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# Recent developments in the application of live feeds in the freshwater ornamental fish culture

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#### Abstract

The industrial development of freshwater ornamental fish culture has been hampered by the lack of suitable live feeds for feeding the fish at the various production stages. This paper reports the recent developments in the applications of the freshwater rotifers (Brachionus calyciflorus), Artemia nauplii, decapsulated Artemia cysts and on-grown Artemia in the freshwater ornamental fish culture. Results demonstrate that the rotifers are an ideal starter feed for dwarf gourami (Colisa lalia), a typical freshwater ornamental fish species with larvae that are too small to ingest Artemia nauplii or Moina at its first feeding. Compared with the conventional yolk food, the use of rotifers as a starter feed significantly improves the growth and survival of the gourami larvae (Days 2-12), and the beneficial effects are extended to the subsequent Artemia-feeding phase (Days 13-32). The freshwater rotifers and Artemia nauplii are also useful in raising Discus larvae in the absence of their parents, which would eliminate the risk of larvae being eaten by the parent fish. Work on decapsulated Artemia cysts indicates that the cysts could be used as a substitute for Artemia nauplii or Moina in freshwater ornamental fish culture. The fry of all the five common ornamental fish species tested (guppy Poecilia reticulata, molly Poecilia sphenops, platy Xiphophorus maculatus, swordtail Xiphophorus helleri and neon tetra Hyphessobrycon herbertaxelrodi) could readily feed on the decapsulated cysts, and their performances in terms of stress resistance, growth and survival are comparable to or better than those fed on Artemia nauplii or Moina. A culture system for production of on-grown Artemia has also been developed specifically for the use in freshwater ornamental fish farms. The system, using diluted artificial seawater of 20% for culture, has a mean production rate of 3 kg/m<sup>3</sup> of water in a 12-day cycle and a production capacity of 8 metric tons of on-grown Artemia a year. With the system, farmers could produce any specific size of on-grown Artemia of up

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to 5 mm to suit the age and size of their fish, by varying the time of harvesting. This characteristic, coupled with the use of bioencapsulation technique to enhance the quality of the on-grown *Artemia*, would make the organism an ideal nursery diet for freshwater ornamental fish. All these results show that the live feeds used in marine foodfish hatchery could be applied to freshwater ornamental fish culture to improve their performance.

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#### 1. Introduction

The success in the hatchery production of fish fingerlings for stocking in the grow-out production system is largely dependent on the availability of suitable live food organisms for feeding fish larvae, fry and fingerlings. The availability of large quantities of live food organisms such as marine rotifer (Brachionus plicatilis and Brachionus rotundiformis) and Artemia nauplii to meet the different stages of fry production has contributed to the successful fry production of at least 60 marine finfish species and 18 species of crustaceans (Dhert, 1996). In contrast, the industrial development of freshwater ornamental fish culture has been hampered by the lack of suitable live feeds for feeding the fish at the various production stages. Currently, inert food items such as egg yolk suspension, milk powder or powdered feeds and natural plankton bloom induced by artificial fertilisation of water are used in larval feeding, and Moina and Tubifex that are cultured in water enriched with organic manure are fed to bigger fish or brooders. There is also no suitable live feed for feeding early fish larvae with small mouth. These traditional practices not only limit the fish stocking density, but also adversely affect fish quality. Many freshwater ornamental fish farmers have shifted from *Moina* to the cleaner *Artemia* nauplii for feeding their young fish. As the nauplii (length of instar-1 Artemia < 0.55 mm) are only half the size of Moina (length < 1.20 mm), it is necessary to look for bigger organisms, both to fill in the size gap, and as a substitute of *Tubifex* for feeding larger fish such as brooders. Furthermore, the high price of Artemia cysts has increased the fish production cost, and cheaper alternative diets with comparable nutritional quality are needed to maintain the cost competitiveness of ornamental fish in the global market.

Many of the modern larviculture technologies used in marine foodfish hatcheries could be adapted for application in the freshwater ornamental fish production. Some of the possible applications have been reported in Dhert et al. (1997). This paper reports the recent developments and discusses the applications of the freshwater rotifers (*Brachionus calyciflorus*), *Artemia* nauplii, decapsulated *Artemia* cysts and on-grown *Artemia* in the freshwater ornamental fish culture, based on the studies conducted in Singapore.

