Insect Protein as a Partial Replacement for Fishmeal in the Diets of Juvenile Fish and Crustaceans

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16.1. INTRODUCTION

16.1.1. The Need for Alternatives to Fishmeal

The availability of land to produce livestock is rapidly dwindling as the human population continues to increase throughout many regions of the world. Similarly, the availability of wild aquatic organisms (fish and crustaceans) is declining due to overharvesting and increased demand by humans (Sargent and Tacon, 1999). Fish farming (cultured fish) could potentially satisfy the increasing demands for animal protein (Naylor et al., 2001). However, rising prices for feed for aquatic organisms destined for human consumption is a reason for concern. One of the main sources of animal protein in commercial feed for juvenile fish and crustaceans is fishmeal, which is dried, ground tissues of undecomposed marine baitfish (herring, anchovy, or menhaden). Fishmeal is becoming less available (Tacon and Metian, 2008; Ng, 2000; Abowei and Ekubo, 2011; van Huis, 2013). The safety of fishmeal has also been under scrutiny. Alternative sources of protein for fish are necessary to decrease dependence on fishmeal. However, the aquaculture industry may need economic incentives to encourage the transition from fishmeal-based feedstuffs to alternative sources of food for fish and crustaceans (Naylor et al., 2009). Researchers are currently investigating the potential for protein from plants as partial or even complete replacements for fishmeal (Le Boucher et al., 2011, 2012); similar research with protein from yeast is ongoing (Peterson et al., 2012). In recent years, the value of insect protein as partial or complete replacements for fishmeal has been studied (van Huis, 2013).

16.1.2. Aims of this Chapter

Although there is some published information on the usefulness of insects as sources of protein in feed for terrestrial livestock (poultry and swine), the purpose of this chapter is to highlight research on insects as sources of protein for juvenile fish and crustaceans (prawn). This chapter attempts to review and synthesize the evidence that protein from insects can partially replace animal protein in fishmeal in feed for juvenile stages of cultured fish. This research is in support of the aquaculture industry and its efforts to provide plentiful fish and prawns for human consumption and remain competitive in a global economy.

16.1.3. Overview of the Content

This chapter presents a review of the scientific literature on insects as important sources of protein to incorporate into fish and prawn diets. In this chapter, the coverage is restricted to insects within three orders—Lepidoptera, Diptera, and Coleoptera. In the Lepidoptera, the focus is on the Oriental silkworm moth Bombyx mori (L.) (Family Bombycidae) and other silkworm moth species (Family Saturniidae). In the Diptera, the focus is on the common house fly Musca domestica (L.) (family Muscidae) and the black soldier fly Hermetia illucens (L.) (family Stratiomyidae). In the Coleoptera, the focus is on the yellow mealworm Tenebrio molitor (L.) and the superworm Zophobas morio (F.) (family Tenebrionidae). This chapter also mentions some challenges and opportunities that could hinder the expansion of the market for insect feed for commercial production of livestock. Topics will include the challenges of creating artificial diets to facilitate rearing insects at a commercial scale and opportunities (or lack thereof) to scale up production of insects to meet the needs of the aquaculture industry. The production of insects of high quality is an important topic to those that rear them professionally (Schneider, 2009).

16.2. MODEL INSECTS AND POTENTIAL AS FEED FOR FISH

16.2.1. Oriental Silkworm Moth and Relatives

The Chinese have reared silkworm moths for centuries for their silk (Zhang et al., 2008). Even today, a sericulture industry for production of silk for human clothing is ongoing in China, India, and elsewhere. The Oriental (mulberry) silkworm moth *B. mori* is used for this trade. Other silkworm moths including the silkworm *Anaphe infracta* Walsingham, muga silkworm *Antheraea assamensis* Helfer, eri silkworm *Samia ricini* (Donovan), and Chinese oak silkworm *Antheraea pernyi* Guérin-Méneville are also reared for their silk. Traditionally, Oriental silkworm larvae are reared on leaves of mulberry *Morus alba* (L.) and close relatives, which have origins in Asia. Due to human transport, the mulberry tree inhabits many countries around the globe in association with silkworm and the sericulture industry. Interestingly, humans consume mulberry

leaves as a vegetable and tea. We eat the fruit (berries) fresh, in preserves, or drink the juice. Only the cocoon of the silkworm moth pupal stage is necessary in silk production. Most growers extract the pupae from cocoons and discard them as waste. Utilization of "waste" silkworm pupae to feed livestock is therefore a good way of capitalizing on the unused product of silk production. This practice of using waste silkworm pupae to support the production of fish is an effective use of resources.

