

# mRNA Expression in European Eel Eggs and Embryos and its Relationship to Hatching Success

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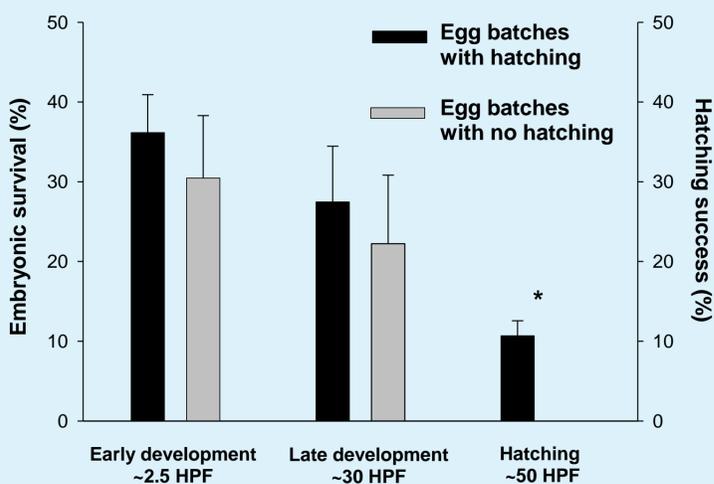
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## 1. Introduction

Aquaculture production of viable European eel larvae has proven feasible<sup>1</sup>, however with highly variable embryonic development and hatching success. Early embryonic development in fishes is catalysed by proteins translated from maternal mRNA incorporated in the oocytes during oogenesis<sup>2</sup>. Variation in quantity of this mRNA may explain this high variation in embryonic development and hatching success.

## 2. Objectives

We analyzed the relative expression of Tubulin  $\beta$ , Insulin-like growth factor 2 (IGF2), Nucleoplasmin (npm2), Prohibitin 2 (PHB2), Phosphatidylinositol glycan biosynthesis class F protein 5 (PIGF5), and carnitine O-palmitoyltransferase liver isoform-like 1 (CPT1), maternal mRNA of these genes have been associated to embryonic development in fishes<sup>3,4,5</sup>. Relative expression of these genes was analyzed at different embryonic developmental stages and compared with hatching success.



**Fig. 1.** Embryonic survival/ hatching success over time. Bars represent means  $\pm$  SEM. The left y-axis shows embryonic survival, and the right y-axis shows hatching success. The HPF  $\times$  egg quality interaction term was significant, therefore the model was decomposed at each HPF and analyzed using a series of t-tests.



Farmed European eel



Stripping of European eel



Incubation flasks



Unfertilized eggs



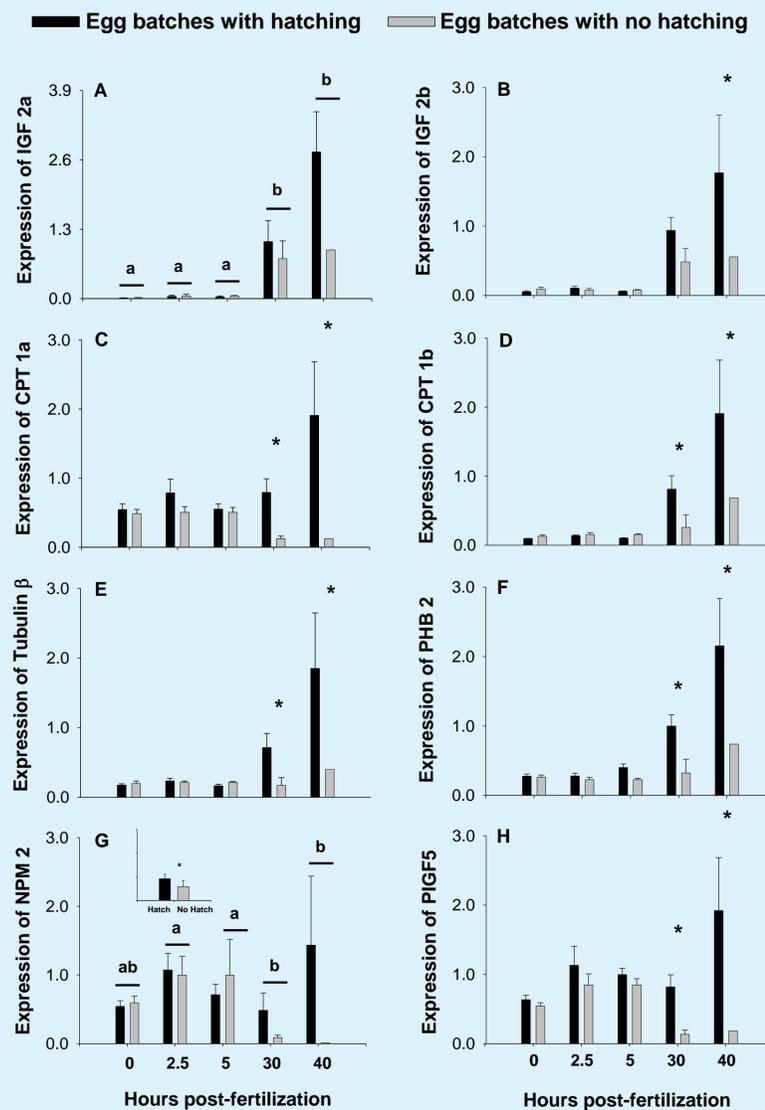
40 HPF embryos



Fume hood

## 3. Materials and methods

- ❑ Samples were taken from 15 batches just before fertilization (0 hours post fertilization; HPF), 2.5 HPF, 5 HPF, 30 HPF, 40 HPF
- ❑ RNA was extracted from samples
- ❑ RNA was transcribed to cDNA by reverse-transcription
- ❑ Relative qPCR was performed
- ❑ Batches were organized into two groups: (i) with hatching larvae (ii) with no hatching (Fig. 1)
- ❑ Expressions data were analyzed by two-way repeated measures ANOVA



**Fig. 2.** Relative gene expression over time. Bars represent means  $\pm$  SEM. For significant HPF  $\times$  egg quality interactions, differences between groups were analyzed by t-tests (shown by an asterisk, panels B, C, D, E, F, and H). For non-significant HPF  $\times$  egg quality interaction main effects were interpreted; time points without a common letter superscript differed (panels A and G). For NPM2 differences in average expression between groups is indicated by an asterisk (panel G).

## 4. Results

- ❑ The HPF  $\times$  egg quality interaction term was significant for IGF2b, CPT1a, CPT1b, Tubulin  $\beta$ , PIGF5, and PHB2 (Fig. 2B, C, D, E, F, and H)
- ❑ No significant differences were found, between the two groups, in expression of any of the genes at 0 HPF, 2.5 HPF, and 5 HPF (Fig. 2)
- ❑ Significant differences were found between the two groups for expression of CPT1a, CPT1b, Tubulin  $\beta$ , PIGF5, and PHB2 at 30 HPF (Fig. 2C, D, E, F, and H)
- ❑ At 40 HPF expression of all genes showed significant differences (Fig. 2)



## 5. Conclusion

This study indicates that maternally incorporated mRNA transcripts, of the analyzed genes, does not govern embryonic development in European eel. However, later in development (30 & 40 HPF) differences in expression, between groups, can be seen for most genes, which points to up-regulation of expression in embryos from batches, which generated hatched larvae.



Mating eels

## 6. Acknowledgements

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