

Review

Development of enriched *Artemia* and *Moina* in larviculture of fish and crustaceans: a review

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ABSTRACT. Inconsistencies in the nutritional values of live food such as *Artemia* and *Moina* are well-known issues. The enrichment of live food is necessary to obtain the optimum nutrients needed for the growth, survival, and immune competence of fish and crustaceans' larvae. The enhanced growth and survival of fish and crustaceans' larvae are vital to continuous aquaculture production. However, enriched live food could be species-specific as various aquatic larval species may respond differently to the enrichment diets. The enrichment of *Artemia* and *Moina* as the "bags of nutrients" has been widely studied and involved various enrichment diets such as commercial diets containing essential fatty acids, highly unsaturated fatty acids, and vitamin C. The use of natural enrichment diets such as yeast, microalgae, and herbal extract, including the common name Chinese chaste tree leaf (*Vitex negundo*), is becoming popular in aquaculture nutritional development. These natural enrichment diets are more economical and environmentally friendly than commercial diets. The compositions of *Artemia* and *Moina* are both affected by the enrichment diets that they consumed, hence directly affecting the growth of the larvae that fed on them. Hence, this review highlights the development of enriched *Artemia* and *Moina* and their effects on the growth performance and the immune competence of fish and crustaceans' larvae.

Keywords: *Artemia*; *Moina*; enrichment; growth; immune; larvae; aquaculture

Aquaculture is among the most sustainable animal protein production systems (Brummett 2013). It plays an important role in providing food security and reducing poverty and malnutrition worldwide (Heck & Béné 2005, Ogello & Munguti 2016, Golden et al. 2017). Therefore, providing sustainable aquaculture practices and productions is important as the demand for protein sources from fish increases with the rapid world population growth. According to Hixson (2014), sustainability of the aquaculture industry includes three main subjects: social, economic, and environmental concerns, whereby environmental sustainability is particularly related to fish nutrition. The focus on fish nutrition can be seen through decades of continuous efforts to develop aquafeed formulations with reasonable cost and expense to enhance the growth of the

cultured fish (Turchini et al. 2019). These formulations fulfill each species' nutrient requirements as aquaculture's diverse management and environmental conditions require different feeding approaches (NRC 1993).

Nutrient requirements are described as the requirement for maximal growth and survival, where the association between fish-diet-feeding has a strong effect in determining the quantitative needs (Izquierdo & Lall 2004). In contrast, the requirement for body maintenance includes the minimum rate of nutrient consumption required to keep the animal's viability and the requirement for fish health, and the requirement for least production cost (Hamre et al. 2013). Interestingly, these definitions cover the aspects of the maximal growth, survival, and health of the fish produced under

economic costs. One of the relevant topics studied over the years is live food usage in larval culture. Live food persists as a necessity in hatcheries for various aquaculture species and is expected to be continuously needed in the future (Dhont et al. 2013). Besides, live food organisms are also preferable to artificial feeds during various fish and crustacean species' larval and early postlarval stages (Das et al. 2007). Although the formulation of microparticulate diets is developing, no artificial larval diet could completely satisfy the requirements of a considerable number of fish and crustacean larval species (Samad et al. 2020). Fish larvae reared on microparticulate diets commonly have slower growth and lower survival rates, possibly caused by diet's low nutritional value, poorly developed digestive systems, or low ingestion rates in the larvae (Holt 2011).

Unlike microparticulate diets, live foods such as zooplanktons possess high digestive enzymes, stimulating larval appetite (Zheng et al. 2018). It is recognized as nutritious living capsules. They possess natural fundamental nutrients and energy sources such as lipids, proteins, vitamins, fatty acids, amino acids, minerals, and carbohydrates required in aquaculture to grow and maintain most cultured species (Mona et al. 2017). Some of the generally used live feeds in aquaculture are rotifers (*Brachionus* sp.), *Artemia* nauplii, *Moina*, copepods and blood worms (Herath & Atapaththu 2013). However, the production of live feed is known to have a possible risk of disease transmission. It needs extra human resources and infrastructure, thus causing an impediment to the hatchery operations and costly (Person 1989, Faulk & Holt 2009).