The idea of using silkworm pupae as food for fish is not novel. Akiyama et al. (1984) supplemented a fishmeal-based diet for chum salmon *Oncorhynchus keta* (Walbaum) with 5% powdered silkworm pupae, dried beef liver, krill meal, or powdered earthworm. Over a 6-week feeding experiment, weight gain, feeding efficiency, and fat content of salmon were greatest when the authors incorporated powdered earthworm rather than any other treatments or control (untreated fishmeal) into the fishmeal-based diet. The protein content of salmon did not differ between treatments in this study (Akiyama et al., 1984). Whether the addition of larger percentages of protein from silkworm pupae would increase its value in the diet of chum salmon is not clear.

Nandeesha et al. (1990) measured the growth of common carp *Cyprinus carpio* (L.) over 140 days on diets containing variable percentages of nondefatted silkworm pupae (41% crude protein) and fishmeal (68% crude protein) in dry pellets, using the methods of Jayaram and Shetty (1981). They used the following treatments: 10% silkworm plus 20% fishmeal, 20% silkworm plus 10% fishmeal, 30% silkworm plus 0% fishmeal, and 0% silkworm plus 25% fishmeal. All diets contained approximately 20%, 40%, and 10% groundnut cake, rice bran, and tapioca flour, respectively. The diet with 30% silkworm plus 0% fishmeal stimulated the most growth of juvenile carp over the test period. The authors concluded that nondefatted silkworm pupae could replace fishmeal in diet formulations as feed for common carp, especially because the texture, flavor, odor, and color of this fish does not change with a change from the standard fishmeal diet (Nandeesha et al., 1990).

In a companion study, Nandeesha et al. (2000) increased the percentage of silkworm pupae in experimental diets. In dry pellets, they formulated diets containing 30%, 40%, or 50% silkworm pupae without any fishmeal; the control diet consisted of fishmeal without any silkworm. All diets contained variable amounts of groundnut cake, rice bran, and tapioca flour to balance the protein content between treatment diets. After a test period of 90 days, the authors discovered no changes in weight gain, food conversion ratio, or protein conversion ratio when they fed carp juveniles any percentage of silkworm-based diet or the control (fishmeal). However, carp retained increasing amounts of protein in tissues with an increase (30–50%) in the percentage of silkworm in the diets. Carp fed the 50% silkworm diet contained more protein (but less fat) than those fed lower percentages of silkworm. Apparently, carp diets can contain up to 50% silkworm pupae without changing the growth and quality of the product (Nandeesha et al., 2000). Adding oil from silkworm pupae or sardines

improved the growth, feed efficiency, and biomass production of common carp (Nandeesha et al., 1999). In outdoor tanks, 9% addition of either oil increased biomass production up to almost 50% over the control (fishmeal without added oil). They did not detect differences between fish oil versus silkworm oil at 3%, 6%, or 9% concentrations in dry feed pellets. The authors concluded their study by stating that adding more fat into diets can incur economic benefits to carp production.

Silkworm (*B. mori*) pupae were a supplement rather than a complete replacement of fishmeal in fish diets (Jintasataporn et al., 2011). Silkworm pupae could replace up to 50% of the protein from fishmeal (equivalent to 14.6% silkworm pupae by weight) in formulated diets for the snakeskin gourami *Trichogaster pectoralis* (Regan), an omnivorous fish. Greater amounts of silkworm pupae (22–30% by weight) caused a decrease in growth and egg production of females, seen after 5 months of feeding. Farmers in Southeast Asia culture this species (and others, such as common carp) in temporary bodies of water in rice fields for human consumption and for export to the aquarium industry (Little et al., 1996).

Commercial production of shrimp (i.e. prawns) relies on fishmeal as feed. A search for alternatives is ongoing. Langer et al. (2011) compared the effects of diets containing silkworm pupae (41%), fishmeal (49%), soybean meal (42%), or earthworm meal (46%) on freshwater prawn Macrobrachium dayanum (Henderson) for 90 days. All diets contained variable amounts of rice bran and mustard oil cake to balance the proximate composition of protein in formulated feeds. The researchers found that the diet containing earthworm meal (rather than any other diet) proved most beneficial for prawn growth, survival, and food conversion ratio. The diet containing silkworm pupae was second best, followed closely by fishmeal. The diet with soybean meal was the least nutritious for freshwater prawn in this study. Biochemical analysis showed that protein content did not differ between diets based on silkworm pupae, fishmeal, or earthworm meal; protein content was greater in all three of these diets when compared to the diet based on soybean. Lipid content was approximately the same for diets containing silkworm pupae versus earthworm meal; lipid content was lowest in the fishmeal diet (Langer et al., 2011).

