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**THE POTENTIAL OF GREEN WATER MEAL AS
AN INGREDIENT IN THE FORMULATED DIET
FOR JUVENILE PACIFIC WHITE SHRIMP,
*Litopenaeus vannamei***

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SYNOPSIS

Steady increase of fishmeal price due to unreliable supply of fishmeal in the world market, coupled with increased demand have led aquaculture nutritionist to search for cheaper and sustainable alternative protein sources that can be incorporated in formulated diets. Green water which consists of green microalgae (mainly *Chlorella* sp.) can be produced easily in freshwater fish culture tank. Dried green water or green water meal GWM contains relatively high crude protein making it a potential candidate for fishmeal substitution. In the present study, the potential of GWM as a substitute of fishmeal in the diets of Pacific white shrimp (*Litopenaeus vannamei*) was evaluated through two feeding trials. In Feeding Trial 1, five isoproteic and isolipidic diets with 0% (control diet), 10% (GWM10), 20% (GWM20), 30% (GWM30) and 40% (GWM40) GWM replacing fishmeal protein were fed to triplicate groups of shrimp, with an average initial weight of 1.73 ± 0.003 g and cultured in a flow-through tank culture system. The shrimps were fed by hand three times daily with amount 7-10% of their body weight for 44 days. In general, growth of the shrimp decreased with increasing level of GWM substitution. Although the 10% replacement of GWM (GWM10) caused significantly lower final body weight (FBW) and weight gain (WG) ($P < 0.05$) compared with the control diet, the dry feed intake (DFI), feed conversion ratio (FCR), and specific growth rate (SGR) of this diet were not statistically different ($P > 0.05$) from the control diet. In Feeding Trial 2, diets were formulated using the same specification with diet's formulation in Feeding Trial 1 and fed to bigger size shrimps with an average initial weight of 6.42 ± 0.02 g and fed at 5-7% of their body weight daily for 56 days to investigate if larger shrimp has better tolerance than the smaller shrimp to GWM as dietary protein source. Similar growth trend with Feeding Trial 1 was observed in this trial. However, shrimp fed diet GWM10 did not show any significant differences in FBW (16.88 ± 0.18 g), WG ($161.14 \pm 3.07\%$), SGR ($1.50 \pm 0.04\%$ /day), DFI (19.68 ± 0.04 g) and FCR (1.89 ± 0.04) from the control diet, indicating an improved utilization of GWM compare to the findings in the Feeding Trial 1. Survival rates in both trials were high (above 95% in Feeding Trial 1 and above 88% for Trial 2) and not affected by the dietary treatments. The whole-body proximate compositions of shrimp fed experimental diets were significantly influenced by the inclusion of GWM with no definite trend in Feeding Trial 1. However, in Feeding Trial 2, the whole-body protein and lipid of the shrimps decreased with the increasing GWM level in the diets. Both trials proved that inclusion of GWM in the diets (GWM10, GWM20, GWM30, and GWM40) produced shrimp with more intense red/orange colour and contained higher total carotenoid concentration compare with the control diet. The present findings suggest that GWM is an acceptable alternative protein source for shrimp which can be included in the diet of Pacific white shrimp at a level of about 10% with advantage of providing carotenoid which will enhance the shrimps colour. Considering GWM as a by-product in an aquaculture system, cost of feed production for shrimp culture can be reduced with the inclusion of GWM in the diets.

