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## Effects of Selected Levels of Water pH on the Growth and Survival of Swordtail (*Xiphophorus helleri*) Larvae

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### Abstract

Larvae of swordtail (*Xiphophorus helleri*) were exposed to pH levels of 5.5, 6.0, 7.0, 8.0, and 8.5 to study growth and survival under laboratory conditions for 42 days. The larvae were fed formulated dry pellets at 7% of their body weight. The growth and survival rates were highest at pH 8.0 and 8.5, while mortality was complete at pH 5.5. In all treatments, the specific growth rate was higher in the first week and decreased in subsequent weeks. Thus, for swordtail larvae, the pH of the water should be between 8.0 and 8.5.

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## Introduction

Water pH is one of the most crucial factors that affect fish health. Fluctuation of pH may cause stress or even mortality. Enzyme systems catalyzing metabolic energy-producing processes usually have an optimum pH. Hence, changes in pH may result in reduced metabolic performance (Heisler, 1986). Diversity of fish species can be severely limited when the pH of a lake or pond falls below 4 or 5 (Charles and Alexander, 1983). Average fish production is normally expected when the water pH ranges 7.5-8.5 (Boyd, 1998; Zweig et al., 1999) while low pH reduces fish growth and reproduction (Lopes et al., 2001). However, the optimum pH range may vary for different species (Zweig et al., 1999).

The ornamental swordtail, *Xiphophorus helleri*, is originally from Central America but is well established in Asian countries. They have many colors and mixtures of colors including red, green, black, and albino, though red is best known. They exhibit sexual dimorphism. Swordtails are extremely popular because they are easy to breed, readily available, have good growth, and are omnivores. The objective of the present study was to investigate the survival and growth of swordtail larvae at varying levels of water pH, so as to determine the optimum water pH for hatchery management.

## Materials and Methods

Larvae of the swordtail, *Xiphophorus helleri*, were procured from a private fish owner at Mumbai, India, and acclimatized to laboratory conditions in a 700-l plastic pool for one week during which they were fed freshly-hatched brine shrimp nauplii to satiation. They were then randomly stocked in 40.5-l experimental glass aquaria in fresh water. Different levels of pH (5.5, 6.0, 7.0, 8.0, and 8.5) were prepared by adding sulfuric acid to lower the pH below 7 or sodium hydroxide to raise it above 7. All treatments were conducted in triplicate.

Twenty larvae were transferred to each tank, maintained with continuous aeration via an air pump, for 42 days. Feed was prepared in the laboratory by mixing the powdered dry ingredients, adding water to prepare a dough, and cooking for 15 min in a pressure cooker (Table 1). After cooling, the dough was pelletized with a hand pelletizer (1 mm matrix) and dried in an oven at 40°C. The dried pellets were stored in air-tight plastic bottles. The feed composition and proximate analysis of the formulated dry pellets were determined according to AOAC (1990).

The larvae were fed thrice daily (10:00, 13:00, 16:00) at 7% of their body weight from one week before the start of experiment and during the 42 days of the experiment. All wastes including uneaten food particles and fecal matter were siphoned daily and 25% of the water was replaced with new

Table 1. Composition of the formulated dry pellets used to feed larvae of swordtail (*Xiphophorus helleri*, Heckel 1848).

| <i>Ingredient</i>            | <i>%</i> |
|------------------------------|----------|
| Fishmeal                     | 50       |
| Groundnut oil cake           | 25       |
| Soybean meal                 | 13       |
| Rice bran                    | 11       |
| Vitamins (premix)*           | 0.3      |
| Cod liver oil                | 0.3      |
| CMC - Starch                 | 0.4      |
| <i>Proximate composition</i> | <i>%</i> |
| Crude protein                | 45.4     |
| Lipid                        | 9.7      |
| Moisture                     | 6.8      |
| Ash                          | 9.5      |
| Carbohydrates                | 28.6     |

\*Vitamin premix contained (per kg feed): 5500 I.U. vitamin A, 1000 I.U. vitamin D<sub>3</sub>, 50 I.U. vitamin E, 10 I.U. vitamin K<sub>3</sub>, 550 mg choline chloride, 100 mg niacin, 20 mg riboflavin, 20 mg thiamin, 50 mg panto-thenic acid, 0.1 mg biotin, 5 mg folacin, 20 mg cyanocobalmin (B<sub>12</sub>), 100 mg vitamin C, 100 mg inositol

water having a similar pH. Length and weight were recorded in ten larvae from each treatment on days 0, 7, 14, 21, 28, 35, and 42. Weight gain was determined as the mean wt x no. of surviving larvae; specific growth rate was calculated as SGR (%) = (ln final wt - ln initial wt)100/t, where t is the duration in number of days.

The pH was checked daily with a pH meter (Control Dynamics pH meter, model APX 175 E/C). Other physicochemical water parameters were determined weekly, as per procedures of APHA (1995), and are shown in Table 2.