16.2.2. House Fly and Black Soldier Fly

The common house fly *M. domestica* has a long history of interactions with humans. It is a nuisance pest and has the capacity to vector diseases, several of which are highly virulent to humans (Nazni et al., 2005; Fasanella et al., 2010). Nevertheless, researchers have proven that house fly maggots are suitable sources of protein for poultry (Ocio et al., 1979; Hwango et al., 2009). Maggots are also good sources of protein for catfish. Fasakin et al. (2003) evaluated methods of processing maggots—hydrolyzed (defatted), not removing fat (full-fatted), and dried in an oven or dried in the sun—on growth and utilization of African catfish *Clarias gariepinus* (Burchell). The authors compared the

diets containing processed maggots with diets containing fishmeal (control). Over a 56 day timeframe, catfish fingerlings were fed the treatments diets. The drying method (oven versus sun-dried) did not affect the suitability of defatted maggots for fish; defatted maggots were not less nutritious than fishmeal. The sun-dried, full-fatted maggots were less nutritious; fish exhibited less growth and weight gain when fed this treatment rather than fishmeal. Fasakin et al. (2003) suggested that defatted maggots provide animal protein comparable in quality to that of fishmeal.

Aniebo et al. (2009) also assessed the potential of house fly maggots, in the form of a meal later compacted into sun-dried pellets, on growth and nutrient utilization of the African catfish *C. gariepinus*. They monitored catfish juveniles fed feed composed of 0%, 50%, or 100% animal protein from maggot meal rather than fishmeal (control) over a 10 week period. Maggot meal contained 48% crude protein. Soybean meal was a major constituent (34–43%) in the three treatment feeds. The authors discovered that feed composed of 50% or 100% animal protein from maggots had no negative effects on fish growth or nutrient utilization when compared to feed composed of fishmeal. Maggot meal (in compacted pellets) is an alternative protein source that should reduce costs and lead to sustainable production of catfish (Aniebo et al., 2009).

In another study with catfish, Sogbesan et al. (2006) compared two techniques to harvest house fly maggots and evaluated the potential of these maggots, formulated into a maggot meal, as dietary protein for catfish. They compared a screen method in which they thinly spread maggot-infested dung on a screen net in the sunlight. In attempts to avoid sunlight, the maggots crawled out of the dung and down into a basin positioned beneath the screen net. The other harvesting technique, the flotation method, involved placing maggot-infested dung in a basin of water; the maggots floated to the surface of the water. The authors used a 3 mm mesh size net to sieve the maggots from the water. The catfish was a hybrid between Heterobranchus longifilis and C. gariepinus. They evaluated the effects of five maggot meal-based diets designed to replace 0%, 25%, 50%, 75%, and 100% of the fishmeal typically present in fish feed. Catfish fingerlings in their experiment were in concrete tanks for 70 days. They found that harvest method (screen vs flotation) had no effect on the quantity of maggots collected within 4h. Catfish fed the diet that replaced 25% (rather than 0%, 50%, 75%, or 100%) of fishmeal with a maggot meal displayed the greatest total final body weight, mean weight gain, feed conversion ratio, and protein efficiency rate. Sogbesan et al. (2006) recommend replacing 25% of fishmeal for maggot meal for better growth performance of hybrid catfish.

Nsofor et al. (2008) compared the growth and food utilization of the catfish *C. gariepinus* when fed diets (pellet form) containing 25% fishmeal, 25% maggot meal, or 12.5% fishmeal plus 12.5% maggot meal for 10 weeks. They discovered that all diets were effective. Fish did not differ significantly in final body weight, specific growth rate, food conversion ratio, or protein efficiency ratio. The authors state that 25% maggot meal could replace the 25% fishmeal in diets

for catfish. Omoyinmi and Olaoye (2012) measured the growth performance of Nile tilapia *Oreochromis niloticus* (L.) fed feed containing protein from house fly *M. domestica*, earthworm *Eudrilus eugeniae* (Kinberg), palm grub *Oryctes rhinoceros* (L.), garden snail *Limocolaria aurora* (Jay), or fishmeal. They subjected tilapia fingerlings to the diets (as crushed pellets) for 70 days. The results revealed that diets with protein from earthworm, garden snail, or house fly were as effective as the fishmeal diet on tilapia growth and survival. Diet with protein from palm grub was less effective than fishmeal (control) for tilapia growth and survival (Omoyinmi and Olaoye, 2012). Earthworm and maggot protein appear to be very suitable alternatives to fishmeal for tilapia growth and development.