Furthermore, the inconsistencies of the nutritional contents in zooplankton as live food are also being debated across different works of literature, and this shortfall could be improved with a feeding routine known as enrichment (Carter 2015). Due to the underdeveloped production protocols of *Moina* in the hatcheries (Samad et al. 2020) and the variations of cysts quality and nutrition compositions in *Artemia* (De Clercq et al. 2005, Vangansbeke et al. 2016), their enrichment protocols and techniques using various enrichment media such as vitamin C (ascorbyl-6), highly unsaturated fatty acids (HUFA) and bio-flocculated microalgae will be further discussed in this review. On the other hand, rotifers and copepods are more generally known and widely discussed in various literature (Ohs et al. 2010, Das et al. 2012, Samad et al. 2020). Therefore, this review will highlight *Artemia* and *Moina*, focusing on their enrichments and effects on growth performance and immune competence of larval fish and crustaceans.

Artemia

The mass culture of *Artemia* is considered a straightforward process. It has the advantages of having enzymatic fortification, which facilitates digestion in developing digestive tracts and has chemical attraction and feeding stimulation through movement (Cahu & Zambonino 2001). Generally, *Artemia* nauplii are often given as prey as soon as it hatches. Their dormant cysts have a long shelf-life, easy to be hatched, and their newly hatched nauplii show potential as larval feed (Carter 2015) (Fig. 1). Despite the fish nutrition industry advancements, *Artemia* nauplii remain as fundamental live food in rearing early stages of crustaceans, fish, and mollusks (Soltanian 2007, Ohs et al. 2010, Le et al. 2018). This organism possesses naturally high α -linolenic acid (ALA) and linoleic acid (LNA) concentrations, and has relatively low arachidonic acid (ARA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (Araújo & Rosa 2016). The components of essential fatty acids, PUFA and HUFA, cannot be synthesized by fish de novo, although some species can convert 18-carbon PUFA to longer HUFA chains (Sargent et al. 1989, Akbary et al. 2011). More than 50 geographical strains are reported (Treece 2000), with seven bisexual species and numerous parthenogenetic populations identified (Asem et al. 2010). Each strain of cyst has a distinct quality, and it might differ significantly between various locations and sources (De Clercq et al. 2005, Vangansbeke et al. 2016). Such variations in quality are due to geographical isolation and peculiar habitat conditions, which lead to variations in phenotypes and diverse physiological, biological, and chemical attributes (Vanhaecke & Sorgeloos 1980).



Figure 1. The image of *Artemia* sp. viewed under a microscope (10x microscope magnification). Photo courtesy to Wizilla Janti Joshua (February 16, 2021).

Previously, *Artemia* was classified as freshwater and marine types micro-crustacean. The freshwater type has a high LNA concentration but a low omega 3 HUFA level (suitable for freshwater fish). In comparison, the marine type has a high content of EPA and omega 3 HUFA (suitable for marine fish and crustaceans) (Watanabe et al. 1980). Discrepancies in the quality and the contents of *Artemia* are still subject to discussions in the scientific research community. Several enrichment techniques boost the content of *Artemia* nauplii before feeding it to fish larvae or crustacean species. To date, nutritional enrichment is a well-established technique in aquaculture and defined as a process for enhancing the nutritional quality of live foods (Kandathil et al. 2019). The ability of *Artemia* to filter enrichment materials makes it a universal live food that is widely accepted in hatcheries of various larval species. Its digestive tract can be loaded in enrichment media with any particles of accepted size (Sorgeloos et al. 2001), making it a living capsule that transports enrichment products which can be manipulated in terms of nutritional requirements for the target organisms and availability of enrichment materials (Palma et al. 2011).

