Recent results from research in microdiet performance

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Molofeed, a Norwegian start-up which is dedicated to developing microdiets that aim to substitute live feed for fish and shrimp larvae in early stages, was involved in the process of improving inert diets and has developed a new generation of microencapsulated feed.

In marine larviculture, microparticulate diets have begun to rule the early larval stage hatcheries and the complete replacement of live feed in marine larval rearing is probably one of the most desired improvements in marine larval fish production. Part of the desire for a full replacement of Artemia nauplii in larval nutrition is the increasing shortage in good quality Artemia cysts coupled with an ever-increasing price and the "natural" deficiencies in common live feed (i.e. rotifers and Artemia nauplii), which need to be boosted with several enrichments to achieve acceptable results. But this approach has its limitations and apparently, technically well-designed microdiets can be used as a "transporter" for any kind of macro-, micronutrients and more.

In this article, Molofeed presents some of the recent results from their research activities in this area.

Methods

Microdiets will become widely accepted if the same performance as in the common live feed protocols are achieved. The major parameters which are considered in hatcheries are the growth rate, occurrence of malformations and survival rates throughout the larval stages. In the process of developing microdiets, specifically these three issues were thoroughly considered and tested in a number of research trials. The results were compared to the performance of larval groups fed with the standard live feed protocol (*Artemia* nauplii). In these trials, seabass larvae were used as the "model species" throughout the research and developmental process of the various recipes. A typical feeding schedule of these trials is depicted (or shown) in Figure 1. Four different feeding regimes were applied with *Artemia* feeding throughout the entire trial period (standard live feeding regime) and compared with an optimized recipe, R 1, amalgamating all knowledge gathered in the developmental process of microdiets.

Another recipe, R 2, designed with higher caloric content in order to support the requirements of larvae specifically in the early ontogenetic stage was used to compare the effects of an early and late weaning schedule.

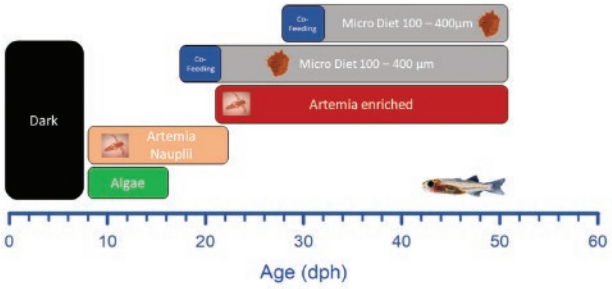

Figure 1. Typical feeding regime in research trials with sea bass larvae. First feeding was 7 days after hatching. First feed was newly hatched *Artemia nauplii* (AF 430, INVE), followed by 48h-old enriched Artemia nauplii (from 21 dph). Complete weaning to microdiets was conducted at 21 dph (early weaning, R 2) and 32 dph respectively (late weaning, R 1 & R 2). A co-feeding period of 3 days was applied in each microdiet group. The rearing trial was finished at 50 dph.

Survival

The survival rate beyond metamorphosis is one of the important indicators of the quality of the rearing conditions and a suitable feeding regime. The survival rate of the late weaning groups fed with Molofeed microdiets (R 1 & R 2) is comparable (Fig. 2) with the standard live feeding protocol (*Artemia* nauplii). The group with early weaning (R 2) shows, however,

a significantly lower survival rate, which is still a typical effect of early weaning. Part of the reasons for significantly lower survival may be attributed to the weak acceptance of microdiets in early larval stages. At this age, there are a significant number of larvae which are not able to switch quickly from live feed to microparticulate feed which resulted in a mortality peak in this treatment shortly after switching to microdiets. Another reason could be the limited digestion capacity in this age which prevents these larvae to digest microdiets as efficiently as in later stages and may contribute to the elimination of those larvae which are "naturally" less viable and disappearing at a later stage anyway. There is certainly the need for further elaboration on how to optimize microdiets for a more successful early weaning.

