



## Review

# Insect meals as emerging protein source in the aquafeed sector: Emphasis on complementary effects to fishmeal, digestibility, carcass characterisation and immunomodulation in aquatic species

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## ABSTRACT

Insect meals received great attention in the aquafeed sector during the past decade due to the unsustainable demand for fishmeal. Desirable nutrient content, especially protein with balanced amino acids is of great importance in the use of insect meals as a viable protein source in aquafeeds. Insect meal could complement about 20-75% of dietary fishmeal in various aquatic species. They have better digestibility compared to low-quality fishmeal, oil meals/cakes, fermented plant proteins, feather meal; blood and meat meal and showed equal performance to high-quality fishmeal. Though partial substitution of fishmeal using insect meals could promote the health status, complete substitution resulted in detrimental effects in some aquatic species. This review concludes that insect meals are of great importance as a sustainable protein source and a viable complement to fishmeal in aquafeeds, thereby, helping to reduce the tremendous demand on fishmeal.

Keywords: Aquafeed, Carcass characterisation, Fishmeal, Immunomodulatory effect, Insect meal

## Introduction

Aquaculture is a pragmatic choice to fulfil the demand for food of the world's ever-increasing human population. FAO (2020) reported that the consumption of fish and fishery products is about 20% of total animal protein (*per capita* intake) in recent times, while it was only 17% in 2017. The increased *per capita* fish consumption (live weight equivalent) from 9 (1961) to 20.5 kg (2018) could be attributed to the expansion of the aquaculture sector. The global aquaculture production was 114.5 million t (Mt) in live weight in 2018 with an estimated value of USD 263.6 billion, which includes finfish (47.2%), crustaceans (8.7%), molluscs (15.7%), aquatic algae (24.5%) and the remaining being other aquatic species. Asian countries, in particular China, Indonesia, India, Vietnam, Bangladesh, Egypt, Norway and Chile contributed about 89% of the total farmed aquatic species. As a result, the aquafeed market is also simultaneously increasing to ensure reliable production of various aquatic species. Industry Report (2020) documented that the global aquafeed market size was valued at USD 63.8 billion and is expected to increase by 4.3% from 2020-2027. Fishmeal is a predominant choice in aquafeed formulations due to their

high protein content with balanced amino acid profiles, good appeal, palatability and digestibility. The reduction of global fishmeal production by 26.5% increased the cost from USD 452 (2000) to USD 1596 (2018) per ton (Jannathulla *et al.*, 2019c). To resolve the tremendous pressure on fishmeal, various proteins originating from both animal and plant sources have been experimented upon in the diet of different aquatic species with varying levels of success. Plant proteins, either individually or in combination, as a primary feed source could not be overlooked in formulations due to certain disadvantages such as, nutrient deficiency (Mousavi *et al.*, 2020), higher content of anti-nutrients (Jannathulla *et al.*, 2018b; 2019a) and poor nutrient assimilation (Dayal *et al.*, 2020). Similarly, the European Union (EU) Commission Regulation restricted use of rendered animal proteins due to the spread of Bovine Spongiform Encephalitis (Title 21 Part 589.2000 of the US Food and Drug Administration, Code of Federal Regulations). This led to the search for a suitable, sustainable and eco-friendly protein source with substantial availability and efficiency in terms of cost and energy conversion potential. Microbial proteins in terms of isolates and whole-cell biomass are being considered as novel protein source, but their high nucleic acid content

and toxicity factors have to be further explored in different aquatic species (Jannathulla *et al.*, 2021). On the other hand, insect-based products as feed ingredient played significant role in animal feed formulation for the past forty years; however, their incorporation into aquafeeds has not received much attention till the last decade. The findings of recent research (Panini *et al.*, 2017a; b; Xiao *et al.*, 2018; Belghit *et al.*, 2019; Mastoraki *et al.*, 2020a; b) suggested that insect meals could be potential protein source in aquafeeds. Earlier reviews were restricted to the nutritional composition of insect meals and their suitability based on dietary requirements of fish. But evaluating the usage of insect meals in the diet of aquatic species, including fish and shrimp, is extremely vital by considering growth performance, digestibility, nutrient balance, carcass characteristics, immune response, economics and sustainability in the production chain and this aspect is primarily discussed in the present review.

#### Permissibility for insect meals use in animal feeds

About 1900 species of edible insects have been identified worldwide, which includes various species of cricket, dragonfly, cockroach, beetle, spider, cicada, bambooworm, silkworm, waterbug, grasshopper, ant and fly larvae. Various studies have already been conducted on the utilisation of insects and their meals as ingredients in several feed industries, including aquaculture, poultry and swine. However, there is a debate still regarding the regulation on the usage of insects and their protein in animal feeds. There are no barriers to utilise insect-based products in developing countries, especially where entomophagy is traditionally practiced. However, the scenario is different in certain countries where their regulation process is either developed, reviewed and/or implemented (Dobermann *et al.*, 2017). As of 2009, the EU Commission Regulation (2009/1069/EC) banned insect meal use in livestock nutrition, since they were considered as a processed animal protein according to PAP act of 2001/999/EC (Toviho and Barsony, 2020). Later, the EU Commission Regulation (2017/893/EC) approved to use insect meals as a feedstuff in the aquaculture sector in 2017 and consequently to pig and poultry in 2019 (Tan *et al.*, 2018). However, Bruni *et al.* (2018) and Wang *et al.* (2018) reported that there is an authorisation by the EU Commission Regulation (893/24/05/2017) for seven insects that included *Hermetia illucens* (black soldier fly), *Musca domestica* (housefly), *Tenebrio molitor* (mealworm or yellow worm), *Alphitobius diaperinus* (lesser mealworm or litter beetle), *Acheta domesticus* (house cricket), *Gryllodes sigillatus* (tropical house cricket or banded cricket) and *Gryllus assimilis* (Jamaican field cricket), as these species are considered as non-pathogenic and do not cause any risk to the health of humans and animals. They also do not act as a vector

and do not transmit any pathogens between the living things, mainly human beings. Similarly, the EU Standing Committee on Plants, Animal, Food and Feed (SCoPaFF) recently allowed using insects as a protein source in the case of fish, mink and pet-food nutrition (Toviho and Barsony, 2020). In addition, the Canadian Food Inspection Agency (CFIA) established a policy to provide information about the safety and nutritional aspects of insect meals. Subsequently, the Federal Food Drug and Cosmetic Act (FFDCA) was amended with respect to use of insect meal (Global Market Insights, 2020). According to EFSA (2015), insect-based products, including whole insects, edible parts, isolates, extracts, meals and/or flours are recognised as novel foods, which are not consumed by human beings to a significant degree. However, recently various EU member countries, in particular UK, Holland, Austria and Belgium created their own legislative regulations through various food safety agencies and authorities with regard to using insect-based products as human food (Mancini *et al.*, 2019). This scenario is not only aiding to produce safe materials but also paves new employment opportunities by motivating the intensification of insect production.

