Title of Research:

17_S05-01

Development of Novel Analytical System of Biological Responses to Chemicals by Deep Phenotyping Method

Principal Investigator:

Hiroyuki Kusuhara (Professor, University of Tokyo, Graduate School of Pharmaceutical Sciences, Department of Molecular Pharmacokinetics)
7-3-1 Hongo, Bunkyo, Tokyo, 113-0033, Japan
(tel) +81-3-5841-4770, (e-mail) kusuhara@mol.f.u-tokyo.ac.jp

Collaborators:

Tadahaya Mizuno (Assistant Professor, University of Tokyo, Graduate School of Pharmaceutical Sciences, Department of Molecular Pharmacokinetics)
7-3-1 Hongo, Bunkyo, Tokyo, 113-0033, Japan
(tel) +81-3-5841-4771, (e-mail) tadahaya@mol.f.u-tokyo.ac.jp

Setsuo Kinoshita (Contract Research Student, University of Tokyo, Graduate School of Pharmaceutical Sciences, Department of Molecular Pharmacokinetics)
7-3-1 Hongo, Bunkyo, Tokyo, 113-0033, Japan

Summary of Research:

Today, organisms are always exposed to chemicals in every situation to some extent. Wide variety of chemicals exist around us and the chemicals affect organisms not one by one but simultaneously like a complex. The fact prompts us to care “multiple exposure”. To achieve the appropriate and correct prediction of multiple exposure of chemicals, it is necessary to comprehend the effects of chemicals even if those are unknown and difficult to be detected. In this study, we tackle with the theme by omic approaches with innovative techniques in both data acquisition and data analysis method.

This study consists of two pillars: (1) a novel acquisition method of proteome profile data by 2-dimensional electrophoresis (2DE) and (2) a new analysis method of profile data, orthogonal linear separation analysis (OLSA). In this 7th term, the last term, we tested the performance of 2DE in detection of chemical features and showed capacity of OLSA to uncover novel aspects of a natural product.

We have carried out 2DE of HeLa cells treated with 6 different anti-cancer drugs under several conditions obtained total 33 proteome profile data. Clustering analysis revealed good separation of each drug group composed of three biological replicates except for Cisplatin and doxorubicin. Of note, combinatorial treatment of a MEK1/2 inhibitor (PD184352) and a microtubule inhibitor (vincristine) constructed a specific cluster and OLSA successfully extracted the combinatorial effect as a specific vector that was not observed in each inhibitor alone.

As for data analysis, we have extended our profile data analysis method to achieve fully unbiased analysis by implementation of parallel analysis. This extension enabled us to estimate the appropriate number of effects to be decomposed, without existing body of biological knowledge. In fact, the basic effects generated by the renewed method were well consistent with gene groups defined in gene ontology, compared with the original one, when Gaussian noise were added to the data. To test the performance of our decomposition analysis approach, Rescinnamine, a natural product was subjected to OLSA. We detected four main effects of the natural product and two of them were actually confirmed in vitro analysis such as luciferase assay and western blotting analysis. All of these data suggest that our platform has the potential to boost deep understanding of combinatorial exposure of chemicals.

Timeline:

March 1, 2019 - February 29, 2020

Topics:

1. Poster presentation at JCIA LRI Annual Workshop 2019 “Development of Novel Analytical

Other issues to be urgently addressed.

System of Biological Responses to Chemicals by Deep Phenotyping Method" (Tokyo, August 30th, 2019)

Publications:

1. Akademi) ICCA-LRI workshop, June 19-20, 2019, Stresa, Italy
2. Journal) Kinoshita S, *Biol Pharm Bull*, 2019