SINOPSIS

Peningkatan berterusan harga tepung ikan kerana bekalan yang tidak menentu di pasaran dunia, ditambah pula dengan permintaan yang semakin meningkat telah menyebabkan pakar pemakanan akuakultur mencari alternatif sumber protein yang lebih murah dan lestari yang sesuai untuk dijadikan ramuan dalam makanan. 'Air hijau' yang terdiri daripada mikroalga hijau (terutamanya *Chlorella* sp.) boleh dihasilkan dengan mudah di tangki ternakan ikan air tawar. Tepung 'Air hijau' (GWM) mengandungi protein yang tinggi menjadikannya sebagai bahan yang berpotensi untuk menggantikan tepung ikan. Dalam kajian ini, potensi GWM sebagai pengganti tepung ikan dalam diet udang putih (*Litopenaeus vannamei*) telah dinilai melalui dua percubaan pemakanan. Dalam Percubaan 1, lima diet isoproteik dan isolipidik dengan 0% (diet kawalan), 10% (GWM10), 20% (GWM20), 30% (GWM30) dan 40% (GWM40) GWM menggantikan protein daripada tepung ikan diberi makan bagi kumpulan triplikat udang dengan purata berat permulaan ialah 1.73 ± 0.003 g dan dikultur di dalam tangki dengan sistem air mengalir. Udadang diberi makan dengan tangan tiga kali sehari dengan jumlah 7-10% daripada berat badan mereka selama 44 hari. Secara umum, pertumbuhan udang menurun dengan peningkatan tahap penggantian GWM. Walaupun penggantian 10% GWM (GWM10) menyebabkan berat badan akhir badan (FBW) dan pertambahan berat badan (WG) yang lebih rendah secara signifikan ($P < 0.05$) berbanding dengan diet kawalan, pengambilan makanan (DFI), nisbah penukaran makanan (FCR), dan kadar pertumbuhan spesifik (SGR) diet ini tidak berbeza dari segi statistik ($P > 0.05$) dari diet kawalan. Dalam Percubaan 2, diet-diet yang telah diformulasikan mengikut spesifikasi yang sama dengan formulasi diet pada Percubaan 1 dan diberi makan kepada udang yang bersaiz lebih besar dengan purata berat permulaan ialah 6.42 ± 0.02 g dengan jumlah 5-7% daripada berat badan mereka setiap hari selama 56 hari bertujuan mengenalpasti sama ada udang yang lebih besar mempunyai toleransi yang lebih baik kepada GWM sebagai sumber protein berbanding udang yang kecil. Trend pertumbuhan yang sama dengan Percubaan 1 telah dilihat pada percubaan ini. Walaubagaimanapun, udang yang diberi makan dengan diet GWM10 tidak menunjukkan sebarang perbezaan signifikan dalam FBW (16.88 ± 0.18 g), WG (161.14 ± 3.07 %), SGR (1.50 ± 0.04 %/day), DFI (19.68 ± 0.04 g) and FCR (1.89 ± 0.04) dengan diet kawalan, menunjukkan penggunaan GWM adalah lebih baik berbanding dengan penemuan pada Percubaan 1. Kadar kemandirian hidup pada kedua-dua percubaan adalah tinggi (melebihi 95% dalam Percubaan 1 dan melebihi 88% dalam Percubaan 2) dan tidak terjejas oleh jenis pemakanan. Komposisi proksimat seluruh badan udang yang diberi makan diet eksperimen adalah dipengaruhi dengan penggunaan GWM tanpa trend tertentu dalam Percubaan 1. Walaubagaimanapun, dalam Percubaan 2, protein seluruh badan dan lipid udang menurun dengan peningkatan kandungan GWM di dalam diet. Kedua-dua percubaan membuktikan bahawa penggunaan GWM dalam diet (GWM10, GWM20, GWM30, dan GWM40) menghasilkan udang yang berwarna lebih merah/oren dan mengandungi jumlah karotenoid lebih tinggi berbanding dengan diet kawalan. Penemuan ini menunjukkan bahawa GWM adalah sumber alternatif protein yang boleh diterima dalam diet udang putih pada paras kira-kira 10% dengan kelebihan karotenoid yang akan meningkatkan warna udang. Memandangkan GWM adalah

produk sampingan akuakultur, kos pengeluaran makanan untuk ternakan udang boleh dikurangkan dengan penggunaan GWM di dalam diet.

INTRODUCTION

Fishmeal is a favourable source of protein in formulated fish or shrimp diets and contributes considerably to the variable production cost in the aquaculture industry. It is considered as the most essential ingredient in aquaculture feed due to the balanced profile of essential amino acid and also provides high palatability compare to other protein sources (Tacon and Akiyama, 1997; Suárez *et al.*, 2009). However, the shortage in world production of fishmeal, coupled with increased demand and competition with terrestrial domestic animals in 2010 and 2011, has further increased fishmeal prices and hence threaten the growth and sustainability of the aquaculture industry (Tacon *et al.*, 2011). Therefore, most of aquaculture nutritionists are focusing on finding cheaper and sustainable alternative protein sources that can be incorporated in formulated diets to reduce the cost of feeding and contribute to the sustainability of aquaculture industry. Finding alternative protein source requires some research priority such as ability to produce in sufficient quantities, understanding of an ingredient's nutritional value, its ability to blend with other ingredients, its effect on pellet stability, and its appeal to the cultured animals (Tacon *et al.*, 2006).

Microalgae is a diverse group of unicellular photosynthetic organism which thrives in aquatic environment (Hemaiswarya *et al.*, 2010). Microalgae is seen as high potential food resources due to the photosynthetic efficiency and high nutritional value (Hanel *et al.*, 2007; Ju *et al.*, 2012; Tongsiri *et al.*, 2010; Macias-Sancho *et al.*, 2014; Badwy *et al.*, 2008). Over the last decades, microalgae received a lot of attention in biotechnology research because it can reach substantially higher productivities compare to traditional crop without require much area or land for the agricultural purposes and presents less or no seasonal environment. Furthermore, the cultivated microalgae provide the source of highly valuable molecules such as protein, pigments, polyunsaturated fatty acids, pharmaceuticals antioxidants and other biologically active compounds (Gouveia *et al.*, 2008). With all of these advantages, the application of microalgae biomass in