Temporal transcript profile of plasticity-related genes in immature zebrafish brain after seizures

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Abstract
In this study, we aimed to investigate the role of genes related to plasticity (karln and bdnf) in the context of epilepsy using the zebrafish seizure-model.

Key words:
Epilepsy, zebrafish, bdnf gene, karln gene

Introduction
It is known that seizures are related to neuroplastic changes in the brain. Zebrafish has become attractive in vivo model for epilepsy studies; however, the transcript profile of plasticity-related genes in immature zebrafish brain after seizures is still unknown. In this study, we aimed to investigate the transcript response of two genes associated with plastic changes in normal and pathological brain conditions, the karln and bdnf \cite{1,2} genes.

Results and Discussion
Larvae at seven days post-fertilization (dpf) were separated in Seizure (LP) and Control (LC) groups. Seven dpf larvae from LS were placed individually in well-plate containing PTZ 15mM. LC was handled in the same condition but in normal bath water. According to their groups (0.05 or 24h after seizure), animals from LP were crioanesthetized and their heads were collected for RNA extraction. We did the same process for the LC. A total of five samples were used for each group, and each sample was composed by pooling 10 heads. The mean ± SEM of karln mRNA levels and the \( p \) values obtained comparing each time-point between the LC and LP groups were the following: LC0.05h 1.014±0.04113 and LP0.05h 0.9112±0.07714 (\( p \geq 0.5 \)); LC24h 0.4729±0.05264 and LP24h 0.4874±0.04331 (\( p \geq 0.5 \)). The mean ± SEM of bdnf mRNA levels and the \( p \) values were the following: LC0.05h 1.028±0.07193 and LP0.05h 1.017±0.1116 (\( p \geq 0.5 \)); LC24h 0.3051±0.02934 and LP24h 0.3223±0.02866 (\( p \geq 0.5 \)). Although no differences were found between LC and LP for both genes investigated in immature zebrafish brain, it is plausible to consider that: (i) the neuroplasticity in immature brain already has a mechanism exceptionally active during the development, so, any changes after seizures would not provide a stimulus beyond what already occurs physiologically; (ii) another possibility is about the time-points investigated. We should consider that differential expression can occur in different time-points we have investigated.

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In a previous study\cite{3}, we found that the bdnf transcript in adult zebrafish brain is up-regulated immediately after induced-seizure, at the time 0.05h. Taking into account this previous result, in adult zebrafish, and the present findings, we can postulate that the response in immature zebrafish brain is different that seem like in adult.

Conclusions
We did not find differences in the transcript levels of the kalrn and bdnf genes in immature zebrafish brain up to 24h after the seizure insult, when compared with no-seizures group. Further investigations are necessary to clarify this question, and the impact of this finding to the currently knowledge that the immature brain is less vulnerable to damage as consequence of seizures\cite{4}.

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Figure 1. Relative quantification of the karln transcripts in zebrafish larvae brain at 0.05h and 24h after pentylenetetrazole-evoked seizures.

Figure 2. Relative quantification of the bdnf transcripts in zebrafish larvae brain at 0.05h and 24h after pentylenetetrazole-evoked seizures.


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