#### 2. Rotifers

The marine rotifers *B. plicatilis* is the most important live food organism for use in larviculture of marine foodfish. Their small size and slow swimming velocity make them an

ideal live food organism for fish larvae that cannot ingest the larger *Artemia* nauplii. Marine rotifers can survive in freshwater for at least 2 h, and have been used for feeding larvae of tilapia *Oreochromis spiluus* (Cruz and James, 1989), gudgeon *Gobio gobio* L. (Kestemont and Awaiss, 1989) and Japanese ornamental carp *Cyprinus carpio* and baitfish *Carassius* sp. (Lubzens et al., 1987). However, marine rotifers sink quickly to the bottom and are therefore not suitable for feeding freshwater fish species with pelagic larvae, unless they are supplied continuously with a peristaltic pump. In this respect, freshwater rotifer such as *B. calyciflorus* is likely to have better potential for application in the larviculture of freshwater ornamental fish. To date, the use of freshwater rotifers is restricted to only a few freshwater foodfish species such as sunshine bass *Morone chrysops* × *Morone saxatilis* (Ludwig, 1994) and gudgeon and Eurasian perch *Perca fluviatilis* (Awaiss, 1991; Awaiss et al., 1992).

The use of the freshwater rotifers B. calyciflorus in the larviculture of freshwater ornamental fish was demonstrated in a recent study using dwarf gourami (Colisa lalia) in Singapore (Lim and Wong, 1997). The B. calyciflorus used in these experiments were produced by batch culture using Chlorella spp. as feed. Dwarf gourami was selected for the experiment because of its small larvae, which measure 2.74 mm total length and cannot ingest macrozooplankton such as *Moina* and *Artemia* nauplii at first feeding. Traditionally, larvae of dwarf gourami are raised in fertilised concrete ponds with earthen bottom (Fernando and Phang, 1994). Due to the difficulties in controlling water quality and the fluctuation in the quality and quantity of live food organisms, the stocking density is low (0.5 larvae/l) and the larvae are fed with egg yolk particles for the first 10 days. The study conducted in indoor tanks (10- and 200-l tanks, stocking densities 10 larvae/l from Day 2 and 30 larvae/l from Day 13) demonstrated that compared with egg yolk particles, the use of the freshwater rotifers as a starter feed significantly improved the growth and survival of dwarf gourami larvae during the rotifer-feeding phase (Days 2-12). These beneficial effects also extended to the subsequent Artemia nauplii feeding phase (Days 13–32). At the Artemia feeding phase, although all the larvae in the egg yolk group and the rotifer group were similarly fed with Artemia nauplii, the growth and survival of the rotifer group continued to be significantly better than those of the egg yolk group in that stage. Since the quantity of feed in the experiments was not limiting, these findings suggested that the quality of a starter feed was crucial to the later stages of development. Fish that were fed sub-optimally in the early stage would continue to suffer from poor performance later. At metamorphosis on Day 32, the overall survival rates of larvae fed rotifers in indoor tanks were 65.1-74.5%, which were about four times those fed egg yolk particles in the outdoor, 100-m<sup>3</sup> ponds (17.5%). The mean total lengths at metamorphosis in the indoor tanks (10.5-11.8 mm) were also significantly higher than in 100-m<sup>3</sup> ponds (8.55 mm, P < 0.05). Based on the stocking density, overall survival rates and the volume of rearing water, the yields of fry derived from larvae fed rotifers were estimated to be 6500 and 7500 fry/m<sup>3</sup> in the indoor tanks, which were much higher than 90 fry/m<sup>3</sup> obtained by farmers in 100-m<sup>3</sup> ponds, with larvae fed with natural plankton supplemented with egg yolk particles.