#### Market potential

The global market of insect meal is increasing year by year and has already crossed USD 112 million. However, the projected growth differs according to surveys. Industry Report (2020) estimated that this sector will grow by 47% CAGR (compound annual growth rate) in the forecast period of 2019-2026 in terms of value, whereas Meticulous Research (2020) reported that the expected growth of this market will be 26.5% from 2020 to 2027 and it is USD 1398.9 million by 2027 in terms of volume. Feed Market (2020) reported that the insect market was valued at USD 687.8 million in 2018 globally and expected to reach USD 1396.4 million by 2024. This enormous growth is driven by various factors, mainly its nutritional value, low environmental impact on their entire life cycle, low risk of transmitting disease, increased emission of greenhouse gas from livestock and poultry. However, it is necessary to flag a standardised regulatory framework, physiological and ethical barriers to enhance the growth of this sector. In the global market, Europe has the largest share and accounted for approximately 45% of the total insect meal in 2018. This is mainly attributed to approvals given by the European Government for rearing insects and consumption of their meals/proteins. The second-largest share is by North America (38.4%) and its market is estimated to exceed USD 73297.9 thousand by 2023. Though Asia-Pacific region has less market share than rest of the world, various insect species are being reared, particularly in China, Korea, India, Thailand,

Malaysia and Vietnam, since these regions have the highest acceptance for insect meals/products. In addition, the Middle East and African countries markets are growing moderately compared to the rest of the world (Market Research Future, 2020). Of all the animal industries, aquaculture segment dominated with a share of 51% as of 2018 across the globe (Research and Markets, 2020). Due to decreased availability and increase in price of fishmeal, insect meals could be a viable option for the aquafeed sector in the future.

### Role of insect meals in aquaculture

Insect meals appear like brownish powder that is enriched protein materials with balanced amino acid profiles, which more closely resemble fishmeal. In addition, the presence of considerable quantities of micronutrients like antioxidants, minerals, vitamins and pigments ( $\beta$ -carotene), makes insect meals a potential feed source in the aquaculture sector and its impact on various aquatic species is discussed in the ensuing paragraphs.

#### *As replacement to fishmeal*

Among all the feed industries, the aquafeed segment consumes the largest quantity of fishmeal produced worldwide due to its richness in terms of nutritional characteristics and certain functional properties like palatability and attractability. Jannathulla *et al.* (2019c) reported that about 70-80% of fishmeal in global production is utilised by the aquafeed industry, in which shrimp consumed about 29%, salmonids 24%, marine fish 23% and others about 24%. The stagnant/reduced availability with escalating price of fishmeal has evoked interest in insect meals in recent times due to the constructive findings of recent studies (Table 1). Mastoraki *et al.* (2020a) investigated the effect of three different insect meals obtained from *T. molitor*, *H. illucens* and *M. domestica* individually by substituting 30% of dietary fishmeal in European seabass (*Dicentrarchus labrax*). Their findings revealed that the substitution of fishmeal using *H. illucens* and *M. domestica* meals exhibited similar growth performance compared to control (no fishmeal substitution), whereas a marginal increase of FCR was observed in the group reared on *T. molitor* meal. This is in agreement with the findings of Gasco *et al.* (2016) who reported that the inclusion level of *T. molitor* meal when exceeded 50% in the diet of European seabass, resulted in deterioration of feed intake. Sanchez-Muros *et al.* (2016) observed that *T. molitor* meal had not only reduced feed efficiency but also showed poor growth in Nile tilapia (*Oreochromis niloticus*) when substituting 50% of fishmeal. The authors suggested that this retarded performance with *T. molitor* meal might be due to higher content of chitin. Song *et al.* (2018) observed about 18%

of chitin in *T. molitor* and it is comparatively less in *H. illucens* (3.85-6.31%; Smets *et al.*, 2020) and *M. domestica* (8.02%; Kim *et al.*, 2016). In general, chitin is similar to fibres in structure and cannot properly be digested by various aquatic species. Excess of chitin hinders lipid absorption and fat digestibility, which might be a reason for poor growth as reported by Mastoraki *et al.* (2020a).

On the contrary, the positive effects of chitin have also been reported in earlier studies wherein it was stated that chitin could be a potential antioxidant (Kaya *et al.*, 2015) and an immunostimulant (Henry *et al.*, 2018), which can increase the abundance of *Bifidobacterium* genus in the gut, thus acting as a prebiotic, enhancing growth and digestibility (Munir *et al.*, 2016). Supporting these findings, improved growth performance was observed in rainbow trout (*Oncorhynchus mykiss*; Belforti *et al.*, 2015) and gilthead seabream (*Sparus aurata*; Piccolo *et al.*, 2017) when they were fed on a diet containing *T. molitor* meal by substituting 66 and 33% of fishmeal, respectively. Similarly, Motte *et al.* (2019) reported that the substitution of fishmeal by 50% using *T. molitor* meal increased weight up to 34% in *Penaeus vannamei* than those fed a control diet containing no *T. molitor* meal (no fishmeal substitution). This is in concordance with the findings of Choi *et al.* (2018) when using a full fat *T. molitor* meal in the same species. On the contrary, Panini *et al.* (2017a) found no improvement with full fat *T. molitor* meal until it was supplemented with amino acids (methionine and lysine) in *P. vannamei*. The incorporation of *T. molitor* meal has shown no impact in blackspot seabream, *Pagellus bogaraveo* (Iaconisi *et al.*, 2017) fed diets when substituting 50% of dietary fishmeal. Fishmeal substitution was increased to 45% in European seabass (Magalhaes *et al.*, 2017), 50% in rainbow trout (Mancini *et al.*, 2018) and 60% in Atlantic salmon (*Salmo salar*) without any deleterious effects on growth performance while using the insect meal of *H. illucens*. In contrast, Kroeckel *et al.* (2012) reported the negative effects in trout (*Psetta maxima*) when the fishmeal substitution was beyond 38% using *H. illucens* meal.