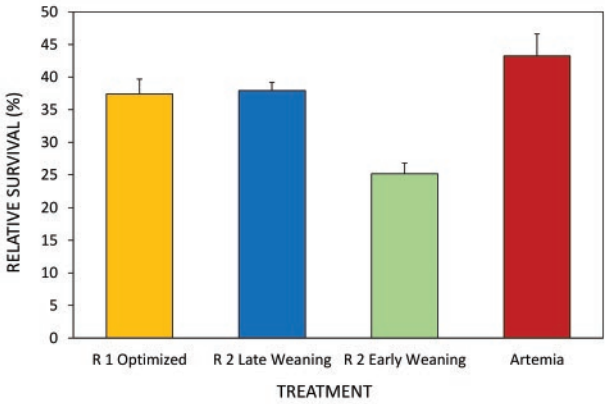

Figure 2. Comparison of the mean mortality for each treatment at the end of the trial (50 dph). The mean values were calculated from 3 replicates for each recipe (all surviving larvae counted at the end of the trial).

Growth rate

The growth rate or weight of the larvae at the end of an experiment is an overall-indicator of the rearing settings. It integrates the entire rearing conditions, such as biotic and abiotic regimes (i.e. water parameter, feeding regime, feed quality and quantity). The results from the experiments clearly depict that the optimized microdiets (R 1) in the late weaning group achieved a higher weight compared to the live feed group at 50 dph (Fig. 3). The groups with early and late weaning fed with the microdiet recipe R 2 achieved the same weight at the end of the trial, demonstrating, that early weaning is not necessarily a disadvantage with properly designed microparticulate feed, considering the growth rate of the larvae.

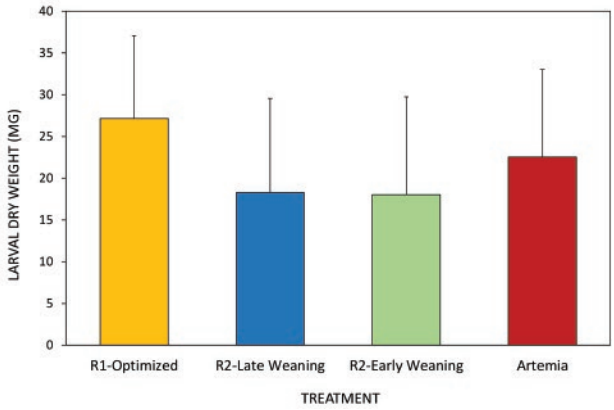

Figure 3. Mean dry weight per larva from the final sampling at 50 dph. Larvae were individually freeze-dried and weight measured with a microbalance. Mean values with standard deviation of 45 individually measured larvae per treatment from 3 replicates (corresponds to 15 individual larvae from each tank).

Skeletal anomalies

Malformations are among the most difficult issues to tackle when feeding marine fish larvae with microdiets. This used to be a major problem, specifically if early weaning (i.e. < 20 dph) was applied. Malformations are an important issue in the rearing of marine fish larvae since the fingerlings which show obvious deviations from the usual morphology are normally not accepted from on-growing farms and hatchery operators put a lot of attention on the occurrence of malformations. The following results provide evidence on the occurrence of malformations in the groups fed with microdiets (optimized recipe R 1, and R 2 for early and late weaning, compared to *Artemia*). Only kyphosis and lordosis were considered.

