Studies on bone deformity in early developmental stage of Nile tilapia *Oreochromis niloticus* induced by environmental conditions

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Highlights:

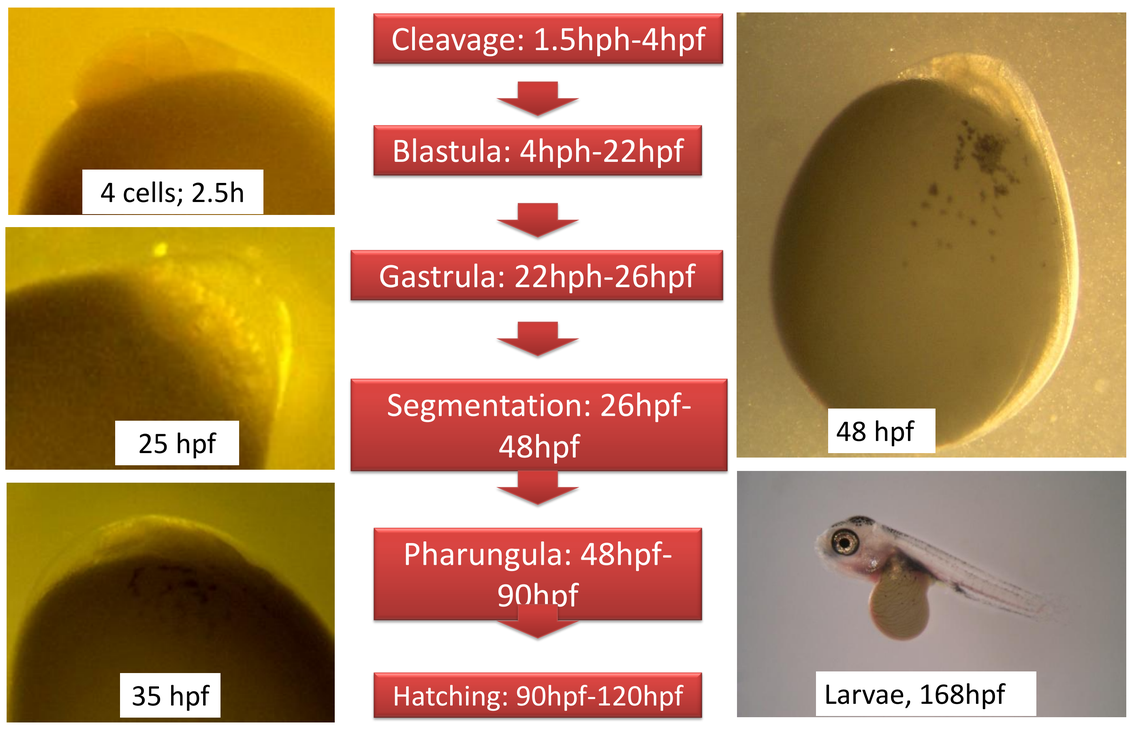
- Abnormal fishes are detrimental to aquaculture in terms of global productivity as of aquatic genetic resources
- Deprivation of oxygen and the onset of acidic pH during somitogenesis lead to spinal deformities
- Paired and/or joint neural and haemal arches are frequent within fry incubated in theses conditions from the 19th vertebrae onward

Introduction:

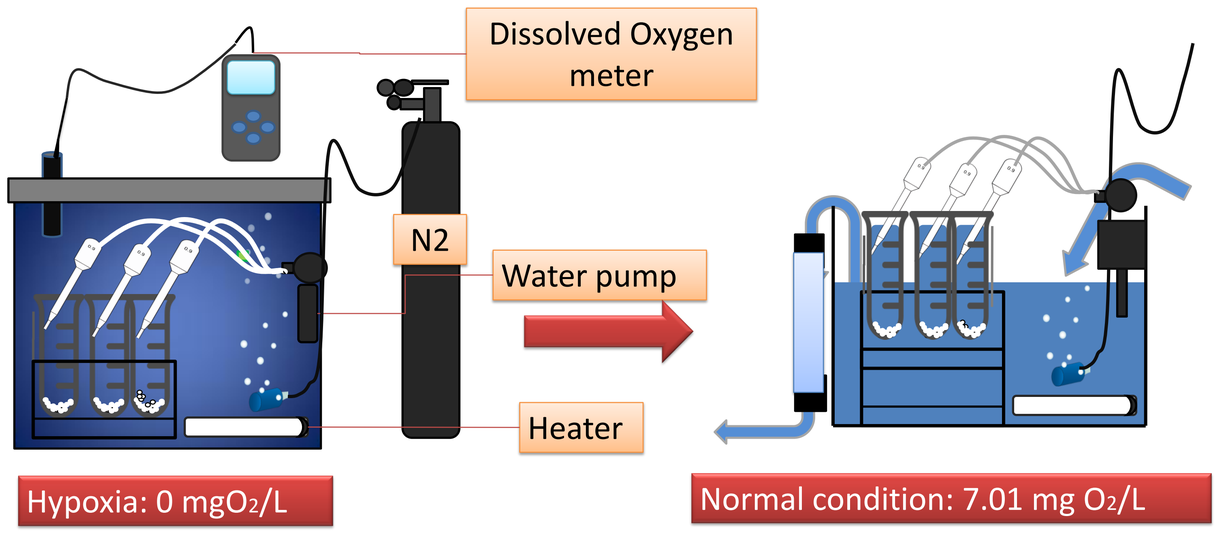
Malformations represent an economic problem due to a lower market value of the products, an increased expenditure on food, a limited growth, supplementary labor for sorting and general handling as well. Hence, there is significant interest in addressing the potential causes of these deformities. The main purpose of this study is to understand the effects of incubation into hypoxia (low oxygen concentration), acidic pH and high level of ammonia on various early life stages of Nile tilapia with regards to the occurrence of spinal malformation.