Moina

Moina is a cladoceran Crustacea and is generally known as water fleas. It is small freshwater zooplankton. *Moina* as live food is commonly and extensively used in commercial hatcheries and fry ornamental fish production (Rasdi et al. 2020a). It has the advantages of having high tolerance to temperature, high reproduction rates, and can forage on phytoplankton and organic wastes (Das et al. 2012). In addition, it is preferred by most fish larvae as its jerking, and whimsical movement makes it a noticeable prey (Mayer & Wahl 1997, Gogoi et al. 2016). In addition, *Moina* possesses digestive enzymes, including proteinases, lipases, amylases, and peptidases which play roles as exoenzymes in the gut of fish larvae (Miah et al. 2013). There are reports on the utilization of *Moina* as the starter food for larvae of milkfish (*Chanos chanos*) and bighead catfish (*Clarias macrocephalus*) (Yilmaz et al. 2006), as well as its effects in increasing the enzyme activity of trypsin, chymotrypsin, and α -amylase in green catfish (*Mystus nemurus*) larvae (Srichanun et al. 2012). It also has been used in rearing the larvae of sutchi catfish (*Pangasius sutchi*) (Potaros & Sitasit 1976), koi carp (*Cyprinus carpio*) (Tay 1973), and bighead catfish (Carreon et al. 1976, Mollah 1983). An adult *Moina* is 700-1000 μm in size, which is longer than a newly hatched brine shrimp (500 μm) (Rottmann et al. 1992) (Fig. 2). The fatty acid content of *Moina* does not fulfill

the requirements for crustacean and fish larval feeds (He et al. 2001). Its nutritional value also is insufficient to boost the survival and the growth of predator larval fish, particularly in HUFAs, which consist of EPA and DHA (Singh et al. 2019).

Most species of *Moina* feed on bacteria, other organisms, algae, and pelagic cladocerans are filter feeders that feed on organic particles from water columns (Manklinniam et al. 2018). Hence, its filter-feeding behavior is utilized in delivering certain nutrients to fish larvae. A report on *Moina mongolica* stated that its EPA content (20:5 ω 3) contains 12.7% of total fatty acids, higher than *Artemia* (2.1%; He et al. 2001). However, most essential amino acids are lower than *Artemia* (Tong et al. 1988). Hence, its filter-feeding behavior is utilized in delivering nutrients to the fish larvae. Apart from that, *Moina* also has been used as a bio carrier of antibacterial drugs and bioencapsulators of probiotics, which boost immune response and system in fish larvae (Wiwattanapatapee et al. 2002, Lashkarboluki et al. 2011). The application of enriched *Moina* will be discussed according to its effects on fish larvae and crustaceans' growth performance and immune competence.

Modification of nutritional compositions through enrichment

Artemia

There are plenty of products or substances that have been utilized for the enrichment of live foods, including emulsions (Leger et al. 1987, Kontara et al. 1991, Clawson & Lovell 1992), microalgae (Watanabe et al. 1978, 1980, 1982, 1983, Olsen et al. 1997), yeasts enriched with different oils (Gatesoupe 1991), bacteria (Gorospe et al. 1996, Gomez-Gil et al. 1998) and liposomes (Hontoria et al. 1993, 1994, Ozkizilcik & Chu 1994, McEvoy et al. 1996). It is important to identify live food's ability to absorb and uphold nutrients from the enrichment product before feeding it to the specific fish larvae species.

Studies on the enrichment of *Artemia* using different components showed some variations in the fatty acids and biochemical contents. For example, *Artemia* enriched for 12 h during the incubation and another 12 h before the harvesting using cod liver oil mixed with ascorbic acid and raw egg yolks (HUFA + vitamin C) showed a higher and consistent amount of individual fatty acids levels except oleic acid (18: n-9) and palmitic acid (16:0), as compared to the newly hatched *Artemia* (Akbari et al. 2011). It proved that the enrichment using different products significantly modified the enriched *Artemia*'s composition (Novelli et al. 2016). A separate report on *Artemia* enriched with commercial products for 24 h with different lipid/protein