St-Hilaire et al. (2007a) evaluated the importance of house fly pupae and black soldier fly H. illucens prepupae in feed for rainbow trout Oncorhynchus mykiss (Walbaum). Treatment diets (feed) involved substituting 0%, 25%, or 50% fly protein for fishmeal (anchovy) and feeding this to juvenile trout for up to 9 weeks. All diets contained soybean meal (16%) and corn gluten meal (8%). Total weight gain of fish did not differ for those fed 25% soldier fly diet and the control (fishmeal). Total weight gain was less for fish fed 50% soldier fly or 25% house fly diet rather than the control. The feed conversion ratio (grams of diet fed to fish/grams of weight gained by fish) was the highest for fish fed the 50% soldier fly diet compared to the other treatments. The protein content (whole body proximate composition) of fish after the end of the experiment did not differ significantly between treatments. The lipid content of fish fed diets containing soldier fly (25% or 50% pupae) did not significantly differ from the control (fishmeal); lipid content in fish fed 25% house fly diet was significantly greater than in fish fed 50% soldier fly diet. Protein from house fly or black soldier fly can replace up to 25% of the fishmeal in fish feed without negatively affecting growth performance and quality of rainbow trout.

Sealey et al. (2011) were concerned that supplementing fish diets with insect protein from the black soldier fly could alter the aroma, taste, and quality of fillets of rainbow trout. In a previous study, adding protein from soldier fly pupae into fish feed altered the fatty acid profile of rainbow trout (St-Hilaire et al., 2007a). Changes to the fatty acid concentration can affect fish flavor and aroma (Turchini et al., 2003). Using black soldier fly prepupae reared on manure from dairy cows, Sealey et al. (2011) replaced either 25% or 50% of fishmeal (anchovy) protein with soldier fly protein or with soldier fly protein enriched with fish offal (discarded visceral organs and fat from rainbow trout). Note that all four diets contained considerable amounts of soybean meal (16%), wheat gluten meal (7.8%), and corn gluten meal (7.0%). They fed rainbow trout the treatment diets (pellet form) for 8 weeks and measured fish growth, fillet taste, and aroma. Replacing fishmeal with either 25% or 50% insect meal from soldier fly protein reduced the growth rate of fish. However, the combination of soldier fly protein with fish offal did not affect growth of fish. The fish that were fed protein from black soldier fly-with or without the fish offal-did not differ in taste or aroma from fish fed protein from fishmeal (Sealey et al., 2011). Bondari and Sheppard (1987) compared diets formulated with either 10% fishmeal, 10% dried prepupae of black soldier fly, or a commercial feed (with an unknown percentage of fishmeal as the animal protein) as dry pellets for the channel catfish *Ictalurus punctatus* (Rafinesque). When they reared catfish juveniles in cages floating on the surface of the water in a 2 ha reservoir, the growth rate (weight gain, total body weight) of males and females was reduced over a 15-week study period when using the 10% soldier fly diet rather than the 10% fishmeal diet or commercial diet. The crude animal protein content was 24%, 30%, and 37.5% in the soldier fly diet, fishmeal diet, and commercial diet, respectively. The authors concluded that black soldier fly could not replace fishmeal as the sole source of animal protein in fish feed.

Kroeckel et al. (2012) examined insect meal derived from prepupae of the black soldier fly as a substitute for fishmeal (herring) for juvenile turbot *Psetta* maxima (L.), a carnivorous flatfish in marine waters. The prepupae were defatted before combining with other components in the feed. Wheat starch and fish oil were key components. They formulated six diets with 0%, 17%, 33%, 49%, 64%, or 76% insect meal. The diets were in pellet form. The authors discovered that growth performance of turbot decreased, generally, as the percentage of insect meal in test diets increased. Fish fed the control diet (no insect meal) showed higher specific growth rate and final body weight than those fed any of the diets containing insect meal. The feed conversion ratio (food intake/weight gain) was approximately the same for diets with 0%, 17%, and 33% insect meal. Greater percentages of insect meal (49%, 64%, and 76%) gave a significantly higher food conversion ratio. The authors concluded that juvenile turbot could have some difficulty digesting chitin (from the soldier fly); they did not detect chitinase activity in fish intestines. They concluded that insect meal derived from black soldier fly can replace only a limited percentage of fishmeal in diets for turbot. Despite these results, the ease with which this fly can be reared on agricultural wastes—effectively converting these wastes into body protein and fat—warrants further study on using it as a protein and fat replacement in food for fish or other livestock (Sheppard et al., 1994; St-Hilaire et al., 2007b; Diener et al., 2011; Kroeckel et al., 2012).

