular silicosa, com números de gêneros e espécies de aproximadamente 250 e 100.000, respectivamente (LEBEAU e ROBERT, 2003). As diatomáceas estão entre os grupos de microalgas relativamente mais ricas em ácidos graxos, principalmente (EPA, 20:5n-3) e (DHA, 22:6n-3), ambos apresentando variação de 5%-35% do total de ácidos graxos poliinsaturados (PATIL et al., 2005;

HEMAISWARYA et al., 2011). Os ácidos graxos poliinsaturados de cadeia longa da família ω-3: ALA (α-linolenico), EPA (eicosapentaenoico) e DHA (docosaexaenoico), e da família ω-6: ARA (araquidônico) e LA (linoleico), são essenciais para uma ótima nutrição, crescimento, tolerância ao stress, para os organismos cultivados (BELL e SARGENT, 2003), porém poucos animais são capazes de sintetizar esses ácidos graxos de cadeia longa e deve obter esses ácidos graxos através da sua dieta (BRETT e MÜLLER-NAVARRA, 1997).

Piña et al. (2005), compararam a sobrevivência, desenvolvimento, comprimento e peso final das três fases de protozoea de *L. vannamei* alimentados com três microalgas *Isochrysis* sp., *Tetraselmis suecica* e *Chaetoceros muelleri*, fornecidas como dietas monoalgal e misturadas, os melhores resultados foram obtidos nos tratamentos com *C. muelleri*, sendo observado mortalidade de 100% nos tratamentos onde só foram ofertados *T. suecica*, atribuído a falta de PUFAs nesta microalga. Kent et al. (2011) verificaram que quando juvenis de *L. vannamei*, foram imersos em tanques contendo monoculturas de *Thalassiosira weissflogii*, *Amphiprora* sp., *Nannochloropsis salina* e *Synechococcus bacillarus*, ingeriram e digeriram as diatomáceas *T. weissflogii* e *Amphiprora* sp., contudo, mesmo com a ingestão de *S. bacillarus* e *N. salina* pelos camarões, não haviam evidência de digestão destas células, significando a importância das diatomáceas como fonte de alimento para estes animais, até mesmo depois da fase larval.

Já em sistemas de bioflocos, Godoy et al. (2011) inoculando as diatomáceas no cultivo de *L. vannamei*, alegaram que as diatomáceas garantiram o melhor desempenho zootécnico dos camarões cultivados. Fato também observado por Khatoon et al. (2009), onde após uma análise bioquímica revelaram que as PLs de *Penaeus monodon* cultivadas em tanques contendo diatomáceas, possuíam alta valor de lipídios, carboidratos e proteínas que garantiram maior crescimento e sobrevivência dos animais cultivados. Ju et al. (2008) obtiveram as maiores taxas de crescimento em *L. vannamei* alimentados com flocos que continham em sua composição biomassa fitoplanctônica (246g Kg), biomassa bacteriana (30g Kg), cinzas (392g Kg), enquanto que o restante (332g Kg) consistia de

detritos e zooplâncton. Análises de pigmentos e de microscopia realizadas pelos autores revelaram respectivamente, que as diatomáceas se prevaleceram com 82,4% com relação às clorofíceas 7,9% e que os gêneros mais representativos foram *Thalassiosira*, *Chaetoceros* e *Navicula*.

3- Referência bibliográfica

ALONSO, M.; LAGO, F.C.; VIEITES, J.M.; ESPIÑEIRA, M. Molecular characterization of microalgae used in aquaculture with biotechnology potential. **Aquaculture International**, v.20, p.847–857, 2012.

ASLAN, S.; KAPDAN, I.K. Batch kinetics of nitrogen and phosphorus removal from synthetic wastewater by algae. **Ecological Engineering**, v.28, p.64–70, 2006.

AVNIMELECH, Y. Control of microbial activity in aquaculture systems: active suspension ponds. **World Aquaculture**, v.34, p.19-21, 1993.

AVNIMELECH, Y.; KOCHVA, M.; DIAB, S. Development of controlled intensive aquaculture systems with a limited water exchange and adjusted carbon to nitrogen ratio. **Aquaculture Bamidgeh**, v.46, p.119-131, 1994.

AVNIMELECH, Y. Carbon/nitrogen ratio as a control element in aquaculture systems. **Aquaculture**, v. 176, p. 227-235, 1999.

AVNIMELECH, Y. Nitrogen control and protein recycling: Activated suspension ponds. **The Advocate,** v. 3, n. 4, p. 23-24, 2000.

AVNIMELECH, Y. **Biofloc Technology - A Practical Guide Book**. 1a ed., Baton Rouge: The world Aquaculture Society. 2009, 182p.

AVNIMELECH, Y. **Biofloc Technology - A Practical Guide Book**. 2a ed., Baton Rouge: The World Aquaculture Society, 2012.

BALLESTER, E.L.C.; ABREU, P.C.; CAVALLI, R.O.; EMERENCIANO, M.; ABREU, L.; WASIELESKY, W. Effect of practical diets with different protein levels on the performance of *Farfantepenaeus paulensis* juveniles nursed in a zero exchange suspended microbial flocs intensive system. **Aquaculture Nutrition**, v.16, p.163–172, 2010.

BALOI, M.; ARANTES, R.; SCHVEITZERB, R.; MAGNOTTI, C.; VINATEA, L. Performance of Pacific white shrimp *Litopenaeus vannamei* raised in biofloc systems with varying levels of light exposure. **Aquacultural Engineering**, v.52, p.39-44, 2013.

BAUER, W.; PRENTICE-HERNANDEZ, C.; TESSER, M.B.; WASIELESKY Jr. W.; POERSCH Substitution of fishmeal with microbial floc meal and soy protein concentrate in diets for the pacific white shrimp *Litopenaeus vannamei*. **Aquaculture**, v.342-343, p.112 -116, 2012.

BELETTINI, F.; DERNER, R.B.; VINATEA, L.A. Utilização das microalgas *Thalassiosira*4 *weissflogii* e *Nannochloropsis oculata* no cultivo de *Litopenaeus vannamei* em sistemas de berçários, sem renovação de água. **Atlântica**, v.33, n.2, p.101-114, 2011.

BELL J.G.; SARGENT, J.R. Arachidonic acid in aquaculture feeds: current status and future opportunities. **Aquaculture**, v.218, p. 491–499, 2003.

BENEMANN, J.R. Microalgae aquaculture feeds. **Journal of Applied Phycology**, v. 4, p.233-245, 1992.

BOROWITZKA, M.A. Microalgae for aquaculture: opportunities and constraints. **Journal of Applied Phycology**, n.9, p.393-401, 1997.

BRITO, L.O.; COSTA, W.M.; OLIVERA, A. Water quality in *Litopenaeus vannamei* nurseries using different fertilization strategies. **World Aquaculture**, v.41, p.22-25, 2010.

BRETT, M.; MÜLLER-NAVARRA, D.C. The role of highly unsaturated fatty acids in aquatic food web process. **Freshwater Biology**, v.38, p.483-499.

BURFORD, M. A.; THOMPSON, P. J.; MCINTOSH, R. P.; BAUMAN, R. H.; PEARSON, D. C. Nutrient and microbial dynamics in high-intensity, zero-exchange shrimp ponds in Belize. **Aquaculture**, v.219, p.393-411, 2003.

CAÑAVATE, J.P.; FERNÁNDES-DÍAZ, C. Pilot evaluation of freeze-dried microalgae in the mass rearing of gilthead seabream (*Sparus aurata*) larvae. **Aquaculture**, v.193, p.257–269, 2001.

CHAMBERLAIN, G. W.; HOPKINS, J. S. Reducing water use and feed cost in intensive ponds. **World Aquaculture,** v. 25, p. 29-32, 1994.

CHIEN, Y.H.; PAN, C.H.; HUNTER, B. The resistance to physical stresses by *Penaeus monodon* juveniles fed diets supplemented with astaxanthin. **Aquaculture**, v. 216, p.177-191, 2003.

COHEN,J.M.; SAMOCHA, T.M.; FOX, J.M.; GANDY, R.L.; LAWRENCE, A.L. Characterization of water quality factors during intensive raceway production of juvenile *Litopenaeus vannamei* using limited discharge and biosecure management tools. **Aquacultural Engeneering**, v.32, p.425-442, 2005.

COSTA, W.C.; FIGUEIREDO, M.B.; CAVALLI, R.O.; GÁLVEZ, A.O. Crescimento populacional de rotíferos *Brachionus plicatilis* Müller,1786, alimentados com microalgas e dieta formulada. **Revista Brasileira de Ciências Agrárias**, v.3, n.2, p.173-178, 2008.

CRAB, R.; DEFOIRDT, T.; BOSSIER, P.; VERSTRAETE, W. Biofloc technology in aquaculture: Beneficial effects and future challenges. **Aquaculture**, v. 356-357, p. 351-356, 2012.

CUZON, G.; GOGUENHEIM, J.; GAXIOLA, G. AQUACOP. 2008. Floc contribution to penaeid intensive culture. p.365-381. Editores L. Elizabeth Cruz Suárez, Denis Ricque Marie,Mireya Tapia Salazar,Martha G. Nieto López, David A. Villareal Cavazos, Juan Pablo Lazo y Ma. Teresa Viana. Avances en Nutrición Acuícola IX. IX **Simposio Internacional de Nutrición Acuícola** 24-27 Noviembre. Universidad Autónoma de Nuevo León, Monterrey, Nuevo León, México. 2008.

DE SCHRYVER, P.; CRAB, R.; DEIFOIRDT, T.; BOON, N.; VERSTRAETE, W. The basics of bio-flocs technology: The added value for aquaculture. **Aquaculture**, v. 277, p.125-137, 2008.

DEHGHAN, M.; JAFARYAN, H.; SAHANDI, J.; JAMALI, H.; ADINEH, H.; FARAMARZI, M. Evaluation of growth and survival rate of artemia parthenogenetica feed with microalgae (*Isochrysis galban*a and *Chlorella vulgaris*) and bakery yeast (*Saccharomyces cerevisiae*). **AACL Bioflux**, v.4, p.463-468, 2011.