Experimental data were subjected to statistical analysis of variance (ANOVA) where differences were considered significant if  $p < 0.05$  (Snedecor and Cochran, 1967). Duncan's multiple range test was used to compare means.

## Results

Gains in both length and weight were highest in the pH 8.0 treatment (Table 3). Survival was  $99.84 \pm 0.80\%$  in the 8.0 treatment,  $99.57 \pm 0.65\%$  in the 8.5 treatment,  $51.62 \pm 0.11\%$  in the 7.0 treatment, and  $48.53 \pm 0.37\%$  in the 6.0 treatment. Larvae did not survive beyond day 14 at pH 5.5. Specific growth rate was highest at day 7 for all treatments (Table 4).

Table 2. Water characteristics.

| <i>pH</i> | <i>Dissolved oxygen (mg/l; avg)</i> | <i>Temperature (°C; avg)</i> | <i>Alkalinity (mg/l; avg)</i> | <i>Phosphorus* (mg/l; max)</i> | <i>Nitrite* (mg/l; max)</i> | <i>Nitrate* (mg/l; max)</i> | <i>Total* ammonia (mg/l; max)</i> |
|-----------|-------------------------------------|------------------------------|-------------------------------|--------------------------------|-----------------------------|-----------------------------|-----------------------------------|
| 5.5       | $6.1 \pm 0.34^a$                    | $27.5 \pm 0.02^a$            | $60.23 \pm 0.41^a$            | $0.0001 \pm 0.00^a$            | $0.01 \pm 0.00^a$           | $0.001 \pm 0.00^a$          | $0.01 \pm 0.00^a$                 |
| 6.0       | $5.9 \pm 0.01^b$                    | $27.5 \pm 0.53^a$            | $60.11 \pm 0.67^a$            | $0.0001 \pm 0.00^a$            | $0.01 \pm 0.00^a$           | $0.001 \pm 0.00^a$          | $0.01 \pm 0.00^a$                 |
| 7.0       | $6.0 \pm 0.82^a$                    | $28.0 \pm 0.22^b$            | $65.01 \pm 0.31^b$            | $0.0001 \pm 0.00^a$            | $0.01 \pm 0.00^a$           | $0.002 \pm 0.00^a$          | $0.01 \pm 0.00^a$                 |
| 8.0       | $6.1 \pm 0.38^a$                    | $27.5 \pm 0.49^a$            | $120.18 \pm 0.77^c$           | $0.0003 \pm 0.00^a$            | $0.02 \pm 0.00^a$           | $0.002 \pm 0.00^a$          | $0.02 \pm 0.00^a$                 |
| 8.5       | $6.2 \pm 0.09^a$                    | $27.5 \pm 0.35^a$            | $126.34 \pm 0.12^d$           | $0.0003 \pm 0.00^a$            | $0.02 \pm 0.00^a$           | $0.003 \pm 0.00^a$          | $0.02 \pm 0.00^a$                 |

Values in a column bearing different superscripts differ significantly ( $p < 0.05$ ).

Table 3. Length and weight of swordtail larvae grown in different water pH levels.

