

# A review on insect meals in aquaculture: the immunomodulatory and physiological effects

Seyedehlaleh Mousavi . Somayeh Zahedinezhad . Jiun-Yan Loh 

Received: 12 April 2020 / Accepted: 24 May 2020 / Published online: 30 May 2020  
© The Author(s) 2020

**Abstract** The depletion of global fishery stock has posted significant effects to the world capture fishery industry. Constant environmental issues and fluctuation of fish meal (FM) costs have pushed the aquafeed industry to seek for alternative protein sources to sustain the huge demand of feed in aquaculture production. Insect meal (IM) has already started to gain global attention as an alternative for FM replacement. Substantial feeding trials have revealed its promising application not only beneficial as in protein replacement but also playing a vital role as a functional ingredient that boosting immunostimulatory effects in a wide range of aquatic animals. Here, we compiled and categorized several major groups of insects, for example black soldier fly, yellow mealworm, housefly, silkworm, alongside a minor group of insect species widely used as the alternative diet in aquaculture. Critically, we discuss the functional properties of the IMs, their immunomodulatory, and physiological effects in aquatic species. Biological parameters include oxidative stress, serum biochemical, hematological parameters, and immune-related genes are also further discussed in this review. In short, we suggest that low levels of IM supplementation could improve survival, immunity, and feed conversion efficiency of targeted aquaculture species. Chitin and its derivatives alongside active substances in insect exoskeleton, such as antimicrobial peptides (AMPs) play an essential role in the immunomodulation of aquatic organisms. Knowledge of this area is still in infancy. Therefore, it is a critical need to further address the mechanism of immunomodulation focusing on the isolated components from IMs.

**Keywords** Alternative protein sources . Antimicrobial peptides . Fish meal . Immunostimulatory . Insect meal

## Introduction

Increasing demand and impermanent supply of fish meal (FM) have resulted in tremendous pressure on wild fishery resources, which endanger the marine ecosystem and global fish stock (Lalander et al. 2015). Many studies have shown that plant protein could be an alternative for FM substitution in aquafeeds. However, some nutritional disadvantages of major plant proteins are often associated with nutrient deficiency, weak nutrient absorption, and most importantly, its usage as a primary food source for mankind cannot be overlooked (Gai et al. 2012). Since the major group of commercial fish species in aquaculture are either carnivorous or omnivorous, therefore, it is crucial to seek for a sustainable and eco-friendly protein source with great availability, and efficient in terms of cost and energy conversion potential such as insect protein (Henry 2015; Xiao et al. 2018).

Insect protein has been gaining a substantial amount of research attention due to its high energy conversion efficiency, good nutritional quality, and benefits in growth and health promotion in fish and shellfish (Barroso et al. 2014; Henry et al. 2015; Xiao et al. 2018). In addition, fast-growing and high

Seyedehlaleh Mousavi, Jiun-Yan Loh (✉)  
Faculty of Applied Sciences, UCSI University, No. 1, Jalan Menara Gading, UCSI Heights, Cheras, 56000 Kuala Lumpur, Malaysia  
e-mail: loh.jiun.yan@gmail.com; lohjy@ucsiuniversity.edu.my

Seyedehlaleh Mousavi  
Department of Fisheries, Faculty of Natural Resources, College of Agriculture & Natural Resources, University of Tehran, Karaj, Iran

Somayeh Zahedinezhad  
Department of Fisheries, Faculty of Natural Resources, Bandar Abbas Branch, Islamic Azad University, Bandar Abbas, Iran



fecundity as demonstrated in most insects is also an important criterion to explore the potential of an insect-based meal. Insect meal (IM) is rich in amino acids (AAs), lipids, vitamins, and minerals, these advantages making them an emerging energy source for agricultural sectors (Premalatha et al. 2011). In less than a decade, research interests on IM have been picked up extensively and developed rapidly in the industry. Thanks to the approval of IM as part of feedstuff ingredients in aquaculture by EU Commission Regulation 2017/893 (Tan et al. 2018). Some companies are now producing it massively as part of feed ingredients, particularly to substitute FM in animal nutrition (Tran et al. 2015). It is important to note that, most IMs contain appreciable amounts of bioactive compounds/chitin with potential antioxidant, antimicrobial, and immunostimulatory features, which could enhance immunity and health status of the cultured species e.g. fish and shrimp (Ogunji et al. 2011; Taufek et al. 2016a; Li et al. 2017; Motte et al. 2019; Oda et al., 2019). Literature suggested that moderate intake of chitin could improve gut health and immunity of culture species (Hoffman et al. 1997; Kim and Rajapakse 2005; Lin et al. 2012; Harikrishnan et al. 2012). Chitin ( $\beta$ -(1-4)-poly-N-acetyl-D-glucosamine), the primary ingredient of arthropods exoskeletons, is the most abundant amino polysaccharide polymer in the insects. Harikrishnan et al. (2012) reported that dietary inclusion of 1.0% chitin and chitosan significantly enhanced hematological parameters e.g. red blood cells (RBCs), white blood cells (WBCs), hemoglobin (HB) levels, lymphocytes, monocytes, and neutrophils in Kelp grouper, *Epinephelus bruneus*, and the disease-resistant against the protozoan parasite, *Philasterides dicentrarchi*. The immunomodulation could be largely attributed to the regulation of the gut microbiome (Loh et al. 2020).

So far, only a limited piece of literature has found on the direct and indirect effects of insects on the immune aspects of aquatic animals. Relevant information on IM-based diets are scarce (Ogunji et al. 2011; Taufek et al. 2016a; Li et al. 2017). Many works of literature are focusing on the effects of FM on the antioxidant and hematological indices of fish (Elia et al. 2018). In teleost immunity, oxidative catabolism can affect the oxidative stress response, and it is one of the major immune responses could be involved in fish disease defend mechanism (Martínez-Álvarez et al. 2005). The antioxidants play a major role in scavenging reactive oxygen species (ROS) and protect cell membranes against damage to these free radicals. Thus, the hematological parameter is very important to monitor the health status and physiological responses of the organisms to environmental stresses (Bhaskar and Rao 1984; Jawad et al. 2004).

In this paper, the immunomodulatory and physiological effects exerted by insect-based diets are critically reviewed with focusing on four major groups, including black soldier fly (*Hermetia illucens*), yellow mealworm (*Tenebrio molitor*), housefly (*Musca domestica*), silkworm (*Bombyx mori*) and a minor group of insect species such as locusts, grasshoppers, termites, crickets, and beetles. Nutritional profiles in particularly protein and lipid contents, nutrient digestibility, and insect productivity are the main factors in determining the selection of IM for aquafeed. Generally, the protein content of IM recommended in the feed production is ranging between 50 – 82% of dry matter generally (Banjo et al. 2006; Rumpold and Schluter 2013). Nonetheless, the nutrient values of the IM such as minerals (e.g. K, Na, Ca, Cu, Fe, Zn, Mn, and P), and vitamins (e.g. B group vitamins, vitamins A, D, E, K, and C) are varied depending on the food sources, species and the processing method (Barroso et al. 2014; Kouřimská and Adámková 2016).

There are some common interpretations of the immunostimulatory effects derived from the IM. According to literature, these health-promoting effects are largely attributed to either direct secretion of antimicrobial peptides (AMPs), or indirect administration to chitin, or other bioactive substances (Ido et al. 2015; Henry et al. 2018). Some studies showed that chitin has a stimulating effect on both cell-mediated and humoral immune response of the fish (Esteban et al. 2000; 2001), which could improve the resistance against infectious pathogens. The immunity-promoting peptides such as AMPs in the IMs are effective in mitigating bacterial infection of the host. The type, amount, and quality of secreted AMPs are dependent on the environmental conditions, the species of insects, the nutrient balance of the diets, and even the sexes of the insects (Vogel et al. 2018). Studies also showed that insects have significant pharmacological properties in enhancing the innate immunity of many species including humans, livestock, and poultry (Akiyama et al. 2000; Schuhmann et al. 2003). It is also interesting to note that, insects show a promising result in treating human chronic diseases, for example osteomyelitis (Sherman and Pechter 1998).

To evaluate the feasibility and practicality of FM substitution or supplementation of IM in aquafeeds, it is of utmost importance to understand the mechanisms of physiological and immunomodulatory effects laying behind the concept. This is the first review aiming to evaluate the effects of IM inclusion on the



physiological and immunomodulatory changes in aquatic animals (e.g. fish, crustaceans, sea cucumber, and bullfrog). The current review suggests that a low level of IM inclusion could provide a high quality of protein source and certain degrees of immunological benefits to the cultured species.

### **Black soldier fly (*Hermetia illucens*)**

Black soldier fly (BSF) is the most important and yet a common farmed insect for the animal feed industry. It has been widely used as a bio-converter in organic wastes management. Simultaneously, it serves as an alternative protein source in livestock and aquaculture (Lalander et al. 2015; Vogel et al. 2018). There are numerous findings on the immunostimulatory effects of dietary BSF on terrestrial animals, for example broilers and swine. However, the findings in aquaculture are still at an early stage. Foysal et al. (2019) showed that poultry-by-product meal + BSF included diet could improve gut microbiota profile, and colonizing ability of beneficial bacteria in the gut of crayfish (*Cherax cainii*). However, the report also showed that an up-regulation in cytokine genes (IL-1 $\beta$ , IL-17F, and TNF- $\alpha$ ), which are associated with inflammatory effects in *Cherax cainii* fed with BSF supplemented diets (Foysal et al. 2019). The immune response of crayfish was largely attributed to the secondary metabolites expressed by the BSF, and the association of immunocompetent cells and mucosal membranes in the intestine. Intestinal mucosal liners are the primary defense system housing for a large variety of intestinal microflora, the inclusion of BSF as part of the diet might modify the composition of the microbiome in the gut (Askarian et al. 2012; Foysal et al. 2019).

It is interesting to note that, BSF larval extract could reduce the number of Gram-negative bacteria, albeit, the same inhibitory effect was not observed on the Gram-positive microbes (Choi et al. 2012). In a study done by Lalander et al. (2015), BSF was effective in reducing the number of *Salmonella* sp., some viruses including reovirus, adenovirus and, enterovirus, as well as some thermo-tolerant coliforms in a waste recycling process. Likewise, no inhibitory effect was reported on Gram-positive enterococcus. Similar findings were also reported by Gabler and Vinnerås (2014), the organic waste recycling using BSF composting reduced the numbers of *S. senftenberg* and *S. typhimurium*. The antimicrobial response could be due to the interaction between the active agent in BSF and the bacterial cell components. For instance, high content of medium-chain fatty acid in BSF (e.g. lauric acid) causes damage to the cell membrane accelerating the entry of antimicrobial compounds into the cytoplasm, and thus kill the bacteria. However, further study is required to elucidate the mechanism behind the microbial inhibition, and specific substances that may contribute as antibacterial agents (Choi et al. 2012; Lalander et al. 2015; Harlystiarini et al. 2019).