Materials and methods:

Depending on water temperature and other parameters, eggs of *Oreochromis niloticus* hatch from 90 hours post fertilization (hpf) to 120 hpf (Fujimura and Okada, 2007).



Hypoxia, through the injection of nitrogen gas into a hermetic experimental tank , was conducted on specific period: 27-33 hpf (Condition I); 33-39 hpf (Condition II); 39-45hpf (Condition III). After artificial insemination, 1559 fertilized eggs were divided into 12 tube tests for 3 replicates of control group and the 3 previously mentioned conditions.

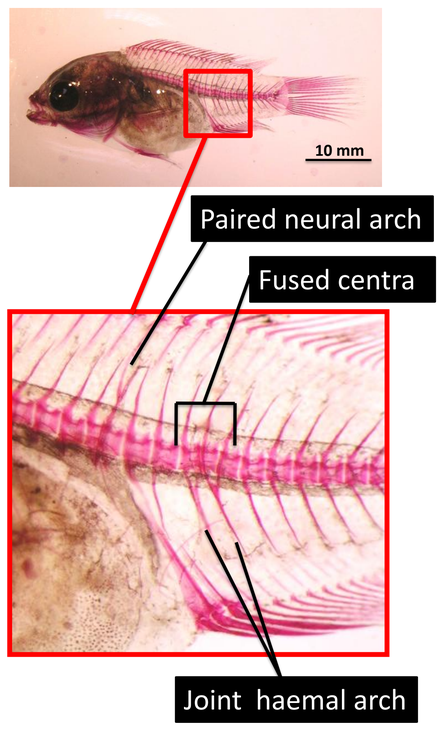


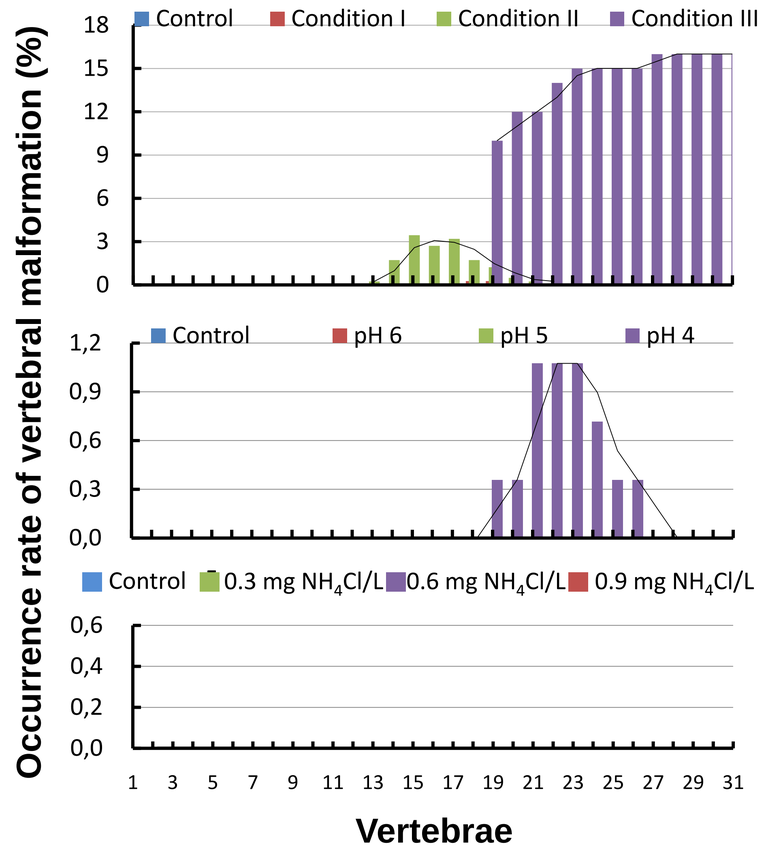
In the second experiment, the onset of **acidic condition** were carried out by adding 1N H_2SO_4 to tap water in order to reach the desired. A total of 1111 fertilized eggs were divided in 3 replicates of 4 treatments and respectively exposed to pH 3.99 ± 0.13; 5.06 ± 0.12; 6.07 ± 0.09 and 7.32 ±0 09 from 22 to 48 hpf which corresponds to the gastrula and segmentation .