Cummins *et al.* (2017) studied the efficacy of *H. illucens* meal at different concentrations (7, 14, 21, 28 and 36%) by substituting protein of fishmeal in *P. vannamei*. They concluded that up to 25% of *H. illucens* meal can be included in shrimp. They suggested that poor performance with higher inclusion of *H. illucens* meal might be due to the deficiency of essential amino acids and the imbalance in the ratio of essential and non-essential amino acids. The performance was higher with *M. domestica* meal over *H. illucens* and *T. molitor* meals, which could substitute about 46-60% of dietary fishmeal in *P. vannamei* (Cao *et al.*, 2012), 75% in Nile tilapia (Ogunji *et al.*, 2008), Asian seabass (*Lates calcarifer*)

Table 1. Rate of fishmeal substitution using various insect meals in the diet of aquatic species

Source of insect meal	Species tested	Inclusion levels tested (%)	Fishmeal level in control diet (%)	Rate of fishmeal substitution (%)	References
<i>H. illucens</i>	<i>P. vannamei</i>	0, 4.5, 7.5, 10.5	15	50	Richardson <i>et al.</i> (2021)
<i>T. molitor</i>	<i>D. labrax</i>	0, 19.5	65	30	Mastoraki <i>et al.</i> (2020a)
<i>H. illucens</i>	<i>D. labrax</i>	0, 19.5	65	30	Mastoraki <i>et al.</i> (2020a)
<i>M. domestica</i>	<i>D. labrax</i>	0, 19.5	65	30	Mastoraki <i>et al.</i> (2020a)
<i>T. molitor</i>	<i>P. palaemon</i>	0, 7.8	26	30	Mastoraki <i>et al.</i> (2020b)
<i>H. illucens</i>	<i>P. palaemon</i>	0, 7.9	26	30	Mastoraki <i>et al.</i> (2020b)
<i>M. domestica</i>	<i>P. palaemon</i>	0, 7.10	26	30	Mastoraki <i>et al.</i> (2020b)
<i>H. illucens</i>	<i>O. mykiss</i>	0, 10, 20, 30	60	50	Terova <i>et al.</i> (2019)
<i>H. illucens</i>	<i>S. salar</i>	-	-	100	Stenberg <i>et al.</i> (2019)
<i>T. molitor</i>	<i>P. vannamei</i>	0, 5.2, 10.3, 15.4, 20.5	25	50	Motte <i>et al.</i> (2019)
<i>T. molitor</i>	<i>O. mykiss</i>	0, 5, 7.5, 15, 25	25	100	Rema <i>et al.</i> (2019)
<i>H. illucens</i>	<i>S. salar</i>	0, 4.91, 9.84, 14.75	10	100	Belghit <i>et al.</i> (2018)
<i>H. illucens</i>	<i>O. mykiss</i>	0, 6.6, 13.2, 26.4	20	50	Dumas <i>et al.</i> (2018)
<i>H. illucens</i>	<i>O. mykiss</i>	-	-	50	Mancini <i>et al.</i> (2018)
<i>T. molitor</i>	<i>P. vannamei</i>	0, 5, 10, 20	20	50	Choi <i>et al.</i> (2018)
<i>H. illucens</i>	<i>P. vannamei</i>	0, 7, 14, 21, 28, 36	25	25	Cummins <i>et al.</i> (2017)
<i>H. illucens</i>	<i>D. labrax</i>	0, 6.5, 13, 19.5	32.4	45	Magalhaes <i>et al.</i> (2017)
<i>T. molitor</i>	<i>S. aurata</i>	0, 25, 50	50	33	Piccolo <i>et al.</i> (2017)
<i>T. molitor</i>	<i>P. bogaraveo</i>	0, 21, 40	61	50	Iaconisi <i>et al.</i> (2017)
<i>M. domestica</i>	<i>L. calcarifer</i>	0, 10, 20, 30	30.3	75	Lin and Mui (2017)
<i>T. molitor</i>	<i>O. niloticus</i>	0, 21, 43	31	50	Sanchez-Muros <i>et al.</i> (2016)
<i>H. illucens</i>	<i>S. salar</i>	0, 5, 10, 20	20	100	Lock <i>et al.</i> (2016)
<i>T. molitor</i>	<i>D. labrax</i>	0, 25, 50	70	50	Gasco <i>et al.</i> (2016)
<i>T. molitor</i>	<i>O. mykiss</i>	0, 25, 50	75	66	Belforti <i>et al.</i> (2015)
<i>M. domestica</i>	<i>C. gariepinus</i>	0, 1.7, 3.3, 5, 6.7, 8.3	26.3	20	Idowu and Afolayan (2013)
<i>M. domestica</i>	<i>C. gariepinus</i>	0, 16.0, 22.8, 35.6, 43.7	31.7	75	Olaniyi and Salau (2013)
<i>H. illucens</i>	<i>P. maxima</i>	0, 16.5, 33.2, 48.6, 64.0, 75.6	68.1	38	Kroeckel <i>et al.</i> (2012)
<i>M. domestica</i>	<i>O. niloticus</i>	0, 15, 25, 35, 45, 500, 68	43	75	Ogunji <i>et al.</i> (2008)

(Lin and Mui, 2017) and African catfish (Olaniyi and Salau, 2013). However, both *M. domestica* and *H. illucens* meals exhibited similar results in the shrimp, *Palaemon adspersus* compared to those reared with a meal of *T. molitor* (Mastoraki *et al.*, 2020b), who suggested that the lower growth by feeding *T. molitor* meal could be attributed to the respective diet, which fails to fulfil the dietary requirement of *P. adspersus*.

Interestingly, 100% fishmeal substitution was reported (Oyelese, 2007) using a meal obtained from *M. domestica* and *T. molitor* in African catfish (*Clarias gariepinus*) and rainbow trout (Rema *et al.*, 2019), respectively. Similarly, Fasakin *et al.* (2003) achieved complete fishmeal substitution using oven dried *M. domestica* meal in clariid catfish (*Clarias gariepinus*). Though salmon growth was comparable with the diet containing *H. illucens* meal by substituting 100% fishmeal (Lock *et al.*, 2016; Belghit *et al.*, 2019), a slight reduction in feed intake was reported. The improved performance reported by Belghit *et al.* (2019) could be due to supplementation

of methionine and lysine. However, the finding of Dumas *et al.* (2018) showed that *H. illucens* meal could substitute only 50% of dietary fishmeal in rainbow trout even after supplementing amino acids (methionine and lysine). This difference might be due to the variation in the dietary amino acids' requirement of the cultured species (Magalhaes *et al.*, 2017), indicating that the species of omnivores can tolerate the inclusion of insect meals and a higher acceptance probably due to their habit of eating insects as a part of their natural food. Cummins *et al.* (2017) stated that the rearing environment also had a significant effect on the result of fishmeal substitution. For example, studies carried out with a green water system, where natural productivity contributes more to the nutrient intake of cultured species, has a higher possibility for the substitution of fishmeal, whereas in studies with a clear water system, where there are no exogenous supplemental nutrients, the substitution is quite challenging as they may not yield the desired results. However, the effect of meals from *A. diaperinus*, *A. domesticus*, *G. sigillatus* and *G. assimilis* does not seem to be addressed adequately.