16.2.3. Yellow Mealworm and Superworm

The yellow mealworm is common in pet stores and other places that sell and market exotic organisms (birds, reptiles, amphibians, and fish) and their nutritional supplies. Yellow mealworms can provide essential amino acids and fats in the diet of humans that consume them (Ghally and Alkoaik, 2009; Oonincx and de Boer, 2012). Unfortunately, published research on the nutritional value and suitability of the mealworm as feed to replace or supplement fishmeal in the diets of fish cultured for human consumption is scant. One study compared the potential of mealworm in the form of a dry powder formulation (mealworm meal) as a partial replacement for fishmeal (Ng, 2000). The authors replaced

0%, 20%, 40%, 60%, 80%, or 100% of the fishmeal with mealworm meal in diets for the African catfish *C. gariepinus*. They were able to replace up to 80% of the fishmeal in the catfish diet without negatively affecting growth rate or feed utilization efficiency (Ng, 2000).

The superworm is also a common insect in pet stores. Similarly, published research on the usefulness and nutritional value of this insect as feed for fish that are destined for human consumption is difficult to find. One study formulated diets (dry pellets) using protein from superworm to replace from 0% to 100% of the protein from fishmeal as feed for the Nile tilapia O. niloticus (Jabir et al., 2012a). At the conclusion of an 8-week feeding trial, they found that fish fed the diets in which protein from superworm replaced 25% or 50% of the fishmeal experienced the greatest weight gain, specific growth rates, feed conversion ratio, and protein efficiency ratio. Higher percentages of superworm in the fish diets resulted in a decrease in the parameters relating to growth. Note that soybean meal and rice bran were major components in all of the diet formulations (ranging from 22% to 31%); the total crude protein in each formulation was nearly the same (ranging from 31% to 34%) in this study (Jabir et al., 2012a). In a companion study, Jabir et al. (2012b) compared the nutrient profiles of diets (dry pellets) containing protein from fishmeal or superworm meal as feed for the red tilapia Oreochromis spp. Superworm meal had lower protein but higher fat content than fishmeal. Seventeen amino acids (including the eight essential amino acids) were found in the superworm and fishmeal diets. Only two amino acids, arginine and tyrosine, were found in higher levels in the superworm meal than in the fishmeal. Based on apparent digestibility coefficients of protein and lipids, the authors discovered that fish had more difficulty digesting the superworm meal than the fishmeal. Slight improvements to the superworm diets are necessary before they can totally replace fishmeal as feed for red tilapia (Jabir et al., 2012b).

16.3. CHALLENGES AND OPPORTUNITIES TO EXPANSION OF MARKET FOR INSECTS AS FEED

16.3.1. Artificial Diets

The desire and necessity to rear insects on artificial diets rather than their natural food to reduce costs has been one of the most important topics in the insect-rearing community (Vanderzant, 1974; Singh, 1977; Cohen, 2004). Researchers are working to develop an artificial diet for silkworms (*B. mori* and relatives) to expedite its rearing. *Bombyx mori* depends on mulberry leaves for proper growth, development, and reproduction. Researchers in China and India have developed crude artificial diets of mulberry leaf powder as food for silkworm moth larvae in attempts to reduce costs of maintaining live plants (Cappellozza et al., 2005). Efforts to increase the suitability of the artificial diet (as compared to fresh mulberry leaves) for silkworm are ongoing. The lack of ascorbic acid (vitamin C) in the powdered diet could limit its suitability for optimum

growth and development of silkworm moth larvae in lieu of fresh mulberry (Cappellozza et al., 2005). Supplementing natural diets (fresh mulberry) with ascorbic acid improves larval growth, development, and cocoon weight in *B. mori* (Singh and Bandey, 2012). The eri silkworm *S. ricini*, which is indigenous to northeast India, performs well on foliage from castor or tapioca (Longvah et al., 2011). There is no crude artificial diet for the eri silkworm.

There is limited need to develop an artificial diet for the common house fly *M. domestica* because researchers have reared it for decades on natural substances such as garbage, waste products, and animal feces under field conditions. Ocio et al. (1979) grew *M. domestica* larvae in residues of municipal organic waste. Aniebo et al. (2008) reared larvae on a mixture of cattle blood and wheat bran. For the sake of laboratory rearing, investigators have formulated several crude artificial diets (Spiller, 1963). For example, Hogsette (1992) showed that *M. domestica* reared well on a mixture of alfalfa meal, wheat bran, and corn meal.

Tomberlin et al. (2002) tested the suitability of three diets as food for black soldier fly larvae in the laboratory. The first diet was a mixture of alfalfa meal, wheat bran, and corn meal, whereas the second contained alfalfa meal, wheat bran, and brewer's dried grain. The third diet contained undisclosed proprietary components. The protein contents in these diets were 15%, 19%, and 15%, respectively. There were no significant differences between diets in terms of their effects on larval development or survivorship. All three diets were suitable for rearing the soldier fly; more than 96% of larvae survived to become prepupae, and up to 27% emerged as adults of slightly female-biased sex ratio (55-60% females). Unfortunately, adult soldier flies were smaller in size when reared on the three diets rather than under natural field conditions in swine and poultry manure (Tomberlin et al., 2002). Black soldier flies mated well inside screen cages $(2 \times 2 \times 4 \text{ m})$ in a small greenhouse $(7 \times 9 \times 5 \text{ m})$, accessible to natural sunlight (Sheppard et al., 2002). Mating even occurred during the winter months in Georgia (the southeastern United States) when cloud cover did not block penetration of sunlight. Adults required water but not food.