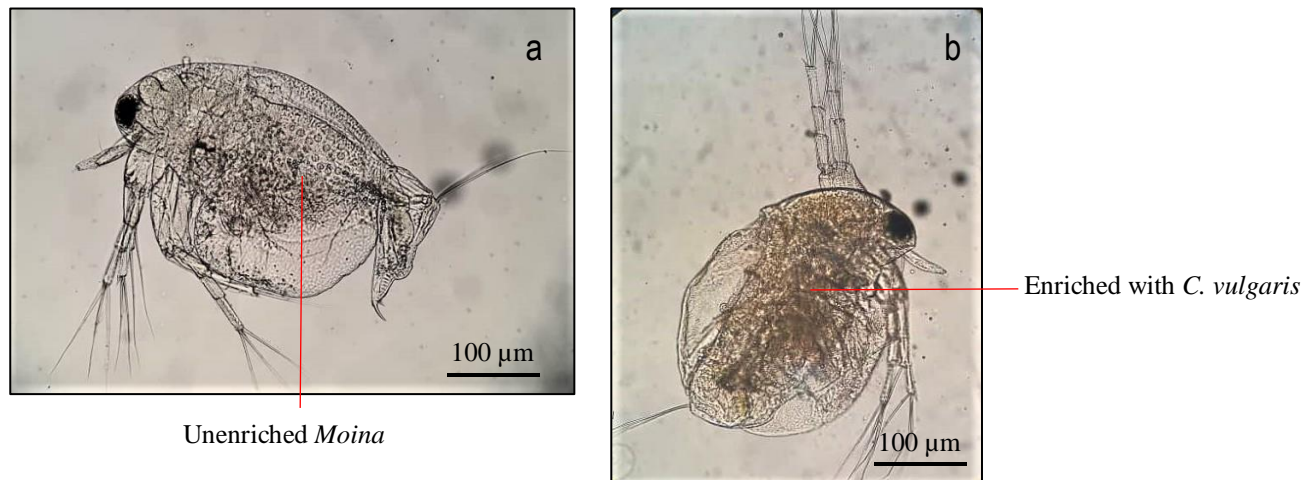


Figure 2. The image of a) unenriched and b) enriched *Moina* sp. It was enriched with *Chlorella vulgaris* viewed under a microscope (10x microscope magnification). Photo courtesy to Wizilla Janti Joshua (May 7, 2021).

contents: DHA Protein Selco (27/29%), Biomar Multigain (13/44%), and Easy Selco DHA (0/67%) showed that *Artemia* enriched with Easy Selco DHA and Biomar Multigain had the highest and the lowest lipid contents of 21.7 and 13.6%, respectively. The study also reported the decreased protein content for all treatments compared to the freshly hatched *Artemia* (Novelli et al. 2016). Apart from DHA Protein Selco, other commercial enrichment diets commonly used to enrich *Artemia* are ALG and A1 Selco (Sorgeloos et al. 1991, Biswas et al. 2006, Figueiredo et al. 2009, Chakraborty et al. 2010). The drawback in these commercial enrichment diets is the high PUFA content which produces detrimental trans fatty acids when exposed to air, light, and high temperature. It could cause larval mortalities (McEvoy et al. 1995, Woollard & Indyk 2003, Chakraborty et al. 2007). The fatty acid composition of many marine microalgae showed more stable PUFA than many commercial enrichment diets (Volkman et al. 1989).

Moina

Compared to *Artemia*, *Moina* is an exceptional choice high in nutrient and protein contents (Loh et al. 2012). *Moina* lacks essential n-3 HUFA (e.g. EPA and DHA), which are needed for the growth and the survival of fish larvae (Singh et al. 2019). Due to this nutritional deficiency, many studies attempted to enrich *Moina* with various enrichment diets, and the effects on fish larvae and crustaceans were studied. Based on the literature, the use of enriched *Moina* is limited to freshwater as it could not survive within a few minutes in seawater (Fushimi & Hashimoto 1969, Nakamoto et al. 2008). Similarly, nutrient deficiency in *Moina* could be solved by manipulating its nutritional values by

taking advantage of its primitive feeding characteristics (Scott & Middleton 1979, Das et al. 2007).

Effects of enriched live food

Growth performance

Artemia

HUFA is the most explored nutrient in *Artemia* because it is vital to the success of fish and crustacean larval rearing (Dhont et al. 2013). *Artemia* nauplii may lack some fundamental n-3 and n-6 PUFA (Daniels et al. 1992, De Barros & Valenti 2003), especially EPA (20:5n-3), DHA (22:6n-3), and arachidonic acid (20:4n-6) (Thin et al. 1999, Chakraborty et al. 2007, Ali et al. 2017). Hence, numerous studies have been done to improve these components in *Artemia* and their effects on fish and crustaceans' larvae. A study by Kamaszewski et al. (2014) reported that the larvae of Russian sturgeon (*Acipenser gueldenstaedtii*) fed with essential fatty acids (EFA) enriched *Artemia* showed an enhanced growth performance in terms of higher body weight and length as well as fatty acid contents. Similar results were achieved in other species of sturgeons, Persian sturgeon (*Acipenser persicus*) (Hafezieh et al. 2009) and the beluga (*Huso huso*) (Jalali et al. 2008). Based on several works of literature, some parts of physiological functions such as immunity, cell adhesion, brain and eye developments, reproduction, ion balance regulation, muscle contraction, vascular tone, and buoyance control are affected by EFA. These functions directly influence the survival and the growth of marine animals (Glencross 2009, Pond & Tarling 2011, Gurr et al. 2016). The EFA are made up primarily from n-3, n-6 and three long-chain PUFA called docosahexaenoic acid (DHA, 22:6n-3), eicosapen-