*Effect on digestibility*

Knowledge on the digestibility parameters of any feed ingredient is important to evaluate its potential because the quality of ingredients depends not only on their nutritional characteristics but also on the bioavailability of nutrients. In addition, the determination of nutrient bioavailability guarantees the formulation of most economic and nutritious diets to fulfil the dietary requirement of cultured species. NRC (2011) stated that dry matter digestibility indicates about overall digestible fractions of both organic and inorganic matters; however, dry matter digestibility varies highly due to the differences in insoluble carbohydrate content of ingredients. Basto *et al.* (2020) estimated the digestibility of both *T. molitor* and *H. illucens* meals in European seabass before and after defatting. Their results indicated that defatted insect meals were superior in their digestibility compared to their respective counterparts. However, the meal of *T. molitor* showed a higher dry matter digestibility (72.4-85.2%) than the group fed *H. illucens* meal (48.3-53.7%). In contrast, *T. molitor* meal has a lower digestibility (45.9%) in *P. vannamei* (Panini *et al.*, 2017a). The wide difference noticed in the performance of *T. molitor* meal would be due to the nutritional variation. The *T. molitor* meal used by Panini *et al.* (2017a) had a higher lipid content than the one used by Basto *et al.* (2020), which might be a reason for reducing digestibility in their study. In addition, the meal of *T. molitor* used by Basto *et al.* (2020) contained a lower content of chitin (4.6-4.8%) and it was 6.1-6.5% in *H. illucens* meal. In addition, the nutrients that are highly sclerotised or that may be found in chitin would also reduce the dry matter digestibility of insect meals, which would have caused poor digestibility even for energy in *P. vannamei* (Panini *et al.*, 2017a). The reduction in nutrient digestibility due to the higher content of chitin was also reported in gilthead sea bream (Piccolo *et al.*, 2017).

Marono *et al.* (2015) suggested that chitin can reduce gut transit time and physically prevent the protein from enzyme degradation resulting in hindered digestibility. Shiao and Yu (1999) studied the effect of dietary chitin at varied levels of inclusions (0, 2, 5 and 10%) in Nile tilapia and observed a lower dry matter digestibility accompanied with the higher inclusion of chitin. However, Fontes *et al.* (2019) reported a higher digestibility of *T. molitor* meal in Nile tilapia compared to other meals obtained from *N. cinerea*, *G. portentosa* and *G. assimilis* (Table 2).

Protein digestibility is mainly dependent on the ability of the aquatic species in hydrolysing protein into tripeptides, dipeptides and free amino acids (NRC, 2011). Basto *et al.* (2020) reported that the meal of *H. illucens* showed a lower digestibility of protein (75.8-87.2%) than the *T. molitor* meals (89.2-92.8%). However, the protein digestibility was 63-88% in turbot (Kroeckel *et al.*, 2012), rainbow trout (Dumas *et al.*, 2018) and Nile tilapia (Fontes *et al.*, 2019). The authors suggested that these differences observed among the studies are probably due to the nutrient variation of insect meals and their processing methodologies. The dietary inclusion of *T. molitor* and *H. illucens* meals by substituting fishmeal (20-25%) showed protein digestibility in the range of 91-92% in European seabass and Asian seabass (Gasco *et al.*, 2016; Magalhaes *et al.*, 2017) and was slightly higher (93-94%) in rainbow trout (Rema *et al.*, 2019), while it was lower than 89% in gilthead seabream and Atlantic salmon (Piccolo *et al.*, 2017; Belight *et al.*, 2019). Terova *et al.* (2019) reported no variations in digestibility parameters in rainbow trout fed *H. illucens* meal at varied levels (0, 10, 20 and 30%) and reported >90% digestibility for protein and energy in all the treatments. Similarly, tilapia fed 30% *H. illucens* meal had comparable dry matter, protein and energy digestibility to those fed control and commercial diets (Davis *et al.*, 2017). This positive effect might be due

Table 2. Apparent digestibility coefficients of dry matter, protein, fat and energy of insect meals in various aquatic species

Source of insect meal	Species tested	ADMD <sup>†</sup>	APD <sup>‡</sup>	AFD <sup>§</sup>	AED <sup>¶</sup>	References
<i>H. illucens</i>	<i>D. labrax</i>	48.3	75.8	91.4	67.2	Basto <i>et al.</i> (2020)
<i>T. molitor</i>	<i>D. labrax</i>	85.2	89.2	94.5	79.2	Basto <i>et al.</i> (2020)
<i>T. molitor</i>	<i>O. niloticus</i>	95.8	85.4	90.6	82.1	Fontes <i>et al.</i> (2019)
<i>H. illucens</i>	<i>O. mykiss</i>	79.6	92.0	-	98.9	Terova <i>et al.</i> (2019)
<i>T. molitor</i>	<i>O. mykiss</i>	84.3	93.8	97.1	84.4	Rema <i>et al.</i> (2019)
<i>H. illucens</i>	<i>S. salar</i>	-	82.0	86.0	-	Belghit <i>et al.</i> (2018)
<i>H. illucens</i>	<i>O. mykiss</i>	77.0	88.0	73.0	-	Dumas <i>et al.</i> (2018)
<i>T. molitor</i>	<i>S. aurata</i>	87.4	87.3	-	89.9	Piccolo <i>et al.</i> (2017)
<i>H. illucens</i>	<i>D. labrax</i>	69.8	91.6	92.3	81.4	Magalhaes <i>et al.</i> (2017)
<i>T. molitor</i>	<i>P. vannamei</i>	45.9	76.1	-	66.5	Panini <i>et al.</i> (2017a)
<i>T. molitor</i>	<i>D. labrax</i>	80.0	92.0	97.0	-	Gasco <i>et al.</i> (2016)
<i>H. illucens</i>	<i>P. maxima</i>	45.2	63.1	78.0	54.5	Kroeckel <i>et al.</i> (2012)

<sup>†</sup>Apparent dry matter digestibility; <sup>‡</sup>Apparent protein digestibility; <sup>§</sup>Apparent fat digestibility; <sup>¶</sup>Apparent energy digestibility

to a lower chitin level in the diet (1.5%) even after including 30% of *H. illucens* meal than the threshold value of 2% (Renna *et al.*, 2017). A similar trend was observed for the inclusion of *H. illucens* meal in Atlantic salmon (Belghit *et al.*, 2019). Basto *et al.* (2020) reported differences in amino acid digestibility between insect meals (*T. molitor* and *H. illucens*) and processing (defatted and full fat) and their findings revealed >90% digestibility for all amino acids in all categories except cystine (49.4%).