Davis (1975) reared yellow mealworm on a mixture of amino acids in proportions occurring in larval tissues as the only source of protein. He commented that his diet was inferior to one based simply on wheat plus brewer's yeast. Zinc and potassium might be essential nutrients to incorporate into an artificial diet (based on casein protein) for normal development of *T. molitor* over multiple generations (Fraenkel, 1958). Culturists have reared yellow mealworms and the superworm on brewer's yeast, rolled oats, white potato, wheat germ, and other stored products for decades. Lipke and Fraenkel (1955) found that corn germ inhibits growth of *T. molitor*. Because of the low cost of many stored products, despite the fact that humans also consume these items, there has been limited progress on developing and refining artificial diets for mealworms and super worms.

Effective and economical artificial diets might facilitate the scale up of rearing operations to provide enough insect protein to support commercial aquaculture. Assessing the quality or health of insects reared on artificial diets against those reared on natural food is essential to any successful mass rearing operation (Cohen, 2004).

16.3.2. Scale-Up of Production

The major concern at this point is increasing the cost-effectiveness of rearing systems for the insect species described previously, considering all components (artificial diets, enclosures, environmental conditions). Designing cages that maximize the production per unit of space, automation of rearing, and use of machinery to replace human labor could reduce some costs associated with rearing on a large scale (Nordlund, 1998; Smith and Nordlund, 1999). Only then can one begin to consider increasing their production on a scale that will meet the needs of the commercial industry. Researchers and industry representatives have begun to engineer rearing factories with specific environmental conditions (light, temperature, humidity) to support large-scale rearing of insects indoors. Published information on the size, design, and engineering of factories that produce the insects discussed in this chapter is limited.

A commercial producer of mealworms (van de Ven Insectenkwekerij) operates a facility in the Netherlands (see Oonincx and de Boer, 2012). At this facility, the producer grows the yellow mealworm and superworm in large quantities and markets them live or freeze-dried to wholesalers, zoos, and import and export companies (http://www.insectenkwekerij.nl/). A commercial producer of the common house fly in Shan Dong Province, China produces 10 and 30 metric tons of dried and live maggots, respectively, per month in a 1000 m² plant (http://shengyang2006.en.busytrade.com). They market their products to the medical and pharmaceutical industries and pet food manufacturers around the globe.

A number of companies in Asia produce and market silkworms, particularly the Oriental silkworm moth *B. mori*. Bratac (http://www.bratac.com.br/bratac/pt/index.php), a company in Parana, Brazil, has rapidly expanded its production of silk. The requisite of feeding on mulberry foliage could hamper the scale-up of production of *B. mori* in some countries where mulberry is less abundant. A potential market for the eri silkworm *S. ricini* is growing in northeast India. This species feeds well on castor and tapioca plants rather than mulberry. Longvah et al. (2011) stated that silk production in India reached approximately 18,475 metric tons from 2006 to 2007. Pupae, which are a waste byproduct of sericulture, represent approximately 60% of the weight of cocoons. These waste pupae could generate 4000 and 2000 metric tons of protein and oil, respectively, each year (Longvah et al., 2011). Therefore, collection and utilization of silkworm pupae could provide added economic benefits to farmers and the sericulture industry if silkworm protein can replace a significant percentage of fishmeal in feed for fish and other livestock.

Researchers have produced the black soldier fly outdoors in sufficient quantities to suggest that scale-up of production to meet the needs of the aquaculture

industry is a possibility. A side benefit of this system of rearing is the management of manure (Diener et al., 2011). The soldier fly ingests some wastes and thereby converts it into proteins and fats that are suitable for consumption by fish and other livestock (Sheppard et al., 1994; St-Hilaire et al., 2007b). The soldier fly can also convert organic waste into biodiesel fuel (Li et al., 2011). No special equipment is necessary to contain the soldier fly; larvae essentially develop in waste in the near vicinity of the livestock. This system of rearing the soldier fly is crude, but it could be economically and environmentally sustainable to the farmer with limited resources.

