Title of Research:

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Development of in vivo fluorescent imaging of neuronal differentiaon in zebrafish for developmental neuotoxicity testing

Principal Investigator:

Yuhei Nishimura (Department of Pharmacogenomics, Mie University Graduate School of Medicine)

Collaborators:

Toshio Tanaka and Reiko Kawase (Department of Pharmacogenomics, Mie University Graduate School of Medicine)

Summary of Research:

The developing brain is extremely sensitive to many chemicals. Exposure to neurotoxicants during development has been implicated in various neuropsychiatric and neurological disorders, including autism spectrum disorder and attention deficit hyperactive disorder. Although rodents have been widely used for developmental neurotoxicity testing, experiments using large numbers of rodents are time-consuming, expensive, and raise ethical concerns. Using alternative non-mammalian animal models may relieve some of these pressures by allowing testing of large numbers of subjects while reducing expenses and minimizing the use of mammalian subjects.

In this study, we have tried to make the three-color zebrafish in which cerulean, a fluorescent protein with maximum excitation (Ex) /emission (Em) at 433/475 nm, would express in neurons, mCitrine, a fluorescent protein with Ex/Em at 516/529 nm, would express in oligodendrocytes, and mCherry, a fluorescent protein with Ex/Em at 587/610 nm, would express in astrocytes. We were able to make one-color zebrafish expressing cerulean in neurons, mCitrine in oligodendrocytes, or mCherry in astrocytes. We were also able to confirm that the zebrafish expressing cerulean in neurons and the zebrafish expressing mCitrine in oligodendrocytes could transmit the phenotype to their offspring. We will cross these two zebrafish to make two-color zebrafish. We will also examine whether zebrafish expressing mCherry in astrocytes could transmit the phenotype to their offspring. Finally, we will cross the two-color zebrafish and the mono-color zebrafish to make three-color zebrafish expression cerulean in neurons, mCitrine in oligodendrocytes, and mCherry in astrocytes.

It has been demonstrated that various developmental neurotoxicants can affect the differentiation of neuron, oligodendrocyte, and astrocytes and that the dysregulated differentiation may cause various neuropsychiatric disorders. The tricolor zebrafish we will develop would make it possible to examine the developmental neurotoxicity of many chemicals and provide a sound basis for human risk assessments.

Timeline:

Nov 2013 ~ Feb 2015

Topics:

Publications:

Zebrafish as a systems toxicology model for developmental neurotoxicity testing.
Nishimura Y, Murakami S, Ashikawa Y, Sasagawa S, Umemoto N, Shimada Y, Tanaka T.
Congenital Anomalies 55(1):1-16 (2015)

In vivo fluorescent imaging of blood-brain barrier disruption in zebrafish using a novel dye.
Nishimura Y, Murakami S, Ashikawa Y, Sasagawa S, Umemoto N, Shimada Y, Tanaka T.
DNT4, Philadelphia, May 2014

Evaluation of developmental neurotoxicity of nicotine using gene knockout in zebrafish
Nishimura Y, Murakami S, Ashikawa Y, Sasagawa S, Kawabata M, Umemoto N, Ariyoshi M, Zhang B, Shimada Y, Tanaka T. The 41st Annual Meeting of Japanese Society of Toxicology, Kobe, July 2014