In general, both methionine and lysine are the most limiting amino acids in the diets of aquatic species, hence an accurate measurement of these amino acids are of paramount importance to formulate an equilibrated diet. The digestibility of lysine was 97.6 and 95.3% in *H. illucens* and *T. molitor* meals, respectively. Digestibility of methionine was higher in full fat meal of *H. illucens* (100.7%) compared to the respective de-fatted one (95.8%), whereas *T. molitor* meal (100.5%) showed higher digestibility for methionine than its counterpart (95.3%) (Basto *et al.*, 2020). Almost similar results were reported in European seabass (Magalhaes *et al.*, 2017) and Atlantic salmon (Belghit *et al.*, 2020) fed on *H. illucens* meal, whereas essential amino acid digestibility was slightly reduced in *P. vannamei* reared on a meal of *T. molitor* (Panini *et al.*, 2017a) (Table 3). Though variations in digestibility have been reported among insect meals, most of the insect meals have better digestibility even compared to low-quality fishmeal (Basto *et al.*, 2020), oil meals/cakes (Jannathulla *et al.*, 2019b), fermented plant proteins (Jannathulla *et al.*, 2018a; b), feather meal (Campos *et al.*, 2018); blood and meat meal (Basto *et al.*, 2020). Similarly, the digestibility of insect meals is almost equal to the high-quality fishmeal, which demonstrates that there is very high potential for the utility of insect meals in aquatic species.

#### Impact on carcass characteristics

The WHO (2013) reported that long-term consumption of fish and fish-derived products reduces the global burden of disease, particularly in children (Mohanty *et al.*, 2014) due to their high quality and rich nutrients. As a result, the Association of Official Analytical Chemists (AOAC) and the Nutrition Labelling

and Education Act (NLEA) devised labelling protocols to aid in the prevention of protein-energy malnutrition (WHO, 2007; FAO/IFAD/WEF, 2013). Hence, producing aquatic species with good quality is imperative and it is noteworthy to assess the nutritional characteristics of farmed species reared on any novel ingredient, including insect meals, as feed mainly influences the carcass composition during farming. Inclusion of *M. domestica* meal in African catfish (Fasakin *et al.*, 2003), Asian seabass (Lin and Mui, 2017) and European seabass (Mastoraki *et al.*, 2020a), *H. illucens* meal in Jian carp (*Cyprinus carp*) (Li *et al.*, 2016), Atlantic salmon (Belghit *et al.*, 2018), European seabass (Mastoraki *et al.*, 2020a) and *T. molitor* meal in blackspot seabream (Iaconisi *et al.*, 2017) and *P. vannamei* (Rahimnejad *et al.*, 2019) had no effect on the nutrient composition of the cultured species. Similar effect was observed by Panini *et al.* (2017b) in *P. vannamei* fed on *T. molitor* meal, but a gradual increase of lipid was reported with increase in the inclusion level of insect meals, which might be due to increasing the dietary lipid content attributed to higher lipid present in the respective insect meals. In addition to the high protein, *T. molitor* is reported to have a very high lipid level compared to other insect meals. Gonzalez-Felix *et al.* (2002) also stated that dietary lipid can increase the lipid deposition in hepatopancreas and muscle in *P. vannamei*. A similar trend was reported in African catfish (Ng *et al.*, 2001) and tilapia (Sanchez-Mouros *et al.*, 2016) when farmed with *T. molitor* meal. In contrast, an increased carcass protein and ash with decreased lipid content was reported by Nandeeshia *et al.* (2002) in common carp fed on insect meal. These inconsistent results might be due to the variations in dietary composition, feeding periods, rearing conditions, animal size and species-specific differences (Rahimnejad *et al.*, 2017).

Fish and fisheries products have a greater satiety effect compared to other animal proteins like chicken, beef and pork. However, the amino acid composition varies from species to species, but their quality mainly depends on having all the essential amino acids in a proper proportion. Mastoraki *et al.* (2020a) observed certain difference in the amino acid composition/retention in European seabass fed on different insect meals (*T. molitor*, *H. illucens* and

Table 3. Apparent digestibility coefficients of essential amino acids of insect meals in various aquatic species

Source of insect meal	Species tested	Arg	His	Ile	Leu	Lys	Met	Phe	Thr	Trp	Val	References
<i>H. illucens</i>	<i>D. labrax</i>	90.9	97.6	92.6	79.1	92.6	92.2	90.3	100.7	92.6	92.1	Basto <i>et al.</i> (2020)
<i>T. molitor</i>	<i>D. labrax</i>	94.4	95.3	93.7	84.7	98.7	100	95.3	93.2	98.0	95.8	Basto <i>et al.</i> (2020)
<i>H. illucens</i>	<i>S. salar</i>	91.0	84.0	84.0	88.0	85.0	90.0	88.0	78.0	-	85	Belghit <i>et al.</i> (2018)
<i>H. illucens</i>	<i>O. mykiss</i>	91.0	91.0	90.0	90.0	92.0	90.0	90.0	70.0	93.0	-	Dumas <i>et al.</i> (2018)
<i>H. illucens</i>	<i>D. labrax</i>	96.7	98.1	88.5	81.5	93.6	93.3	93.6	93.1	-	91.9	Magalhaes <i>et al.</i> (2017)
<i>T. molitor</i>	<i>P. vannamei</i>	86.1	72.7	77.9	76.8	86.4	76.7	74.6	84.9	-	75.4	Panini <i>et al.</i> (2017a)

*M. domestica*). The variation in the structural and metabolic role might be a reason for obtaining such findings. Khalil and Khan (1995) documented that lysine is one of the most limiting amino acids in various food materials, in particular, a cereal-based diet, but it is essentially required for optimal growth, especially for children. This amino acid is relatively high in fish-based products and its level was further increased in gilthead seabream and rainbow trout due to the inclusion of *T. molitor* meal (Iaconisi *et al.*, 2019). They also observed a similar trend for leucine in both the fish. De Bandt and Cynober (2006) documented that leucine plays an important role in muscle protein synthesis and has an excellent therapeutic effect in various stress conditions of burn, trauma and sepsis. Similarly, alanine was found to be increased in gilthead sea bream and rainbow trout reared on *T. molitor* meal, but the increase was more abundant in rainbow trout. Alanine has a variety of roles in chronic renal failure and nervous system disorders concerning the implication of human nutrition (Vuzelov *et al.*, 1999).