DRAAISMA, R.B.; WIJFFELS, R.H.; SLEGERS, P.M.; BRENTNER, L.B.; ROY, A.; BARBOSA, M.J. Food commodities from microalgae. **Opinion in biotechnology**, v.24, p. 169-177, 2013. SEMI- INTENSIVE MONOCULTURE OF FRESHWATER SHRIMPS IN NIGERIA: A REVIEW.

ELEZUO, K.O. Biotic communities and feeding habits of *Litopenaeus vannamei* (Boone 1931) and *Litopenaeus stylirostris* (Stimpson 1974) in monoculture and polyculture semi-intensive ponds. **Continental Journal Biological Sciences**, v.4, n.2, p.13 - 21, 2011.

FRÓES, C.; FÓES, G.; KRUMMENAUER, D.; POERSCH, L.H.; WASIELESKY Jr. Densidade de estocagem na engorda de camarão-branco cultivado em sistema de biofloco. **Pesquisa Agropecuária Brasileira**, v. 48, n.8, p. 878-884, 2013.

FRÓES, C.N. Aprimoramento das técnicas de manejo do cultivo do camarão branco *Litopenaeus vannamei* (Boone) em sistema de bioflocos. 2012. 69p. **Dissertação (Doutorado) -** Universidade Federal do Rio Grande do Sul, Rio Grande do Sul.

FURTADO, P.S.; POERSCH, L.H.; WASIELESKY Jr., W. Effect of calcium hydroxide, carbonate and sodium bicarbonate on water quality and zootechnical performance of shrimp *Litopenaeus vannamei* reared in bio-flocs technology (BFT) systems. **Aquaculture**, v. 321, p. 130-135, 2011.

GAGNEUX-MOREAUX, S.; MOREAU, C.; GONZALEZ, J.L.; COSSON, R.P. Diatom artificial medium (DAM): a new artificial medium for the diatom *Haslea ostrearia* and other marine microalgae. **Journal of Applied Phycology**, v.19, n.5, p.549-556, 2007.

GODOY, L.C.; ODEBRECHT, C.; BALLESTER, E.L.C.; MARTINS, T.G.; WASIELESKY, W. Effect of diatom supplementation during the nursery rearing of *Litopenaeus vannamei* (Boone, 1931) in a heterotrophic culture system. **Aquaculture International**, v.20, n.3, p.559-569, 2011.

GOUVEIA L.; BATISTA, A.P.; MIRANDA, A.; EMPIS, J.; RAYMUNDO, A. *Chlorella vulgaris* biomass used as colouring source in traditional butter cookies. **Innovative Food Science & Emerging Technologie**s, v.8, p.433-436, 2007.

GRIFFITHS, M.J.; HILLE, R.P.; HARRISON, S.T.L. Lipid productivity, settling potential and fatty acid profile of 11 microalgal species grown under nitrogen replete and limited conditions. **Journal Applied Phycology**, v.24, p.989-1001, 2012.

HARGREAVES, J.A. Photosynthetic suspended-growth systems in aquaculture. **Aquaculture Engineering**, v.34, p.344-363, 2006.

HEMAISWARYA, S.; RAJA, R.; KUMAR, R.R.; GANESAN, R.; ANBAZHAGAN, C. Microalgae: a sustainable feed source for aquaculture. **World Journal Microbiology Biotechnology**, v.27, p.1737-1746, 2011.

HOPKINS, J.E.; HAMILTON, R.; SANDIFER, P.; BROWDY, C.; STOKES, A. Effect of water exchange rate on production, water quality, effluent characteristics and nitrogen budgets of intensive shrimp ponds. **Journal of the World Aquaculture Society,** v. 24, n. 3, p. 304-320, 1993.

INTERAMINENSE, J.A.; CALAZANS, N.F.; VALLE, B.C.; VOGELEY, J.L.; PEIXOTO, S.; SOARES, R.; FILHO, V.L. *Vibrio* spp. Control at Brine Shrimp, *Artemia*, Hatching and Enrichment. **Journal of the World Aquaculture Society**, v..45, n. 1, p. 65-74, 2014.

JU, Z.Y.; FORSTER, I.; CONQUEST, L.; DOMINY, W. Enhanced growth effects on shrimp (*Litopenaeus vannamei*) from inclusion of whole shrimp floc or floc fractions to a formulated diet. **Aquaculture Nutrition,** v. 14, p. 533–543, 2008.

JU, Z.Y.; FORSTER, I.P.; DOMINY, W.G. Effects of supplementing two species of marine algae or their fractions to a formulated diet on growth, survival and composition of shrimp (*Litopenaeus vannamei*). **Aquaculture**, v. 292, p. 237–243, 2009.

KENT, M.; BROWDY, C.L.; LEFFLER, J.W. Consumption and digestion of suspended microbes by juvenile Pacific white shrimp *Litopenaeus vannamei*. **Aquaculture**, v. 319, n.3-4, p. 363-368, 2011.

KHATOON, H.; BANERJEE, S.; YUSOFF, F.M.; SHARIFF, M. Evaluation of indigenous marine periphytic *Amphora, Navicula* and *Cymbella* grown on substrate as feed supplement in *Penaeus monodon* postlarvae hatchery systems. **Aquaculture Nutrition**, v.15, p.186-193, 2009.

KHATOON, H.; BANERJEE, S.; YUSOFF, F.M.; SHARIFF, M. Use of microalgal-enriched *Diaphanosoma celebensis* Stingelin, 1900 for rearing *Litopenaeus vannamei* (Boone, 1931) postlarvae. **Aquaculture Nutrition**, v.19, p.163–17, 2013.

KRUMMENAUER, D.; PEIXOTO, S.; CAVALLI, R.; POERSCH, L.H.; WASIELESKY, W. Super intensive culture of White shrimp, *Litopenaeus vannamei,* in a biofloc technology system in Southern Brazil at different stocking densities. **Journal of the World Aquaculture Society,** v. 42, n.5, p.726-733, 2011.

KRUMMENAUER, D.; SEIFERT JUNIOR, C.A.; POERSCH, L.H.; FOES, G.K.; LARA, G.R.; WASIELESKY Jr. W. Cultivo de camarões marinhos em sistema de bioflocos: Análise da reutilização da água. **Atlantica**, v.34, p.103-111, 2012.

KRUMMENAUER, D.; SAMOCHA, T.; POERSCH, L.P.; LARA, G.; WASIELESKY Jr, W. The reuse of water on the culture of Pacific White Shrimp, *Litopenaeus vannamei*, in BFT system. **Journal of the World Aquaculture Society**, v. 45, n.1, p.3-14, 2014.

KUBAN, F.J.; WILKENFELD, J.; LAWRENCE, A. Survival and growth of *Penaeus setiferus* and *Penaeus aztecus* larvae fed *Artemia* beginning at the protozoea two substage versus the mysis one substage. **World Mariculture Society**, n.14, p. 38-42, 1983

LAING, I.; HELM, M.M. Factors affecting the semi-continuous production of *Tetraselmis suecica* (Kylin) Buth in 200 L vessels. **Aquaculture**, v.22, p.137–148.

LAVÍN, P.L.; LOURENÇO, S.O. An evaluation of the accumulation of intracellular inorganic nitrogen pools by marine microalgae in batch cultures. **Brazilian Journal of Oceanography**, v.53, p. 55-68, 2005.

LEBEAU, T.; ROBERT, J. M. Diatom cultivation and biotechnologically relevant products. Part II: Current and putative products. **Applied Microbiology and Biotechnology, v.** 60, p. 624-632, 2003.

LEZAMA-CERVANTES, C.; PANIAGUA-MICHEL, J. Effects of constructed microbial mats on water quality and performance of *Litopenaeus vannamei*. **Journal Aquaculture Engineering,** v.42, p.75-81, 2010.

MAKRIDIS, P.; COSTA, R.A.; DINIS, M.T. Microbial conditions and antimicrobial activity in cultures of two microalgae species. *Tetraselmis chuii* and *Chlorella minutissima*, and effect on bacterial load of enriched *Artemia* metanauplii. **Aquaculture**, v.255, p.76–81, 2006.

MARTÍNEZ-CÓRDOVA, L.R.; CAMPAÑA-TORRES, A.; MARTÍNEZ-PORCHAS, M.; LÓPEZ-ELÍAS, J.A.; GARCÍA-SIFUENTES, C.O. Effect of alternative mediums on production and proximate composition of the microalgae *Chaetoceros muelleri* as food in culture of the copepod *Acartia* sp. **Latin American Journal of Aquatic Research**, v.40, n.1, p.169-176, 2012.

MARTÍNEZ-CÓRDOVA, L.R.; PEÑA-MESSINA. Biotic communities and feeding habits of *Litopenaeus vannamei* (Boone 1931) and *Litopenaeus stylirostris* (Stimpson 1974) in monoculture and polyculture semi-intensive ponds. **Aquaculture Research**, v. 36, n. 11, p. 1075-1084, 2005.

MOSS, S.M; ARCE, B.J.; ARGUE, C.A.; OTOSHI, F.; CALDERON, R.O.; TACON, A.G.J. 2001. **Greening of the blue revolution:** efforts toward environmentally responsible shrimp culture. In: BROWDY, C.L.; JORY, D.E. The New Wave: Proceedings of the Special Session on Sustainable Shrimp Culture. The World Aquaculture Society, Baton Rouge, Louisiana, USA. 2001. p. 1–19.

MULLER-FEUGA, A. Microalgae for aquaculture. 2004. In: RICHMOND, A. (Ed). **Handbook of microalgal culture**: biotechnology and applied phycology. Oxford: Blackwell Science, p.352–364.

NIU, J.; TIAN, L.X.; LIU, Y.J. Effect of dietary astaxanthin on growth, survival and stress tolerance of postlarval shrimp, *Litopenaeus vannamei*. **Journal of the World Aquaculture Society**, v.40, p.795–802, 2009.

NORIEGA-CURTIRS, P. Primary productivity and related fish yield in intensely manured fishponds. **Aquaculture**, v.17, p.335–344, 1979.