taenoic acid (EPA, 20:5n-3), and arachidonic acid (ARA, 20:4n-6) (Mejri et al. 2021).

The enrichments of *Artemia* were also tested using several natural ingredients. The herbal extract from the Chinese chaste tree leaf (*Vitex negundo*) was used to enrich *Artemia*. The study by Arulvasu et al. (2012) reported that 6 h enrichment with 2.5 mg mL⁻¹ of the herbal extract L⁻¹ gave the maximum specific growth rate and survival rate (95.83%) to common molly (*Poecilia sphenops*) fry as compared to unenriched *Artemia* (89.58%). The use of natural plant extracts in aquatic animals has an increasing demand for sustainable and eco-friendly aquaculture (Lewis & Ausubel 2006). However, the toxicity levels of herbal extract should be evaluated beforehand. *V. negundo* leaf extract with the concentration of 2.5 mg mL⁻¹ has been recommended to enrich *Artemia* before feeding it to the fish fry. In prawn culture, river prawn *Macrobrachium americanum* larvae fed with *Artemia* enriched with *Chaetoceros calcitrans* showed the most enhanced growth and survival, followed by the larvae group fed with *Artemia* enriched with *Tetraselmis suecica*, thus suggesting that it might be due to the variations in the nutritional quality of the ingested nauplii (Méndez-Martínez et al. 2018). All treatments showed the abundance of LNA and LOA (PUFA group) and EPA (HUFA group), with the highest amounts of fatty acids detected in *C. calcitrans*-enriched *Artemia* nauplii. A total of 90-95% microalgae composition represented in dry weight comprises lipids, proteins, carbohydrates, and minerals (Voltolina & López-Elías 2002), possibly with 60% protein (Lora-Vilchis et al. 2004, Kotrbacek et al. 2015). In addition, microalgae at the exponential phase of culture growth could supply up to 30-40% protein, 10-20% lipid, and 5-15% carbohydrate content if offered as enrichment diets to *Artemia* (Brown & Blackburn 2013). This nutrient composition represents an energy source with a high benefit to cost ratio (Becker 2013). The combination of probiotics and microalgae studied by Kandathil et al. (2020) revealed that *Artemia* enriched with bio-flocculated algae (*Chlorella vulgaris* flocculated with *Lactobacillus acidophilus* and *Bacillus subtilis*) affected the seven-day-old freshwater fish catla (*Gibelion catla*) fry group. The effect was indicated by the clear accumulation of probiotics inside the digestive tract and the significant increases in lipid and ash contents compared with the fries group fed with unenriched *Artemia*. Regarding the accumulation of the probiotics, the growth performance of the fry group fed with enriched *Artemia* also showed better growth performance. This result indicated that probiotics flocculated in the algae used in enriching *Artemia*, altered the composition of the intestinal microflora,

promoted nutrient digestibility, boosted absorption quality, and increased enzyme activity in fish (Lara-Flores et al. 2003, Balcazar et al. 2006).

