Survival and Growth on different Zebrafish Strains fed with a combination of Dry and Saltwater Rotifer Diets



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Introduction

In last few years, the number of **zebrafish facilities** using the **saltwater rotifer**, *Brachionus plicatilis*, as first feeding, has increased considerably. Zebrafish, like many other fish species in aquaculture can be considered a **live food** dependent fish, specifically during their **first larval stages**¹. However, it has been demonstrated that zebrafish larvae could be also reared without live food with a significant growth and a high survival². In this trial we analysed the effects of three different diets on **survival and growth** rates using larvae from three different **wild-type** strains and a **mutant** strain during premetamorphic and metamorphic stages (from 5 dpf- days post fertilization- until 14 dpf).

Materials & Methods



Rearing trials

Embryos from 10 pairs of adult zf wild-type strains (**ABxTL, AB, Tue**) and one double homozygous mutant strain, **Tranac'** (mpv17^{b18/b18}; mitfa^{w2/w2}) were incubated at 28 °C in Petri dishes from days 0 to 4 postfertilization. **On day 5**, 25 random hatched larval fish per group, were transferred into a **3L tank** connected to the **recirculating** flow-through system. Three tanks for each feed condition were set up to perform **triplicated sampling** for each strain (Figure **1**).





A. SURVIVAL

No significant differences were found between the control and diet groups in survival data for two of the wildtype strains at the end of the trial but we discovered significant differences on survival for one of the wildtype strains and *the mutant strain* between control and Diet B (Figure 2).



Figure 2. Survival of Zebrafish Strains. Values represent the mean \pm SD (n = 25). Differences were analysed using One-way ANOVA and Dunnett's post-test. Significant differences respect control: *, p < 0.05; **, p < 0.01.







Figure 1.	A. Feeding schedule. Schematic representation of the feeding schedule.
5-9dpf:	1. Rotifer Diets: Diet A (50 rot/mL/day), Diet B (250 rot/mL/day)
	2. Control Diet (10 rot/mL/day + dry diet (5% body weight)
10-14dpf:	1. Rotifer Diets: Diet A (25 rot/mL/day) + Artemia (2 art/mL/day), Diet B
	(125 rot/mL/day) + Artemia (2 art/mL/day)
	2. Control Diet (10 rot/mL/day) + dry diet (5%)+ Artemia (2 art/mL/day)

B. Overview of the rearing trial. Images of the rearing trial.

Growth and survival measurements

At 15 dpf each replicate tank was removed from the system and larvae were counted and photographed using a digital camera (Nikon D3200 ©). The total length of survival larvae was performed by analysing the photographs with Fiji ImageJ software. All statistical analyses were carried out in GraphPad Prism software (version 6.0)

B. GROWTH

Interestingly, after 14 dpf there were **no significant differences** in growth for the **wildtype** and the **mutant** strain that depended on diet. In contrast, control-fed larval fish were significantly longer than the diet A-fed fish in two of the wildtype strains (Figure 3).



Figure 3. Growth of Zebrafish Strains. Values represent the mean \pm SD (n= 25). Differences were analysed using One-way ANOVA and Dunnett's post-test. Significant differences respect control: **, p < 0.01; ****, p < 0.001.

Conclusions

These results suggest that **rotifers** are suitable as a **first feed** for **larval zebrafish** but it is widely known that rotifers and artemia are deficient in some essential nutrients. **Powdered dry food** would facilitate in **compensating** for this **imbalance**.

Further investigation is required into **standardization** of **larval feeding protocols** but these results suggest that **further refinement** depending on **zebrafish strain** is essential to achieve reliable results in scientific research.

References

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