



Uroz Ljubobratovi , Endre Balogh , Svetlana Lengyel, Gyula Kovács, Ágnes Adorján, Endre Janurik, István Csengeri, András Rónyai

National Agricultural Research and Innovation Centre, Fisheries Institute NAIK HAKI, Szarvas, Hungary

Introduction

Techniques for stickiness elimination in pikeperch, *Sander lucioperca*, eggs have been documented in several studies (Demska-ak and ak 2005; Kucharczyk et al., 2007; K iztçan et al. 2012; arski et al. 2013). Testing described methodologies in our hatchery lead to unsuccessful egg incubation, either in terms of de-adhesion or hatching rate (Ljubobratovi et al., 2013). Variability of previously reported outcomes with usage of tannic acid for the mentioned purpose could be due to the different water quality parameters as it plays a significant role in the reaction between phenol groups and proteins (Ozidal et al., 2013). From the previous study it was concluded that milk has a positive impact on hatching and embryo development while in the term of de-adhesion it needs further improvement of used technique. Kaolin as the clay mineral with fine texture could be used for this purpose. In this study influence of water hardness on tannic acid bath, improvement of milk treatment with kaolin bath and preliminary testing of kaolin itself as the de-adhesion substance has been investigated.

Materials and Methods

For the purpose of the study six females have been stripped. Eggs of three females have been used for tannic acid treatments and the eggs of the rest females have been used for the milk and kaolin treatments. In that sense, each replication of each treatment was originating from different female. Four treatments were tested in triplication while the two treatments (K-3 and K-4) were preliminary tested without replicates, as follows:

T-C . eggs were bathed in tannic acid solution in clean hatchery water (15_dH water hardness);

T-EDTA . 400 . eggs were bathed in tannic acid solution in 400mg L⁻¹ EDTA solution in hatchery water (9_dH water hardness);

T-EDTA . 800 . eggs were bathed in tannic acid solution in 800mg L⁻¹ EDTA solution in hatchery water (3_dH water hardness);

MK- eggs were stirred in milk solution (1L 3.5% milk in 7L of hatchery water) for 30 minutes and bathed in 25cc L⁻¹ kaolin solution for three minutes;

K-3 and K-4 . in these treatments after 30 minutes stirring in clean hatchery water the eggs were bathed in 25cc L⁻¹ kaolin solution for 3 and 4 minutes, respectively.

Statistical analyses was based on the one-way ANOVA and the Welch's Test for parameters with normal distribution and parameters with unknown distribution were analysed with the Kruskal-Wallis Test (P<0.05).

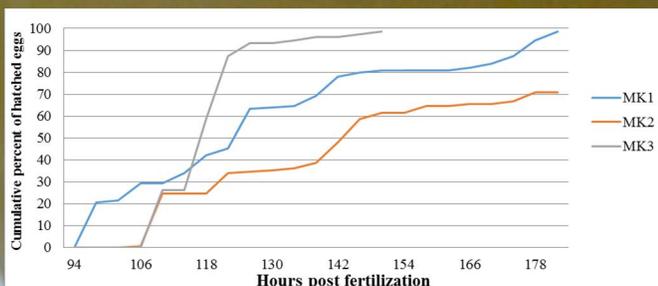


Figure 1. Hatching dynamics in three replications of milk and kaolin (MK) treatment.

Table 1. The effect of different de-adhesion methods on fertilization, hatching and deformity rate

Parameter	T-C	T-EDTA-400	T-EDTA-800	MK	K-3*	K-4*
Fertilization rate (%)	66.8±7.2 ^a	66.5±10.0 ^a	63.7±11.9 ^a	-	-	-
Hatching rate (%)	5.5±1.3 ^a	3.5±1.5 ^a	2.7±14.3 ^a	89.3±16.2 ^b	67	85
Hatching period (%)	49±22 ^a	12±4 ^b	60±4 ^a	65±23 ^a	84	72
Deformity rate (%)	33.1±4.5 ^{ab}	62.9±35.8 ^a	36.1±12.7 ^{ab}	3.6±1.8 ^b	16.0	36.7

*treatments without replications

Results and Discussion

In term of adhesiveness elimination all treatments were effective. Results of the study are summarized in Table 1. Hatching dynamics for MK and K treatments are presented in Figures 1 and 2.

Results obtained in this study are indicating that the method with tannic acid can lead to variable outcomes among hatcheries either from the reason of different water quality parameters or differences among commercial products. It seems that reaction between phenol groups and ions during the tannic acid bath does not have significant impact on the hardening of egg chorion. Combined treatment with milk and kaolin for egg de-adhesion can be described as successful and reliable for effective stickiness removal and obtaining high hatching rate, while the preliminary testing of kaolin implies rather simple technique which needs further investigation.



Figure 2. Hatching dynamics in K-3 and K-4 treatments

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