The use of freshwater rotifers is likely to have an important impact to the freshwater ornamental fish industry. The application of the rotifers would enable intensive larviculture of freshwater ornamental fish species with small larvae, which would eventually lead to exponential increase in the yield of the fry, as demonstrated in the study using dwarf gourami. Similar results were also obtained with white cloud mountain minnow,

Tanichthys albonubes (Lim et al., 1997). A list of freshwater ornamental fish with small larvae that require small zooplankton such as rotifers for feeding is given in Table 1. Besides, the availability of the small live food organisms would also facilitate breeding of new fish species with small larvae that could not be raised economically using the existing extensive culture method. This would eventually enhance the number of fish species for commercial production.

It should be noted that the densities of freshwater rotifers (100–120/ml, Lim, 2001a) obtained from mass cultures using dry food, and even with algae, are far below the routine densities of 600/ml obtained with marine rotifers *B. plicatilis* using artificial diets (Dhert,

Table 1 List of freshwater ornamental fish species with small larvae that require small zooplankton such as rotifers for fry production

Family	Common name	Scientific name	
Characidae	Head and tail light tetra	Hemigrammus ocellifer	
	Glowlight tetra	Hemigrammus erythrozonus	
	Silver-tipped tetra	Hasemania nana	
	Black neon tetra	Hyphessobrycon herbertaxelrodi	
	Lemon tetra	Hyphessobrycon pulchripinnis	
	Flame tetra	Hyphessobrycon flammeus	
	Black tetra	Gymnocorymbus ternetzi	
	Red-eyed tetra	Moenkhausia oligolepis	
	Neon tetra	Paracheirodon innesi	
	Penguin fish	Thayeria boehlkei	
	Serpae tetra	Hyphessobrycon serpae	
	Bloodfin tetra	Aphyocharax anisitsi	
	Red-nose tetra	Hemigrammus bleheri	
	Black phantom tetra	Megalamphodus megalopterus	
Melanotaeniidae	Red Australian rainbow	Glossolepis incisus	
Belontiidae	Fighting fish	Betta splendens	
	Paradise fish	Macropodus opercularis	
	Dwarf gourami	Colisa lalia	
	Blue gourami	Trichogaster trichopterus	
	Golden gourami	Trichogaster trichopterus	
	Pearl gourami	Trichogaster leeri	
Cyprinidae	Zebra danio	Brachydanio rerio	
• 1	Leopard danio	Brachydanio frankei	
	White cloud mountain minnow	Tanichthys albonubes	
	Silver shark	Balantiocheilos melanopterus	
	Red-finned shark	Epalzeorhynchos frenatus	
	Rainbow shark	Epalzeorhynchos erythrurus	
	Red-tailed black shark	Epalzeorhynchos bicolor	
Cyprinodontidae	Black-finned pearl fish	Cynolebias nigripinnis	
J 1	Common lyretail	Aphyosemion australe	
Cichlidae	Brown Discus	Symphysodon aequifasciata axelrodi	
	Angelfish	Pterophyllum scalare	
Callichthyidae	Bronze catfish	Corydoras aeneus	
	Albino aeneus catfish	Corydoras aeneus var.	
	Peppered corydoras	Corydoras paleatus	

1996). This could strongly limit the application of freshwater rotifers as starter diet in the ornamental fish industry. To cut down the production cost and the facilities required for culture, further research to improve the culture technique of freshwater rotifers is warranted.

# 3. Artemia nauplii

Artemia nauplii are a standard larval diet after the rotifer-feeding phase for almost all the marine foodfish species. Due to their convenience as an off-the-shelf feed and requiring only 24 h of incubation from cysts, Artemia nauplii are the most widely used live food organism for the fry production of marine as well as freshwater fish and crustaceans (Van Stappen, 1996). In freshwater ornamental fish culture, Artemia nauplii may be used as intermediate feed for larvae when Moina is too big to be ingested by the larvae. A major drawback in feeding Artemia nauplii to freshwater fish is that the nauplii die after 30–60 min in freshwater, and must therefore be fed to the fish intermittently every 2–3 h (Merchie, 1996). This could be overcome by adopting the cold storage technique, which preserves the nutritional quality and energy content of Artemia nauplii at temperatures near 4 °C (Léger et al., 1983). This technique allows not only a constant supply of high quality nauplii but also more frequent feeding to the freshwater fish larvae.