PATIL, V.; REITAN, K.I.; KNUTSEN, G.; MORTENSEN, L.M.; KÄLLQVIST. Microalgae as source of polyunsaturated fatty acids for aquaculture. **Current Topics in Plant Biology,** v 6, p. 57–65, 2005.

PATIL, V.; KALLQVIST, T.; OLSEN, E.; VOGT, G.; GISLEROD, H.R. Fatty acid composition of 12 microalgae for possible use in aquaculture feed. **Aquaculture International**, v.15, p.1-9, 2007.

PEÑAFLORIDA, V.D. Interaction between dietary levels of calcium and phosphorus on growth of juvenile shrimp, *Penaeus monodon*. **Aquaculture**, v. 172, n.281–289, 1999.

PERNET, F.; MALET, N.; PASTOUREAUD, A.; VAQUER, A.; QUÉRÉ, C.; DUBROCA, L. Marine diatoms sustain growth of bivalves in a Mediterranean lagoon. **Journal of Sea Research**, v.68, p. 20–32, 2012.

PETTERSEN, A.K.; TURCHINI, G.M.; JAHANGARD, S.; INGRAM, B.A.; CRAIG, D.H. Effects of different dietary microalgae on survival, growth, settlement and fatty acid composition of blue mussel (*Mytilus galloprovincialis*) larvae. **Aquaculture**, v.309, p.115-124, 2010.

PIÑA, P.; VOLTOLINA, D.; NIEVES, M. ROBLES, M. Survival, development and growth of the Pacific White shrimp *Litopenaeus vannamei* protozoea larvae, fed with monoalgal and mixed diets. **Aquaculture**, v.253, p. 523-530, 2006.

PRIMAVERA, J.H. Overcoming the impacts of aquaculture on the coastal zone. **Ocean & Coastal Management**, v.49, p.531–545, 2006.

PULZ, O.; GROSS, W. Valuable products from biotechnology of microalgae. **Applied Microbiology and Biotechnology**, v.65, p.635–48, 2004.

RAY, A. J.; SEABORN, G.; LEFFLER, J. W.; WILDE, S. B.; LAWSON, A.; BROWDY, C. L. Characterization of microbial communities in minimal exchange, intensive aquaculture systems and the effects of suspended solids management. **Aquaculture**, v. 310, p. 130-138, 2010.

RICHMOND, A. **Handbook of microalgal culture:** biotechnology and applied phycology. Blackwell Science Ltda, 2004.

ROMERO, R.; YUFERA, M. Contribution of gut content to the nutritional value of *Brachionus plicatilis* used as prey in larviculture. **Aquaculture**, v.364-365, p.124-126, 2012.

SAMOCHA, T.M.; WILKENFELD, J.S; MORRIS, T.C.; CORREIA, E.S.; HANSON, T. Intensive raceways without water exchange analyzed for white shrimp culture. **Global Aquaculture Advocate**, v.13, p.22–24, 2010.

SAMOCHA, T.M.; SCHVEITER, R.; KRUMMENAUER, D.; MORRIS, T.C.; WOODRING, S. Recent advances in super-intensive raceway systems for production of marketable-size *Litopenaeus vannamei* under no water exchange. **The Practical Asian Aquaculture Network, Thailand**, v.2, p.20–23, 2011.

SCHNEIDER, O.; SERETI, V.; EDING, E.H.; VERRETH, J.A.J. Analysis of nutrient flows in integrated intensive aquaculture systems. **Aquaculture Engineering,** v.32, p. 379-401, 2005.

SILVA, K.R.; WASIELESKY Jr. W.; ABREU, P.C. Nitrogen and phosphorus dynamics in the biofloc production of the Pacific white shrimp *Litopenaeus vannamei*. **Journal of the World Aquaculture Society**, v. 44, n. 1, p. 30-41, 2013.

SILVA, A.F.; LARA, G.R.; BALLESTER, E.C.; KRUMMENAUER, D.; ABREU, P.C.; WASIELESKY JR, W. Efeito das altas densidades de estocagem no crescimento e sobrevivência de *Litopenaeus vannamei* na fase final de engorda, cultivados em sistemas de bioflocos (BFT). **Ciência Animal Brasileira**, v.14, n.3, p.279-287, 2013.

SOARES, R.; PEIXOTO, S.; WASIELESKY, W.; D'INCAO. Effect of different food items on the survival and growth of *Farfantepenaeus paulensi*s (Pérez-Farfante 1967) postlarvae. **Aquaculture Research**, v.37, p.1413-1418, 2006.

SPOLAORE, P.; JOANNIS-CASSAN, C.; DURAN, E.; ISAMBERT, A. Commercial applications of microalgae. **Journal of Bioscience and Bioengineering**, v. 101, n.2, p.87-96, 2006.

TACON, A. G. J.; CODY, J.; CONQUEST, L.; DIVAKARAN, S.; FORSTER, I. P.; DECAMP, O. Effect of culture system on the nutrition and growth performance of Pacific white shrimp *Litopenaeus vannamei* (Boone) fed different diets. **Aquaculture Nutrition,** v. 8, p. 121-137, 2002.

THOMPSON, F.L.; ABREU, P.C.; WASIELESKY, W. Importance of biofilm for water quality and nourishment in intensive shrimp culture**. Aquaculture**, v.203, n.3, p. 263-278, 2002.

TIMMONS, M.B.; LOSORDO, T.M. Aquaculture water reuse systems: engineering design and management. Elsevier, Amsterdam, Netherlands, 1994.

VENERO, J. A.; McABEE, B.; LAWSON, A.; LEWIS, B. L.; STOKES, A. P.; LEFFLER, J. W.; BROWDY, C.L. Greenhouse-enclosed superintensive shrimp production: Alternative to traditional ponds in U.S, **Advocate**, v. 12, p. 61-63, 2009.

VIAU, V.E.; DE SOUZA, D.M.; RODRIGUEZ, E.M.; WASIELESKY, W.; ABREU, P.C.; BALLESTER, E.L.C. Biofilm feeding by postlarvae of the Pink shrimp *Farfantepenaeus brasiliensis* (Decapoda, Penaidae). **Aquaculture Research**, v.44, n.5, p.783-794, 2013.

WASIELESKY, W.; ATWOOD, H.; STOKES, A.; BROWDY, C.L. Effect of natural production in a zero exchange suspended microbial floc based super-intensive culture system for white shrimp *Litopenaeus vannamei*. **Aquaculture**, v.258, p. 396-408, 2006a.

WASIELESKY,W.; ATWOOD, H.; KEGL, R.; BRUCE,J.; STOKES, A.; BROWDY, C.L. Efeito do pH na sobrevivência e crescimento do camarão branco Litopenaeus vannamei em cultivos superintensivos. In: Aquaciência 2006. **Anais**. Bengo Gonçalves/RS, 2006b

YIN, X.W.; MIN, W.W.; LIN, H.J.; CHEN, W. Population dynamics, protein content, and lipid composition of *Brachionus plicatilis* fed artificial macroalgal detritus and *Nannochloropsis* sp.diets. diets. **Aquaculture**, v.380-383, p.62-69, 2013.

YUSOFF, FM.; ZUBAIDAH, M.S.; MATIAS, H.B.; KWAN, T.S. Phytoplankton succession in intensive marine shrimp culture ponds treated with a commercial bacterial product. **Aquaculture Research**, v. 33, p. 269-278, 2002.

ZHOU, X.X.; WANG, Y.B.; LI, W.F. Effect of probiotic on larvae shrimp (*Penaeus vannamei*) based on water quality, survival rate and digestive enzyme activities. **Aquaculture**, v. 287, p. 349-353, 2009.

4- Artigo científico

Os resultados obtidos do trabalho experimental dessa dissertação são apresentados no artigo intitulado: Effect of addition of *Navicula* sp. on plankton composition and postlarvae growth of *Litopenaeus vannamei* reared in culture tanks in zero water exchange (manuscrito), que se encontra anexado.

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Effect of addition of *Navicula* sp. on plankton composition and postlarvae growth of *Litopenaeus vannamei* reared in culture tanks in zero water exchange

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ABSTRACT. The aim of this study was to evaluate the effect of the addition of *Navicula* sp. on plankton composition and postlarvae growth of *Litopenaeus vannamei* reared in culture tanks in zero water exchange systems. Four treatments were considered: zero water exchange (ZWE); ZWE with the addition of feed (ZWE-F); ZWE with the addition of *Navicula* sp. (ZWE-N) and ZWE with the addition of feed and *Navicula* sp. (ZWE-FN), all in triplicate. Shrimp of 17.7 ± 0.02 mg were stocked at a density of 2500 shrimp m^{-3} and the microalgae were added on the 1st, 5th and 15th days at a density of 5 x 10^4 cell.mL^{-1}. The shrimp were fed a commercial feed composed of 42% crude protein four times a day except in the ZWE treatment. For data analysis we used Cochran, Shapiro-Wilk, ANOVA, Tukey and Student-t tests ($P < 0.05$). The most frequent genera were: *Anabaena, Arcella, Asplanchma, Bosmina, Brachionus, Cylindrotheca, Daphnia, Fragilaria, Hemiaulus, Keratella, Orthoseira, Oscillatoria, Phymatodocis, Rhabdonema, Skeletonema, Sckizothrix* and *Ulothrix.* Significant differences between treatments were observed for TAN, NO_2-N, alkalinity, final weight, weight gain, final biomass, biomass gain, feed conversion ratio, specific growth rate and survival. The ZWE-FN treatment showed better production parameters, indicating the benefits of addition of *Navicula* sp as a natural food source for postlarvae *L. vannamei* in zero water exchange systems.

Keywords: *Navicula*, Phytoplankton, Zooplankton, Cyanobacteria, Shrimp, Zero water exchange.

Efecto de la adición de *Navicula* sp. sobre la composición del plancton y el crecimiento de las postlarvas de *Litopenaeus vannamei* criadas en tanques de cultivo sin recambio de agua

RESUMEN. El objetivo de este estudio fue evaluar el efecto de la adición de *Navicula* sp. en la composición del plancton y el crecimiento de las postlarvas de *Litopenaeus vannamei* en estanques de cultivo sin recambio de agua. Se realizaron cuatro tratamientos: sin recambio de agua (ZWE); ZWE más adición de ración alimenticia (ZWE-F); ZWE más la adición diatomea *Navicula* sp.