In a feed formulation study, Hu et al. (2017) showed that only up to 20% of dietary FM can be replaced with BSF without posing significant negative effects on blood parameters of juvenile yellow catfish (*Pelteobagrus fulvidraco*). The increase of lysozyme (LZM) activity encourages stimulation of the fish immunity system and may contribute to the resistance against infectious pathogens (Misra et al. 2006). Nurin et al. (2018) suggested that dietary inclusion of BSF extract could activate the immuno-competence hematological parameters in challenged common carp (*C. carpio*) with *A. hydrophila*. Also, 8 % BSF extract helps to increase the total blood count ( $1.95 \times 10^5$  cell/ml), and the differential counts of leukocytes (81.55%; 16.42% and 0.33% for lymphocytes, monocytes, and neutrophils, respectively) compared to the control groups ( $1.63 \times 10^5$  cell/ml in total leukocytes, TLC). This finding also showed that BSF can enhance the immune system of *C. carpio*, and it could play an important role in disease prevention.

The occurrence of oxidative stress is always associated with the onset of various types of diseases in many animals including fish (Martínez-Álvarez et al. 2005). There is an increasing number of studies are focusing on the antioxidants in fish's health (Nan et al. 2013; Park et al. 2014; Taufek et al. 2016b; Elia et al. 2018). However, only a few works have been reported on the effects of dietary inclusion of BSF on fish oxidative stress (Li et al. 2017; Hu et al. 2017; Elia et al. 2018). A recent study showed that the inclusion of defatted BSF meal in Jian carp (*Cyprinus carpio* var. *Jian*) diets enhanced serum catalase (CAT) but had no effect on serum superoxide dismutase (SOD) - an antioxidant (Li et al. 2017). CAT-SOD antioxidant enzymatic mechanism is the first line of defense against ROS. ROS is an essential oxidative byproduct of the normal metabolic process. Indeed, oxidative stress is induced by imbalanced production of ROS, or ineffective antioxidant mechanisms to control its negative effects (Monaghan et al. 2009). If ROS could not be suppressed by antioxidants (activated by antioxidative enzymes), it could adversely



affect DNA, proteins, and cell membrane lipids as a result (Pamplona and Costantini 2011). This leads to immunosuppression and thus affect the health status of the hosts.

Elia et al. (2018) demonstrated dietary inclusion of BSF meals (< 20%) upregulated oxidative stress biomarkers (SOD and CAT) in rainbow trout (*Oncorhynchus mykiss*). Likewise, dietary inclusion of BSF meals boosted the activity of serum immune-related enzymes such as LZM and SOD in yellow catfish - *P. fulvidraco*. The most remarkable immunity improvement was observed at 25% FM replacement by BSF meals (Xiao et al. 2018). However, in another study, replacement levels of defatted BSF meals in juvenile Jian carp diets beyond 75% resulted in oxidative stress, and intestinal histopathological damage (Li et al. 2017). Christian et al. (2020a) reported the feed consumption of Siberian sturgeon (*Acipenser baerii*) juveniles was decreased when the concentration of BSF meals increased. Fifty percentage replacement of FM with BSF meals could adversely affect the growth performance of sturgeon, and the fish refused to consume diet when the experimental diet was replaced with 100% BSF. This probably caused by changes in oxidative stress biomarkers e.g. higher SOD activity was found in sturgeon's liver and kidney when the fish administrated to > 50% BSF meals (Christian et al. 2020b). Another study by Magalhães et al. (2017) demonstrated that a high level of BSF inclusions in European sea bass, *Dicentrarchus labrax* dietary was associated with a significant decline in total cholesterol (TCHO) concentration. Despite a low level of dietary inclusion of BSF enhances the immune system ability of fish, but high inclusion levels lead to an inhibition of superoxide formation, and higher levels of nitric oxide (NO), both of which are responsible to high oxidative stress (Hu et al. 2017; Li et al. 2017).

A study showed that ethoxyresorufin-o-deethylase (EROD), glutathione s-transferase (GST), and glutathione (GLU) in the BSF could enhance protecting effects against the oxidative process in *O. mykiss* (Elia et al. 2018). The authors opined that amino acid or lipid profiles in BSF meal may explain the alteration of antioxidant balance in *O. mykiss* fed with IM. For instance, a lower level of essential amino acids (EAAs) in BSF meals may affect the catalytic activity of seleno-dependent glutathione peroxidase (SeGPx) activity. Furthermore, arginine and lysine are known to be the key AAs in synthesizing NO, which is a signaling and an antimicrobial factor in immunity responses (Yaghoubi et al. 2017). However, a study showed that *C. carpio* treated with crystalline lysine - supplemented diet, the results showed an enhancement of the fish body crude protein but did not show any significant difference in antioxidant indices (Zhang et al. 2013; Ji et al. 2015).

Evidence showed that the composition of fatty acids, immunomodulatory effects (e.g. constitutive and inducible immunity), and antimicrobial activity of IM are diet-dependent (Barroso et al. 2014; Vogel et al. 2018). For instance, it has been reported that feeding BSF larvae on diets supplemented with sunflower oil induced the expression of AMPs and maximized the number of these peptides (Vogel et al. 2018). Paul et al. (2017) reported that Orthopterans e.g. *Acheta domesticus* and *Conocephalus discolor* contained different kinds of linolenic acid. Diet modification can improve the essential fatty acids (EFAs) of these insects, which in turn, beneficial to the health-promoting factors.

### **Yellow mealworm (*Tenebrio molitor*) and superworm (*Zophobas morio*)**

Yellow mealworm (*Tenebrio molitor*) and superworm (*Zophobas morio*) are another group of common insect-based diet for captive reptiles, birds and small animals. These darkling beetle's larvae have been used as replacement feed in many fish species. Some reports show that *T. molitor* improved the digestibility and growth indices in the fish e.g. *D. labrax* when compared with those fed with FM (Henry 2015; 2018). While 25% of FM replacement by *Z. morio* in the fish diet showed an insignificant adverse effect on body composition and feed utilization in Nile Tilapia (*Oreochromis niloticus*) (Jabir et al. 2011; Jabir et al. 2012a; 2012b). Like BSF, a study on yellow mealworm (YMW) and superworm (SW) on immunological and health status aspects in aquaculture is still lacking. Only a few studies discussed the oxidative stress and inflammatory responses (e.g. ceruloplasmin, myeloperoxidase, and NO) in fish after feeding with diets containing these beetles' larvae (Henry et al. 2018).

According to Tang et al. (2012), mice administrated YMW demonstrated strong disease resistance against several pathogens, suggesting that YMW can be potentially used as bioactive peptides and natural antibacterial agents, through stimulation of non-specific phagocytic functions and elevation of hemolysin. In a mice study, high contents of serum NO could play a significant role in immune regulating, especially



to acid phosphatase (ACP) and alkaline phosphatase (AKP), all of these are the important indices for biological detoxification (Tang et al. 2012). Likewise, biochemical indices of the serum also an important consideration in fish health. Like those findings in BSF, Sankian et al. (2018) noticed a significant fall in the TCHO concentration in mandarin fish (*Siniperca scherzeri*) fed with YMW contents in the diets.

LZM is another bioindicator for non-specific immunity. LZM is present naturally in the insect hemolymph, which acts with defense peptides synergistically in pathogenic inhibition (Lin and Shiao 2003; 2005; 2007). This implies the presence of pathogens in the body will lead to high levels of hemolymph. Due to its mucolytic nature, LZM mediates resistance against pathogens by cleaving the linkages in the bacterial cell wall (Saurabh and Sahoo 2008). In a study on yellow catfish, Su et al. (2017) found that high activity of LZM was noticed in the fish when fed with YMW-based diets, and subsequently challenged by *Edwardsiella ictaluri*. However, the concentration of LZM present in the fish is dose-dependence (Sankian et al. 2018). In other words, LZM concentration is corresponding to the amount of YMW in the diet. Evidence showed that when European sea bass *D. labrax* administrated with an increasing amount of YMW in their diet, the activities of serum LZM and trypsin inhibition are significantly higher, which also corresponding to the anti-parasitic activity (Henry et al. 2018).

Inclusion of YMW in the fish dietary could also increase the level of serum SOD, nitric acid, malondialdehyde (MDA) and the activities of ceruloplasmin, myeloperoxidase and glutathione peroxidase (GPx) in the fish (Su et al. 2017; Sankian et al. 2018; Henry et al. 2018). All these antioxidant indices have critical roles in eliminating excessive superoxide radicals to maintain the homeostasis of ROS in the fish species (Sankian et al. 2018). These findings agreed with Su et al. (2017), who reported a noticeable up-regulating of hepcidin (HFE) gene in the liver of yellow catfish when fed with YMW supplemented diets. The gene expression of immunoglobulin M (IgM), major histocompatibility complex (MHC) and Interleukin-2 (IL-2) were also noticeably high when YMW supplemented diets were tested on the yellow catfish. These biochemical indices are crucial to the response of bacterial invasion, tissue damage, and immunological reactions (Rauta et al. 2012; Arenas-Ramirez et al. 2015; Su et al. 2017).

Shrimp has naturally lacked an adaptive immune system; this makes the innate immune system is critically important for them to fight against diseases. In a study conducted on *Litopenaeus vannamei*, partly replacement of FM (50%) with YMW in dietary preparation led to an up-regulated expression of immune genes, including  $\beta$ -1, 3-glucan binding proteins (BGBP), prophenoloxidase (proPO), and crustin, which are the important genes in promoting disease-resistant against white spot syndrome virus (WSSV) (Choi et al. 2018). BGBP is an activating factor to proPO cascade, it plays a key role in the shrimp immunity system. While, activated proPO is important in recognizing non-self-invaders, hemocyte communication, and melanin production in the shrimps (Vargas-Albores and Yepiz-Plascencia 2000; Bachère 2000). Crustin is another important AMP in shrimps, which is an essential requirement in the cellular defense. Choi et al. (2018) suggested that the activation of these immune genes mentioned above is partly due to the increased amount of circulating hemocytes. They postulated that the inclusion of YMW in the diet possibly enhanced the levels of adenosine triphosphate (ATP), and the total hemocyte count (THC), which involve substantially in the phagocytosis and defense proteins releasing. Motte et al. (2019) also showed that dietary inclusion of defatted YMW improved the survival rates when *L. vannamei* challenged against *Vibrio parahaemolyticus*. At the same time, THC and phenoloxidase (PO) activities were found significantly higher when dietary YMW was included in the shrimp feeding regime, this suggesting that immunity and disease resistance in shrimp was greatly enhanced with YMW diet. PO activity is the main indicator of the immunity system in many crustaceans, it was found 25 times higher in the shrimps fed with YMW compared to those fed purely on FM. The authors also speculated chitin, polysaccharides, nucleotides, AMPs, and/or melanin pigments in insects could play some roles in the immunological benefits to shrimps. A study on a closely related group of mealworms also showed that inclusion of SW meal in *L. vannamei* larval feeding improved non-specific immune indices e.g. SOD, CAT, ACP, and AKP (Zhao et al. 2011).