Moina

Singh et al. (2019) reported that the larval group of climbing perch (*Anabas testudineus*) fed with HUFA and vitamin C-enriched *Moina* showed the highest weight gain and significantly higher mean specific growth rate than the unenriched *Moina*. Fish cannot synthesize vitamin C (ascorbic acid) (Chatterjee 1973, Dabrowski 1990). The biosynthesis of collagen and steroid hormones is often associated with vitamin C. Vitamin C enhances larval immune response and resistance to environmental stressors and toxicants (Dabrowski & Blom 1994). The effects of HUFA-enriched *Moina* were also studied on the postlarvae of giant freshwater prawn *Macrobrachium rosenbergii* (Das et al. 2007). The emulsions containing sunflower oil, cod liver oil, and MaxEPA capsules were used as lipid sources. The study found that all emulsions increased the levels of EPA and DHA in *Moina* and significantly improved the growth rates, the survival rates, and the fatty acid composition of postlarvae. This result suggested that lipid levels in *Moina* can be boosted like that in *Artemia* (Watanabe et al. 1982, Leger et al. 1987, Dhert et al. 1990, Velazquez 1996) through an enrichment process. Besides, an investigation was done on black tiger shrimp (*Penaeus monodon*) larvae to study the effects of *Moina* enriched with a formula comprised of yeast, canola oil, *Nannochloropsis* sp., and *Chlorella* sp. (Rasdi et al. 2021). The highest specific growth rate and survival rate were recorded in larvae fed with yeast enriched *Moina* of 17.22 ± 0.10 and 91.78 ± 1.67%, respectively. The study also recorded the best result as compared to other enrichment formulas in terms of the biochemical composition of shrimp larvae with 64.04 ± 0.40% of protein, 4.91 ± 2.43% lipid, 16.89 ± 2.75% moisture, and 10.38 ± 2.05% ash. Yeast such as *Saccharomyces cerevisiae* has been proven to boost the immune responses, growth performance, and diseases resistance in various fish species such as in flounder (Harikrishnan et al. 2011), common carp (Gopalakannan & Arul 2010), grouper (Chiu et al. 2010) and Nile tilapia (Abdel-Tawwab et al. 2008).

Moina has also been proposed to replace *Artemia* nauplii as the live food for marine finfish (Fushimi & Hashimoto 1969, Oka et al. 1982, Fermin 1991, Fermin & Bolivar 1994). However, it was not made possible as it would die within a few minutes in seawater (Fushimi & Hashimoto 1969, Nakamoto et al. 2008). Kotani et al. (2016) recommended the solution of this issue by freezing *Moina macrocopa* cultured with freshwater

Chlorella sp. as feed and nutritionally enriching it with a commercial enrichment diet, DC DHA SELCO (INVE Technologies NV, Dendermonde, Belgium) for 24 h. The frozen *M. macrocopa* was fed to the larvae and the juvenile red sea beam (*Pagrus major*). The performance of the larvae group was compared to the larvae groups fed with DC DHA SELCO (INVE Technologies NV, Dendermonde, Belgium) enriched *Artemia* nauplii and wild zooplankton. The result showed that the enriched *Artemia* and wild zooplankton gave better growth to the larvae as they contained more DHA than EPA than the frozen enriched *M. macrocopa*, which contained more EPA than DHA (Kotani et al. 2016). Aside from the enriched *Moina* effects on fish and crustacean larvae, Rasdi et al. (2020b) demonstrated that *Chlorella* sp. enriched *Moina* fed to juvenile siamese fighting fish (*Betta splendens*) had the highest specific growth and survival rates, as well as protein (81.22%) and lipid (21.44%) contents compared to fish fed with *Moina* enriched with yeast (control), palm kernel cake and egg yolk. Generally, microalgae are promising ingredients that possess good nutritional contents and enhance fish immune response (Reyes-Becerril et al. 2013, Sarker et al. 2018). They are the most common live food for herbivorous zooplankton, including cladocerans and rotifers (Agadjihouede et al. 2014). On top of protein which is the essential amino acid that provides energy sources to zooplankton, other important nutrients including PUFA, pigment, sterols, and vitamins are also found in microalgae (Rasdi et al. 2020b).

Immune competency

The enrichment of *Artemia* using EFA and 1 g of vitamin C had promoted the highest survival rate in 120 day-old sailfin molly larvae (*Poecilia latipinna*) with up to $93.2 \pm 3.0\%$. The 10-day-old larvae showed the highest resistance, up to $91.5 \pm 4.7\%$, when subjected to a high-temperature stress test (Mousavi-Sabet et al. 2015). Many pieces of literature supported the addition of vitamin C aids in decreasing the negative stress effects in larvae and thus increased their survival (Dhert & Sorgeloos 1995, Merchie et al. 1997, Gapasin et al. 1998, Lim et al. 2002). Other than EPAs and vitamin C, Abdollahi et al. (2019) showed that *Artemia franciscana* enriched with β -carotene from the microalga *Dunaliella salina* had the highest amounts of lysozyme, alkaline phosphate activity of the mucus, and total immunoglobulin of the platyfish (*Xiphophorus maculatus*). The first-line defense against potential pathogens is regulated by immunoglobulin; meanwhile, fish skin mucus possesses various innate immune factors, including lysozymes, glycoproteins, and proteases (Shephard 1993, Magnadóttir 2006). The evaluation of enriched *Artemia* using probiotics

