Consequences of brain aromatase knock-out on cell proliferation, differentiation and behavior in zebrafish

Cassandra Malleret\textsuperscript{1}, Mélanie Blanc-Legendre\textsuperscript{2}, Laëtitia Guillot\textsuperscript{1}, Xavier Cousin\textsuperscript{2}, Thierry D. Charlier\textsuperscript{1}, and Elisabeth Pellegrini\textsuperscript{1}

\textsuperscript{1}Institut de recherche en santé, environnement et travail (Irset) – Université d’Angers, Université de Rennes, École des Hautes Études en Santé Publique [EHESP], Institut National de la Santé et de la Recherche Médicale, Structure Fédérative de Recherche en Biologie et Santé de Rennes – 9 avenue du professeur Léon Bernard 35000 Rennes, France

\textsuperscript{2}Station IFREMER de Palavas-les-Flots – Institut français de Recherche pour l’Exploitation de la Mer – Chemin de Maguelone 34250 Palavas-les-Flots 525 Avenue, France

Résumé

Amongst its pleiotropic physiological effects, 17B-estradiol (E2) regulates neuroplasticity. The key enzyme involved in E2 synthesis from testosterone is aromatase. In the brain of teleost fish, aromatase B (AroB) is encoded by the \textit{cyp19a1b} gene. Previous data from the laboratory have shown that, in zebrafish, \textit{cyp19a1b} is strongly expressed in radial glial cells that actively divide to generate newborn cells, illustrating the central role of E2 in neuroplasticity.

The aim of this work is to investigate the impact of AroB deletion on neurogenesis and behavior using specific AroB-KO juvenile and adult zebrafish.

Swimming activity was investigated in 1 month juveniles and several behavioral traits were studied in 3 month adults. Statistical analyses revealed that AroB-KO males were hypoactive compared to WT counterparts, and AroB-KO females showed a similar trend. In addition, AroB-KO males displayed decreased boldness and aggressivity compared to their WT counterparts while females were not statistically impacted.

Proliferation and differentiation of dopaminergic and serotonergic neurons were also investigated in juvenile and adult development stages by immunofluorescence and by RT-qPCR.

In adults, our results highlighted region-dependent effects of AroB deletion on cell proliferation in both male and female AroB-KO compared to WT. While an increase in the number of proliferating cells was observed in the olfactory bulbs of AroB-KO animals, the other regions of the telencephalon and diencephalon showed a statistically significant decrease. qPCR performed on adult brains showed a significant decrease in PCNA transcript expression, a marker cell proliferation, in both AroB-KO adult males and females compared to wild-type. No change in the number of dopaminergic and serotonergic neurons was observed in adult male and female olfactory bulbs, telencephalon and diencephalon.

In juveniles, proliferation was quantified in three regions, the ventral pallium of the telencephalon (equivalent to amygdala) the medial pallium of the telencephalon (equivalent to
hippocampus and the diencephalic nucleus of posterior recess. A significant decrease in cell proliferation was described in the nucleus of posterior recess in AroB-KO animals compared to WT whereas no differences were observed in ventral pallium and medial pallium. A significant decrease of PCNA expression was also highlighted by qPCR in the whole brain of AroB-KO juveniles compared to WT animals. For the serotonergic neurons, we observed a significative increase of the number of serotonergic neurons in the paraventricular organs of AroB-KO animals compared to WT in juveniles. The effect of cyp19a1b gene deletion on dopaminergic neurons is under analysis.

Our results suggest that an alteration in local E2 production impacts brain proliferation and is associated with a modification of behavior.

A BRB-Seq study is currently under taken to identify genes affected by AroB deletion in juveniles and adults. Quantification of monoamines and their metabolites by HPLC will also improve the knowledge about the link between AroB and behavior. This work was supported by the French National Research Agency (FEATS project; ANR-19-CE34-0005-05).

**Mots-Clés:** aromatase, estradiol, zebrafish, behavior, neurogenesis