Like many insects, YMW's exoskeleton contains a substantial amount of chitin (Su et al. 2017; Li et al. 2017; Magalhães et al. 2017; Gasco et al. 2018; Henry et al. 2018). Low-level dietary inclusion of chitin had been reported to improve the immunomodulatory effects in fish (Hoffman et al. 1997; Kim and Rajapakse 2005; Lin et al. 2012). This could be due to that various cell surface receptors of chitin help to induce the generation of cytokine, and chemokine and thus activate the innate immune cells (Schuhmann et al. 2003; Fu et al. 2009; Ido et al. 2015; Henry 2018). Insect chitin also plays an essential role as a prebiotic, by



promoting growth factors and/or bacterial counts of the gut (Olsen et al. 2006; Sankian et al. 2018). Askarian et al. (2012) demonstrated that supplementation of chitin altered the structure of intestinal microbiota and increased the abundance of *Staphylococcus* sp., *Lactobacillus* sp., *Bacillus* sp. and *Acinetobacter* sp. in the gut of Atlantic salmon (*Salmo salar* L.). Some studies showed YMW diets could modulate microbiota, and the dominant bacteria in the gut systems of sea bream, sea bass, rainbow trout, and shrimp (Choi et al. 2018; Gasco et al. 2018). However, high inclusion of YMW (> 25%) in gilthead sea bream (*Sparus aurata*) indicated that ineffective in nutrients absorption, which in turn, causes unfavorable growth performance and nutrient utilization (Piccolo et al. 2017). Ng et al. (2001) also reported that full substitution of YMW (100%) in catfish, *Clarias gariepinus* reduced growth performance and protein utilization in the fish. While moderate supplementation of YMW (< 25%) in dietary preparation could be benefiting the cultured species. This clearly demonstrated in a study conducted on European sea bass (*Dicentrarchus labrax*) juveniles, the authors found that supplementation of YMW (~1%) improved protein level, protein digestibility and digestible EAAs in the fish (Ana et al. 2020). In another study, Doğankaya (2017) reported dietary FM replacement at 25% by SW showed a better growth performance on *O. mykiss* fingerlings compared to 0% and 50% of FM substitution, while 100% IM substitution harmed the fish overall performance. Thus, the complete substitution of FM with YMW and SW may not be a perfect plan, despite some health benefits exerted by these IMs.

### Housefly (*Musca domestica*)

Housefly maggot is an emerging candidate for IM in aquaculture farming. In a recent study done on bullfrog *Rana catesbeiana*, dietary inclusion of housefly maggot meal (HFMM) leads to enhanced activities of SOD, CAT, and total antioxidant capacity (T-AOC) (Li et al. 2019).

Under oxidative stresses, most of the vertebrates produce enzymatic and non-enzymatic antioxidant substances to protect themselves from these stresses. Therefore, antioxidant ability in the liver and intestine is an important indication of the overall health status of the hosts. Wen et al. (2015) showed that the increasing level of HFMM in Bagrid catfish, *Pelteobagrus fulvidraco* resulted in an enhancement of plasma and hepatopancreas (exocrine pancreatic tissue which separated from the liver by a connecting tissue) SOD activities (Wen et al. 2015). While feeding carp, *Cyprinus carpio* with HFMM containing diets could increase CAT activity in the liver, but no changes occurred in the level of GST (Ogunji et al. 2011). Liu et al. (2010) and Chao et al. (2012) also demonstrated that feeding *L. vannamei* with diets containing HFMM, and incorporated with lecithin, chitosan, and vitamins could significantly boost up the serum activities of SOD, AKP, ACP, peroxidase (POD), and MDA (Cao et al. 2012). Ming et al. (2013) found that adding 2.5% of HFMM to black carp, *Mylopharyngodon piceus* diet leads to an elevated level of serum LZM, complement 3 and 4 (C3 and C4), GPx, AKP, SOD, CAT, and SOD, as well as CAT activities in the liver. The contents of MDA in the serum and liver were reduced though. According to Liu et al. (2013), 4.2% dietary HFMM significantly improved swim bladder inflation (SBI), serum activities of SOD, GPx, and CAT, alongside the serum activities of SOD, GPx, and CAT. While, adding 5% dietary HFMM could significantly improve the activities of hepatic SOD, GPx, and CAT as well as LZM, C3, and C4. Generally, adding 4.2-5.0% HFMM to *M. piceus* diets could enhance the antioxidative capability and non-specific immunity in the fish (Liu et al. 2013).

Incongruent with the previous studies, findings of Li et al. (2019) revealed that increasing the amount of HFMM in bullfrog diets could enhance the activity of serum LZM. However, a study done by Lin and Mui (2017) showed that LZM activity in Asian sea bass (*Lates calcarifer*) was suppressed when the fish was fed with a diet containing 300 g kg<sup>-1</sup> HFMM. In crustacean, antibacterial peptide (ABPs) extracts from HFMM improve some immune-related indicators in *L. vannamei*. The maximum levels of phagocytic rate and hepatic LZM were observed when ABPs extracts were given at 3 g kg<sup>-1</sup>, while, higher serum activities of PO, POD, AKP, LZM, and T-AOC were reported with 2 – 3 g kg<sup>-1</sup> ABPs extracts (Chen et al. 2010). These differences might be due to the type of crude HFMM and extracts used in different studies. In this context, further investigation is needed to clarify the controversial roles of LZM activity, and its metabolic pathway in fish and crustacean models.

Heat shock protein 70 (HSP70) gene expression is another stress-inducing indicator which influenced by dietary composition. HSP70 helps to translate proteins, facilitate cellular or organellar transport of



proteins, and to maintain protein homeostasis in cell levels (Hemre et al. 2004; Cellura et al. 2006). Li et al. (2019) revealed when *R. catesbeiana* fed with HFMM, the expression of HSP70 in the liver was enhanced. Similar results were observed in *C. carpio*, when the fish were treated with silkworm pupae (SWP) meal (Ji et al. 2015) and defatted BSF (Li et al. 2017). Again, the related study in this area is still very scarce, more research is needed in HSP70 gene regulation in aquatic species.

In terms of hematological enhancement, Ido et al. (2015) showed that adding low levels of HFMM in red sea bream (*Pagrus major*) diets improved peritoneal leukocyte phagocytic activity. This activated innate immune system protected the fish against *Edwardsiella tarda* - a prevalent pathogenic agent in fish species. The authors attributed this effect to the AMPs which include defensin, cecropin, attacin, and dipterocin. Adding 2.5% of HFMM in the *M. piceus* diet resulted in high resistance against *A. hydrophila* as well as enhanced growth and antioxidant capacity (Ming et al. 2013). Okore et al. (2016) also demonstrated that the inclusion of dietary HFMM at a high level (e.g. 30 – 45%) enhanced the total count of WBCs (increased to  $3.8 \times 10^4 \text{ mm}^3$ ) of African catfish (*C. gariepinus*). The elevation of WBCs will lead to immunity enhancement and disease resistance in fish and animals (Akinwande et al. 2004).

Innate (non-specific) immunity is an immediate response of the specific immunity. It has an essential role in the immune system of teleost (Anderson 1992; Ibrahim et al. 2010). Phagocytosis, for example, has been recognized as one of the most substantial aspects of innate immunity in the defense system of the host against pathogenic invaders (MacArthur and Fletcher 1985). Wang et al. (2017) demonstrated that dietary replacement of HFMM at  $180 \text{ g kg}^{-1}$  showed a positive effect in flesh quality (e.g. higher water-holding capacity and instrumental texture) and health status of *O. niloticus*, however, a higher level of HFMM inclusion ( $>180 \text{ g kg}^{-1}$ ) could suppress the innate immunity of tilapia, causing significantly lower physiological performances in terms of survival rate, specific growth rate, weight gain, and also higher feed conversion rate (FCR). A report by Ido et al. (2015), demonstrated that the fork length, body weight, FCR, peritoneal leukocyte and phagocytic activity of red sea bream (*Pagrus major*) were improved after 6 months of feeding with a low level of HFMM. A recent study by Alofa et al. (2020) also showed that 20% of FM could be substituted by 25% of HFMM in the diet of Nile tilapia without posting any adverse effects on the growth performance and FCR. Like BSF and YMW mentioned early, partial replacement of HFMM could promote health status in many aquatic species. However, the complete substitution of FM by HFMM resulted in detrimental effects in some species, for instance, full replacement of FM by HFMM caused a decrease in macrophage phagocytosis in the head kidney of *O. niloticus* (Wang et al. 2017).

### Silkworm (*Bombyx mori*) pupae

Literatures showed that silkworm pupae (SWP) have been used as target IMs for some aquatic organisms, including African catfish (Kurbanov et al. 2015), chum salmon (Akiyama et al. 1984), Nile tilapia (Salem et al. 2008), common carp (Xu et al. 2018), rainbow trout (Shakoori et al. 2016), snakeskin Gourami (Jintasataporn et al. 2011) and Pacific white shrimp (Rahimnejad et al. 2019). Evidence showed that polysaccharides such as silkrose or dipterose, extracted from silkworm possess immunostimulatory effects that could improve the health status of mammals and aquatic species (Motte et al. 2019). Replacing SWP (68 – 90%) in Jian carp (*C. carpio*) diets had shown a reduction in the concentrations of plasma high-density lipoprotein cholesterol (HDLc), low-density lipoprotein cholesterol (LDLc), and TCHO in fish (Ji et al. 2015). These biochemical properties are some important indices to the stress-inducing factors. Some authors claimed the immunostimulatory effects of IM is partly attributed to chitin (Su et al. 2017; Li et al. 2017; Magalhães et al. 2017; Gasco et al. 2018; Henry et al. 2018), which is predominantly found in silkworm as in the form of chitosan (Paulino et al. 2006). The decreased concentrations of plasma TCHO and triglyceride (TG) in the plasma and liver of Jian carp presumably induced by chitin, and chitosan (Xia et al. 2011; Sankian et al. 2018). Chitin and its derivatives play a modulatory role in lipid metabolism through influencing the enterohepatic cycle of the bile acid, routine digestion, and absorption of lipids, as well as the process of generating fatty acids in the liver (Koide 1998; Xia et al. 2011).