Bacillus subtilis and *Lactobacillus plantarum* against vibriosis (*Vibriosis anguillarum*) in European sea bass (*Dicentrarchus labrax*) larvae was carried out by Touraki et al. (2012). The study proved that the most efficient protection with the highest survival rates against vibriosis in fish larvae was offered by *Artemia* enriched with *B. subtilis*, up to 86.7% compared to the untreated larvae group (36.7%). The treatment for bacterial diseases, especially in fish larvae, usually involves antibiotics administered in *Artemia* as the carrier (Duis et al. 1995, Touraki et al. 1996, 1999) or through antibiotics bath or immersion (Samuelsen 2003). Hence, the use of probiotics is seen as a promising measure to curb pathogens in fish culture (Kesarcodi-Watson et al. 2008).

As for culture feed *Moina*, the highest survival (by percentage) of climbing perch (*Anabas testudineus*) larvae was achieved by feeding it with HUFA-enriched *Moina* (28.67 ± 1.45) as compared to the unenriched *Moina* group (12.00 ± 0.58) after 15 days of treatment (Singh et al. 2019). The study indicated that enrichment using HUFA alone is enough to enhance the immune competency of the larvae. A different study with an engaging result was obtained from sea beam larvae which showed improved survival when fed with frozen enriched DC DHA SELCO *M. macrocopa* (Kotani et al. 2016). According to Koven et al. (1990), Bessonart et al. (1999), Estévez et al. (1999), and Koven et al. (2001), EPA, ARA, and n-3 HUFA are recognized to boost the survival of finfish larvae and these components might be the determining factors that affect the survival of the red sea beam larvae. Table 1 summarizes the effects of enriched *Artemia* and *Moina* on the growth, survival, and immune competence of fish and crustaceans' larvae. The role of live feed as the carriers for various enrichment products to different larval stages of aquatic animals could be more beneficial than direct administration of the enrichment products such as probiotics, DHA, and HUFA. For instance, the direct administration of probiotics (a mixture of *Bacillus licheniformis*, *B. subtilis*, and *B. pumilus*) into the water was reportedly did not give any significant increase in immunity, survival, and growth of Eurasian perch (*Perca fluviatilis* L.) larvae, hence suggested that such administration of probiotics was not sufficient to enhance the digestive mechanisms of the larvae (Mandiki et al. 2011).

Prospects and conclusion

The enrichment of *Artemia* and *Moina* with various enrichment media can be seen to affect the growth, survival, and immune competence of fish and crustaceans' larvae. It is also reported to affect the juvenile of some fish species. This study can enhance

Table 1. Effects of enriched *Artemia* and *Moina* on growth and survival of different fish larvae and prawn species.

