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Incubator for individual eggs of fish

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ABSTRACT

We have constructed and tested a device for incubation of individual eggs of fish. We measured the diameter of fertilized and water hardened eggs of vendace (*Coregonus albula*). Each egg developed in a separate incubator, which was a modified Pasteur pipette. Incubation of 315 eggs resulted in 249 hatched embryos (eleutheroembryos). The technique enabled identification of each eleutheroembryo hatching from a given egg. Example of relationship between external diameter of egg and the length of eleutheroembryo showed potential applications of the technique in studies on fish reproduction biology.

INTRODUCTION

Numerous studies have been directed at describing the relationship between the size of fish eggs and a variety of important early life history characteristics including survival to hatch (Johnston et al. 2007), morphometrics (e.g. total length etc.) (Luczynski et al. 1986), vertebral counts (Lindsey and Ali 1971), growth rate (Springate and Bromage 1985; Wallace and Aasjord 1984), resistance to starvation (Rana 1985), etc. In most cases the investigators did not identify eleutheroembryos emerging from individual eggs of known size; instead, they reported correlation between mean size of eggs and mean size (or other parameters) of eleutheroembryos (and subsequent larvae).

In this study we measured individual eggs of vendace [*Coregonus albula* (L.)] and then the individual eleutheroembryos that hatched from each egg. Successful incubation of over 300 eggs and examples of relationships between external diameter of the egg and the dimensions of the eleutheroembryo and its yolk sac proved that such a technique may be applied in case of a variety of fish species [as vendace is a fragile species, with long incubation time, high oxygen demand, etc. (Luczynski 1985)].

MATERIAL AND METHODS

Vendace eggs were stripped from live females into a plastic dish, fertilized by milt from live males, and allowed to water-harden at the temperature of 5-6°C.

External diameter of each individual egg was measured using ocular micrometer (magnification 21X) to an accuracy

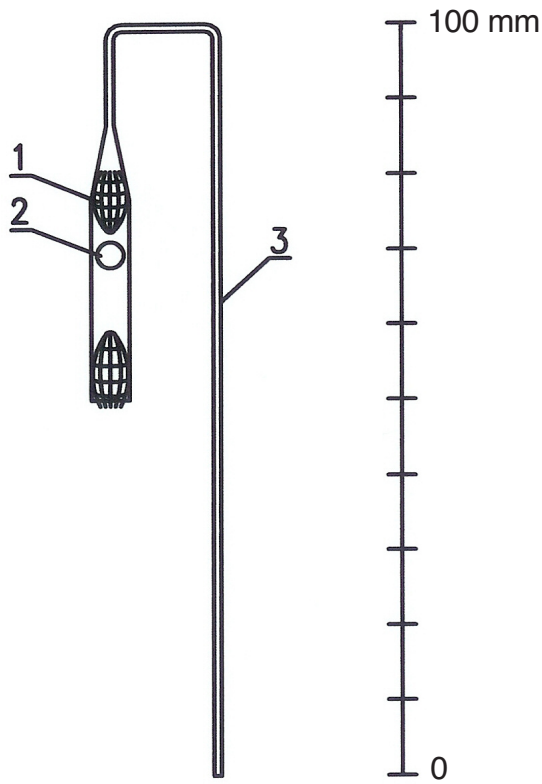

Figure 1. Incubator for individual eggs of fish—a modified Pasteur pipette, hung over the edge of the water tank (wide end inside the tank), with tulle stoppers (1) keeping the fish egg (2) inside the incubator. A capillary tube (3) causes the water flow through the incubator (see: Figure 2).

of 0.01mm. After hatching each eleutheroembryo was measured for body length (caudal length, CL), total length (TL) as well as yolk-sac length and height using the same method ascribed for the eggs.

An incubator for individual egg (Figure 1) was a modified (designed and manufactured by R.B.) Pasteur pipette, whose work depends on the capillary forces. Water drops falling from the narrow tip of the incubator (Figure 2) signalled that the incubator was working and the egg was well supplied with water. The flow through the incubator ranged between 0.5 and 10.0ml·min^{-1} (the average for 30 incubators was 2.9ml·min^{-1}). Such a flow of well oxygenated water secured successful incubation of vendace eggs. Incubators were hung over the edge of the water tank (Figure 2). The edge had numbered furrows which enabled identification of individual incubators. Water in the tank was mixed mechanically, because aeration could result in a blockage of the capillaries by tiny air-bubbles.

At first, a small piece of a tulle (a "tulle stopper") was placed inside the incubator close to the narrow piece of the pipette (Figure 1). The egg was placed in the incubator using a pipette ordinarily used in fish hatcheries. Then the wide piece of the incubator was closed with another tulle stopper.