The dietary inclusion of hydrolysates defatted SWP extracts (25%: 50%; SWP: FM) showed an enhancement of serum SOD and CAT activities in mirror carp (*Cyprinus carpio var. specularis*) (Xu et al. 2018). Similarly, hepatic SOD activity and MDA content in *C. carpio* fed with diets containing 25 – 50% *B. mori* chrysalis oil showed significant improvement after the feeding trials (Chen et al. 2017). Free radicals



induce oxidative stress that can cause damage to tissues and cells. As mentioned earlier, serum SOD and CAT, as the main protecting factors against oxidative stress, these enzymes maintain the levels of superoxide anions ( $O_2^-$ ) and hydrogen peroxide ( $H_2O_2$ ) inside the cells by enhancing oxidation resistance in fish (Xu et al. 2018). MDA is the products of a lipoxidation process, it contributes to the maintenance of homeostatic functions by acting as biological markers in oxidative stress, and tissue damage (Mateos et al. 2005; Surapaneni and Venkataramana 2007; Ji et al. 2015; Zhou et al. 2017). However, a high concentration of SWP (> 75%) in the diet replacement could cause adverse effects to oxidative stress, and subsequently, leads to lipid peroxidation damage to the hepatopancreas, and cause shrinkage of hepatopancreatic cells (Ji et al. 2015).

In general, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) has a direct relationship to hepatopancreatic damage and necrosis (Chen et al. 2017). In several studies on fish models, SWP inclusion in dietary supplementation could improve hepatic AST and ALT and non-specific immunity of mirror carp. SWP also showed positive protective effects against the fish liver's cells (Zhang et al. 2013). Substitution of FM with SWP level up to 50%, showed a significant increase in serum AST and ALT activities, meanwhile reducing serum glutathione (GLU) content and hepatic SOD activity in *C. carpio* (Ji et al. 2012; Zhang et al. 2013; Ji et al. 2015). With that, the authors concluded that replacing 50% FM with defatted SWP in the diets could improve the overall health status and growth performance of *C. carpio*.

The spleen is a major filtration organ of blood-borne antigens, it also plays an important function in immunopoiesis in the finfish immune system. Indeed, the spleen index (SI) has a direct indication of the immunity of fish (Ren et al. 2001). Replace FM with more than 40g kg<sup>-1</sup> fermented SWP meal mixture in mirror carp diets could significantly increase SI level (Zhou et al. 2017), and protection against disease WSSV in crayfish (*Procambarus clarkia*) (Wei and Xu 2005). This finding agreed with a previous study done on mice, which suggesting spleen weight increment after the mice were treated with SWP diets (Nan et al. 2013). However, increase the replacement levels of SWP in the diet may result in a reduction of MDA in mirror carp (Zhou et al. 2017).

In contrast to MDA, dietary FM replacement (up to 70%) with SWP in *C. carpio* diets resulted in the up regulation of hepatic HSP70 gene. HSP70 is an important indicator of disease occurrence *de facto*. This protein is often expressed at a very low level in a normal condition, high expression of HSP70 indicates the organism is under stresses caused by various factors such as pesticides, heavy metals, desiccation, diseases and parasites (Cellura et al. 2006; Ijaiya and Eko 2009; Jesus et al. 2013).

A study on innate immunity of sea cucumber, *Apostichopus japonicus* revealed that replacing dietary FM with 5% of silkworm meal slightly reduced phagocytosis, but enhanced LZM activity, while serum AKP did not show any significant difference among the treatment groups (Sun et al. 2014). Following the stimulation of phagocytic amoebocytes and  $O_2^-$  could generate other oxidants with a noticeable level of antimicrobial and cytotoxic functions in the animals (Coteur et al. 2002). AKP is an important component of the lysosomal enzyme. It significantly affects the lysis of phagocytized foreign matters in the coelomocytes, and enhance *A. japonicus* resistance against pathogens, immune status, as well as cytothysis (Li 2007; Wang et al. 2009; Zhang et al. 2010).

Another study on innate immunity of finfish, Shakoori et al. (2015) demonstrated that WBCs were noticeably high in *O. mykiss* when treated with dietary supplementation of SWP. As the level of WBCs increases, it would enhance the immune response towards the pathogens (Shakoori et al. 2015). In contrast, the lowering of RBCs and HB indicate signs of anemia (Ijaiya and Eko 2009). According to Zhang et al. (2019), peptidoglycan recognition proteins (PGRPs) in insects corresponding with the peptidoglycan (PGN) in bacterial cell walls. PGRPs participate in immune modulation as well as the pathogen clearance process. The authors also found that the presence of transmembrane protein, BmPGRP-S5 in *B. mori* larval hemocytes exhibited some activities of phagocytosis towards *E. coli* and *Bacillus megaterium*. Generally, SWP shows some promising results in immunity and oxidative stress enhancement, primarily due to the presence of AMPs as well as chitin, which both stimulating the effect on macrophage activity (Taufek et al. 2016b; Li et al. 2019).

## Other insects

Apart from the four main insect species discussed here, there is also a growing trend of research focusing on the replacement of FM with a variety of IMs in aquaculture feeds. Various studies have been done on other species as well (Table 1).



**Table 1** List of minor groups of insect meal used in aquaculture

No	The scientific name of insect	Common name	Target fish	Observations	Reference(s)
1	<i>Acheta domesticus</i>	House cricket	Walking catfish ( <i>C. batrachus</i> )	Among different animal and plant protein ingredients for replacing FM, dietary inclusion of dried <i>Acheta</i> sp. led to the lowest specific growth and survival rate.	Reeta et al. 2011b; 2011c
2	<i>Acheta domesticus</i>	House cricket	Tilapia ( <i>O. niloticus</i> )	Data on growth, food utilization and survival showed better performance at 10% inclusion of <i>Acheta</i> sp. meal, comparing to control (40% FM and no grasshopper meal).	Okoye and Nnaji 2005
3	<i>Acheta domesticus</i>	House cricket	African catfish ( <i>C. gariepinus</i> )	A diet containing 10% <i>Acheta</i> sp. meal and 30% FM yielded the best data on growth and nutrient utilization indices while 100% survival rate was reported with diets containing 30% <i>Acheta</i> sp. meal and 10% FM.	Nnaji and Okoye 2005
4	<i>Acheta domesticus</i>	House cricket	Red hybrid tilapia ( <i>Oreochromis</i> sp.)	Diet with 60% <i>A. domesticus</i> + 40% rice bran showed the best result in survival and growth rates. However, the increasing amount of <i>Acheta</i> sp. meal negatively affected liver histology.	Lee et al. 2017
5	<i>Acheta domesticus</i> + <i>Hermetia illucens</i>	House cricket + black soldier fly	Not determined	Dietary replacement of FM with cricket and BSF meal can result in suitable mineral composition (phosphorus and potassium) and leaching effect on pellets as the level of FM substitution increased to 75%.	Irungu et al. 2018
6	<i>Gryllus bimaculatus</i>	Field cricket	African catfish ( <i>C. gariepinus</i> )	It is possible to replace up to 100% FM in fish feed by <i>Gryllus</i> sp. without affecting body composition and feed utilization. Data on TP, WBC counts, survival rate, LZM, and globulin showed dietary CM could enhance the innate immune system and disease resistance against <i>A. hydrophila</i> .	Taufek et al. 2018a; 2018b
7	<i>Gryllus assimilis</i>	Field cricket	Tilapia ( <i>Oreochromis</i> sp.)	Data on antioxidant enzyme activities, hematological responses, and growth performance showed <i>Gryllus</i> sp. could be an adequate alternative to FM. Dietary inclusion of <i>Gryllus</i> sp. did not adversely affect growth indices as well as the digestibility of nutrients and AAs.	Taufek et al. 2016a; 2016b
8	<i>Poekilocerus pictus</i>	Grasshopper	Walking catfish ( <i>C. batrachus</i> )	The dietary inclusion of CM did not adversely affect growth indices as compared to commercial diet preparation.	Alfaro et al. 2019
9	<i>Zonocerus variegatus</i> L.	Variegated grasshopper	African catfish ( <i>C. gariepinus</i> )	The 100% substitution of dietary FM with <i>Poekilocerus</i> sp. meal did not affect hematological indices but led to adverse effects on the gills structure and ovarian steroidogenesis. Up to 25% of dietary FM replacement with <i>Zonocerus</i> sp. meal did not lead to any negative effect on nutrient utilization and growth.	Reeta et al. 2011a; 2011c Alegbeye et al. 2012



Table 1 continued

No	The scientific name of insect	Common name	Target fish	Observations	Reference(s)
10	<i>Locusta migratoria</i>	Migratory Locust	Tilapia ( <i>O. niloticus</i> )	Data on nutrient digestibility, hematological indices and growth performance showed up to 25% of FM can be replaced with <i>Locusta</i> sp. meal.	Emehinaiyé 2012
11	<i>Macrotermes subhyalinus</i>	Mendi termite	Vundu ( <i>Heterobranchus longifilis</i> )	Nutritional and economic evaluations showed 50% dietary inclusion of <i>Nasutitermes</i> sp. meal yields the best results in terms of profitability and sustainability.  Increasing levels of a mixture of de-winged reproductive termite – soybean meals, enhanced body protein, and lipid levels.	Sogbesan and Ugwumba 2008
12	<i>Macrotermes nigeriensis</i>	Mendi termite	African catfish ( <i>C. gariepinus</i> )	Dietary ratio of 75:25 ( <i>Macrotermes</i> sp.: soybean meal, respectively) yielded the best results in growth performance, nutrient utilization, and flesh quality.	Sadiku and Tiarniyu 2007
13	<i>Nasutitermes</i> sp.	Subterranean termite	Freshwater prawn ( <i>Macrobrachium rosenbergi</i> )	<i>Nasutitermes</i> sp. meal was not suitable as the sole protein source but can be used for AA supplementation since it can support growth and survival.	Serrano and Poku 2014
14	Not determined	Termite + other protein sources	Not determined	Data on AAs composition showed termite at 5.88% yields the best results. Then instead maggot, earthworm and chicken viscera meals were advisable as FM alternatives.	Djissou et al. 2018
15	<i>Oryctes rhinoceros</i>	Rhinoceros beetles	Climbing perch ( <i>Anabas testudineus</i> ), Goldfish ( <i>Carassius auratus</i> ), European carp ( <i>Cyprinus carpio</i> ), Oscar ( <i>Astronotus Ocellatus</i> )	<i>Oryctes</i> sp. powder is suitable for goldfish and carp on wheat mixtures. Supplemented pellets with <i>Oryctes</i> sp. powder are comparable with commercial fish feed.	Kamarudin et al. 2007
16	<i>Oryctes rhinoceros</i>	Rhinoceros beetles	Mud catfish ( <i>C. gariepinus</i> ) and Vundu ( <i>H. longifilis</i> )	Dietary FM can completely be replaced by <i>Oryctes</i> sp., however, in terms of growth performance and nutrient utilization, the most adequate level of FM replacement was 25%.	Fakayode and Ugwumba 2013



As shown in Table 1, only a limited study emphasizing the effects of IM diets on the immune-related responses or oxidative indices of fish (Reeta et al. 2010; Zhao et al. 2011; Taufek et al. 2016a; 2016b; Taufek et al. 2018a; 2018b). As we discussed earlier, the stress level in mammals and fish is closely associated with hematocrit percentage of packed cell volume (PCV), TLC, and HB (Ido et al. 2015; Nurin et al. 2018). Reeta et al. (2010) showed some hematological markers in the immunity of *C. batrachus* including HB%, PCV%, TLC, and complete blood count (CBC). This indicated that the dietary inclusion of grasshopper (*P. pictus*) could enhance the immune system and protection against pathogens. Similarly, cricket meal (CM) supplemented diets enhanced immunity and antioxidant in the African catfish. However, with high dietary inclusion of CM, it could lower down the oxidative activity as well as WBC counts (Taufek et al. 2016b). Despite CM lowering oxidative activity and WBC counts, fish fed with 100% CM replacement diet showed higher activity of GST without posting any negative effect on the growth parameters (Taufek et al. 2016b). GST plays a major role in detoxifying products of oxidative stress to some easily dischargeable products, via catalyzing the conjugation of some metabolites and products of lipoperoxidation. In the same study, the authors also demonstrated that CM could improve the immune response and protection against *A. hydrophila*. The author ascribed the immunostimulatory effect of CM is possibly due to the presence of antimicrobial activity of chitin, but not AMPs (Taufek et al. 2016b).