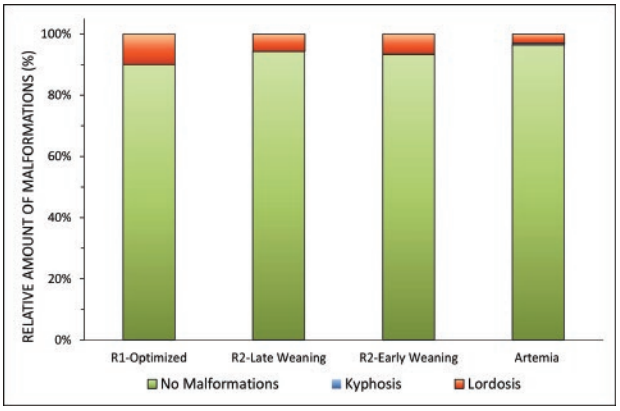

Figure 4. Relative amount of malformations for all treatments. Mean value of 3 replicates for each treatment. About 100 larvae per replicate were examined for malformations (about 300 per treatment). Lordosis is the most prominent malformation for all treatments. Kyphosis was mainly visible in the live feed group, but in a small amount.

The results demonstrated, that the rate of malformed larvae in the microdiet groups is close to the live feed group and even the early weaning did not result in a higher rate of malformations at the end of the trial. It is obvious that lordosis was the most prominent deformation, kyphosis was not an issue, with the exception of a small segment in the *Artemia* group. A small amount of malformations is usually inevitable under aquaculture conditions and can be considered as "white noise".

In order to demonstrate, that the shape and size of the larvae fed with R 1 are similar to the Artemia group examples of photos from the final evaluation of the larval size and occurrence of malformations are presented (Fig. 5 a,b). There is almost no difference in the morphology, size and shape of the larvae.

Figure 5 a-b. Arrangement of larvae at the end of a rearing trial for the measurement of individual length and rating of malformations. The example shows part of the about 100 larvae arranged per tank, in this case R 1 (left) compared with the live feed group (right, *Artemia*).

Technical quality of microdiets

The technical quality has a significant impact on the overall performance of microdiets and is often an underestimated issue. The Molofeed products use a specific and patented microencapsulation technology which reduces leaching and results in an excellent technical quality of the product. The technical properties of Molofeed microdiets help to minimize the deterioration of the water quality due to remaining food particles in the rearing tanks. The Molofeed microdiets do not disintegrate in the water and excessive feed can easily be removed from the tank bottom. The uniformity of the particle size and shape and the surface structure are quality indicators and can facilitate acceptance and feeding success (Fig. 6a).

In addition, the behavior of microdiets under humid air conditions, such as is common in tropical areas is an important feature when considering the use of microdiets under these conditions. The behavior of the Molofeed products was tested in a tilapia hatchery in Malawi (Fig. 6b). It was demonstrated that even under high air humidity, the microdiets maintain their powder-like structures and stay pourable.

Molofeed achievements

In summary, the results which were presented here demonstrate that replacement of live feed with Molofeed microdiets from 28 dph yields reasonable results which are comparable with the performance of the live feed group. Even early weaning has yielded in this trial comparable good results, although the trade-off is apparently still a higher mortality rate but the weight was the same in both treatments for R 2. Further research and improvements in the Molofeed microdiet recipes will show, if the deficiencies for early weaning can be diminished. It would be a great advantage for hatchery operators, if early weaning (e.g. around day 18 in seabass) is a reasonable option in the future.

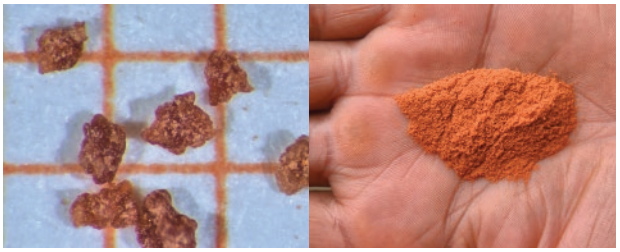

Fig. 6a (left) and Fig 6b (right). Visual inspection of a microdiet batch(left). In this picture, the size class 200 – 400µm was monitored. Scale size is 0.5mm. The special feature of Molofeed microdiets is the capsule-like character. There are some variabilities in the surface of the capsules, which disappear in the re-hydration process. Under high air humidity, the Molofeed products maintain their pourable character, thus making it suitable also for feeding larvae under tropical conditions.

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