The use of *Artemia* nauplii has been demonstrated in a recent study on the culture of fry and adults of the guppy (*Poecilia reticulata*) (Lim et al., 2002a). Results showed that while there was no difference in stress resistance, the growth performances of the guppy fry and adults fed *Artemia* nauplii for 4 weeks were significantly better than those fed *Moina* (Table 2). Guppy adults fed *Artemia* nauplii also survived significantly better than

Table 2
Performance of guppy fry and adults fed different diets for 4 weeks

Parameters	Brine cysts	Dried cysts	Artemia nauplii	Moina
Guppy fry <sup>1</sup>				
Stress index (at 30 ‰) <sup>2</sup>	$210.5 \pm 3.11^{a}$	$221.8 \pm 5.25^{b}$	$247.0 \pm 6.88^{c}$	$253.5 \pm 4.51^{\circ}$
Wet weight (mg)	$92.0 \pm 1.04^{a}$	$122.3 \pm 4.94^{b}$	$91.9 \pm 8.51^{a}$	$76.7 \pm 4.71^{c}$
Dry weight (mg)	$24.9 \pm 1.54^{a}$	$37.0 \pm 2.87^{b}$	$23.9 \pm 2.51^{a,c}$	$19.1 \pm 2.58^{c}$
Total length (mm)	$20.6 \pm 0.47^{a}$	$21.7 \pm 0.46^{b}$	$20.2 \pm 0.51^{a}$	$20.1 \pm 0.66^{a}$
Survival rate (%)	$65.3 \pm 4.33^{a}$	$60.6 \pm 1.11^{a,b}$	$58.0 \pm 2.71^{b}$	$57.0 \pm 3.72^{b}$
Guppy adults <sup>3</sup>				
Stress index (at 35 ‰) <sup>2</sup>	$225.5 \pm 4.80^{a}$	$242.8 \pm 9.91^{a}$	$233.5 \pm 17.75^{a}$	$246.5 \pm 7.33^{a}$
Wet weight (mg)	$323.3 \pm 19.06^{a}$	$343.9 \pm 15.48^{a}$	$323.1 \pm 14.43^{a}$	$260.3 \pm 15.76^{b}$
Dry weight (mg)	$112.6 \pm 10.97^{a}$	$119.5 \pm 6.12^{a}$	$110.1 \pm 2.62^{a}$	$78.0 \pm 2.44^{b}$
Total length (mm)	$30.2 \pm 0.25^{a}$	$31.1 \pm 0.53^{a}$	$30.2 \pm 0.52^{a}$	$29.0 \pm 0.62^{b}$
Survival rate (%)	$100.0\pm0^a$	$99.8 \pm 0.50^{a}$	$99.5 \pm 1.00^{a}$	$98.0 \pm 0.82^{b}$

Values are means  $\pm$  standard deviations from four replicates. Values within each row that do not share the same superscript are significantly different (P<0.05).

<sup>&</sup>lt;sup>1</sup> Initial fish weight and length of fry were 6.2 mg and 9.6 mm, respectively.

<sup>&</sup>lt;sup>2</sup> Stress indices were obtained by salinity stress test reported in Lim et al., in press. The higher the stress index shown, the lower the stress resistance.

<sup>&</sup>lt;sup>3</sup> Initial fish weight and length of adult fish were 148.0 mg and 22.7 mm, respectively.