## Conclusions

Various insect species have been explored as alternative feeds for cattle, poultry, swine, and fishes for the past two decades. The trend is growing primarily due to substantial protein demand, and other nutritional contents e.g. EFA and functional peptides. In this review, we have carried out comprehensive coverage by highlighting the potential of IM as immunostimulatory agents, and health promoters in aquaculture. IM was found effectively improving the immunological status of aquatic animals, including humoral immune responses, immunocompetence, resistance against pathogens, immune genes expression, and antioxidant responses.

Based on the literature, high dietary inclusion of IM in aquafeeds may lead to immunosuppression, for example, phagocytic activity reduction. Therefore, complete FM replacement with IM in aquafeeds is remaining controversial. However, we suggest that partial replacement of FM with IM could offer a superior way to reduce the heavy dependence of FM in aquafeed manufacturing. On top of that, important substances e.g. AMPs and chitin in IM could serve as functional ingredients, in which the immune-stimulating and antioxidant properties could be fully explored to enhance the overall health status of the cultured species. Moreover, the immune-stimulating and antioxidant properties of AMPs in the insects could offer a natural alternative to synthetic drugs and antibiotics in aquaculture. In the effort of natural alternative exploitation, more works of genetic-based study on screening and extracting AMPs genes in IM is required to understand the roles of AMPs, and their effects on fish physiological and immunological indices. Field trial and *in vivo* assessments are needed to verify the effectiveness of IM in disease suppression and management.

## Future considerations and remarks

IM is an emerging protein source for various animal production industries. The potential of IM could not be underestimated. However, the understanding of physiological and immunomodulatory effects in aquaculture is still limited. Thus, it is important to explore IM from different biological aspects. Here, we summarized a few proposed areas for future studies:

- (i) Optimize the insect's nutritional values through standard feeding regimes;
- (ii) Explore the variety of insects that potentially can be used in aquafeed development;
- (iii) Understand the roles of chitin in immune induction and suppression. Further investigations are required on the digestibility, toxicity, and nutritional contents of chitin, this will help to clarify the mechanism of immunomodulation;
- (iv) Investigate the adaptive immune response of subsequent generations, e.g. cultured species gaining beneficial effects via IM administration may able to pass down genetic materials to their offspring, which could then enhance overall immunity at the later stages;
- (v) Optimize the production of insect mills; more research is needed to enhance the productivity of IMs to



- meet the market demands and to increase the cost-efficiency;
- (vi) Study the possibility of disease transmission e.g. pathogenic bacteria, viruses, parasites, and fungus, from unhealthy insects that used for aquafeed production. In other words, biosecurity of IM as a feed ingredient should be strictly monitored;
- (vii) Understand the mechanisms of action in AMPs, chitin and functional peptides on the immune stimulation through advanced technologies e.g. gene expression studies, agglutination assays, complement fixation, enzyme immunoassays, precipitation tests, Western blot tests, etc.;
- (viii) Improve the downstream processing of IM through low-temperature drying, de-fattening, and ensilaging.

**Conflict of interest** The authors declare no conflicts of interest.

**Authors' contribution** Mousavi S contributed to the information searching, drafting, and manuscript writing; Zahedinezhad S contributed to the editing and proofreading of the manuscript; Loh JY contributed to the writing framework, compilation, editing and improvement of the manuscript. All authors read and approved the final manuscript.

**Acknowledgments** The authors would like to thank the Faculty of Applied Sciences, UCSI University for the visiting scholar, and postdoc opportunity granted to Dr. Mousavi S under the supervision of Dr. Loh JY. The authors also express their sincere thanks to Dr. Gunasekaran B and Dr. Yam HC for their valuable comments during the manuscript preparation.

**Ethical approval** This article does not contain any studies with animals performed by any of the authors.

**List of abbreviations** AA: Amino acids; ABP: Antibacterial peptide; ACP: Acid phosphatase; AKP: Alkaline phosphatase; ALB: Albumin; ALT: Alanine aminotransferase; AMP(s): Antimicrobial peptide(s); AST: Aspartate aminotransferase; ATP: Adenosine triphosphate; BGBP:  $\beta$ -1, 3- glucan binding proteins; BSF: Black soldier fly; C3: Complement 3; C4: Complement 4; CAT: Catalase; CM: Cricket meal; CBC: Complete blood count; COS: Chito-oligosaccharides; DNA: Deoxyribonucleic acid; EAA(s): Essential amino acid(s); EFA(s): Essential fatty acid(s); EORD: Ethoxyresorufin O-deethylase; FA(s): Fatty acid(s); FCR: Feed conversion ratio; FM: Fish meal; GLO: Globulin; GLU: Glutathione; GPx: Glutathione peroxidase; GST: Glutathione S-transferase;  $H_2O_2$ : Hydrogen peroxide; HB: Hemoglobin; HDLc: High-density lipoprotein cholesterol; HFE: hepcidin; HFE: Housefly larvae; HFMM: Housefly maggot meal; HLS: Honey locust seed; HSP70: Heat shock protein 70; IgM: Immunoglobulin M; IL-2: Interleukin-2; IM: Insect meal; LA: Linoleic acid; LDLc: Low-density lipoprotein cholesterol; LNA: Linolenic acid; LZM: Lysozyme; MDA: Malondialdehyde; MHC: Major histocompatibility complex; NO: Nitric oxide;  $O_2^-$ : superoxide anions; PCV: Packed cell volume; PGN: peptidoglycan; PGRP(s): peptidoglycan recognition protein(s); PO: Phenoloxidase; POD: Peroxidase; ProPO: Prophenoloxidase; EROD: Ethoxyresorufin O-deethylase; RBC(s): Red blood cell(s); ROS: Reactive oxygen species; SBI: Swim bladder inflation; SCO: Silkworm chrysalis oil; SeGPx: Seleno-dependent glutathione peroxidase; SI: Spleen index; SOD: Superoxide dismutase; SWP: Silkworm pupae; T-AOC: Total antioxidant capacity; TBARS: Thiobarbituric acid reactive substances; TCHO: Total cholesterol; TG: Triglyceride; THC: Total Hemocyte Count; TLC: Total leukocyte; TP: Total protein; WBC(s): White blood cell(s); WSSV: White spot syndrome virus; YMW: Yellow mealworm

## References

- Akinwande AA, Moody FO, Sogbesan OA, Ugwumba AAA, Ovie SO (2004) Haematological response of *Heterobranchus longifilis* fed varying dietary protein levels. Proceedings of the 19th Annual Conference of the Fisheries Society of Nigeria, Ilorin, Nigeria, pp 715-718
- Akiyama T, Murai T, Hirasawa Y, Nose T (1984) Supplementation of various meals to fish meal diet for chum salmon fry. *Aquaculture* 37: 217-222
- Akiyama N, Hijikata M, Kobayashi A, Yamori T, Tsuruo T, Natori S (2000) Anti-tumor effect of N-beta-alanyl-5-S-glutathionyl dihydroxyphenyl alanine (5-S-GAD), a novel anti-bacterial substance from an insect. *Anticancer Res* 20(1A): 357-362
- Alegbeleye WO, Obasa SO, Olude OO, Otubu K, Jimoh W (2012) Preliminary evaluation of the nutritive value of the variegated grasshopper (*Zonocerus variegatus* L.) for African catfish *Clarias gariepinus* (Burchell. 1822) fingerlings. *Aquac Res* 43(3): 412-420
- Alfaro AO, Núñez WL, Marcia J, Fernández IM (2019) The Cricket (*Gryllus assimilis*) as an alternative food versus commercial concentrate for tilapia (*Oreochromis* sp.) in the nursery stage. *J Agric Sci* 11(6): 97-104
- Alofa CS, Adite A, Abou Y (2020) Evaluation of spirulina (*Spirulina platensis*) wastes and live housefly (*Musca domestica*) larvae as dietary protein sources in diets of *Oreochromis niloticus* (Linnaeus 1758) fingerlings. *Aquat Res* 3(1): 24-35
- Ana B, Elisabete M, Luisa MPV (2020) Nutritional value of different insect larvae meals as protein sources for European sea bass (*Dicentrarchus labrax*) juveniles. *Aquaculture* 521: 735085. <https://doi.org/10.1016/j.aquaculture.2020.735085>
- Anderson DP (1992) Immunostimulants, adjuvants, and vaccine carriers in fish: applications to aquaculture. *Annu Rev Fish Dis* 2: 281-307
- Arenas-Ramirez N, Woytschak J, Boyman O (2015) Interleukin-2: biology, design and application. *Trends Immunol* 36(12): 763-777
- Askarian F, Zhou Z, Olsen RE, Sperstad S, Ringø E (2012) Culturable autochthonous gut bacteria in Atlantic salmon (*Salmo salar* L.) fed diets with or without and chitin. Characterization by 16S rRNA gene sequencing, ability to produce enzymes and *in vitro* growth inhibition of four fish pathogens. *Aquaculture* (1-8): 326-329
- Bachère E (2000) Shrimp immunity and disease control. *Aquaculture* 191(1-3): 3-11
- Banjo AD, Lawal OA, Songonuga EA (2006) The nutritional value of fourteen species of edible insects in southwestern Nigeria. *Afr J Biotechnol* 5(3): 298-301
- Barroso FG, de Haro C, Sánchez-Muros MJ, Venegas E, Martínez-Sánchez A, Pérez-Bañón C (2014) The potential of various insect