(ZWE-N) y ZWE más adición ración alimenticia y más la *Navicula* sp. (ZWE-FN), todos con tres repeticiones. Los camarones con peso de 17,7 ± 0,02 mg fueron sembrados a una densidad de 2500 camarones m^{-3}, las microalgas fueron adicionadas el 1º, 5º y 15º dias de cultivo a una densidad de 5 x 10^4 cel.mL^{-1}. Los camarones se alimentaron con una ración comercial con 42% de proteína cruda cuatro veces al día. Para los análisis estadísticos se utilizaron las pruebas de Cochran, Shapiro Wilk, ANOVA, Tukey y t de Student (P <0,05). Los géneros más frecuentes fueron: *Anabaena, Arcella, Asplanchma, Bosmina, Brachionus, Cylindrotheca, Daphnia, Fragilaria, Hemiaulus, Keratella, Orthoseira, Oscillatoria, Phymatodocis, Rhabdonema, Skeletonema, Sckizothrix* and *Ulothrix.* Se encontraron significativas observadas entre los tratamientos para TAN, NO_2-N, alcalinidad, peso final, ganancia de peso, biomasa final, ganancia de biomasa, el índice de conversión, tasa de crecimiento específico y supervivencia. El tratamiento ZWE-FN mostró los mejores parámetros de producción, resaltando los beneficios de la adición de la *Navicul*a sp con fuente de alimento natural para el postlarvas *L. vannamei* sin recambio de agua.

Palabras clave: *Navicula*, Fitoplancton, Zooplancton, Cyanobacteria, Camarones, Sin recambio de agua.

INTRODUCTION

Large quantities of formulated feed with high animal protein content can cause eutrophication in aquaculture systems, increasing the nutrient load in effluents (Tacon *et al*., 2002). Their use increases production costs (Audelo-Naranjo *et al*., 2012) and can result in an insufficient supply of some essential nutrients (Crab *et al*., 2007), thus becoming a limiting factor in intensive systems. To minimize or reduce this nutrient deficiency, organic and inorganic fertilizers can be added to the cultivation systems to promote growth of the microbial community, which is a food source (Brito *et al*., 2009a,b; Asaduzzaman *et al*., 2010; Lara-Anguiano *et al*., 2013). Shrimp can feed on natural biota such as phytoplankton, zooplankton and bacteria present in culture systems (Otoshi *et al*., 2011). This biota can supply some of the shrimps' nutritional needs (Martínez- Cordova & Enríquez-Ocaña, 2007), and improve the activity of digestive enzymes (Xu *et al*., 2012).

In intensive farming systems with Pacific white shrimp (*Litopenaeus vannamei*), microalgae (through photosynthesis) and the other constituents of the microbial community can play an important role in recycling nutrients (Audelo-Naranjo *et al*., 2012; Sánchez *et al*., 2012), decreasing the anoxic zones in ponds and alleviating the nutrient load in wastewater (Martínez-Porchas *et al*., 2010), while providing a nutrition source for shrimp in semi-intensive (Otoshi *et al*., 2011) and intensive systems (Sánchez *et al*., 2012).

Depending on the species and culture conditions, benthic diatoms contain an average of 32 to 38% crude protein (Gordon *et al*., 2006). However, Khatoon *et al*. (2009) found that *Navicula sp.*, grown in a Conway culture medium contain 494 g of crude protein, 259 g of lipids and 111 g of carbohydrates per kilogram of dry matter, and the profile of polyunsaturated fatty acids includes 82 g of EPA and 22 g of DHA for each kilogram of total fatty acids. Despite the importance of diatoms, little attention has been paid to them in zero water exchange systems, mainly due to the reduced availability of light and the predominance of heterotrophic bacteria.

In zero or minimal exchange systems, the main forms of nitrogen removal are photosynthetic and heterotrophic bacterial activities (Cohen *et al*., 2005; Becerra-Dorame *et al*., 2011). For this reason, in zero or minimal water exchange, it is necessary to know the components of the natural community and understand the role of each one in the entire ecosystem (Avnimelech, 2009; Crab *et al*., 2012).

In this respect, the aim of this study was to evaluate the effect of the addition of the benthic diatom *Navicula* sp. on the plankton composition and postlarvae growth of *Litopenaeus vannamei* reared in culture tanks in zero water exchange.

MATERIALS AND METHODS

Experimental Conditions

An indoor trial was conducted for 20 days at the Sustainable Mariculture Laboratory (LAMARSU) of the Fisheries and Aquaculture Department (DEPAq) of the Rural Federal University at Pernambuco (UFRPE), Recife, Brazil (08°01'00.16¨S, 034°56'57.74"W). The experimental design was completely randomized with four treatments: zero water exchange (ZWE); ZWE with the addition of feed (ZWE-F); ZWE with the addition of *Navicula* sp. (ZWE-N) and ZWE with the addition of feed and *Navicula* sp. (ZWE-FN), all in triplicate.

Five days prior to stocking shrimp, water from a matrix tank (TAN 0.12 mg L^{-1}, NO_2-N 2.26 mg L^{-1}, alkalinity 100 mg $CaCO_3$ L^{-1} and settleable solids 27 mL L^{-1}) was mixed and equally distributed to fill twelve black-plastic tanks (50cm x 35cm x 23cm) up to approximately 50% of the volume, and completed with 50% sea water (with a salinity of 35 g L^{-1}, and which was filtered and treated with a chlorine solution of 10 mg L^{-1}, then dechlorinated and aerated for 48 h).

Aeration was supplied with three airstones from a 2-HP blower. There was no water exchange during the experimental period, but dechlorinated freshwater was added to compensate for evaporation. The light intensity was kept at ~ 1000 lux using a fluorescent lamp with a 12-hour light/dark photoperiod.

Shrimp stocking, feeding, and addition of organic carbon

Specific pathogen-free postlarvae (17.7 ± 0.02 mg) of *L. vannamei* were obtained from a commercial laboratory (Potiporã, Barra de Sirinhaém, PE, Brazil) and stocked at a density of 2,500 shrimp m^{-3}. The postlarvae were fed four times a day (at 0800, 1100, 1400 and 1700h), with a commercial shrimp feed with 42% crude protein (Aquavita Premiun, Guaraves, Paraíba, Brazil) based on Van Wyk's table (1999) and adjusted daily according to estimated shrimp consumption, mortality rate and leftover feed. Molasses (40% organic carbon) was added once a day to establish a 12:1 C:N ratio in the experimental units throughout the culture period, assuming that 50% of the amount of feed is organic carbon and 1 kg of the 42% crude protein feed with 6.25%-N has 67.2 g of nitrogen, there is a need for 306.4g organic carbon, or 766.1 g of molasses (Samocha et al. 2007; Avnimelech 2009).

Shrimp performance parameters

Shrimp weight was monitored at the end of the experiment, when biomass gain, specific growth rate (SGR), mean final weight, weekly growth, feed conversion ratio (FCR), survival and yield were determined based on the following equations: Biomass gain (g) = final biomass (g) – initial biomass (g); SGR (% day^{-1}) = 100 x [ln final weight (g) – ln initial weight (g)] / time (days); Final weight (g) = final biomass (g) / survival; Weekly growth (g $week^{-1}$) = biomass gain (g) / times (weeks) of culture; FCR = feed supplied (dry weight)/ biomass gain; Survival (%) = (number of individuals at the end of the evaluation period / initial number of individuals stocked) x 100; Yield (Kg m^{-3}) = final biomass (kg) / volume of experimental unit (m^3).

Diatom addition

The benthic diatoms (*Navicula sp.*) were obtained from LAMARSU-DEPAq-UFRPE and cultured in a Conway medium (Walne, 1966) containing g L^{-1} $FeCl_3.6H_2O$ 1.30; $MnCl_2.4H_2O$ 0.36; H_3BO_3 33.6; EDTA 45.0; $NaH_2PO_4.2H_2O$ 20.0; $NaNO_3$ 100.0; $ZnCl_2$ 1.1; $CoCl_2.6H_2O$ 1.0; $(NH_4)6M_{O7}O_{24}.4H_2O$ 0.45; CuSO4.5H2O 1.0; $Na_2SiO_3.5H_2O$ 2.0; vitamins B12 0.1 and B1 1.0, which was used in a 1.0 mL L^{-1} solution, maintained in water with 30 g L^{-1} salinity, pH 7.9, temperature 25 ± 1°C and the light intensity was kept at ~ 2000 lux using a fluorescent lamp with a 12-hour light/dark photoperiod. Diatoms were added on days 1, 5, 10 and 15 of cultivation in the experiment in the (ZWE-N) and (ZWE-FN) tanks at a concentration of 5 x 10^4 $mL^{-1,}$ corresponding to an addition of approximately 400 mL of microalgae to the tanks.

Water quality monitoring

Dissolved oxygen and temperature were monitored with a DO meter (YSI model 55, Yellow Springs, Ohio, USA) twice a day (8:00 and 16:00 h). Salinity (YSI 30 model 30/50, Yellow Springs, Ohio, USA), pH (pH meter YSI model 100, Yellow Springs, Ohio, USA), total ammonia nitrogen (TAN), nitrite-nitrogen (NO_2-N) and alkalinity ($CaCO_3$) were monitored every five days using a spectrophotometer (ALFAKIT- AT10P, Brazil) and a compact alkalinity kit (ALFAKIT, Brazil), respectively.

Phytoplankton, Zooplankton and Cyanobacteria monitoring

Vertical water sampling was performed at the start and end of the experiment using plastic bottles with a volume of 500 mL for phytoplankton, zooplankton and Cyanobacteria collection. The water was filtered through a cylindrical-conical net (mesh: 15 µm for phytoplankton and Cyanobacteria, 50 µm for zooplankton) to 10 mL, to obtain a 50-fold concentration. The phytoplankton, zooplankton and Cyanobacteria were fixed with formalin (4%), buffered with borax (1%) and stored in 10-mL plastic recipients. A Sedgewick-Rafter chamber and stereomicroscope with magnification of 800 x were used for identification and quantification of the phytoplankton, zooplankton and Cyanobacteria samples, respectively (Pereira-Neto *et al.*, 2008).