those fed *Moina*, but this was not observed in the case of guppy fry. Another study on freshwater ornamental fish in Singapore has demonstrated the use of Artemia nauplii in the feeding of Brown Discus (Symphysodon aeuifasciata axelrodi) (Lim and Wong, 1997). In Discus, larvae are dependent on the body slime of their parents as a nutrient during the first 2 weeks of exogenous feeding (Degen, 1986). The study showed that Brown Discus larvae could be raised in the absence of the parent fish through feeding with B. calyciflorus for 4 days (Days 4-7), followed by Artemia nauplii for a week (Days 8-14) (Lim and Wong, 1997). Their growth and survival rates were comparable with those that rely on parental feeding. In Discus breeding, many farmers and hobbyists have attempted to raise the larvae separated from their parents (Wattley, 1985; Petrovicky, 1988), in order to reduce cannibalism on the larvae and to shorten the breeding interval. One commonly used strategy is smuggling valuable larvae to a pair of spawners of lower quality, which act as foster parents and feed the young Discus (Degen, 1986; Fernando and Phang, 1994). However, there is always the risk that the foster parents eat the larvae (Wattley, 1985). A second strategy is raising the larvae artificially using egg yolk food, which is either smeared around the waterline (Wattley, 1985) or coated on pieces of Plexiglas shaped like fish, serving as mock parents (Petrovicky, 1988). This strategy is tedious and laborious as the egg food must be freshly prepared, applied anew at each feeding, and given to the fry at regular intervals throughout the day (Petrovicky, 1988). In addition, it requires change of tank water 2 h after each feeding (Wattley, 1985). The two existing strategies are therefore not practical for commercial application. Apart from eliminating the risk from being eaten by the parent fish, the feeding using freshwater rotifers and Artemia nauplii offers other advantages. As rotifers could be kept alive in tanks, only one feeding per day is needed. It is cleaner, causes no pollution in the larval tanks, hence water change after each feeding is not required. Feeding using rotifers and Artemia nauplii is therefore more practical for use in commercial breeding of Discus. Application of the method in Discus breeding would shorten the breeding interval and lead to a higher yield of fry. The results obtained so far indicated that Artemia nauplii, despite their marine origin, have a good potential for application in the freshwater ornamental fish culture.

### 4. Decapsulated Artemia cysts

Decapsulated *Artemia* cysts have been found to be a good quality diet comparable with freshly hatched *Artemia* nauplii for the larvae of marine shrimps and freshwater prawns, such as *Metapenaeus monoceros* (Royan, 1980), *Penaeus monodon* (Mock et al., 1980), *Penaeus indicus, Metapenaeus ensis, Metapenaeus endeavouri* and *Macrobrachium rosenbergii* (Bruggeman et al., 1980). For fish species, good results with the decapsulated cysts were obtained in the larviculture of freshwater foodfish species such as *Clarias gariepinus* (Verreth et al., 1987) and carp (Vanhaecke et al., 1990). Its use for marine species is, however, not successful, as the early larvae do not possess the enzymes necessary for digestion of the embryonic membranes (Dhert, 1991). Decapsulated cysts have a good potential for application in freshwater ornamental fish culture. Due to their smaller diameter (about 200 μm), decapsulated cysts may be used for feeding larvae of

certain ornamental fish species that cannot feed directly on *Artemia* nauplii. The cysts contain an average of 30% more energy than newly hatched *Artemia* nauplii. Dried decapsulated cysts, when treated appropriately, can be stored for years, and hence may be fed off the shelf without the need for hatching. Since a considerable amount of *Artemia* cysts in the world market has a low commercial value due to their low hatching rate (Ribeiro and Jones, 1998), they could easily be transformed to useful feed for freshwater ornamental fish and result in saving to the feed cost of the industry.