The capacity to manage fish wastes using the house fly and soldier fly should help facilitate the scale-up potential of rearing systems of both species (see Sheppard et al., 1994; Diener et al., 2011). On the other hand, we rarely consider techniques for managing insect wastes (feces) in connection with scale up of insect production. Elaborate techniques may or may not be necessary in this regard, depending on the habitat of the insect. Clearly, more research on this topic is necessary. Feces from terrestrial species (mealworms) could be sieved away from developing larvae and their food medium. For insects living within aquatic systems or very humid terrestrial systems, special filtration systems may be necessary to separate insect waste from fish waste. Feces can be air-dried, disinfected to eliminate pathogens (e.g. bacteria), then used as a biodegradable fertilizer for plants in terrestrial systems. One company in southern California sells insect feces to gardeners for this purpose (see http://www.pchydro.com/news_article/article_id/50).

16.4. QUALITY CONTROL AND PRODUCTION

16.4.1. Maintenance of Long-Term Insect Colonies

Techniques that could prevent the decline or eventual deterioration of the colony are necessary in rearing all species highlighted in this review. Methods used to prevent decline in colonies may include manipulative breeding between hybrids to restore and maintain normal growth and reproduction of populations in culture. Mating between siblings should be discouraged (Mackauer, 1976; Roush and Hopper, 1995). Introducing wild (field) adults into well-established colonies to mate with domesticated adults could prevent inbreeding depression and any subsequent reduction in fitness. (Careful screening of wild adults is necessary to prevent inadvertent spread of pathogens and parasitoids into the colonies.) Periodic monitoring of colonies for apparent undesirable changes in behavior of adults (compared to wild adults) is necessary (Boller, 1972; Huettel, 1976).

16.4.2. Colony Hygiene and Preventing Disease Transmission

As mentioned briefly in the introduction of this chapter, the production of high-quality insects is an important topic to those that rear them professionally (Schneider, 2009). Ensuring that insects are produced using the highest

standards of care and nutrition (food sources) is profitable to everyone involved. Rearing professionals must maintain healthy, disease-free colonies in laboratories or rearing facilities. Most insects held in confinement at the moderate to high densities necessary for mass production can potentially harbor pathogens. Consequently, culturists must routinely check the health of their colonies and maintain hygienic rearing conditions. In this chapter, the common house fly—more than the other species—is probably most often revered as a vector of pathogens that can harm humans (Banjo et al., 2005; Fasanella et al., 2010). Culturists must use constant vigilance in ensuring that adult house flies do not escape from rearing facilities. The inadvertent transmission of disease is not a concern for the black soldier fly; they do not vector human pathogens. In fact, the black soldier fly can limit populations of the house fly through competitive exclusion under natural conditions in the field (Sheppard, 1983; Bradley and Sheppard, 1984). The black soldier fly can also help reduce infectious bacterial populations in chicken and cow manure (Erickson et al., 2004; Liu et al., 2008).

The other species highlighted in this chapter—yellow mealworm, superworm, and several species of silkworm—are not known to vector human pathogens. However, they all have their own complement of parasites and pathogens (Barnes and Siva-Jothy, 2000; Valtonen et al., 2010; Guo-Ping and Xi-Jie, 2011; Isaiarasu et al., 2011), and several could reduce the health of a colony if left unchecked. The effects of feeding diseased or contaminated insects—whether in a natural form or formulated into an insect meal or pellet—to fish is unknown. We know very little about the interactions of insect pathogens and fish health, so this subject requires research. The likelihood that fish would become contaminated after feeding on diseased/unhealthy insects and therefore become unfit for human consumption is very remote, but it cannot be ruled out entirely (see Noonin et al., 2010).

16.5. CONCLUSIONS AND RECOMMENDATIONS

16.5.1. Synthesis

The reliance on fishmeal arose, in part, from a need to supply mass quantities of animal protein in the diet to meet the nutritional requirements of fish in culture. Today, rising costs, unreliable availability, and questionable quality (safety) of fishmeal has encouraged many feed production companies to search for alternative sources of animal protein to incorporate into fish feeds. In this review, I have attempted to demonstrate that all model insects have merits as potential sources of protein to incorporate into feed for fish destined for human consumption. The model species examined in this review have positive and negative aspects that could affect their adoption as sources of insect protein in feed for fish and crustaceans on a large scale. The differences in protein and fat content between these species are important considerations. It varies between species and between stages of development within a species. Most research shows that

the yellow mealworm and the superworm are at the high end of the scale for these nutrients; the Oriental silkworm is at the low end (see Finke, 2002). However, other factors such as ease of rearing, availability of artificial diets to scale-up rearing, potential of vectoring infectious diseases, and societal approval are just as important. The black soldier fly is a prime candidate for large-scale production. It is easily reared in the same outdoor facilities alongside fish and has an added benefit of recycling animal wastes. Although it is not known to vector human pathogens, societal acceptance of this fly—or any fly species, for that matter—is low, particularly in the more affluent societies around the globe. Thus, we will need to re-educate the public on the value of insects and their potential role in aquaculture in the near future.