- species for use as food for fish. *Aquaculture* 422: 193-201
- Bhaskar BR, Rao KS (1984) Influence of environmental variables on haematological ranges of milkfish, *Chanos chanos* (Forsk.), in brackish water culture. *Aquaculture* 83(1-2): 123-136
- Cao JM, Yan J, Huang YH, Wang GX, Zhang RB, Chen XY, Wen YH, Zhou TT (2012) Effects of replacement of fish meal with housefly maggot meal on growth performance, antioxidant and non-specific immune indexes of juvenile *Litopenaeus vannamei*. *Shuichan Xuebao* 36(4): 529-537
- Cellura C, Toubiana M, Parrinello N, Roch P (2006) HSP70 gene expression in *Mytilus galloprovincialis* hemocytes is triggered by moderate heat shock and *Vibrio anguillarum*, but not by *V. splendidus* or *Micrococcus lysodeikticus*. *Dev Comp Immunol* 30(11): 984-997
- Chen B, Cao J, Chen PJ, Zhao H, Lan H, Zhu X (2010) Effects of antibacterial peptides of *Musca domestica* on growth performance and immune related indicators in *Litopenaeus vannamei*. *Shuichan Xuebao* 17(2): 258-266
- Chen H, Tian J, Wang Y, Yang K, Ji H, Li J (2017) Effects of dietary soybean oil replacement by silkworm, *Bombyx mori* L., chrysalis oil on growth performance, tissue fatty acid composition, and health status of juvenile Jian carp, *Cyprinus carpio* var. Jian. *J World Aquacult Soc* 48(3): 453-466
- Choi WH, Yun JH, Chu JP, Chu KB (2012) Antibacterial effect of extracts of *Hermetia illucens* (Diptera: S. tratiomyidae) larvae against Gram negative bacteria. *Entomol Res* 42(5): 219-226
- Choi IH, Kim JM, Kim, NJ, Kim JD, Park C, Park JH, Chung TH (2018) Replacing fish meal by mealworm (*Tenebrio molitor*) on the growth performance and immunologic responses of white shrimp (*Litopenaeus vannamei*). *Acta Sci Anim Sci* 40. <https://doi.org/10.4025/actascianimsci.v40i1.39077>
- Christian C, Manuela R, Carola L, Alessio B, Marta G, Marco M, Sihem D, Achille S, Francesco G, Antonia CE, Marino P, Laura G (2020a) First insights on black soldier fly (*Hermetia illucens* L.) larvae meal dietary administration in Siberian sturgeon (*Acipenser baerii* Brandt) juveniles. *Aquaculture* <https://doi.org/10.1016/j.aquaculture.2019.734539>
- Christian C, Laura G, Ilaria B, Vanda M, Katia V, Marino P, Paolo P, Maria CB, Danila RF, Achille S, Antonia CE, Ambrosius JMD, Francesco G (2020b) Could dietary black soldier fly meal inclusion affect the liver and intestinal histological traits and the oxidative stress biomarkers of Siberian sturgeon (*Acipenser baerii*) juveniles? *Animals* 10(1): 155
- Coteur G, Warnau M, Jangoux M, Dubois P (2002) Reactive oxygen species (ROS) production by amoebocytes of *Asterias rubens* (Echinodermata). *Fish Shellfish Immunol* 12(3): 187-200
- Djissou AS, Odjo I, Godome T, Koshio S, Fiogbe ED (2018) Amino acids composition of maggot, earthworm, termite and chicken viscera meals used as proteins sources in fish feeding. *IJA* 8(1): 114-118
- Doğankaya L (2017) Effects of fish meal substitution with super worm (*Zophobas morio*) meal on growth performance of rainbow trout fingerlings. *Turk J Fish Aquat Sci* 32(1): 1-7
- Elia AC, Capucchio MT, Caldaroni B, Magara G, Dörr AJM, Biasato I, Gai F (2018) Influence of *Hermetia illucens* meal dietary inclusion on the histological traits, gut mucin composition and the oxidative stress biomarkers in rainbow trout (*Oncorhynchus mykiss*). *Aquaculture* 496: 50-57
- Emehinaiy PA (2012) Growth performance of *Oreochromis niloticus* fingerlings fed with varying levels of migratory locust (*Locusta migratoria*) meal. Dissertation, Federal University of Agriculture, Abeokuta, Ogun State
- Esteban MA, Mulero V, Cuesta A, Ortuno J, Meseguer J (2000) Effects of injecting chitin particles on the innate immune response of gilthead seabream (*Sparus aurata* L.). *Fish Shellfish Immunol* 10(6): 543-554
- Esteban MA, Cuesta A, Ortuno J, Meseguer J (2001) Immunomodulatory effects of dietary intake of chitin on gilthead seabream (*Sparus aurata* L.) innate immune system *Fish Shellfish Immunol* 11(4): 303-315
- Fakayode O, Ugwumba A (2013) Effects of replacement of fishmeal with palm grub (*Oryctes rhinoceros* Linnaeus, 1758) meal on the growth of *Clarias gariepinus* (Burchell, 1822) and *Heterobranchus longifilis* (Valenciennes, 1840) fingerlings. *J Fish Aquat Sci* 8: 101-107
- Foyosal MJ, Nguyen T, Chaklader MR, Siddik M, Tay CY, Fotedar R, Gupta SK (2019) Marked variations in gut microbiota and some innate immune responses of freshwater crayfish, marron (*Cherax cainii*, Austin 2002) fed dietary supplementation of *Clostridium butyricum*. *PeerJ* 7:e7553. <https://doi.org/10.7717/peerj.7553>
- Fu P, Wu J, Guo G (2009) Purification and molecular identification of an antifungal peptide from the hemolymph of *Musca domestica* (housefly). *Cell Mol Immunol* 6(4): 245
- Gabler F, Vinnerås B (2014) Using black soldier fly for waste recycling and effective *Salmonella* spp. reduction. Dissertation, Swedish University of Agricultural Sciences, Sweden]
- Gai F, Gasco L, Daprà F, Palmegiano GB, Sicuro B (2012) Enzymatic and histological evaluations of gut and liver in rainbow trout, *Oncorhynchus mykiss*, fed with rice protein concentrate based diets. *J World Aquacult Soc* 43(2): 218-229
- Gasco L, Finke M, Van Huis A (2018) Can diets containing insects promote animal health? *J. Insects Food Feed* 4(1): 1-4
- Harlystiarini H, Mutia R, Wibawan IWT, Astuti DA (2019) In vitro antibacterial activity of black soldier fly (*Hermetia Illucens*) larva extracts against gram-negative bacteria. *Bul Peternak* 43(2): 125-129
- Harikrishnan R, Kim JS, Balasundaram C, Heo MS (2012) Dietary supplementation with chitin and chitosan on haematology and innate immune response in *Epinephelus bruneus* against *Philasterides dicentrarchi*. *Exp Parasitol* 131: 116-124
- Hemre GI, Deng DF, Wilson RP, Berntssen MH (2004) Vitamin A metabolism and early biological responses in juvenile sunshine bass (*Morone chrysops* × *M. saxatilis*) fed graded levels of vitamin A. *Aquaculture* 235(1-4): 645-658
- Henry M, Gasco L, Piccolo G, Fountoulaki E (2015) Review on the use of insects in the diet of farmed fish: past and future. *Anim Feed Sci Tech* 203: 1-22
- Henry MA, Gasco L, Chatzifotis S, Piccolo G (2018) Does dietary insect meal affect the fish immune system? The case of mealworm, *Tenebrio molitor* on European sea bass, *Dicentrarchus labrax*. *Dev Comp Immunol* 81: 204-209
- Hoffman J, Johansen A, Steiro K, Gildberg A, Stenberg E, Bøgvold J (1997) Chitoooligosaccharides stimulate Atlantic salmon, *Salmo salar* L., head kidney leukocytes to enhanced superoxide anion production in vitro. *Comp Biochem Phy B* 118(1): 105-115
- Hu J, Wang G, Huang Y, Sun Y, He F, Zhao H, Li N (2017) Effects of substitution of fish meal with black soldier fly (*Hermetia illucens*) larvae meal, in yellow catfish (*Pelteobagrus fulvidraco*) diets. *Isr J Aquacult-Bamid IJA* 69.2017.1382, 9 p
- Ibrahim, M. D., Fathi, M., Mesalhy, S., & El-Aty, A. A. (2010). Effect of dietary supplementation of inulin and vitamin C on the growth, hematology, innate immunity, and resistance of Nile tilapia (*Oreochromis niloticus*). *Fish Shellfish Immunol* 29(2): 241-246