A recent study on guppy adults and the fry of five common freshwater ornamental fish species, viz. guppy, molly (P. sphenops), platy (Xiphophorus maculatus), swordtail (Xiphophorus helleri) and black neon tetra (Hyphessobrycon herbertaxelrodi), demonstrated that decapsulated cysts could be used as a substitute for Artemia nauplii or Moina in freshwater ornamental fish culture (Lim et al., 2002a). The study has shown that for both guppy fry and adults, the performance in terms of growth, survival and stress resistance of fish fed decapsulated Artemia cysts is better than or comparable with those fed Artemia nauplii or Moina (Table 2). Similar results were also obtained in the fry of all the other four ornamental fish species tested, viz. platy (initial wet weight 7.6 mg), swordtail (8.8 mg), molly (15.9 mg) and tetra (19.0 mg). Examination of the fatty acid profiles has revealed that the fatty acid compositions in terms of 20:5 (n-3) (eicosapentaenoic acid, EPA), 22:6 (n-3) (docosahexaenoic acid, DHA), total (n-3) HUFA and total fatty acid methyl ester (FAME) of dried cysts and brine cysts, are higher than those in Artemia nauplii and Moina. Hence the better performance in fish fed decapsulated cysts could at least be partly attributed to the superior fatty acid composition of the cysts, which corresponds to the higher energy content in Artemia cysts than in their nauplii (Vanhaecke et al., 1983). In particular, the higher stress resistance observed in cyst-fed fry could be associated with their higher (n-3) HUFA, which is also known to reduce the effects of stress (Menasveta, 1994). These results may also imply that when fry are given more adequate energy levels in their feeds, they will not only grow and survive better, but may also have better stress resistance. The same study also compares the performance of brine cysts and dried cysts using guppy fry and tetra fry. Results have revealed that in both species, fry fed brine cysts tend to display a higher level of stress resistance, while those fed dried cysts tend to fare better in growth. In addition, the more buoyant dried cysts are more suitable than the fast sinking brine cysts for feeding pelagic fish fry such as Tetra fry. The higher stress resistance in fish fed brine cysts compared to those fed dried cysts is not associated with the ascorbic acid (AA) content, since both brine cysts and dried cysts are deficient in AA. The fatty acid profiles of the two cyst diets and the fry fed the respective diets have also revealed that the better growth obtained in fry fed dried cysts compared to those fed brine cysts could be due to the higher (n-3) HUFA, including EPA and DHA, in the dried cysts. The lower contents in brine cysts could be due to their further metabolism after processing, as they are still alive while the dried cysts are dead.

The results obtained so far have demonstrated that the fry of all the five freshwater ornamental fish tested could readily accept decapsulated *Artemia* cysts and their performance is comparable or better than those fed *Artemia* nauplii or *Moina*. The availability of the low-cost and more hygienic decapsulated *Artemia* cysts will provide the ornamental fish industry with a suitable feed substitute for *Artemia* nauplii or *Moina*. The direct use of

decapsulated cysts may also open a new area of application for low-hatch or no-hatch cysts in the ornamental fish industry, and hence a substantial saving in the feed cost. Use of the decapsulated cysts is likely to have a positive impact in terms of farm hygiene and feed cost to the ornamental fish industry.

# 5. On-grown Artemia

The bigger and older on-grown Artemia could be a good alternative live feed for use in the hatchery of ornamental fish. The fast growth and the nonselective feeding behaviour of the Artemia make it worth to consider their culture to a bigger size using cheap agricultural by-products as feeds (Dhert, 1991). Dhert et al. (1993) developed a simple culture system for juvenile and adult Artemia as feeds for post-larval P. monodon and Lates calcarifer. Compared with Artemia nauplii, the use of the larger on-grown Artemia in the aquaculture industry is still not popular, because of the lack of supply. To facilitate the supply of on-grown Artemia for use in the ornamental fish industry, a pilot culture system for the production of on-grown Artemia in freshwater ornamental fish farms has been developed in Singapore (Lim et al., 2001). The system had 21 culture units, each consisting essentially of three components: an oval-shaped raceway, an airwater lift system and two waste collectors. Using artificial seawater at 20 % for culture and at a mean production rate of 3 kg/m<sup>3</sup> of water in a 12-day cycle, the system had a production capacity of 8 metric tons wet weight of on-grown Artemia a year. Costbenefit analysis showed that with a capital investment of US\$82,000 and an annual cost of production of US\$81,000, the system achieved a high internal rate of return of 88% over a 10-year period and a short payback period of 1.23 years. The culture system is a cheap alternative to the more sophisticated super-intensive system used in Europe. Compared with the complex automated system, the system is cost effective, simple and easy to set up and operate. As the system occupies only a small land area and uses diluted artificial seawater for culture, the freshwater ornamental farmers will have no problem to integrate Artemia production using the culture system into their farm operation to increase farm profitability.