The phytoplankton and Cyanobacteria were identified following Hoek *et al*. (1995) and Stanford (1999), and concentrations were estimated following Pereira-Neto *et al.* (2008) and expressed as cells mL^{-1}. The zooplankton was identified following Boltovskoy (1999) and concentrations were estimated following APHA (2005) and expressed as org mL^{-1}.

Statistical analysis

A parametric one-way ANOVA was used to analyze production parameters, after confirming homocedasticity (Cochran $P < 0.05$) and normality (Shapiro-Wilk $P < 0.05$). Tukey's test ($P < 0.05$) was performed to compare and rank means from the four treatments. Water quality parameters, phytoplankton, zooplankton and Cyanobacteria density were analyzed by performing repeated ANOVA measures. Data analyses were performed using ASSISTAT Version 7.7 (Assistat Analytical Software, Campina Grande, Paraiba, Brazil).

RESULTS

The mean values of dissolved oxygen, temperature, pH and salinity determined in the four treatments were not significantly different ($P > 0.05$) (Table 1). However, significant differences ($P < 0.05$) were detected for TAN, NO_2-N and alkalinity (Table 1).

The phytoplankton population was composed of 35 genera at the start of the experiment and 28 genera at the end. The most frequent genera were *Fragilaria, Orthoseira, Rhabdonema* and *Skeletonema* at the start and *Cylindrotheca, Hemiaulus, Skeletonema, Phymatodocis and Ulothrix* at the end (Table 2). The zooplankton population was composed of 7 genera at the start and 13 at the end. The most frequent genera were *Daphnia* and *Brachionus.* at the start and *Arcella, Bosmina, Daphnia, Asplanchma, Brachionus and Keratella* (Table 3). The Cyanobacteria were composed of 13 genera at the start and 11 at the end. The most frequent genera were *Anabaena, Oscillatoria* and *Sckizothrix* at the start and at the end(Table 4). However, no significant differences ($P > 0.05$) were detected for phytoplankton, zooplankton and Cyanobacteria density.

The shrimp survival rates were all above 87% during the 20-day experimental period in ZWE-FN and ZWE-F. However in ZWE and ZWE-N the survival rates were below 50%. The shrimp FCR in ZWE-FN was significantly lower ($P < 0.05$) than the ZWE-F. Shrimp performance parameters (final weight, final biomass, weight gain, biomass gain and SGR in the ZWE-FN were significantly higher ($P < 0.05$) than in the other treatments (Table 5).

DISCUSSION

The water quality parameters of dissolved oxygen, pH, salinity and TAN were within the ranges suggested by Van Wyk & Scarpa (1999) for marine shrimp. However, temperature and NO_2-N for all treatments and alkalinity, with the exception of ZWE-FN, were different than that recommended. The water temperature was lower in all treatments, yet presented no influence on growth and feed consumption, because growth and FCA rates were good.

The NO_2-N levels found in this study did not cause great problems when salinity was between 20-35 g L^{-1} (Wasielesky *et al*., 2006). However, Cohen *et al.* (2005), studying a zero water exchange system, observed an exponential increase in NO_2-N levels during the growth period, causing shrimp mortality. The ZWE-FN had the highest concentration of TAN among the treatments. A zero water exchange system can have sudden changes of TAN and NO_2-N and accumulate NO_3-N, because of a variation in the microbial biomass during the culture period (Cohen *et al*., 2005), even with a higher C:N ratio (15-20:1) (Gao *et al*., 2012).

Khatoon *et al*. (2009) observed higher TAN and NO_2-N concentrations in the control than in the groups treated with the addition of diatoms during the culture of *Penaeus monodon*. Sanchez *et al*. (2012) observed significant differences in concentrations of NO_2-N in tanks with and without the addition of diatoms in cultivation of *L. vannamei*. However, Godoy *et al*. (2012), when comparing tanks receiving bioflocs, tanks with addition of diatoms and mixed tanks (bioflocs and diatoms), noted significant differences in water quality variables. The diatoms can probably absorb part of the

nutrients provided in autotrophic microbial-based-systems, but in heterotrophic microbial-based-systems the accumulation of particles reduces the penetration of light, which in turn likely reduces the nutrient absorption rates of the diatoms. Castillo-Soriano *et al*. (2013) showed that heterotrophic and nitrifying bacteria are the main factors responsible for the transformation of TAN and NO_2-N in heterotrophic microbial-based-systems.

Levels of $CaCO_3$ less than or equal to 100 mg L^{-1} and pH under 7 for long periods can affect the performance of shrimp in zero water exchange systems (Furtado *et al*., 2011). The alkalinity levels in the ZWE-FN was at the recommended level, which probably contributed to the better growth of shrimp. The higher alkalinity may be related to phytoplankton production since microalgae take in CO_2 from the water column during photosynthesis, leading to $CO_2 + H_2O = HCO_3^- + H^+$, thus making more bicarbonate ions available in the water column (Van Wyk & Scarpa 1999; Becerra-Dórame *et al*., 2011).

Cyanobacteria were the most abundant organisms, followed by Heterokontophyta and Chlorophyta. However, *Microcystis* and *Merismopedia* (Cyanobacteria) were not observed in the ZWE-N and ZWA-FN treatments at the end of the experiment. The data in the literature on the quantity and composition of phytoplankton in shrimp farming systems are extremely variable. Maia *et al.* (2011, 2013), studying intensive culture systems in Brazil, reported densities above 400,000 cells mL^{-1}. These amounts may vary according to the fertilization regime and environmental conditions (temperature and salinity), which can favor undesirable blooms of Pyrrophyta and Cyanobacteria (Campos *et al*., 2007). Ray *et al*. (2010) and Becerra-Dórame *et al*. (2012) found a predominance of Cyanobacteria in relation to other plankton groups, in zero water exchange systems. The prevalence of Cyanobacteria in shrimp culture is probably related to the accumulation of phosphorus and eutrophication of the culture environment, as documented by Emerenciano *et al*. (2011), who found an increase in the concentration of phosphorous in systems with zero water exchange.

In zero water exchange systems, zooplankton can be part of the microbial aggregate (Ray *et al*., 2010), however, factors such as the addition of feed and *Navicula* sp appear not to influence the development of zooplankton, because its composition was very similar in the all treatments. The higher Rotifera density observed, in comparison with other zooplankton groups, is probably related to the adaptation of these organisms to higher levels of nutrients and solids. Casé *et al.* (2008) found a higher rotifer density with increased availability of organic matter in shrimp ponds. Similar results were reported in zero water exchange systems by Anand *et al.* (2013) and Campos *et al*. (2009). Other zooplankton groups, such as Copepoda, Cladocera and Protozoa were found in biofloc systems (Anand *et al,*. 2013; Emerenciano *et al.*, 2013).

Shrimp prefer diatoms over other microalgae groups (Jú et *al*., 2008, 2009). Even in intensive culture systems, the microbial community may play an important role in nutrient cycling (Sánchez *et al*., 2012) providing important nutritional compounds, such as essential amino acids and highly unsaturated fatty acids that are essential to shrimp survival and growth (Jú et *al*., 2008, 2009; Khatoo *et al*., 2009). Increased natural productivity can cause a positive productive response in the shrimp postlarvae (Becerra-Dórame *et al*., 2011). According to Porchas-Cornejo *et al*. (2012) shrimp in the enhanced ponds consumed 68% natural foods and 32% formulated feed, while shrimp in unenhanced ponds consumed 42% natural foods and 58% formulated feed.

Our results illustrate the beneficial effects of a bacterial and *Navicula* sp. consortium on growth of shrimp postlarvae in a zero water exchange system. Similar results indicating the beneficial effects of diatoms were observed by Moss and Pruder (1995) with the use of pennate and centric diatoms, which improved growth of *L. vannamei* in intensive systems; Otoshi *et al*. (2011) with higher growth percentages (22 - 390%) in tanks with high concentrations of diatoms, especially of the genera *Navicula* sp. in a semi-intensive system and Khatoon *et al*. (2009) which found a significantly higher growth rate of *P. monodon* (postlarvae) shrimp reared in tanks containing substrate coated with *Amphora*, *Navicula* and *Cymbella*. The final weights (242 – 348 mg) at 20 days were higher than those found by Becerra-Dórame *et al*. (2011) in autotrophic (72 mg) and heterotrophic (93 mg) microbial-based-systems at 28 days and Kim *et al*. (2014) in heterotrophic (132 mg) microbial-based-systems at 14 days with *L. vannamei* postlarvae, this demonstrates high natural productivity in the experimental tanks in our study. The SGR (14.87% day^{-1}) in ZEW-FN were significantly higher as compared to Becerra-Dórame *et al*. (2011) in autotrophic (5.59% day^{-1}) and heterotrophic (6.22% day^{-1}) microbial-based-systems. This is similar to that observed by Banerjee *et al*. (2010), who found a significantly higher SGR (~ 15% day^{-1}) for shrimp *P. monodon* (postlarvae) reared with additional *Bacillus pumilus* and periphytic microalgae.

The survival rate was highest in ZEW-F (87%) and ZEW-FN (96%) indicating that shrimp of this species need commercial feed for their survival and growth. Becerra-Dórame *et al*. (2011) (76%) and Kim *et al*. (2014) (91.5%) found higher survival rates in heterotrophic microbial-based-systems. Khatoon *et al*. (2009) found that the use of diatoms increased the survival rate and growth of postlarvae, because the biochemical composition of the shrimp raised in tanks with substrates coated with mixed diatoms had significantly higher protein, lipids, PUFA, and EPA and DHA content than those reared in control tanks.

The lower FCR (0.99) in ZEW-FN showed that *Navicula* sp are a significant food source for postlarvae shrimp. Sánchez *et al*. (2012) reported that microalgae present in the culture system significantly improved weight gain and FCR of shrimp, thus potentially reducing the feed cost

associated with shrimp production. Lower FCR in a zero water exchange system was also observed by Silva *et al*. (2009) (0.8 – 1.2), Becerra-Dórame *et al*. (2011) (0.65 – 0.69) and Becerra-Dórame *et al*. (2012) (0.54 – 0.61).