- Iido A, Iwai T, Ito K, Ohta T, Mizushige T, Kishida T, Miura C, Miura T (2015) Dietary effects of housefly (*Musca domestica*) (Diptera: Muscidae) pupae on the growth performance and the resistance against bacterial pathogen in red sea bream (*Pagrus major*) (Perciformes: Sparidae). *Appl Entomol Zool* 50(2): 213-221
- Ijaiya AT, Eko EO (2009) Effect of replacing dietary fish meal with silkworm (*Anaphe infracta*) caterpillar meal on performance, carcass characteristics and haematological parameters of finishing broiler chicken. *Pak J Nutr* 8(6): 850-855
- Irunge FG, Mutungi CM, Faraj AK, Affognon H, Tanga C, Ekese S, Nakimbugwe D, Fiaboe KKM (2018) Minerals content of extruded fish feeds containing cricket (*Acheta domesticus*) and black soldier fly larvae (*Hermetia illucens*) fractions. *Int Aquat Res* 10: 101-113
- Jabir MDAR, Razak SA, Vikineswary S (2011) Effects of amino acid supplementation in super worm based diets on growth performance and feed utilization of juvenile Nile tilapia. *Int Fish Symposium 2011*: 72-75
- Jabir MDAR, Razak SA, Vikineswary S (2012a) Chemical composition and nutrient digestibility of super worm meal in red tilapia juvenile. *Pak Vet J* 32(4)
- Jabir MDAR, Razak SA, Vikineswary S (2012b) Nutritive potential and utilization of super worm (*Zophobas morio*) meal in the diet of Nile tilapia (*Oreochromis niloticus*) juvenile. *Afr J Biotechnol* 11(24): 6592-6598
- Jawad LA, Al-Mukhtar MA, Ahmed HK (2004) The relationship between haematocrit and some biological parameters of the Indian shad, *Tenuulosa ilisha* (Family Clupeidae). *Anim Biodiv Conserv* 27(2): 47-52
- Jesus TF, Inácio A, Coelho MM (2013) Different levels of hsp70 and hsc70 mRNA expression in Iberian fish exposed to distinct river conditions. *Genet Mol Biol* 36(1): 61-69
- Ji H, Cheng XF, Li J, Zhang JL, Liu C (2012) Effect of dietary replacement of fish meal protein with silkworm pupae on the growth performance, body composition, and health status of *Cyprinus carpio* var. *specularis* fingerlings. *Shuichan Xuebao* 36: 1599-1611
- Ji H, Zhang JL, Huang JQ, Cheng XF, Liu C (2015) Effect of replacement of dietary fish meal with silkworm pupae meal on growth performance, body composition, intestinal protease activity and health status in juvenile Jian carp (*Cyprinus carpio* var. Jian). *Aquac Res* 46(5): 1209-1221
- Jintasataporn O, Chumkam S, Jintasataporn O (2011) Substitution of silkworm pupa (*Bombyx mori*) for fish meal in broodstock diets for snakeskin gourami (*Trichogaster pectoralis*). *J Agri Sci Tech-Iran* 1(8): 1341-1344
- Kamarudin N, Moslim R, Arshad O, Wahid MB, Chong A (2007) Potential of utilizing rhinoceros beetles (*Oryctes rhinoceros*) as an ornamental fish feed supplement. *J Oil Palm Res* 19: 313-318
- Kim SK, Rajapakse N (2005) Enzymatic production and biological activities of chitosan oligosaccharides (COS): A review. *Carbohydr Polym* 62(4): 357-368
- Koide SS (1998) Chitin-chitosan: properties, benefits and risks. *Nutr Res* 18(6): 1091-1101
- Kouřimská L, Adámková A (2016) Nutritional and sensory quality of edible insects. *NFS J* 4: 22-26
- Kurbanov AR, Milusheva RY, Rashidova SSh, Kamilov BG (2015) Effect of replacement of fish meal with silkworm (*Bombyx mori*) pupa protein on the growth of *Clarias gariepinus* fingerling. *Int J Fish Aquat Stud* 2(6): 25-27
- Lalander CH, Fidjeland J, Diener S, Eriksson S, Vinnerås B (2015) High waste-to-biomass conversion and efficient *Salmonella* spp. reduction using black soldier fly for waste recycling. *Agron Sustain Dev* 35(1): 261-271
- Lee SW, Tey HC, Wendy W, Zahari MW (2017) The effect of house cricket (*Acheta domesticus*) meal on growth performance of red hybrid tilapia (*Oreochromis* sp.). *Int J Aquat Sci* 8(2): 78-82
- Li JY (2007) The immunological characters and pathogenic study of cultured *Apostichopus Japonicus*. Dissertation, Ocean University of China, Qingdao (in Chinese)
- Li S, Ji H, Zhang B, Zhou J, Yu H (2017) Defatted black soldier fly (*Hermetia illucens*) larvae meal in diets for juvenile Jian carp (*Cyprinus carpio* var. Jian): Growth performance, antioxidant enzyme activities, digestive enzyme activities, intestine and hepatopancreas histological structure. *Aquaculture* 477: 62-70
- Li X, Rahimnejad S, Wang, L., Lu, K., Song, K., & Zhang, C. (2019). Substituting fish meal with housefly (*Musca domestica*) maggot meal in diets for bullfrog *Rana (Lithobates) catesbeiana*: Effects on growth, digestive enzymes activity, antioxidant capacity and gut health. *Aquaculture* 499: 295-305
- Lin YH, Shiau SY (2003) Dietary lipid requirement of grouper, *Epinephelus malabaricus*, and effects on immune responses. *Aquaculture* 225(1-4): 243-250
- Lin YH, Shiau SY (2005) Dietary vitamin E requirement of grouper, *Epinephelus malabaricus*, at two lipid levels, and their effects on immune responses. *Aquaculture* 248(1-4): 235-244
- Lin YH, Shiau SY (2007) The effects of dietary selenium on the oxidative stress of grouper, *Epinephelus malabaricus*, fed high copper. *Aquaculture* 267(1-4): 38-43
- Lin S, Mao S, Guan Y, Lin X, Luo L (2012) Dietary administration of chitoooligosaccharides to enhance growth, innate immune response and disease resistance of *Trachinotus ovatus* Fish Shellfish Immun 32(5): 909-913
- Lin YH, Mui JJ (2017) Evaluation of dietary inclusion of housefly maggot (*Musca domestica*) meal on growth, fillet composition and physiological responses for barramundi, *Lates calcarifer*. *Aquac Res* 48(5): 2478-2485
- Liu L, Li S, Chen J, Zhang C, Wang H (2010) Effect of fresh housefly larva on growth and immunity in Pacific white leg shrimp *Litopenaeus vannamei*. *Fisheries Science (Dalian)* 29(12): 721-724
- Liu L, Ye JY, Wu CL, Zhang YX, Shao XP, Jiang ZQ, Ming JH, Liu P (2013) Effects of Maggot meal on non-specific immunity and antioxidative capability of black carp (*Mylopharyngodon piceus*). *J Zhejiang Ocean Univ (Natural Science)* 2: 103-108 (in Chinese)
- Loh JY, Chan HK, Yam HK, In LLA, Lim CSY (2020). An overview of the immunomodulatory effects exerted by probiotics and prebiotics in grouper fish. *Aquacult Int* 28:729-750
- MacArthur JI, Fletcher TC (1985) Phagocytosis in fish. In: *Fish immunology*. Academic Press] pp. 29-46
- Magalhães R, Sánchez-López A, Leal RS, Martínez-Llorens S, Oliva-Teles A, Peres H (2017) Black soldier fly (*Hermetia illucens*) pre-pupae meal as a fish meal replacement in diets for European seabass (*Dicentrarchus labrax*). *Aquaculture* 476: 79-85
- Martínez-Álvarez RM, Morales AE, Sanz A (2005) Antioxidant defenses in fish: biotic and abiotic factors. *Rev Fish Biol Fisher* 15(1-2): 75-88
- Mateos R, Lecumberri E, Ramos S, Goya L, Bravo L (2005) Determination of malondialdehyde (MDA) by high-performance liquid chromatography in serum and liver as a biomarker for oxidative stress: Application to a rat model for hypercholesterolemia and evaluation of the effect of diets rich in phenolic antioxidants from fruits. *J Chromatogr B* 827(1): 76-82
- Ming J, Ye J, Zhang Y, Yang X, Wu C, Shao X, Liu P, (2013) The influence of maggot meal and L-carnitine on growth, immunity,



- antioxidant indices and disease resistance of black carp (*Mylopharyngodon piceus*). J Chinese Cereals Oils Assoc 28: 80-86
- Misra CK, Das BK, Mukherjee SC, Pattnaik P (2006) Effect of multiple injections of  $\beta$ -glucan on non-specific immune response and disease resistance in *Labeo rohita* fingerlings. Fish Shellfish Immun 20(3): 305-319
- Monaghan P, Metcalfe NB, Torres R (2009) Oxidative stress as a mediator of life history trade-offs: mechanisms, measurements and interpretation. Ecol Lett 12(1): 75-92
- Motte C, Rios A, Lefebvre T, Do H, Henry M, Jintasatoporn O (2019) Replacing fish meal with defatted insect meal (Yellow mealworm *Tenebrio molitor*) improves the growth and immunity of pacific white shrimp (*Litopenaeus vannamei*). Animals 9(5): 258. <https://doi.org/10.3390/ani9050258>
- Nan LU, Liao XY, Weng XC, Song HS (2013) Preparation of antioxidant peptides from silkworm pupae protein and evaluation of activities in vitro. J Shanghai Univ 19(2): 215-220 (in Chinese)
- Ng WK, Liew FL, Ang LP, Wong KW (2001) Potential of mealworm (*Tenebrio molitor*) as an alternative protein source in practical diets for African catfish, *Clarias gariepinus*. Aquac Res 32: 273-280
- Nnaji CJ, Okoye FC (2005) Substituting fish meal with grasshopper meal in the diet of *Clarias gariepinus* fingerlings. In: 19th Annual Conference of the Fisheries Society of Nigeria (FISON), Ilorin, Nigeria, pp 30-36
- Nurin FN, Maftuch, Yanuhar U (2018) Larvae of *Hermetia illucens* promotes the immunocompetence of haematology and muscle histopathology of common carp (*Cyprinus carpio*) challenged with *Aeromonas hydrophila*. IJSTR 7(4): 126-131
- Oda KS, Elisabeth H, Luisa P, Nina SL, Erik-Jan L, Marit E, Ikram B (2019) Effect of dietary replacement of fish meal with insect meal on *in vitro* bacterial and viral induced gene response in Atlantic salmon (*Salmo salar*) head kidney leukocytes. Fish Shellfish Immun 91: 223-232
- Ogunji JO, Nimptsch J, Wiegand C, Schulz C, Rennert B (2011) Effect of housefly maggot meal (maggemeal) diets on catalase, and glutathione S-transferase in the liver and gills of carp *Cyprinus carpio* fingerling. Int Aquat Res 3: 11-20
- Okore OO, Ekedo CM, Ubiaru PC, Uzodinma K (2016) Growth and haematological studies of African catfish (*Clarias gariepinus*) juveniles fed with housefly larva (*Musca domestica*) as feed supplement. IJAES 2(3): 21-30
- Okoye FC, Nnaji JC (2005) Effects of substituting fish meal with grasshopper meal on growth and food utilization of the Nile tilapia, *Oreochromis niloticus* fingerlings. In: 19th Annual Conference of the Fisheries Society of Nigeria (FISON), Ilorin, Nigeria, pp 37-44
- Olsen RE, Suontama J, Langmyhr E, Mundheim H, Ringø E, Melle W, Hemre GI (2006) The replacement of fish meal with Antarctic krill, *Euphausia superba* in diets for Atlantic salmon, *Salmo salar*. Aquac Nutr 12(4): 280-290
- Pamplona R, Costantini D (2011) Molecular and structural antioxidant defenses against oxidative stress in animals. Am J Physiol Regul Integr Comp Physiol 301(4): R843-R863
- Park K, Choi J, Nam S, Kim S, Kwak K, Lee S, Nho S (2014) Antioxidant activities of black soldier fly, *Hermetia illucens*. J Sericultural Entomolog Sci 52(2): 142-146
- Paul A, Frederich M, Megido C, Alabi T, Malik P, Uyttenbroeck R, Francis F, Blecker C, Haubruge E, Lognay G, Danthine S (2017) Insect fatty acids: A comparison of lipids from three orthopterans and *Tenebrio molitor* L. larvae. J Asia-Pac Entomol 20(2): 337-340
- Paulino AT, Simionato JI, Garcia JC, Jorge Nozaki J (2006) Characterization of chitosan and chitin produced from silkworm chrysalides. Carbohydr Polym 64: 98-103
- Piccolo C, Iaconisi V, Marono S, Gasco L, Loponte R, Nizza S, Bovera F, Parisi G (2017) Effect of *Tenebrio molitor* larvae meal on growth performance, *in vivo* nutrients digestibility, somatic and marketable indexes of gilthead sea bream (*Sparus aurata*). Anim Feed Sci Tech 226: 12-20
- Premalatha M, Abbasi T, Abbasi SA (2011) Energy-efficient food production to reduce global warming and ecodegradation: The use of edible insects. Renew Sust Energ Rev 15(9): 4357-4360
- Rahimnejad S, Hu SC, Song K, Wang L, Lu KL, Wu R, Zhang CX (2019) Replacement of fish meal with defatted silkworm (*Bombyx mori* L.) pupae meal in diets for Pacific white shrimp (*Litopenaeus vannamei*). Aquaculture 510: 150-159
- Rauta PR, Nayak B, Das S (2012) Immune system and immune responses in fish and their role in comparative immunity study: a model for higher organisms. Immunol Lett 148(1): 23-33
- Reeta J, Rakhi S, Johri PK (2010) Effect of different formulated plant and animal diet on hematology of *Clarias batrachus* Linn. under laboratory conditions. Biochem Cell Arch 10(2): 283-291
- Reeta J, Rakhi S, Johri PK (2011a) Impact of formulated plant and animal supplemented diets on nutritional efficiency, growth and body composition in juveniles of *Clarias batrachus* in experimental tanks. J Exp Zool India 14(1): 59-68
- Reeta J, Rakhi S, Johri PK (2011b) Studies on ovarian activity in formulated feed treated *Clarias batrachus* Linn. J Exp Zool India 14(1): 111-115
- Reeta J, Rakhi S, Johri PK (2011c) Histopathological examination of the gill, liver, kidney, stomach, intestine, testis and ovary of *Clarias batrachus* Linn. during the feeding on different formulated feeds. J Exp Zool India 14(1): 77-79
- Ren ZL, Huo QG, Zeng H, Guo Q (2001) Effect of oxidized fish oil on the performance and muscular histological structure of carps. Acta Zoonutrimenta Sinica 1(13): 59-64
- Rumpold BA, Schlüter OK (2013) Nutritional composition and safety aspects of edible insects. Mol Nutr Food Res 57(5): 802-823
- Sadiku SSS, Tiamiyu LO (2007) Wing reproductive termite (*Macrotermes nigeriensis*)–soybean (*Glyxine max*) meals blend as dietary protein source in the practical diets of *Heterobranchius bidorsalis* fingerlings. Pak J Nutr 6(3): 267-270
- Shakoori M, Gholipour H, Naseri S, Khara H (2016) Growth, survival, and body composition of rainbow trout, *Oncorhynchus mykiss*, when dietary fish meal is replaced with silkworm (*Bombyx mori*) pupae. Arch Pol Fisheries 24: 53-57
- Salem MFI, Khalafalla MME, Saad IAI, El-Hais AMA (2008) Replacement of fish meal by silkworm *Bombyx mori* pupae meal in Nile tilapia, *Oreochromis niloticus* diets. EJNF 11(3): 611-624
- Sankian Z, Khosravi S, Kim YO, Lee SM (2018) Effects of dietary inclusion of yellow mealworm (*Tenebrio molitor*) meal on growth performance, feed utilization, body composition, plasma biochemical indices, selected immune parameters and antioxidant enzyme activities of mandarin fish (*Siniperca scherzeri*) juveniles. Aquaculture 496: 79-87
- Saurabh S, Sahoo PK (2008) Lysozyme: an important defense molecule of fish innate immune system. Aquac Res 39(3): 223-239
- Schuhmann B, Seitz V, Vilcinskis A, Podsiadlowski L (2003) Cloning and expression of gallerimycin, an antifungal peptide expressed in immune response of greater wax moth larvae, *Galleria mellonella*. Arch Insect Biochem Physiol 53(3): 125-133



