Feasibility study of cyclodextrins as active pharmaceutical ingredients for the treatment of GM1-gangliosidosis

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GM1-gangliosidosis is a rare lysosomal storage disorder characterized clinically by a wide range of variable neurovisceral, ophthalmological and dysmorphic features. Without enough functional β-galactosidase, GM1-gangliosides cannot be degraded in lysosomes, and accumulate to toxic levels in many tissues and organs, particularly in the brain. In spite of several approaches for the treatment of GM1-gangliosidosis, such as enzyme replacement therapy, gene therapy, chemical chaperone therapy, there is no treatment available for patients with ganglioside storage diseases. Therefore, development of novel drugs for GM1-gangliosidosis is needed.

Cyclodextrins (CyDs) are cyclic oligosaccharides that are widely used in the pharmaceutical field. CyDs can extract cell membrane components such as cholesterol and phospholipids from lipid rafts, which contain high concentrations of cholesterol and glycosphingolipids including GM1-gangliosides. Meanwhile, the systemic administration of 2-hydroxypropyl-β-CyD (HP-β-CyD) to mice lacking Niemann–Pick disease type C (NPC) protein was beneficial probably due to the ability to modify the internal environment of the endosomal/lysosomal compartment. Thus, we hypothesized that CyDs are useful for treatment of GM1-gangliosidosis. Therefore, in the present study,
we investigated the effects of various CyDs on the GM1-ganglioside level accumulated in EA1 cells, fibroblasts from patients with GM1-gangliosidosis [1].

We examined the effects of CyDs on the GM1-ganglioside levels in EA1 cells using FITC-labeled cholera toxin B-subunit, which can bind to GM1-gangliosides specifically. Herein, we used the two experimental conditions, i.e. (1) 10 mM CyD and 1 h treatment and (2) 1 mM CyD and 24 h treatment. In our preliminary study, under the former conditions, we confirmed that CyDs extracted membrane components from the EA1 cells. Under the latter conditions, we revealed that CyDs except for M-β-CyD did not extract the membrane components. The treatment with 10 mM CyDs for 1 h did not significantly change fluorescence intensity derived from FITC. Meanwhile, the treatment with 1 mM CyDs for 24 h decreased the fluorescent intensity, especially the prominent lowering effects of methyl-β-CyD and dimethyl-α-CyD were shown with statistical difference, compared to those of the other CyDs. The similar lowering effects of CyDs on the fluorescence intensity derived from Alexa®488-conjugated CTB (Alexa-CTB) were observed in EA1 cells using a confocal laser scanning microscopy. Hence, the lowering effect of CyDs on the GM1-ganglioside levels in the cells was highly unlikely to be associated with the extracting ability of CyDs on plasma membrane components of the cells. Collectively, these results suggest that CyDs impair the GM1-ganglioside levels in EA1 cells in a different way from the extraction ability of CyDs on membrane components.

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REFERENCE