Figure 2. A series of incubators hanging over the edge of the water tank; water drops out from the capillary tube endings.

Hanging of the pipette over the edge of the water tank usually started the water flow through the incubator.

The system required a regular maintenance, and each incubator had to be controlled at least twice daily. In case of any trouble with the air-bubble (or whatever cause of the water flow discontinuation), a pipette equipped with a narrow rubber pipe on one end and with a small rubber bulb on the other helped solving the trouble.

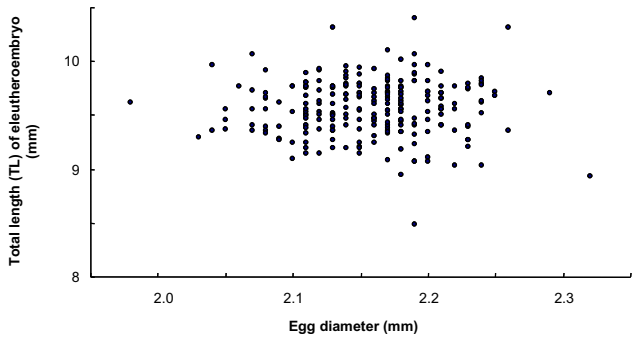

Figure 3. Data on the total length (TL) of vendace (*Coregonus albula*) eleutheroembryos hatched from 249 individual eggs of known external diameter.

RESULTS AND CONCLUSIONS

The incubation of individual eggs of vendace worked well until the day 68 of the experiment, when the water supply to the laboratory was discontinued during the night. The incubators worked until the water level in the tank dropped and the water flow through the incubators was disrupted. With no water flow, the embryos, already well advanced in their development, experienced hypoxia. Under hypoxic conditions vendace embryos tend to hatch prematurely (Luczynski et al. 1993). Due to that, 111 eleutheroembryos hatched overnight in response to the stress of hypoxia (in total 249 embryos hatched from 315 eggs incubated in the experiment).

It is known that the environmental conditions of egg incubation can move the event of embryos' hatching into earlier or later stages of their development (Luczynski et al. 1993). In case of this experiment, it meant that numerous embryos have hatched not because they were mature enough in their development, but because they experienced the hypoxia stress. This resulted in mass hatching of embryos differently advanced in their development, and prevented seeking for the relationships between the egg diameter and the length of hatching eleutheroembryo and the size of its yolk sac. Accordingly, Figure 3 shows only an example of a possible relationship between the size of the individual eggs and dimensions of emerging eleutheroembryos.

Such relationships could be examined if the experiment went well until its end; in case of our study, however, the failure in the water supply caused premature hatching of embryos and terminated the experiment. The conclusion is that the most important feature of the incubation system is the tank(s) containing the water reserve big enough to supply the incubators over the time intervals between successive maintenance services.

REFERENCES

Johnston, T.A., M.D. Wiegand, W.C. Leggett, R.J. Pronyk, S.D. Dyal, K.E. Watchorn, S. Kollar, J.M. Casselman. 2007. Hatching success of walleye embryos in relation to maternal and ova characteristics. Ecology of Freshwater Fish 16: 295-306.

Lindsey, C.C., M.Y. Ali. 1971. An experiment with medaka, *Oryzias latipes*, and a critique of the hypothesis that teleost egg size controls vertebral count. Journal of the Fisheries Research Board of Canada 28: 1235-1240

Luczynski, M. 1985. Survival of *Coregonus albula* (L.) (Teleostei) embryos incubated at different thermal conditions. Hydrobiologia 121: 51-58.

Luczynski, M., W. Wojtaszek, A. Kirklewska. 1986. The influence of egg size and thermal conditions of incubation on the body length and the yolk sac volume of vendace (*Coregonus albula* L.) eleutheroembryos. Acta hydrochimica et hydrobiologica 14: 175-183.

Luczynski, M., P. Brzuzan, P. Czerkies. 1993. The hatching process in Coregoninae embryos--A review. In: Fish Ecotoxicology and Ecophysiology (ed. T. Braunbeck, W. Hahnke, H. Segner), pp. 199-208. VCH Verlagsgesellschaft mbH, Weinheim.

Rana, K.J. 1985. Influence of egg size on the growth, onset of feeding, point-of-no-return, and survival of unfed *Oreochromis mossambicus* fry. Aquaculture 46: 119-131.

Springate, J.R.C., N.R. Bromage. 1985. Effects of egg size on early growth and survival in rainbow trout (*Salmo gairdneri Richardson*). Aquaculture 47: 163-172.

Wallace, J.C., D. Aasjord. 1984. An investigation of the consequences of egg size for the culture of Arctic charr, *Salvelinus alpinus* (L.). Journal of Fish Biology 24: 427-435.