In freshwater ornamental fish culture, the availability of bigger *Artemia* would certainly cater to the demand for live food organisms bigger than *Moina*. The size and form of a live food organism primarily determine its food value for a particular fish species. While a small food organism is desirable for fish larvae, the use of larger organisms is more beneficial as long as the size of the food organism does not interfere with the ingestion mechanism of the predator (Merchie, 1996). Fish would take a long time to attain satiation if fed smaller live food organisms, and this would result in poor growth due to inefficient feeding and waste of energy. The on-grown *Artemia* in the culture system developed in Singapore grow from 0.45 mm at inoculation to an average length of about 5 mm in 12 days. This size range is considered suitable for all sizes of freshwater ornamental fish species of up to 10 cm total length. By varying the harvesting time during the 12-day cycle, it is possible to obtain *Artemia* of any specific size within the size range for feeding, which would ensure a better energy balance in food uptake and assimilation. The flexibility in tailoring the prey size according to the age and size of the fish to be fed is

limited with other smaller live food organisms such as rotifers and *Moina*. The advantages of using on-grown *Artemia* for feeding to achieve better growth performance in ornamental fish have been demonstrated in a recent feeding experiment using Discus juveniles. Results revealed that Discus juveniles displayed a better feeding response to the on-grown *Artemia* than the other two diets, *Moina* and frozen bloodworms. Fish fed on-grown *Artemia* attained satiation within 15–30 min, and by then most of the fish displayed a bulging belly. This was however not observed in fish fed on *Moina* or bloodworms. In terms of wet weight, the *Artemia*-fed group grew significantly faster than the *Moina*-fed group, which in turn grew faster than the bloodworm-fed group (Table 3). The mean survival rate of fish fed on-grown *Artemia* was 12% higher than the group fed *Moina*, but it had no significant difference from the two groups fed *Moina* and bloodworms, respectively. The more effective food uptake, arising from bigger and more suitable size of on-grown *Artemia*, could have at least partly contributed to the better growth in the *Artemia*-fed group.

The fatty acid profile of on-grown Artemia obtained from the culture system in Singapore has also been studied (Lim et al., 2001) and the results compared with those of Artemia nauplii, Moina and bloodworms. The results showed that although the ongrown Artemia was deficient in 18:3 (n-3) (linolenic acid, LNA), their 18:2 (n-6)(linoleic acid, LLA), DHA and EPA were the highest among all the four diets tested. Due to lack of published data, it is not known whether the levels of these fatty acids in food organisms would be important for freshwater ornamental fish. On the other hand, the 20:4 (n-6) (arachidonic acid, ADA) and total (n-6) HUFA in the on-grown Artemia were also higher than in the other three food organisms. ADA might be essential to the maturation and spawning of freshwater ornamental fish. A recent study on the fatty acid profiles of common feed items used by the industry for maturation such as beef heart and Tubifex found unusually high ADA levels (Ako et al., 1999). Tamaru et al. (2000) reported that both the ADA/EPA ratio and the ADA/total fatty acids ratio in the broodstock diet of Armoured Catfish (Corydoras aeneus) were directly correlated with the egg production (number of ova/spawn) of the fish. ADA has also been shown to participate in gonadotropin-releasing hormone stimulation of gonadotropin in goldfish (Chang et al., 1989).

The continuous, nonselective feeding behaviour of on-grown *Artemia* also makes the organism an ideal booster diet, as its nutritional quality could be tailored to suit the fish requirements through bioencapsulation. This characteristic has been demonstrated by

Table 3
Mean and standard deviations of body weight, total length and survival rate of Discus juveniles after feeding on the respective feeds for 2 weeks

Parameters	Fish feeds			
	On-grown Artemia	Moina	Frozen bloodworms	
Wet weight (g)	$0.85 \pm 0.01^{a}$	$0.81 \pm 0.01^{b}$	$0.75 \pm 0.01^{\circ}$	
Total length (mm) Survival rate (%)	$3.48 \pm 0.03^{\mathrm{a}}$ $90.0 \pm 13.2^{\mathrm{a}}$	$3.44 \pm 0.04^{a,b} 78.3 \pm 10.4^{a}$	$3.37 \pm 0.04^{b}$ $91.7 \pm 2.89^{a}$	

Means with different superscripts are significantly different at P < 0.05.