According to Otoshi *et al*. (2011) and Kent *et al*. (2011), *L. vannamei* has a good ability to utilize the microbial community present in aquaculture systems as a food source. Xu *et al*. (2012) showed that the accumulation of microorganisms in the form of flocs substantially contributes to nourishment of the shrimp. However, the availability of these microbial aggregates alone is not enough for the satisfactory growth of shrimp. Similar results were observed by Emerenciano *et al*. (2007, 2011).

In intensive systems a beneficial microbial community should be developed and sustained (Ray *et al*., 2010). But it is difficult to maintain high densities of diatoms in bioflocs systems, because of competition with bacteria for nutrients, reduction in light and higher levels of suspended matter (Godoy *et al*., 2012). The addition of *Navicula* sp appears to boost the postlarval growth of *L. vannamei* in zero water exchange systems. Nevertheless, the data obtained in the ZWE-F and ZWE-FN treatments showed that even with plentiful natural food, shrimp of this species need commercial feed for their survival and growth, but the presence of benthic diatoms appears to increase the efficiency of the use of the commercial feed in systems with zero water exchange, because the FCR was significantly lower in the ZWE-FN than in the ZWE-F treatment.

CONCLUSION

The addition of the benthic diatom *Navicula* sp. increased the growth of postlarvae *L. vannamei* and improved the FCR in a zero water exchange system. These diatoms provide a significant natural food source for shrimp in their early stage. However, further studies related to the density and frequency of adding *Navicula* sp., or other diatoms are needed to improve control over Cyanobacteria and increase the shrimp growth rate in zero water exchange systems.

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REFERENCES

American Public Health Association (APHA). 2005. Standard methods for the examination of water and wastewater. American Public Health Association, Washington, 560 pp.

Anand, P. S. S., Kumar, S., Panigrahi, A., Ghoshal, T. K., Dayal, J. S., Biswas, G., Sundaray, J.K., De, D., Ananda, R. R., Deo, A. D., Pillai, S. M. & Ravichandran, P. 2012. Effects of C:N ratio and substrate integration on periphyton biomass, microbial dynamics and growth of *Penaeus monodon* juveniles. Aquacult. Int., doi 10.1007/s10499-012-9585-6.

Asaduzzaman, M., Rahman, S. M. S., Azim, M. E., Islam, M. A., Wahab, M. A., Verdegem, M. C. J. & Verreth, J. A. J. 2010. Effects of C/N ratio and substrate addition on natural food communities in freshwater prawn monoculture ponds. Aquaculture, 306: 127–136.

Audelo-Naranjo, J. M., Voltolina, D. & Romero-Beltrán, E. 2012. Cultural of white shrimp (*Litopenaeus vannamei* Boone, 1931) with zero water exchange and no food addition: An eco-friendly approach. Lat. Am. J. Aquat. Res., 40 (2): 441-447.

Avnimelech, Y. 2009. Biofloc Technology – A Practical Guide Book. The World Aquaculture Society, Baton Rouge, 182 pp.

Banerjee, S., Khatoon, H., Shariff, M. & Yusoff, F. Md. 2010. Enhancement of *Penaeus monodon* shrimp postlarvae growth and survival without water exchange using marine *Bacillus pumilus* and periphytic microalgae. Fish. Sci., 76: 481–487

Becerra-Dorame, M. J., Martinez-Porchas, M., Martinez-Cordova, L. R., Rivas-Vega, M., Lopez-Elias, J. A. & Porchas-Cornejo, M. A. 2012. Production response and digestive enzymatic activity of the Pacific white shrimp *Litopenaeus vannamei* (Boone, 1931) intensively pregrown in microbial heterotrophic and autotrophic-based systems. The Scientific World Journal., 2012: Article ID 723654, 6 pages.

Becerra-Dorame, M. J., Martinez-Cordova, L. R., Lopez-Elias, J. A. & Martinez-Porchas, M. 2011. Evaluation of autotrophic and heterotrophic microcosm-based systems on the production response of *Litopenaeus vannamei* intensively nursed without *Artemia* and with zero water exchange. Isr. J. Aquacult.-Bamidgeh., IIC :63.2011.620, 7 pages.

Boltovskoy, D. 1999. South Atlantic zooplankton. In: Bradford-Grieve, J. M., Markhaseva, E. L. & Rocha, C. E. F. (eds.). Copepoda. Backhuys Publishers, Leiden, pp. 869-1098.

Brito, L. O., Dantas, D. M. M., Pereira-Neto, J. B., Soares, R. & Olivera, A. 2009a. Efeito de duas estratégias de fertilização na composição do fitoplâncton no cultivo de *Litopenaeus vannamei*. Arq. Ciên. Mar., 42 (1): 30-35.

Brito, L. O., Pimentel, L. F., Marcelino, S. C., Melo, V. F., Costa, W. M. & Oliveira, A. 2009b. Efeito de duas estratégias de fertilização na composição do zooplâncton no cultivo de *Litopenaeus vannamei.* Rev. Bras. Enga. Pesca., 4 (1): 57-66.

Campos, S. S., Silva, U. L., Lúcio, M. Z. T. & Correia, E. S. 2009. Natural food evaluation and water quality in zero water exchange culture of *Litopenaeus vannamei* fertilized with wheat bran. Aquacult. Int., 17: 113–124.

Campos, A. A. B., Maia, E. P., Costa, W. M., Brito, L. O. & Olivera, A. 2007. Descrição dos principais grupos fitoplanctônicos do afluente e efluente em fazenda de criação do camarão marinho *Litopenaeus vannamei* com sistema de recirculação parcial de água. Bol. Inst. Pesca., 33(1): 113-119.

Casé, M., Leça, E. E., Leitão, S. N., Santanna, E. E., Schwamborn, R. & Junior, A. T. M. 2008. Plankton community as an indicator of water quality in tropical shrimp culture ponds. Mar. Pollut. Bull., 56: 1343-1352.

Castillo-Soriano, F. A., Ibarra-Junquera, V., Escalante-Minakata, P., Mendoza-Cano, O., Ornelas-Paz, J. J., Almanza-Ramírez, J. C. & Meyer-Willerer, A. O. 2013. Nitrogen dynamics model in zero water exchange, low salinity intensive ponds of white shrimp, *Litopenaeus vannamei*, at Colima, México. Lat. Am. J. Aquat. Res., 41 (1): 68-79.

Cohen, J. M., Samocha, T. M., Fox, J. M., Gandy, R. L. & Lawrence, A.L. 2005. Characterization of water quality factors during intensive raceway production of juvenile *Litopenaeus vannamei* using limited discharge and biosecure management tools. Aquacult. Eng., 32: 425-442.

Crab, R., Defoirdt, T., Bossier, P. & Verstraete, W. 2012. Biofloc technology in aquaculture: Beneficial effects and future challenges. Aquaculture, 356-357: 351-356.

Crab, R., Avnimelech, Y., Defoirdt, T., Bossier, P. & Verstraete, W. 2007. Nitrogen removal techniques in aquaculture for a sustainable production. Aquaculture, 270: 1-14.

Emerenciano, M., Cuzon, G., Paredes, A. & Gaxiola, G. 2013. Evaluation of biofloc technology in pink shrimp *Farfantepenaeus duorarum* culture: growth performance, water quality, microorganisms profile and proximate analysis of biofloc. Aquacul. Int., 21 (6): 1381-1394.

Emerenciano, M., Ballester, E. L. C., Cavalli, R. O. & Wasielesky W. 2011. Effect of biofloc technology (BFT) on the early postlarval stage of pink shrimp *Farfantepenaeus paulensis*: growth performance, floc composition and salinity stress tolerance. Aquacult Int., 19 (5): 891-901.

Emerenciano, M. G. C., Wasielesky Jr, W., Soares, R. B., Ballester, E. C., Izeppi E. M. & Cavalli, R. O. 2007. Crescimento e sobrevivência do camarão-rosa (*Farfantepenaeus paulensis*) na fase de berçário em meio heterotrófico. Acta. Sci. Biol. Sci., 29 (1): 1-7.

Furtado, P. S., Poersch, L. H. & Wasielesky, W. 2011. Effect of calcium hydroxide, carbonate and sodium bicarbonate on water quality and zootechnical performance of shrimp *Litopenaeus vannamei* reared in bio-flocs technology (BFT) systems. Aquaculture, 321: 130–135.

Gao, L., Shan, H. W., Zhang, T. W., Bao, W. Y. & MA, S. 2012. Effects of carbohydrate addition on *Litopenaeus vannamei* intensive culture in a zero-water exchange systems. Aquaculture, 342: 89 – 96.

Godoy, L. C., Odebrecht, C., Ballester, E. L. C., Martins, T, G. & Wasielesky, W. 2012. Effect of diatom supplementation during the nursery rearing of *Litopenaeus vannamei* (Boone, 1931) in a heterotrophic culture system. Aquacult. Int., 20 (3):559-569.

Gordon, N., Neori, A., Shpigel, M., Lee, J. & Harpaz, S. 2006. Effect of diatom diets on growth and survival of the abalone *Haliotis discus* hannai postlarvae. Aquaculture, 252: 225–233.

Hoek, C., Mann, D. & Jahns, H. M. 1995. Algae: An Introduction to Phycology. Cambridge University Press, Cambridge. 623 pp.

Jú, Z. Y., Forster, I. P. & Dominy, W. G. 2009. Effects of supplementing two species of marine algae or their fractions to a formulated diet on growth, survival and composition of shrimp (*Litopenaeus vannamei*). Aquaculture, 292: 237–243.

Jú, Z. Y., Forster, L., Conquest, L., Dominy, W., Kuo, W. C. & Horgen, F. D. 2008. Determination of microbial community structures of shrimp floc cultures by biomarkers and analysis of floc amino acid profiles. Aquacul. Res., 39: 118-133.