- Serrano AE, Poku G (2014) Nutritive value of termite as fish meal supplement in the diet of freshwater prawns (*Macrobrachium rosenbergii* de Man) juveniles. *ELBA Bioflux* 6(2): 63-71
- Shakoori M, Gholipour H, Naseri S (2015) Effect of replacing dietary fish meal with silkworm (*Bombyx mori*) pupae on hematological parameters of rainbow trout *Oncorhynchus mykiss*. *Comp Clin Path* 24(1): 139-143
- Sherman RA, Pechter EA (1988) Maggot therapy: a review of the therapeutic applications of fly larvae in human medicine, especially for treating osteomyelitis. *Med Vet Entomol* 2(3): 225-230
- Sogbesan AO, Ugwumba AAA (2008) Nutritional evaluation of termite (*Macrotermes subhyalinus*) meal as animal protein supplements in the diets of *Heterobranchius longifilis* (Valenciennes, 1840) fingerlings. *Turk J Fish Aquat Sc* 8(1): 149-157
- Su J, Gong Y, Cao S, Lu F, Han D, Liu H, Xie S (2017) Effects of dietary *Tenebrio molitor* meal on the growth performance, immune response and disease resistance of yellow catfish (*Pelteobagrus fulvidraco*). *Fish Shellfish Immunol* 69: 59-66
- Sun Y, Chang AK, Wen Z, Li Y, Du X, Li S (2014) Effect of replacing dietary fish meal with silkworm (*Bombyx mori* L.) caterpillar meal on growth and non-specific immunity of sea cucumber *Apostichopus japonicus* (Selenka). *Aquac Res* 45(7): 1246-1252
- Surapaneni KM, Venkataramana G (2007) Status of lipid peroxidation, glutathione, ascorbic acid, vitamin E and antioxidant enzymes in patients with osteoarthritis. *Indian J Med Sci* 61: 9-14
- Tan SW, Lai KS, Loh JY (2018) Effects of food wastes on yellow mealworm *Tenebrio molitor* larval nutritional profiles and growth performances. *EIMBO* 2(1): 1-6
- Tang Q, Dai Y, Zhou B (2012) Regulatory effects of *Tenebrio molitor* Linnaeus on immunological function in mice. *Afr J Biotechnol* 11(33): 8348-8352
- Taufek NM, Aspani F, Muin H, Raji AA, Razak SA, Alias Z (2016a) The effect of dietary cricket meal (*Gryllus bimaculatus*) on growth performance, antioxidant enzyme activities, and haematological response of African catfish (*Clarias gariepinus*). *Fish Physiol Biochem* 42(4): 1143-1155
- Taufek NM, Muin H, Raji AA, Razak SA, Yusof HM, Alias Z (2016b) Apparent digestibility coefficients and amino acid availability of cricket meal, *Gryllus bimaculatus*, and fishmeal in African catfish, *Clarias gariepinus*, diet. *J World Aquacult Soc* 47(6): 798-805
- Taufek NM, Muin H, Raji AA, Md Yusof H, Alias Z, Razak SA (2018a) Potential of field crickets meal (*Gryllus bimaculatus*) in the diet of African catfish (*Clarias gariepinus*). *J Appl Anim Res* 46(1): 541-546
- Taufek NM, Simarani K, Muin H, Aspani F, Raji AA, Alias Z, Razak SA (2018b) Inclusion of cricket (*Gryllus bimaculatus*) meal in African catfish (*Clarias gariepinus*) feed influences disease resistance. *J Fish*. doi: 10.17017/jfish.v6i2.2018.264
- Tran G, Heuzé V, Makkar HPS (2015) Insects in fish diets. *Anim Front* 5(2): 37-44
- Vargas-Albores F, Yepiz-Plascencia G (2000) Beta glucan binding protein and its role in shrimp immune response. *Aquaculture* 191(1-3): 13-21
- Vogel H, Müller A, Heckel DG, Gutzeit H, Vilcinskas A (2018) Nutritional immunology: diversification and diet-dependent expression of antimicrobial peptides in the black soldier fly *Hermetia illucens*. *Dev Comp Immunol* 78: 141-148
- Wang T, Sun Y, Jin L, Xu Y, Wang L, Ren T, Wang K (2009) Enhancement of non-specific immune response in sea cucumber (*Apostichopus japonicus*) by *Astragalus membranaceus* and its polysaccharides. *Fish Shellfish Immunol* 27(6): 757-762
- Wang L, Li J, Jin JN, Zhu F, Roffeis M, Zhang XZ (2017) A comprehensive evaluation of replacing fishmeal with housefly (*Musca domestica*) maggot meal in the diet of Nile tilapia (*Oreochromis niloticus*): growth performance, flesh quality, innate immunity and water environment. *Aquac Nutr* 23(5): 983-993
- Wei KQ, Xu ZR (2005) Effect of white spot syndrome virus envelope protein Vp28 expressed in silkworm (*Bombyx mori*) pupae on disease resistance in *Procambarus clarkii*. *Shi Yan Sheng Wu Xue Bao* 38(3): 190-198 (in Chinese)
- Xia WS, Liu P, Zhang JL, Chen J (2011) Biological activities of chitosan and chitooligosaccharides. *Food Hydrocol* 25(2): 170-179
- Xiao XP, Jin P, Zheng LY, Cai MM, Yu ZN, Yu J, Zhang JB (2018) Effects of black soldier fly (*Hermetia illucens*) larvae meal protein as a fishmeal replacement on the growth and immune index of yellow catfish (*Pelteobagrus fulvidraco*). *Aquac Res* 49(4): 1568-1577
- Xu XX, Ji H, Yu HB, Zhou JS (2018) Influence of replacing fish meal with enzymatic hydrolysates of defatted silkworm pupa (*Bombyx mori* L.) on growth performance, body composition and non-specific immunity of juvenile mirror carp (*Cyprinus carpio* var. specularis). *Aquac Res* 49(4): 1480-1490
- Yaghoubi M, Torfi Mozanadeh M, Marammazi JG, Safari O, Gisbert E (2017) Effects of dietary essential amino acid deficiencies on the growth performance and humoral immune response in silvery black porgy (*Sparidentex hasta*) juveniles. *Aquac Res* 48(10): 5311-5323
- Wen YH, Huang YH, Wang GX, Mo WY, Chen XY, Mi HF, Cao JM (2015) Effect of replacement of fish meal with maggot meal on antioxidant indexes, digestive enzyme activities, foregut and hepatopancreas histological structure of *Pelteobagrus fulvidraco*. *Feed Industry* 4: 29-35
- Zhang Q, Ma H, Mai K, Zhang W, Liufu Z, Xu W (2010) Interaction of dietary *Bacillus subtilis* and fructooligosaccharide on the growth performance, non-specific immunity of sea cucumber, *Apostichopus japonicus*. *Fish Shellfish Immunol* 29(2): 204-211
- Zhang JL, Yu P, Huang JQ, Hong JI, Qiu LJ, Yang K (2013) Effects of fish meal replacement by defatted silkworm pupae on growth performance, body composition and health status of Jian carp (*Cyprinus carpio* var. Jian). *Chinese J Animal Nutri* 25 (7): 1568-1578
- Zhang RN, Li CT, Ren FF, Ye MQ, Deng XJ, Yi HY, Cao Y, Yang WY (2019) Functional characterization of short-type peptidoglycan recognition proteins (PGRPs) from silkworm *Bombyx mori* in innate immunity. *Dev Comp Immunol* 95: 59-67
- Zhao HY, Zhao B, Zhang L, Su YJ (2011) Application of *Zophobas morio* protein powder to *Panaeus vannamei* larva feed. *J Tianjin Agric Univ* 18: 20-23
- Zhou JS, Chen YS, Ji H, Yu EM (2017) The effect of replacing fish meal with fermented meal mixture of silkworm pupae, rapeseed and wheat on growth, body composition and health of mirror carp (*Cyprinus carpio* var. specularis). *Aquac Nutr* 23(4): 741-754

#### Publisher's Note

IAU remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