| pH                 | Day       |                        |                        |                        |                        |                        |                        | Total gain (%)           |
|--------------------|-----------|------------------------|------------------------|------------------------|------------------------|------------------------|------------------------|--------------------------|
|                    | 0         | 7                      | 14                     | 21                     | 28                     | 35                     | 42                     |                          |
| <i>Length (cm)</i> |           |                        |                        |                        |                        |                        |                        |                          |
| 5.5                | 0.75±0.13 | 0.85±0.05 <sup>a</sup> | 1.29±0.10 <sup>b</sup> | *                      | *                      | *                      | *                      | *                        |
| 6.0                | 0.75±0.13 | 0.87±0.07 <sup>a</sup> | 1.14±0.13 <sup>b</sup> | 1.33±0.17 <sup>b</sup> | 1.51±0.18 <sup>b</sup> | 1.98±0.20 <sup>b</sup> | 2.01±0.39 <sup>c</sup> | 168.00±3.12 <sup>a</sup> |
| 7.0                | 0.75±0.13 | 0.92±0.10 <sup>a</sup> | 1.38±0.15 <sup>b</sup> | 1.56±0.18 <sup>b</sup> | 1.92±0.21 <sup>b</sup> | 2.93±0.29 <sup>c</sup> | 3.73±0.81 <sup>d</sup> | 397.33±3.59 <sup>b</sup> |
| 8.0                | 0.75±0.13 | 0.95±0.14 <sup>a</sup> | 1.45±0.20 <sup>b</sup> | 1.84±0.32 <sup>b</sup> | 2.01±0.73 <sup>c</sup> | 3.52±0.97 <sup>d</sup> | 4.48±0.98 <sup>e</sup> | 497.32±4.10 <sup>c</sup> |
| 8.5                | 0.75±0.13 | 0.98±0.17 <sup>a</sup> | 1.79±0.29 <sup>b</sup> | 2.93±0.37 <sup>c</sup> | 3.76±0.71 <sup>d</sup> | 4.10±0.82 <sup>e</sup> | 5.51±1.12 <sup>f</sup> | 634.67±4.85 <sup>d</sup> |
| <i>Weight (g)</i>  |           |                        |                        |                        |                        |                        |                        |                          |
| 5.5                | 0.40±0.10 | 0.55±0.01 <sup>a</sup> | 0.59±0.02 <sup>a</sup> | *                      | *                      | *                      | *                      | *                        |
| 6.0                | 0.40±0.10 | 0.57±0.12 <sup>a</sup> | 0.60±0.14 <sup>a</sup> | 0.66±0.17 <sup>a</sup> | 0.69±0.19 <sup>a</sup> | 0.74±0.20 <sup>a</sup> | 0.99±0.22 <sup>b</sup> | 147.50±2.52 <sup>a</sup> |
| 7.0                | 0.40±0.10 | 0.59±0.13 <sup>a</sup> | 0.63±0.16 <sup>a</sup> | 0.68±0.19 <sup>a</sup> | 0.74±0.20 <sup>a</sup> | 0.87±0.23 <sup>a</sup> | 1.95±0.25 <sup>b</sup> | 387.52±2.86 <sup>b</sup> |
| 8.0                | 0.40±0.10 | 0.60±0.15 <sup>a</sup> | 0.65±0.17 <sup>a</sup> | 0.72±0.20 <sup>a</sup> | 0.95±0.22 <sup>a</sup> | 1.63±0.27 <sup>b</sup> | 2.45±0.30 <sup>c</sup> | 512.51±3.71 <sup>c</sup> |
| 8.5                | 0.40±0.10 | 0.62±0.17 <sup>a</sup> | 0.68±0.21 <sup>a</sup> | 0.98±0.27 <sup>a</sup> | 1.60±0.29 <sup>b</sup> | 2.85±0.31 <sup>c</sup> | 3.60±0.38 <sup>d</sup> | 800.00±3.89 <sup>d</sup> |

Values in rows with different subscripts significantly differ ( $p < 0.05$ ) as determined by ANOVA and Duncan's test comparison of mean values.

\* No survival

Table 4. Weekly specific growth rate (SGR) of swordtail larvae grown in different pH.

| pH  | Day                    |                        |                        |                        |                        |                        |
|-----|------------------------|------------------------|------------------------|------------------------|------------------------|------------------------|
|     | 7                      | 14                     | 21                     | 28                     | 35                     | 42                     |
| 5.5 | 5.55±0.32 <sup>a</sup> | 1.00±0.47 <sup>b</sup> | *                      | *                      | *                      | *                      |
| 6.0 | 5.65±0.37 <sup>a</sup> | 0.73±0.05 <sup>b</sup> | 1.36±0.77 <sup>c</sup> | 0.63±0.43 <sup>b</sup> | 0.99±0.69 <sup>b</sup> | 2.15±0.83 <sup>d</sup> |
| 7.0 | 5.75±0.69 <sup>a</sup> | 0.93±0.38 <sup>b</sup> | 1.09±0.52 <sup>c</sup> | 1.20±0.65 <sup>c</sup> | 2.31±0.86 <sup>d</sup> | 3.77±0.10 <sup>e</sup> |
| 8.0 | 5.79±0.70 <sup>a</sup> | 1.14±0.27 <sup>b</sup> | 1.46±0.67 <sup>b</sup> | 2.06±0.89 <sup>c</sup> | 3.34±0.69 <sup>d</sup> | 4.31±0.03 <sup>e</sup> |
| 8.5 | 6.26±0.25 <sup>a</sup> | 1.36±0.65 <sup>b</sup> | 1.96±0.98 <sup>b</sup> | 3.30±0.79 <sup>c</sup> | 4.67±0.64 <sup>d</sup> | 5.23±0.95 <sup>e</sup> |

Values in rows with different subscripts significantly differ ( $p < 0.05$ ) as determined by ANOVA and Duncan's test comparison of mean values.

\* No survival

## Discussion

pH values below 7 reduce fish growth and reproduction (Lopes et al., 2001). High alkaline pH likewise decreases growth (Boyd, 1998; Zweig et al., 1999). Larval stages are the most sensitive to pH in silver catfish (Lopes et al., 2001) and flagfish (Graig and Baksi, 1977). The decreased oxygen carrying capacity in low blood pH can be an important cause of fish mortality at low pH levels. Trout had considerably reduced oxygen consumption at levels lower than pH 8 and sodium loss when exposed to low pH levels for a few hours (Packer and Dunson, 1970). We therefore believe that the mortality of fish grown in pH 5.5 might have been due to deprivation of oxygen, coupled with loss of body sodium. In conclusion, while alkaline water of pH 8.0 or 8.5 was most suitable for growth and survival of swordtail fry in the current study, literature

suggests that the preferred pH may vary with the age of the fish. Hence, the suitable pH level for other stages of swordtail should be investigated.

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