Title of Research:

20-1-11

Proposal of a new AOP for the neurotoxicity and developmental neurotoxicity assessment of glutamate receptor binding agonists that cause learning and memory impairment.

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Summary of Research:

This study proposes an adverse outcome pathway (AOP) in which the molecular initiating event (MIE) is the binding of compounds to glutamate receptors and the key event (KE) is drebrin loss. The adverse event (AO) is the learning and memory impairment caused by morphological changes in dendritic spines. Drebrin is an actin-binding protein that governs dendritic spine formation of CNS neurons and is responsible for the morphological plasticity of dendritic spines associated with learning and memory. The subcellular localization of drebrin is determined by glutamate receptor activity, and when drebrin is lost, the learning and memory mechanism does not function properly. In this study, we have established an experimental system using frozen hippocampal neurons prepared from rat embryo. We will construct an in vitro method to evaluate compounds that cause learning and memory impairment, and will replace animal experiments for neurotoxicity. We have developed an image processing algorithm for quantitative analysis of neuron count, dendrite length, and drebrin clusters from high-content image data using a confocal image cytometer. In particular, the brightness distribution analysis of drebrin clusters is highly sensitive. We have started to develop a machine learning platform for AI. From the images of immunocytochemical staining, we will clarify the indices for quantitatively evaluating the structural changes of neurons, and provide SOPs for culture techniques and analysis methods. In the future, we are planning to build an experimental system using neurons derived from human iPS cells.

Timeline:

March 1, 2020 - February 28, 2021

Topics:

Oral presentation at JCIA LRI Annual Workshop 2020 “Proposal of a new AOP for the neurotoxicity and developmental neurotoxicity assessment of glutamate receptor binding agonists that cause learning and memory impairment.” (On-line, August 21st, 2020)

Publications:

1. Shogo Mase, Yuko Sekino, Tomoaki Shirao, Toshinari Mitsuoka, “Optimization of algorithms to quantify changes in drebrin distribution induced by glutamate stimulation of cultured neurons for a confocal quantitative image cytometer.” The 63rd Annual Meeting of the Japanese Society for Neurochemistry, Web, September, 2020

2. Toshinari Mitsuoka, Shogo Mase, Noriko Koganezawa, Yuichi Kato, Tomoaki Shirao, Yuko Sekino “Assessment of CB agonist CP55940 in maturity for rat hippocampal neurons using a high-throughput immunocytochemical assay and image digital analysis.” The 94th Annual Meeting of the Japanese Pharmacological Society, Web＋On Site, Sapporo, March 2021.