Live feed	Enrichment component(s)	Target species	Effects on fish larvae	References	
<i>Artemia</i>	Essential fatty acids (EFA)	Russian sturgeon larvae (<i>Acipenser gueldenstaedtii</i> L.)	Enhanced growth performance in terms of higher body weight and length and fatty acid contents	Kamaszewski et al. (2014)	
	Herbal extract from <i>Vitex negundo</i> leaf	Common molly fry (<i>Poecilia sphenops</i>)	Maximum specific growth rate and survival rate (95.83%) as compared to unenriched <i>Artemia</i> (89.58%)	Arulvasu et al. (2012)	
	Microalgae <i>Chaetoceros calcitrans</i>	Giant freshwater prawn larvae (<i>Macrobrachium americanum</i> L.)	Most enhanced growth and survival of larvae and the highest amounts of fatty acids detected in enriched <i>Artemia nauplii</i>	Méndez-Martínez et al. (2018)	
	Bio-flocculated algae <i>Chlorella vulgaris</i> flocculated with <i>Lactobacillus acidophilus</i> and <i>Bacillus subtilis</i>	Catla fry (<i>Catla catla</i>)	Clear accumulation of probiotics inside the digestive tract, significant increases in lipid and ash contents, and enhanced growth performance of fry	Kandathil et al. (2020)	
	EFA and 1 g of vitamin C	Sailfin molly larvae (<i>Poecilia latipinna</i> L.)	Promoted the highest survival rate	Mousavi-Sabet et al. (2015)	
	β -carotene from the microalga <i>Dunaliella salina</i>	Platyfish (<i>Xiphophorus maculatus</i>)	Gave the highest amounts of lysozyme, alkaline phosphate activity of the mucus, and total immunoglobulin	Abdollahi et al. (2019)	
	Probiotics <i>Bacillus subtilis</i>	European sea bass larvae (<i>Dicentrarchus labrax</i> L.)	Most efficient protection with the highest survival rates against vibriosis (<i>Vibriosis anguillarum</i>)	Touraki et al. (2012)	
	Highly unsaturated fatty acids (HUFA) and vitamin C	Climbing perch larvae (<i>Anabas testudineus</i> L.)	Highest weight gain, significantly higher mean specific growth rate, and survival	Singh et al. (2019)	
	<i>Moina</i>	HUFA (sunflower oil, cod liver oil, and MaxEPA capsules)	Postlarvae of giant freshwater prawn (<i>Macrobrachium rosenbergii</i>)	Increased the levels of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in <i>Moina</i> and significantly improved the growth rates, survival rates, and fatty acid composition of postlarvae	Das et al. (2007)
		Yeast, canola oil, <i>Nannochloropsis</i> sp. and <i>Chlorella</i> sp.	Black tiger shrimp larvae (<i>Penaeus monodon</i>)	Highest specific growth rate and survival rate were recorded in larvae fed with yeast enriched <i>Moina</i> of 17.22 ± 0.10 % and 91.78 ± 1.67 %	Rasdi et al. (2021)
	<i>Moina macrocopa</i> cultured with freshwater <i>Chlorella</i> sp. as feed + enrichment with commercial enrichment diet, DC DHA SELCO (Note: The <i>Moina macrocopa</i> was frozen after enrichment)	Larvae and the juvenile of red sea bream (<i>Pagrus major</i>)	Enriched <i>Artemia</i> and wild zooplankton gave better growth to the larvae as they contained more DHA than EPA compared to the frozen enriched <i>M. macrocopa</i> . However, improved survival was recorded in the larvae group fed with frozen enriched <i>M. macrocopa</i>	Kotani et al. (2016)	
	<i>Chlorella</i> sp.	Siamese fighting fish (<i>Betta splendens</i>) juvenile	Highest specific growth and survival rates, and also protein (81.22%) and lipid (21.44%) contents	Rasdi et al. (2020b)	

the knowledge necessary for selecting the best media for enriching live foods of cultured larval species concerning its specific impact on certain species. Natural enrichment diets such as microalgae and yeast seem to be more promising than commercial diets. It is more cost-saving and environmentally friendly, thus offering sustainable aquaculture practices. Live feed enhances the appetite of fish and crustacean larvae. The enrichment diets could boost extra beneficial effects to their growth, survival, and immune competency compared to adding them separately by considering the undeveloped digestive tract of the larvae. A proper guideline for live food enrichment could be established and accessible to farmers or hatchery operators. It can also be a future market to be developed and introduced commercially to enhance and boost the growth performance and survival of fish and crustaceans' larvae. In return, it can help fish farmers and hatchery operators estimate operating costs to produce enriched live foods that carry maximum benefits to the growth (Duis et al. 1995, Touraki et al. 1996, 1999) or through antibiotics bath or immersion (Samuelsen 2003). Hence, probiotics are a promising measure to curb pathogens in fish culture (Kesarodi-Watson et al. 2008) survival and the immune competence of larvae during the rearing period. Hence, further studies on the species-specific enriched live food with specified enrichment media and techniques are recommended.

ACKNOWLEDGMENT

This work was financially supported by the Ministry of Higher Education (MOHE) Malaysia through the Malaysia-Japan SATREPS-COSMOS (JPMJSA 1509) project.

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Received: October 18, 2021; Accepted: February 16, 2022