As in lipids, insect meals also influence the body fatty acid composition of aquatic species especially, C16:0, C18:1 and C18:2 as they are rich in these constituents. The content of SFA was not affected by the inclusion of full fat *H. illucens* meal in European seabass (Gasco *et al.*, 2016) and rainbow trout (Iaconisi *et al.*, 2018), whereas Mancini *et al.* (2018) reported a low level of SFA in rainbow trout when fed on de-fatted *H. illucens* meal. This difference might be due to the extremely high content of C12:0 in a meal obtained from *H. illucens*. The level of MUFA, in particular, C18:1 increased proportionately with the level of a meal derived from *T. molitor* in tilapia (Sanchez-Muros *et al.*, 2016) and *P. vannamei* (Panini *et al.*, 2017b). However, the higher inclusion of *T. molitor* meal showed a reverse trend for PUFA. This is in agreement with the findings of Mastoraki *et al.* (2020a) in European seabass fed on *T. molitor*, *H. illucens* and *M. domestica* included diets and Iaconisi *et al.* (2017) and Sanchez-Muros *et al.* (2016) in blackspot seabream and Nile tilapia, respectively fed on full fat *T. molitor* meal. This could be due to the low level/absence of C20:5 and C22:6 fatty acids in the insect meals. However, Mancini *et al.* (2018) reported no difference in PUFA content in rainbow trout reared on both full fat and de-fatted *H. illucens* meal. This is possibly due to the PUFA content being maintained at a level that is high enough to fulfil the dietary requirement of rainbow trout by keeping a higher volume of fishmeal and fish oil in their diets. A similar effect was described by Skalli and Robin (2004) in European seabass. Though the fatty acid composition of diets has directly reflected on the body composition of the cultured species, the concentration of certain fatty acids like C20:4, C20:5 and C22:6 can be retained more in the body during the culture. Gonzalez-

Felix *et al.* (2002) suggested that this phenomenon is due to the selective retention capability of aquatic species. The sparing and retention effects of fatty acids have already been demonstrated in various aquatic species (Xu *et al.*, 1994; Deering *et al.*, 1997; Khan *et al.*, 2018). Similarly, the n-3/n-6 ratio decreased from 1.5-0.5 in *P. indicus* (Ouraji *et al.*, 2009) and 0.69-0.52 in *P. vannamei* (Panini *et al.*, 2017b) and was 0.74 in tilapia (Sanchez-Muros *et al.*, 2016) due to the inclusion of insect meal. Though insect meal reduced this ratio, it is still within the optimal concentration (0.25-1) described according to the human nutritional perspective (Simopoulos, 2002). The major limitation of using insect meals in aquafeed is that they lack in PUFA, in particular C20:5 and C22:6. It is therefore suggested to switch over to a finisher diet (no fishmeal substitution) for a certain period prior to harvesting to restore these fatty acids (C20:5 and C22:6) as they are important in human nutrition. Khan *et al.* (2018) suggested a period of 30 days in both laboratory and yard experimental conditions for penaeid shrimps, whereas, a period of 16-20 weeks was reported by Bell *et al.* (2004) in salmon fish for feeding the finisher diet.

#### *Immunomodulatory effects*

In aquatic species, it is necessary to understand the mechanisms behind the inclusion of insect meals on their immunomodulatory and physiological effects, because the major immune responses are mainly involved in disease defence mechanisms. Similarly, the haematological parameters give an idea about the health status and physiological responses of the fish. Mousavi *et al.* (2020) suggested that the inclusion of insect meals can provide a high-quality protein and has an immunological benefit in farmed animals to a certain degree. Foysal *et al.* (2019) found an improved profile of gut microbiota in crayfish (*Cherax cainii*) when reared on a diet containing *H. illucens* meal and is in agreement with Askarian *et al.* (2012), who observed an alteration in the microbiome composition of the gut, having a positive effect on a primary defence system of Atlantic salmon. Foysal *et al.* (2019) also observed an up-regulation of various cytokine genes such as IL (interleukins)-1 $\beta$ , IL-8, IL-10, IL-17F and TNF (tumor necrosis factor)- $\alpha$  in crayfish fed *H. illucens* meal. These genes are mainly associated with the immunity and inflammation of aquatic species. This is in agreement with the findings of Su *et al.* (2017) while including *T. molitor* meal in the diet of yellow catfish resulting in an up-regulation for MHC (major histocompatibility complex) II, IL-1, CypA (cyclophilin), Ig (immunoglobulin) M and HE (hepcidin) genes. The authors suggested that these expressions could be attributed to the presence of chitin. Chitin generally increase immunity in aquatic species by activating innate immune cells and inducing cytokines

and chemokine production through a different cell surface receptor (Lee *et al.*, 2017). Cuesta *et al.* (2008) reported that the major cellular innate immune activities were increased in gilthead seabream due to prior incubation with chitin particles. This result corroborates with the findings of Esteban *et al.* (2001), who found an enhanced immunity in gilthead seabream with increase in the dietary content of chitin (0, 25, 50 and 100 mg kg<sup>-1</sup>). Similarly, the immune system and disease resistance increased in mrigal fish (*Cirrhina mrigala*) reared on a diet enriched with 1% chitin (Mari *et al.*, 2014). In contrast, Stenberg *et al.* (2019) observed down-regulation of TLR (toll-like receptor)-22 and C/EBP (ccat-enhancer binding protein)- $\beta$  genes in Atlantic salmon fed *H. illucens* meal due to the presence of chitin. Notwithstanding, antimicrobial peptides present in the insect meals would also be a reason for increasing the immune response of the aquatic species. Moon *et al.* (1994) observed inhibition of Gram positive bacteria growth due to the injection of *E. coli* and antibacterial protein (tenecin-1) obtained from the haemolymph of *T. molitor*. Similarly, the expression of immune genes, including  $\beta$ -1,3 BGBP ( $\beta$ -glucan binding protein), proPO (prophenol oxidase) and crustin was reported by Choi *et al.* (2018) in *P. vannamei* fed on *T. molitor* meal, which could be attributed to increase in the circulatory haemocytes induced by the dietary inclusion of insect meal. Activated haemocytes play numerous roles in enhancing the innate immunity of the aquatic species. Wang *et al.* (2018) documented that protein, including antimicrobial proteins and crustin, mainly involved in defence mechanism is enhanced by the activated haemocytes. Activated haemocytes also eliminate microbes and other foreign particles by engaging in phagocytosis process (Choi *et al.*, 2018).

Stenberg *et al.* (2019) reported almost eighteen different immune stimulation genes involved in inflammatory response and other metabolic processes in Atlantic salmon fed with diets containing *H. illucens* meal (Table 4). These genes are more closely related to the immune-inflammatory effects, which suggest that the immunity and disease resistance of aquatic species is greatly enhanced with the intake of insect meals. Similarly, the immune system is stimulated by dietary administration of *H. illucens* meal by increasing the secretion of lysozyme, which resulted in enhanced resistance of rohu (*Labeo rohita*) against infectious pathogens (Misra *et al.*, 2006). Nurin and Maftuch (2018) reported that the inclusion of 8% *H. illucens* meal activated the immune-competency in common carp challenged with *Aeromonas hydrophila* by increasing the total blood count from  $1.63 \times 10^5$  to  $1.95 \times 10^5$  cells ml<sup>-1</sup>. Similarly, the total count of WBC increased to  $3.8 \times 10^4$  mm<sup>3</sup> in African catfish when fed higher level of *M. domesticus* meal (30-45%) (Okore,