Kent, M., Browdy, C. L. & Lefler, J. W. 2011. Consumption and digestion of suspended microbes by juvenile pacific white shrimp *Litopenaeus vannamei*. Aquaculture, 319: 363 – 368.

Khatoon, H, Banerjee, S., Yusoff, F. M. & Shariff, M. 2009. Evaluation of indigenous marine periphytic *Amphora*, *Navicula* and *Cymbella* grown on substrate as feed supplement in *Penaeus monodon* postlarvae hatchery systems. Aquacul. Nutr., 15: 186-193.

Kim, S. K., Pang, Z., Seo, H. C., Cho, T. R., Samocha, T. & Jang, I. K. 2014. Effect of bioflocs on growth and immune activity of Pacific white shrimp, *Litopenaeus vannamei* postlarvae. Aquacul.. Res., 45, 362–371.

Lara-Anguiano, G. F., Esparza-Leal, H. M., Sainz-Hernández, J. C., Ponce-Palafox, J. T., Valenzuela-Quiñónez, W., Apun-Molina, J. P. & Klanian, M. G. 2013. Effects of inorganic and organic fertilization on physicochemical parameters, bacterial concentrations, and shrimp growth in *Litopenaeus vannamei* cultures with zero water exchange. J. World. Aquacult Soc., 44 (4): 499-510.

Maia, E. P., Modesto, G. A., Brito, L. O., Olivera, A., Gesteira, T. C. V. 2013. Effect of commercial probiotic on bacterial and phytoplankton concentration in intensive shrimp farming (*Litopenaeus vannamei*) recirculation systems. Lat. Am. J. Aquat. Res., 41(1): 126-137, 2013.

Maia, E. P., Olivera, A. & Brito, L. O. 2011. Brazilian shrimp farms for *Litopenaeus vannamei* with partial and total recirculation systems. Int. J. Aqu. Sci., 2 (1):16-26, 2011.

Martinez-Cordova L.R. & Enríquez-Ocaña, L.F. 2007. Study of benthic fauna in the discharge lagoon of a shrimp farm with special emphasis on the polychaetes. J. Biol. Sci., 7: 12-17.

Martínez-Porchas M., Martínez-Córdova L. R., Porchas-Cornejo M. A. & López-Elías, J.A. 2010. Shrimp polyculture: a potentially profitable, sustainable, but uncommon aquacultural practice. Rev. Aquacult., 2:73-85.

Moss, S. M. & Pruder, G. D. 1995. Characterization of organic particles associated with rapid growth in juvenile white shrimp, *Penaeus vannamei* Boone, reared under intensive culture conditions. J Exp Mar Biol Ecol., 187 (2): 175-191

Otoshi, C. A., Moss, D. & Moss, S. M. 2011. Growth - enhancing effect of pond water on four size clases of Pacific white shrimp, *Litopenaeus vannamei*. J. World. Aquacul. Soc., 42 (3): 417-422.

Pereira- Neto, J. B., Dantas, D. M. M., Olivera, A. & Brito, L. O. 2008. Avaliação das comunidades planctônica e bentônica de microalgas em viveiros de camarão (*Litopenaeus vannamei*). Bol. Inst. Pesca., 34 (4): 543-551.

Porchas-Cornejo, M. A., Martínez-Porchas, M., Martínez-Córdova, L. R., Ramos-Trujillo, L. & Barraza-Guardado, R. 2012. Consumption of natural and artificial foods by shrimp (*Litopenaeus vannamei*) reared in ponds with and without enhancement of natural productivity. Isr. J. Aquacult.-Bamidgeh., IJA_64.2012.709, 7 pages.

Ray, A. J., Seaborn, G., Leffler, J. W., Wilde, S. B., Lawson, A. & Browdy, C. L. 2010. Characterization of microbial communities in minimal exchange, intensive aquaculture systems and the effects of suspended solids management. Aquaculture, 310: 130-138.

Samocha, T. M., Patnaik, S., Speed, M., Ali, A. M., Burger, J. M., Almeida, R. V., Ayub, Z., Harisanto, M., Horowitz, A. & Brock, D. L. 2007. Use of molasses as carbon source in limited discharge nursery and grow-out systems for *Litopenaeus vannamei*. Aquacult. Eng., 36: 84–191.

Sánchez, D. R., Fox, J. M., Gatlin, D. & Lawrence, A. L. 2012. Dietary effect of squid and fish meals on growth and survival of Pacific white shrimp *Litopenaeus vannamei* in the presence or absence of phytoplankton in an indoor tank system. Aquacult. Res., 43: 1880–1890.

Silva, U. L., Melo, F. P., Soares, R. B., Spanghero, D. B. N. & Correia, E. S. 2009. Efeito da adição do melaço na relação carbono/nitrogênio no cultivo de camarão *Litopenaeus vannamei* na fase berçário. Acta. Sci. Biol. Sci., 31 (4): 337-343.

Stanford. C. 1999. A guide to phytoplankton of aquaculture ponds: Collection, analysis and identification. Department of Primary Industries, Queensland, 55pp.

Tacon, A.G., Cody, J. J., Conquest, L. D., Divakaran, S., Forster, I. P. & Decamp, O. E. 2002. Effect of culture system on the nutrition and growth performance of Pacific white shrimp *Litopenaeus vannamei* (Boone) fed different diets. Aquacul. Nutr., 8: 121-137.

Van Wyk, P. 1999. Nutrition and feeding of *Litopenaeus vannamei* in intensive culture systems. In: Van Wyk, P., Davis-Hodgkins, M., Laramore, R., Main, K. L., Mountain, J. & Scarpa, J. (eds.). Farming marine shrimp in recirculating freshwater systems. Florida Department of Agriculture and Consumer Services. Harbor Branch Oceanic Institute, Florida, pp. 125-139.

Van Wyk P. & Scarpa J. **(**1999) Water Quality Requirements and Management. In: Van Wyk, P., Davis-Hodgkins, M., Laramore, R., Main, K. L., Mountain, J. & Scarpa, J. (eds.). Farming marine shrimp in recirculating freshwater systems. Florida Department of Agriculture and Consumer Services. Harbor Branch Oceanic Institute, Florida, pp. 141–162.

Xu, W. J., Pan, L. Q., Zhao, D. H. & Huang, J. 2012. Preliminary investigation into the contribution of bioflocs on protein nutrition of *Litopenaeus vannamei* fed with different dietary protein levels in zero water exchange culture tanks. Aquaculture, 350-353: 147-153.

Walne, P. R. 1966. Experiments in the large scale culture of the larvae of *Ostrea edulis*. Fish. Inv., 25 (4): 1-53.

Wasielesky, W., Emereciano, M., Ballester, E., Soares, R., Cavalli, R. & Abreu, P. C. 2006. Cultivos em meios com flocos microbianos: um novo caminho a ser percorrido. Panorama Aquicult., 16 (96): 14-23.

Table 1. Water quality parameters during the culture (20 days) of *Litopenaeus vannamei* postlarvae reared in zero exchange water, with and without the addition of feed and/or diatoms.

Parameters / Treatments[1]	**Salinity (ppt)**	**Temperature (ºC)**	**DO (mg L^{-1})**	**pH**	**TAN (mg L^{-1})**	**NO_2–N (mg L^{-1})**	**Alkalinity (mg $CaCO_3$ L^{-1})**
ZWE	27.0 ± 0.10a	25.0 ± 0.10a	6.6 ± 0.03a	7.4 ± 0.13a	0.10 ± 0.09b	2.71 ± 0.15a	96.7 ± 8.1b
ZWE-F	27.0 ± 0.06a	25.0 ± 0.10a	6.2 ± 0.07a	7.4 ± 0.06a	0.32 ± 0.04b	2.56 ± 0.22a	87.3 ± 7.6b
ZWE-N	27.0 ± 0.07a	25.0 ± 0.12a	6.5 ± 0.04a	7.4 ± 0.05a	0.40 ± 0.02b	2.76 ± 0.05a	99.3 ± 4.41ab
ZWE-FN	26.9 ± 0.01a	24.5 ± 3.15a	6.1 ± 0.12a	7.4 ± 0.08a	1.07 ± 0.21a	1.52 ± 0.26b	131.3 ± 9.75a

[1]The data correspond to the mean ± standard deviation. Mean values in same row with different superscript differ significantly ($P < 0.05$). ¥Results from repeated measures ANOVA and Tukey test; Zero water exchange (ZWE); ZWE with the addition of feed (ZWE-F); ZWE with the addition of *Navicula* sp. (ZWE-N) and ZWE with the addition of feed and *Navicula* sp. (ZWE-FN); dissolved oxygen (DO), total ammonia nitrogen (TAN) and nitrite (NO_2–N).

Table 2. Phytoplankton composition during the culture (20 days) of *Litopenaeus vannamei* postlarvae reared in zero water exchange, with and without the addition of feed and/or diatoms.