Most studies show that insect protein can be formulated into a dry powdered meal or into pellets. Researchers mix plant protein with insect protein, along with various lipids, into the meal or pellets. The combination of the insect and plant-based nutrients into the same feed does not appear to affect its acceptance by fish, at least not for omnivorous species such as catfish and carp. These fish accept the insect meal or pellets as readily as live or whole insects. This suggests that the procedures used to process the insect-based feeds do not destroy essential nutrients that fish require for growth and reproduction.

The innate feeding preferences of fish and prawn may determine the suitability of insects as protein in feed. Research shows that omnivorous species (e.g. carp and catfish) that typically feed on insects found on the bottom of lakes, ponds, and other temporary or permanent bodies of water survive on feed containing a considerable percentage of insect protein. Therefore, omnivorous fish (especially freshwater species) are less dependent on fishmeal. For example, fishmeal represents a small percentage (less than 5%) of the total composition of commercial feed for channel catfish farmed in the United States (Li et al., 2008; Robinson and Li, 2012). In 2007, fishmeal only represented 5% of the total composition of feed for Chinese carp (Tacon and Metian, 2008; Naylor et al., 2009). Highly carnivorous species (e.g. trout and salmon) also eat insects, particularly in the juvenile stages, but research shows their growth and feeding efficiency declines when their feed contains a considerable amount of insect protein. Highly carnivorous fish and marine fish generally have a high requirement (400-500 g/kg dietary dry matter) for protein rich in essential amino acids (Sargent and Tacon, 1999). These fish can easily fulfill this requirement with the consumption of fishmeal.

Several crude artificial diets are available to rear some insects that show promise as sources of protein to support the aquaculture industry. However, more research is necessary to refine these diets. A systems approach to rearing of target insects is necessary; this would encompass artificial diets, efficient design of enclosures, and automation (mechanization) of essential operations to decrease the costs associated with human labor. Establishment of insect factories that can produce the massive quantities of insects needed to support the increasing demands of the aquaculture industry is also necessary.

16.5.2. Future Research

- 1. More research in the area of feeding preferences of fish that show some affinity for insect protein (in comparison to species that do not) is necessary. We need to determine if some fish can adapt after several generations of exposure to diets that contain more protein derived from insect meal rather than fishmeal. There is reason to believe that a carnivorous species could adapt to an insect-based diet. Note that carnivorous fish can potentially adapt to a plant-based diet after a few generations of artificial selection (see Le Boucher et al., 2011, 2012).
- 2. The potential of producing insects to meet the increasing needs of the fish industry is unclear. Knowledge on this subject has rarely progressed beyond experimental stages in small plots or enclosures using juvenile fish. More research is necessary to prove that insects can be produced effectively and efficiently to support the nutrition of juvenile and adult fish to meet the demands of this burgeoning market.
- 3. Expansion of the efforts to promote insect protein to niche markets, such as producers/sellers of exotic fish and other pets, is encouraged. The pet industry may be a near-term (rather than long-term) option to using insect protein rather than fishmeal in artificial diets designed for tropical and temperate fish sold as pets. Pet stores sell insects (e.g. mealworms) as live food for birds, reptiles, amphibians, and fish (Barker et al., 1998; Finke, 2002). Formulation of insect protein into a meal or dry pellet to reduce costs and increase shelf life (compared to live food) would be advantageous. This idea would be problematic, however, for species that only accept live prey (e.g. many amphibians in the adult stage).
- **4.** Development of cost-effective artificial diets to rear insects on a large scale, accompanied by design of machinery to automate rearing, will be a prerequisite to expanding the market for insect protein. Automation could potentially reduce labor costs.
- 5. Disclosure of information on the size, design, and engineering of rearing facilities (factories, warehouses) is necessary. Presently, very few companies that produce and market insects as feed for vertebrates furnish details on the design of their rearing facilities in the published literature. Financial incentives might be necessary to encourage companies to disclose and share information.
- 6. More collaboration between the feed industry, government and academic researchers, and local farmers is necessary to identify novel sources of insect protein and design rearing systems that are efficient, cost-effective, and sustainable. For example, Neptune Industries (http://www.neptuneindustries.net) was collaborating with Mississippi State University to patent Ento-Protein, a product formulated with protein from laboratory-reared insects (http://www.dafvm.msstate.edu/landmarks/07/fall/8-9.pdf). The company intended to market Ento-Protein as a replacement for fishmeal.

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