2016). The oxidative stress biomarkers such as superoxide dismutase (SOD), catalase (CAT) and lysozyme (LZM) were upregulated due to the inclusion of *H. illucens* meal as fishmeal substitute. Lin and Shiau (2007) stated that LZM present in insect meal of *T. molitor* acts with defence peptides synergistically in inhibiting pathogenic activity in grouper (*Epinephelus malabaricus*). This result is corroborated with the findings of Zhao *et al.* (2011) and Cao *et al.* (2012), who also found an improvement in non-specific immune indices such as SOD, CAT, acid phosphatase (ACP) and alkaline phosphatase (AKP) in *P. vannamei* reared on *T. molitor* and *M. domesticus* meals. However, the immunity rate varied from species to species based on the rate of inclusion of insect meal. It was at <20% in rainbow trout (Elia *et al.*, 2018), 25% in yellow catfish (*P. fulvidraco*) (Xiao *et al.*, 2018), 75% in jian carp (Li *et al.*, 2017) and at 4.2% in *P. vannamei* (Liu *et al.*, 2013). Similarly, the protective effect against the oxidative process in rainbow trout fed on a diet with *H. illucens* meal was primarily by production of ethoxyresorufin-o-deethylase (EROD), glutathione s-transferase (GST) and glutathione (Elia *et al.*, 2018). In contrast, a higher level of inclusion of *H. illucens* meal inhibited SOD and nitric oxide (NO) formation, both of which are responsible for high oxidative stress (Li *et al.*, 2017). Though these differences might be due to the type of insects used to prepare the meals, further studies are warranted to clarify the controversial roles and their metabolic pathway in both fish and crustacean models.

The dietary administration of *T. molitor* meal increased SOD and NO, in addition to increasing malondialdehyde (MDA) and the activities of ceruloplasmin, myeloperoxidase and glutathione peroxidase in yellow catfish (Su *et al.*, 2017) and European sea-bass (Henry *et al.*, 2018). Sankian *et al.* (2018) opined that these indices play a crucial role in eliminating excessive superoxide radicals to maintain homeostasis in fish. The expression of HSP (heat shock protein)-70 and 27 are enhanced in Atlantic salmon fed a meal of *H. illucens* (Stenberg *et al.*, 2019). It is an important index and is mainly involved in translating proteins, transporting cellular and organellar proteins and maintaining protein homeostasis at the cellular level. However, the information related to HSP in aquatic species is scanty as of date. Overall, partial substitution of fishmeal using insect meals could promote the health status of several aquatic species. However, complete substitution resulted in detrimental effects in some of the aquatic species (Wang and Shelomi, 2017).

#### Challenges encountered with insect meals

Insect meals generally contain high levels of lipids; hence, the occurrence of rancidity (lipid oxidation) would be more leading to reduced shelflife of the product. It

Table 4. Relative expressions of various immune related genes in aquatic species reared on diets containing insect meals at varied levels of inclusion by substituting fishmeal

Source of insect meal	Inclusion level tested	Species tested	Reported genes	Primer used (5'-3')	Relative expression	References
<i>H. illucens</i>	0, 20, 40, 60	<i>A. percula</i>	GR	CGGTCACCTGCTACGTCTTCA	2.8-3.1	Vargas-Abundez <i>et al.</i> (2019)
			HSP70	ACGGAGAGTCGATTTTCGATG	1.4-2.0	
<i>H. illucens</i>	-	<i>S. salar</i>	IL-1 $\beta$	GTATCCCATCACCCCATCAC	0.01-0.5	Stenberg <i>et al.</i> (2019)
			IL-10	GGCTTCCCTGTTGGACGAAG	0.01-0.6	
			IN-8	GAGCGGTCAGGAGATTTGTC	0.001-0.5	
			TNF- $\alpha$	GCGGAGCATACCACTCCTCT	0.005-0.4	
			TLR22	AAAGGATGAGGACCCGATG	0.08-0.2	
			TLR3	GTTTCATGGTCAATTACAGTAGG	0.1-0.3	
			C/EBP $\beta$	CGCGTGGAGCAGCTGTCAAGA	0.1-0.4	
			P38MAPK	GGCACACAGACGATGAGATG	0.1-0.5	
			Cd36	GGATGAACTCCCTGCATGTGA	0.001-0.3	
			LOX5	ACTAAGTTTGTGCTTCGG	0.1-0.4	
			COX2	GGAGGCCTACTCCAACCTATT	0.01-0.4	
			PTGES	TCCAGCCAATGTCTTAGT	0.3-0.6	
			PTGDS	ATCCCAGGCCGCTTAC	0.05-0.4	
			GPx1	TCTCCTGCCATAACGCTTGA	0.3-0.7	
			Mn-SOD	CCACGTCCATGCCTTTGG	0.2-0.6	
			Cu/Zn-SOD	GAAGCTGACGGGAGAGATCG	0.3-0.7	
			HSP70	CCCCTGTCCCTGGGTATTG	0.3-0.7	
			HSP27	GCACATGGGCCCTCTGACTAT	0.1-0.4	
			<i>H. illucens</i>	0, 31.5	<i>C. cainii</i>	
IL-8	-	1.2-1.4				
IL-10	-	1.6-1.7				
IL-17F	-	2.4-2.7				
TNF- $\alpha$	-	1.2-1.5				
<i>T. molitor</i>	0, 5, 10, 20	<i>P. vannamei</i>	$\beta$ -1, 3 BGBP	-	1.8-6.0	Choi <i>et al.</i> (2018)
			proPO	-	2.0-14.0	
			Crustin	-	0.9-20.0	
<i>T. molitor</i>	0, 9, 18, 27	<i>P. fulvidraco</i>	MHC II	CTGAAGCAGGCGCTAAACAC	1.4-2.5	Su <i>et al.</i> (2017)
			IL-1	CTTGAGAAACGGACCCGGTG	1.0-1.8	
			CypA	TCTGTCCATGGCTAACGCAG	0.9-1.8	
			IgM	TTGGACTCGTGATGCCAAGG	0.8-1.3	
			HE	GCCGTCAGCGTCATCATCG	0.9-1.3	

GR-Glucocorticoid receptor; HSP-Heat-shock proteins; IL-Interleukins; TNF-Tumour necrosis factor; TLR-Toll-like receptor; C/EBP-ccat-enhancer binding protein; MAPK-Mitogen activated phosphokinase; Cd-Fatty acid translocase protein; LOX-Lipoxygenase; COX-Cyclooxygenase; PTGES-Prostaglandin E synthase; PTGDS-Prostaglandin D synthase; GP-Glutathione per-oxidase; SOD-Superoxide dismutase; BGBP-  $\beta$ -glucan binding protein; proPO-Prophenol oxidase; MHC-Major histocompatibility complex; Cyp-Cyclophilin; IgM-Immunoglobulin M; HE-Hepcidin.

is, therefore, suggested to include certain additives of antioxidants during the processing of insect meals. Freccia *et al.* (2020) reported that developing pre-processing and manufacturing procedures to extract the excess lipid from the insect meals, which would be more helpful to overcome the above issue and this extracted lipid can be used as a lipid source in feed or by any other industry already using animal byproducts. An effort made by Davis

*et al.* (2017) already described a method to reduce the lipid content by both commercial oil press and hexane extraction processing in *H. illucens*, which has initial fat content of 30%. Their findings revealed that the oil press could reduce about 10% of lipid and the remaining portion was left in the meal itself, whereas the hexane extraction method reduced the fat content to 5-8%, which additionally had a positive outcome in the protein content of the meal.

