

## Webinar - "Zebrafish Nutrition and Husbandry: Where to next?" (July 15<sup>th</sup>, 2015)

Skretting & Boston Children's Hospital hosted a webinar to discuss recent advances in zebrafish feeding and what challenges lay ahead. The session was moderated by Christian Lawrence, fish facility manager at Boston Children's Hospital. Below is a list of questions asked to the speakers on the chat board during the seminar.

### QUESTIONS AND COMMENTS FROM CHAT BOARD

- 1) Can you define "reproductive success"?

*Reproductive success refers to the outcome of any spawning event where females and males are "set-up" into mating tanks for typically 24hrs. A successful cross results in embryo production. Reproductive success is expressed as a ratio or percentage of successful crosses as a function of the total crosses that were set up in the study.*

- 2) Some effort to catch a prey can be good exercise for development of muscle in young fish.

*While this may be true from a physiological standpoint, considering the relatively high density of live prey in the husbandry tank, and the relatively slower swimming speed of a rotifer/Artemia next to a zebrafish, there is likely no reduction in the swimming activity of a zebrafish capturing a rotifer/Artemia relative to a feed particle falling through the water column. It is a known natural behavior of fish to increase swimming activity as prey density decreases, and, therefore, more time with high prey density in the tank will result in less exercise. Conversely, when feeding only 1-2 times per day with manufactured diet, the period of time with low feed presence will result in greater swimming activity. In direct feeding trials using Artemia as a control, we have found Gemma Micro fed fish to grow faster and larger (in length) which would indicate sufficient muscle growth without live prey.*

- 3) Is it common Artemia as a vector of fungi?

*Artemia can be surface disinfected to remove contaminants with a decapsulation process which uses hypochlorite. However, this does not prevent fungus from growing on embryo surfaces, along with hatched or unhatched embryos during incubation.*

- 4) Which manuals was that \*(referred to by one of the speakers when talking about live feed production)?

*FAO Manual on Culture of Live Feeds: <ftp://ftp.fao.org/docrep/fao/003/w3732e/w3732e00.pdf>*

*Plankton Culture Manual: <http://www.amazon.com/Plankton-Culture-Manual-Sixth-Edition/dp/0966296044>*

- 5) Does the Gemma Micro fed fish have the same life/fecundity span?

*In several studies that directly evaluated Gemma Micro versus control feed regimes, the data indicates that Gemma Micro fish will spawn for the same duration as controls in a given time period. In a study soon to be published, fish were crossed for more than one year (30 crosses) during which time Gemma Micro fish produced 30% more embryos and had more consistent end of life spawning.*

- 6) Fatter fish normally die earlier

*Overfeeding fish results in obesity. Fatter fish is not the result of one feed type over another, but total caloric intake. That being said, we have seen no evidence of this claim.*

- 7) Did egg production continue to improve as the female aged on the GM diet?

*In direct trials, egg production is greater at every spawning event and then plateaus and remains consistent until females reach the end of their reproductive lifespan. Again this is a context specific issue. Diet is only one of a wide number of factors that will impact this.*

- 8) Is there an article associated with the embryo production numbers shown in the last talk?

*The data in this talk was taken from two references:*

*C. Lawrence et al. / Aquaculture 368–369 (2012) 103–108.*

Barton 2015. *Aquaculture America*. ZHA special session. This paper is in the process of publication.

- 9) Any published studies with Gemma and egg quality, and not just # of embryos?

*In both above referenced studies (#9), egg quality is indicated by viable embryos assessed prior to hatching. In Lawrence et al 2012, the mean number of viable eggs ranged from 70-87% with no significant difference separating 1 feed/day from the controls. For the most part, the quantity data was only presented in the seminar due to time constraints.*

- 10) Can we get the guide for the care and use of laboratory animals?

<http://grants.nih.gov/grants/olaw/guide-for-the-care-and-use-of-laboratory-animals.pdf>

- 11) What is the fastest you can grow zebrafish to fertility?

*Most labs using Gemma Micro report egg production within 60days. More advanced facilities which approach feeding methods and practices can have reproduction in 45days. This is very rare, and again is context specific. Densities, feed type, feed delivery, tank shape, etc.*

- 12) If you feed with GEMMA MICRO nothing else is needed?

*Correct. Gemma Micro can be used as a stand-alone diet for all life stages of zebrafish from first feeding through reproduction.*

- 13) What about the "live prey" or "cage enrichment" argument for artemia?

*The live prey argument pre-supposes that a live prey item provides superior "behavioral enrichment" to a prepared diet. Firstly, fish use more than just visual senses to hunt and capture prey. They also use olfactory senses for this function, and Gemma Micro provides both visual presence (feed particles in contrast to their environment), and olfactory cues for attraction. If you watch a tank of fish compete for food pellets that are designed to cover all areas of the water column, you will see that they must engage in the same "hunting" behavior as those fed with Artemia or rotifers. So while it is possible that a live prey could enhance the welfare of laboratory fish by eliciting natural prey seeking/foraging/hunting behaviors, it is also possible that a processed pellet will do the same thing. At present there is no published data supporting either premise.*

- 14) If I feed Gemma Micro, don't I need to feed hatchfry?

*If we understand the question, it appears that you are asking if you must feed hatchfry encapsulon (another commercially available microdiet) prior to feeding fish GM. The answer to this is no. You can feed zebrafish a wide variety of different items prior to weaning them onto GM. These items must meet a certain number of requirements to be successful (size, palatability, nutritional content, stability, biological availability, etc.), but there is certainly no stipulation that you use one diet over another prior to the introduction of GM.*

- 15) What about feeding with Gemma Micro twice a day and Artemia once a day.

*The use of Artemia for zebrafish is completely unnecessary from a nutritional standpoint when using Gemma Micro. In fact, the nutritional contribution of Artemia in this program would be insignificant when you consider that each particle of Gemma Micro 300 (~23µg, dry weight) is about 10 times more concentrated nutrition than an Artemia (~2µg, dry weight).*

- 16) What about fish performance GM and Artemia?

*In trials that we are aware, Gemma Micro has only been evaluated as a stand-alone feed versus controls which includes competitor dry diet(s) plus Artemia. Results have consistently shown performance criteria are the same, and in some cases better in Gemma Micro (stand-alone) treatments.*

- 17) What embryo numbers are people averaging/female?

*This will depend on how the cross is set up (communal spawning, single crosses). Range can be 50-500 embryos/female irrespective of feed program. In Lawrence et al. 2012, mean number of embryos produced by feeding Gemma Micro 1x per day (w/out Artemia) was 400 (+/-25 sd).*