Division/ Genera	Initial culture	Final Culture			
		ZWE	ZWE-F	ZWE-N	ZWE-FN
Dinophyta (cells mL^{-1})	**1.37**	**5.38a**	**6.15a**	**2.69a**	**3.46a**
Gymnodinium	0.23	1.54	3.08	1.92	1.15
Peridinium	0.21	3.08	0.77	0.77	1.15
Scrippsiella	0.93	0.77	2.31	0.00	1.15
Heterokontophyta (cells mL^{-1})	**1828.95**	**3546.74a**	**3218.17a**	**3514.92a**	**3683.04a**
Biddulphia	0.06	0.00	0.00	0.00	0.00
Characiopsis	0.03	0.00	0.00	0.00	0.00
Chloridella	9.31	1.92	5.77	3.46	1.92
Cocconeis	0.18	0.00	0.00	0.00	0.00
Coscinodiscus	0.08	0.00	0.38	0.38	0.38
Cyclotela	0.08	1.54	13.47	1.15	0.00
Cylindrotheca	26.82	1412.74	1994.46	1495.85	1488.92
Cymbella	0.93	0.00	0.38	0.00	0.38
Diatoma	14.00	28.09	61.17	47.32	50.40
Diploneis	0.00	0.00	0.00	0.00	0.00
Fragilaria	555.17	0.00	7.31	21.54	38.47
Hemiaulus	49.58	663.67	150.70	736.77	939.13
Navicula	101.89	17.31	13.85	116.19	36.55
Ophiocytium	0.03	0.00	0.00	0.00	0.00
Orthoseira	192.79	14.11	31.55	0.00	0.00
Rhabdonema	454.45	0.00	1.92	0.00	0.00
Skeletonema	421.57	1402.36	934.90	1090.72	1121.88
Synedra	1.78	5.00	2.30	1.54	5.00
Tetracyclus	0.13	0.00	0.00	0.00	0.00
Thalassiosira	0.03	0.00	0.00	0.00	0.00

Triceratium	0.03	0.00	0.00	0.00	0.00
Chlorophyta (cells mL^{-1})	**1310.67**	**2726.22a**	**2873.73a**	**1896.35a**	**1572.98a**
Actinastrum	0.06	0.00	0.00	0.00	0.00
Botryococcus	14.98	106.19	35.39	13.85	30.78
Characium	0.03	0.00	0.00	0.00	0.00
Haematococcus	0.71	1.15	1.15	0.77	1.15
Koliella	0.00	6.16	2.31	146.58	8.85
Micrasterias	0.00	0.38	0.00	0.77	0.00
Mychonastes	344.77	747.92	706.37	0.00	173.13
Phymathodocis	92.30	1064.17	860.80	1029.55	634.81
Planctonema	291.58	126.96	459.76	288.55	173.13
Spirogyra	145.02	121.96	73.10	9.62	71.37
Spirotaenia	2.49	0.00	0.00	0.00	0.00
Tetradesmus	0.00	0.00	0.77	0.00	0.00
Ulothrix	418.73	551.32	734.07	548.63	479.76
Euglenophyta (cells mL^{-1})	**3.05**	**3.08a**	**6.93a**	**1.92a**	**2.31a**
Euglena	0.79	0.77	0.77	0.00	0.77
Trachelomonas	2.26	2.31	6.16	1.92	1.54
Total phytoplankton (cells mL^{-1})	**3,144**	**6,281a**	**6,104a**	**5,415a**	**5,261a**

[1]The data correspond to the mean. Mean values in same row with different superscript differ significantly ($P < 0.05$). [¥]Results from repeated measures ANOVA; Zero water exchange (ZWE); ZWE with the addition of feed (ZWE-F); ZWE with the addition of *Navicula* sp. (ZWE-N) and ZWE with the addition of feed and *Navicula* sp. (ZWE-FN).

Table 3. Zooplankton composition during the culture (20 days) of *Litopenaeus vannamei* postlarvae reared in zero water exchange, with and without the addition of feed and/or diatoms.

Division/ Genera	Initial culture	Final culture			
		ZWE	ZWE-F	ZWE-N	ZWE-FN
Protozoa (org mL^{-1})	**0.30**	**0.31a**	**0.41a**	**0.31a**	**0.55a**
Arcella sp.	0.22	0.25	0.21	0.21	0.43
Leprotintinnus sp.	0.08	0.05	0.20	0.10	0.13
Cladocera (org mL^{-1})	**0.43**	**0.89a**	**0.97a**	**1.16a**	**1.07a**
Bosmina sp.	0.09	0.39	0.40	0.47	0.53
Daphnia sp.	0.35	0.50	0.58	0.69	0.54
Cirripedia (ind./mL)	**0.00**	**0.16a**	**0.25a**	**0.10a**	**0.28a**
Nauplios	0.00	0.16	0.25	0.10	0.28
Copepoda (org mL^{-1})	**0.13**	**0.27a**	**0.51a**	**0.96a**	**0.13a**
Clausocalanus sp.	0.00	0.10	0.11	0.39	0.00
Euterpina sp.	0.13	0.12	0.17	0.26	0.11
Harpaticoida sp	0.00	0.04	0.23	0.31	0.02
Rotifers (org mL^{-1})	**0.62**	**1.54a**	**1.08a**	**1.51a**	**1.62a**
Asplanchna sp.	0.06	0.39	0.31	0.44	0.63
Brachionus sp.	0.56	0.43	0.27	0.45	0.43
Euchlanis sp.	0.00	0.08	0.14	0.04	0.09
Filinia sp.	0.00	0.23	0.10	0.32	0.11
Keratella sp.	0.00	0.40	0.26	0.27	0.35
Total zooplankton (org mL^{-1})	**1.48**	**3.16a**	**3.21a**	**4.05a**	**3.65a**

[1]The data correspond to the mean. Mean values in same row with different superscript differ significantly ($P < 0.05$). [¥]Results from repeated measures ANOVA; Zero water exchange (ZWE); ZWE with the addition of feed (ZWE-F); ZWE with the addition of *Navicula* sp. (ZWE-N) and ZWE with the addition of feed and *Navicula* sp. (ZWE-FN).

Table 4. Cyanobacteria composition during the culture (20 days) of *Litopenaeus vannamei* postlarvae reared in zero water exchange, with and without the addition of feed and/or diatoms.

Genera	**Initial culture**	**Final Culture**			
		ZWE	**ZWE-F**	**ZWE-N**	**ZWE-FN**
Anabaena	25.47	153.51	185.83	48.48	122.35
Aphanocapsa	529.54	937.98	2300.71	575.18	2004.85
Dactylococcopsis	17.05	25.01	19.24	33.86	21.55
Gloeothece	2.76	0.00	0.00	0.00	0.00
Merismopedia	28.27	0.00	19.24	0.00	0.00
Microcystis	6.41	134.66	192.37	0.00	0.00
Oscillatoria	542.28	5454.80	4969.61	5251.62	4816.10
Plectonema	38.12	22.89	114.46	34.34	57.23
Pseudanabaena	59.96	205.06	155.05	120.04	175.05
Schizothrix	838.57	3123.27	2369.96	3758.08	1599.72
Spirulina	18.52	90.03	74.64	20.39	143.50
Radiocystis	0.00	0.00	0.00	0.00	9.62
Synechocystis	0.45	0.00	0.00	0.00	0.00
Total Cyanobacteria (cells mL^{-1})	**2,107a**	**10,147a**	**10,401a**	**9,841a**	**8,949a**

[1]The data correspond to the mean. Mean values in same row with different superscript differ significantly ($P < 0.05$). [¥]Results from repeated measures ANOVA; Zero water exchange (ZWE); ZWE with the addition of feed (ZWE-F); ZWE with the addition of *Navicula* sp. (ZWE-N) and ZWE with the addition of feed and *Navicula* sp. (ZWE-FN).

Tabela 5. Shrimp production parameters during the culture (20 days) of *Litopenaeus vannamei* postlarvae reared in zero water exchange, with and without feed and/or diatoms.

Parameters / Treatments	**Final weigth (mg)**	**Final biomass (mg)**	**Weight gain (mg)**	**Biomass gain (mg)**	**SGR (% day^{-1})**	**Survival (%)**	**FCR**
ZWE	242 ± 31.2b	10056 ±1297c	224 ±31.2b	8286 ± 1297c	13.05 ± 0.65b	41.5 ± 0.7b	-
ZWE-F	272 ± 7.5b	23693 ± 658b	254 ±7.57b	21923 ± 658b	13.66 ± 0.13b	87.0 ± 18.0a	1.2 ± 0.11a
ZWE-N	256 ± 31.5b	11278 ± 1386c	238 ±31.5b	9508 ± 1386c	13.34 ± 0.61b	44.0 ± 2.82b	-
ZWE-FN	348 ± 41.5a	33440 ± 3992a	330 ± 41.5a	31670 ± 3992a	14.87 ± 0.61a	96.0 ± 1.41a	0.99 ± 0.22b

[1]The data correspond to the mean of 3 replicates ± standard deviation. Mean values in same row with different superscript differ significantly ($P < 0.05$). [¥]Results from one-way ANOVA, Tukey test and Student's t-test. Zero water exchange (ZWE); ZWE with the addition of feed (ZWE-F); ZWE with the addition of *Navicula* sp. (ZWE-N) and ZWE with the addition of feed and *Navicula* sp. (ZWE-FN); SGR (% day^{-1}) = 100 x [ln final weight (g) – ln initial weight (g)] / time and FCR = amount of feed consumed / biomass.

4. 1- Normas da Revista [Latin American Journal of Aquatic Research INSS: 0718-560x]

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Authors:
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In English and Spanish, 250 words maximum, indicating the main results or findings presented in the text.

Keywords:
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- Introduction
- Material and Methods
- Results
- Discussion
- Conclusions (optional)
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a) Entries in the reference list should follow this format: Author(s). Year of publication. Article title. Abbreviated journal name (See: Journal Title Abbreviations), Journal volume (and number in parentheses): First and last page numbers.

- Coelho,V., R.A. Cooper & S. Rodrigues. 2000. Burrow morphology and behaviour of the mud shrimp Upogebia omissa (Decapoda, Thalassinidea, Upogebiidae). Mar. Ecol. Progr. Ser., 200: 229-240.

b) Book references should indicate: Author(s). Year of publication. Book title. Editorial, City, Pages.

- Thurman, H. & A. Trujillo. 2002. Essentials of oceanography. Prentice Hall, New Jersey, 524 pp.

c) Articles published in books should indicate: Author(s). Year of publication. Article title. Editor(s). Book title. Editorial, City, Pages.

- Brummet, R.E. & B.A. Costa-Pierce. 2002. Village-based aquaculture ecosystems as a model for sustainable aquaculture development in Sub-Saharan Africa. In: B. Costa-Pierce (ed.). Ecological aquaculture: evolution of the blue revolution. Blackwell Science, Oxford, pp. 145-160.

d) Articles published on Internet should indicate: Author(s). Year of publication. Article title. Web site. Date reviewed.

- Walker, J.R.1997. MLA-Style citations of Internet sources. [http://www.cas.usf.edu/english/walker/janice.html]. Reviewed: 24 January 2008.

e) References to articles or books published in CD-Rom should indicate: author(s), year of publication, (CD-ROM), article or book titles, editorial, city.

- Retamal, M.A. 2000. (CD-ROM). Decápodos de Chile. ETI-Universidad de Concepción. Springer-Verlag, Berlin.
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