Lim et al. (2001), who reported that the HUFA content in on-grown Artemia could be enhanced through bioencapsulation using an oil-emulsified product rich in docosahexaenoic acid or DHA (DHA-Selco, supplied by INVE Aquaculture, Belgium). In another study, Lim et al. (2002b) reported that although the AA content of on-grown Artemia fed rice bran and defatted soybeans during culture was undetectable, the content can be effectively enhanced through bioencapsulation using ascorbyl palmitate, following the technique reported by Merchie et al. (1995) (Fig. 1). Raising the vitamin C content in the on-grown Artemia also resulted in a concomitant increase in the incorporated AA level in the guppy. Hence bioencapsulated Artemia may also be used for nutritional prophylaxis, that is, to enhance the stress resistance and disease resistance of ornamental fish by incorporating products such as vitamin C, or immunostimulants in the on-grown Artemia for fish feeding. This is likely to lead to improvement in post-shipment survival, which has been shown in the case of guppy fed formulated diet boosted with vitamin C supplement (Lim, 2001b). Due to the small size of the brooders of ornamental fish, which are less than 100 g in most species, the enriched on-grown Artemia may also be used to enhance the nutritional quality of the broodstock. Hence other possible applications include boosting the on-grown Artemia with (a) essential nutrients such as (n-3) HUFA to improve growth and survival, reduce incidence of deformity and increase vigour, (b) pigments or colour enhancer to obtain better colour or pigmentation, (c) therapeutic drugs for fish disease treatment and (d) vaccines for fish immunisation and (e) appropriate hormones to induce sex reversal, maturation, and spawning.

Availability of the on-grown *Artemia* would offer ornamental fish farmers and exporters the possibility to apply the bioencapsulation technique to improve their fish performance and quality. In addition, the effective bioencapsulation characteristics of ongrown *Artemia* also make the organism a useful tool for larval nutrition study on freshwater ornamental fish.

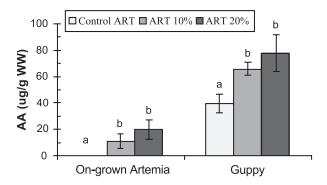


Fig. 1. Vitamin C contents ( $\mu$ g/g wet weight) in on-grown *Artemia* bioencapsulated with different levels of ascorbyl palmitate (ART 10: 10% AP added to booster emulsion; ART 20: 20% AP added) and in whole-body tissue of the guppy fed the respective *Artemia* for 20 days. Value represents mean of three replicates and its standard deviation. Any two bars marked with different alphabet letters within the same group indicate significant difference between means ( $P \le 0.05$ ).

#### 6. Conclusions

The experimental results presented in this paper have demonstrated that many of the live feeds used for fry production of marine species could also be applied successfully in the freshwater ornamental fish culture. The freshwater rotifer B. calyciflorus could be used as a suitable live feed for feeding early larvae with small mouth. Use of the rotifers would enable intensive larviculture of the freshwater ornamental fish, and would lead to better larval performance and an exponential increase in the production yield. Results have also demonstrated that Artemia nauplii, despite their marine origin, have a good potential for application in the freshwater ornamental fish culture. Compared with Moina, the use of Artemia nauplii for feeding would result in significant improvement in the growth performance of the guppy adults and fry, and better survival rate in the adult fish. With combined feeding using B. calvciflorus and Artemia nauplii, feeding of Brown Discus larvae becomes less tedious and more practical for use in commercial breeding of the Discus, and this would also eliminate the risk of larvae being eaten up and shorten the breeding interval, thereby leading to higher yield of fry. All the feeding experiments on guppy adults and fry of five common freshwater ornamental fish species have also revealed that decapsulated Artemia cysts could be used as a substitute for Artemia nauplii or Moina. Apart from being a hygienic off-the-shelf feed, the direct use of the cysts also signifies a new area of application for low-hatch cysts in the ornamental fish industry, with concomitant saving in the feed cost. On-grown Artemia may be used for feeding bigger ornamental fish such as brooders. The availability of on-grown Artemia would not only offer farmers and exporters a better alternative live food organism for feeding to their fish, but more importantly, the possibility of enhancing the fish performance and quality through bioencapsulation. The use of all the live feeds discussed in this paper is likely to have a positive impact to the ornamental fish industry.

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