Though insects contain <20% carbohydrates, the major quantity is occupied by chitin and it is mainly embedded in the exoskeleton of insects. It ranged from 2.7 to 49.8 mg kg<sup>-1</sup> on a wet/fresh weight basis and is almost equal to a range of 11.6-137.2 mg kg<sup>-1</sup> of dry weight (Toviho and Barsony, 2020). Several advantages of chitin have already been reported in terrestrial animals and its effect on finfish in terms of antimicrobial activity, antioxidant activity, growth-promoting and immune-stimulation effects as detailed by Abdel-Ghany and Salem (2020). In contrary, Maghalhaes *et al.* (2017) documented that chitin reduces the digestibility of both lipids and proteins as it prevents the digestive enzymatic activities on these substrates and make them inaccessible to the fed animals. Therefore, its removal prior to dietary administration may improve the digestibility of insect meals in shrimp. Besides nutritional benefits associated with insect-based products, food safety concerns with regard to human health such as biosafety, bioaccumulation and long-term effect of consuming products produced with insect meals needs to be addressed, because they may carry exogenous and endogenous risk factors to human health. Insects generally have the ability to accumulate potential hazards like contaminants, pathogens, heavy metals, allergens and pesticides (Van der Spiegei *et al.*, 2016). Belluco *et al.* (2013) found thiaminase in *T. molitor*, which has an anti-nutrient property during consumption and the authors also observed a high bacterial count in *G. sigillatus*. Similarly, certain anti-nutrients (hydrocyanide, oxalate, phytate, saponin and tannin) and heavy metals (cadmium, lead, mercury and arsenic) have been reported in insect-based products (Nishimune *et al.*, 2000; Adeduntan, 2005; Zhang *et al.*, 2009; Banjo *et al.*, 2013; Musundire *et al.*, 2016). It is reported that about 1-3.2% of adult and 0.1-5.7% of children have food allergy in Europe (Nwaru *et al.*, 2014) and about 19% of people have sensitised skin allergy in Belgium (Francis *et al.*, 2019) with products from *A. domesticus* and *T. molitor*. Similarly, due to consumption of insect-based products, fatal reaction is reported to be about 18 (Ji *et al.*, 2008) and 7.6% (Barenes *et al.*, 2015) in consumers from China and Laos, respectively. Allergic reactions due to insect-based products have also been reported in other parts of the world, in particular India and Africa (Chakravorty *et al.*, 2011; Kung *et al.*, 2011).

EFSA (2015) reported that prevalence and the level of contaminants in insect meals and other insect-based products are influenced by several factors, primarily species, production method, stage of harvest and substrate used. However, the feed/substrate used in the rearing process is the principal route for exposing food safety hazards from insect-based products to humans. FAO (2013) observed mycotoxins, particularly aflatoxins in the edible part of insects reared on a feed/substrate

contaminated with *Fusarium*, *Aspergillus* and *Penicillium* species. Kachapulula *et al.* (2018) observed aflatoxicoses development in insects and its level exceeds the regulatory limit of 10 µg kg<sup>-1</sup>, which has acute and chronic effects on both human and animal immune status. It is therefore important to have a concern on the above-mentioned factors, especially the selection of quality hazard-free feed/substrate to improve the food safety aspects of insect-based products (Van der Fels-Klerx, 2018). However, it is practically possible only in controlled production conditions, but not with wild harvesting. In this context, certain technologies, especially microbial fermentation and enzyme treatments can be utilised to obtain diverse insect-based products with unique quality/properties in recent times (Castro-Lopez *et al.*, 2020). Islam and Yang (2017) reported that fermented *T. molitor* using *Lactobacillus plantarum* and *Saccharomyces cerevisiae* improved the growth performance and increased the immune responses against *E. coli* and *Salmonella* spp. in broiler chicks. Similarly, several ways have been suggested to control the effect of hazards present in the insects. For example, selective farming can reduce the chemical hazards and microbial and parasitic contaminations are controlled by an effective cooking process (Belluco *et al.*, 2013). The authors also suggested labelling the allergic hazards on the package to avoid the usage by the respective susceptible concern. Likewise, certain regulatory challenges need to be addressed worldwide to enhance utilisation of insect meals. The most important difficulties are in constructing rearing infrastructure, breeding sufficient quality parent stock and achieving consistent standard of the products. In addition, the legislation for the new revolutionary products will be quite challenging to the industries rearing insects and producing insect-based products. Similarly, the science behind using insect meals and other insect-based products as a feed source in aquatic species should be developed further, which could help in the growth of the industry worldwide in future.

## Conclusion

Insect meals provide high amount of nutrients, especially protein and also have considerable quantity of micronutrient like minerals and vitamins, with great economic and environmental advantages. Similarly, certain components present in insect meals make them functional ingredients, by which they enhance the immune status of fed animals. In addition, the anti-microbial properties of insect meals offer a natural alternative to synthetic drugs and other antibiotics used in the aquaculture sector. These positive scenarios make insect meals a potential protein source in the diets of various aquatic species. They can complement about 20-75% of dietary fishmeal and in certain cases, 100% fishmeal substitution was

reported with proper supplementation of amino acids. This helps to reduce the formulation cost in the face of the volatile commodity market of protein ingredients and the limited supply of fishmeal. Most of the insect meals showed a superior effect compared to plant proteins and certain animal proteins, and performed equally to high-quality fishmeal. However, a higher dietary inclusion of certain insect meals causes immune suppression and reduction in phagocytic activity in the cultured animals. These controversial reports warrant further field trials, especially genetic-based studies and *in vivo* assessments for better comprehension of their roles and the effects on physiological and immunological parameters of aquatic species along with their effectiveness in disease suppression and management. Likewise, challenges present in insect meals in terms of nutritional aspects (high lipid, chitin, hazards) can effectively be controlled by modern technologies invented in recent times. Though insects and insect-based products are categorised under GRAS (generally recognised as safe) list in various countries and communities, there is still hesitation among the consumers in terms of their biosafety. Similarly, there are legal issues related to legislation, which need to be addressed by the respective policymakers based on scientific evidence to bring this budding industry to the next level in the future.

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