In the last experiment, NH_4Cl were added at 3 different concentrations: 0.3 mg/L; 0.6 mg/L; 0.9 mg/L NH_4Cl to sustain **high level of ammonia**. The exposure of 1200 fertilized eggs divided in 3 replicates of the 3 conditions was maintained from gastrula period (22hpf) until hatching (5 dpf).
Larvae were then reared until 19 days post hatching and their total length, standard length, head length, body height were measured before double staining with alcian blue and alzarin red.

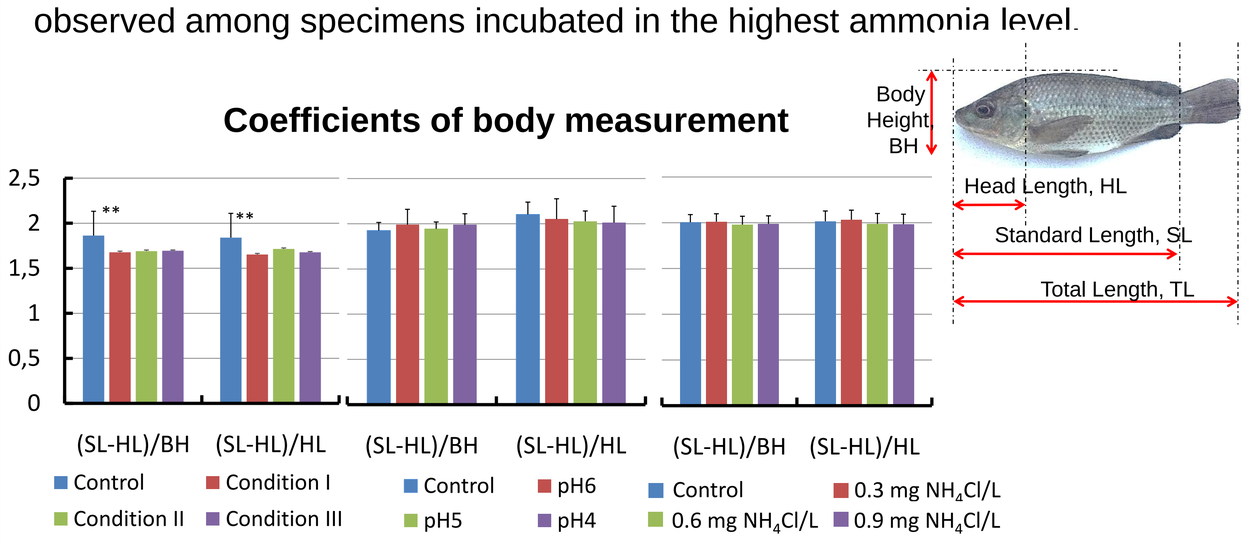
Results:

Double staining revealed higher incidence of vertebral deformation within fry incubated in hypoxia. It was commonly observed from the 19th vertebrae onward. No spinal deformity was however observed with fry from the high ammonia incubation.





The measurement showed that fry of Nile tilapia incubated in hypoxia presented a substantial significative compacted body compared to control condition (Tukey test, p <0,01). No significant difference were observed between fry within the acidic and high ammonia treatments. However, fish incubated in acidic water presented a shorter trunk, which was positively correlated with the level of pH. Along with this, a delay in growth was observed among specimens incubated in the highest ammonia level.



Conclusion:

These results showed that only a deprivation of oxygen during somitogenesis lead to spinal deformities. However, as the ammonia analysis by spectrophotometer indicated a concentration of 0.03± 0.01 mg NH_3-N/L, 0.09 ± 0.02 mg NH_3-N/L and 0.17 ± 0.02 mg NH_3-N/L within our last experiment, it is suggested to increase the concentration of NH_4Cl in order to witness more relevant data. Furthermore, future research should be carried out on a case-by-case assessment with regards to the other parameters interfering with the onset of low pH, high ammonia level or genetics studies as well.

Reference:

Fujimura K., Okada K. (2007): Development of the embryo, larva and early juvenile of Nile tilapia *Oreochromis niloticus* (Pisces: Cichlidae). Developmental staging system. Development, Growth & Differentiation 49, 301-324

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