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学位論文の題名	Protective effect of Irsogladine against aspirin-induced mucosal injury in human induced pluripotent stem cell-derived small intestine （ヒト人工多能性幹細胞由来小腸におけるアスピリン誘発粘膜傷害に対するイルソグラジンの保護効果） Medicina. 2022 ;59(1):92.
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Abstract

Background: Acetylsalicylic acid (ASA) is widely used for preventing cerebrovascular and cardiovascular diseases. Low-dose-aspirin (LDA) exerts antithrombotic effects by suppressing cyclooxygenase-1 activity, which leads to decreased prostaglandin (PG) production. As PGs play an important role in maintenance of the gastrointestinal (GI) epithelium by upregulating mucosal blood flow, a decrease in PG synthesis can cause GI mucosal damage Gastrointestinal (GI) tract injury is one of the major complications of aspirin use, potentially leading to severe GI bleeding. However, no drugs for preventing aspirin-induced small intestinal injury have been developed.

Objectives: The aim of this study was to establish a human experimental model for investigating aspirin-induced small intestinal mucosal injury. In addition, we evaluated the protective effect of Irsogladine against aspirin-induced small intestinal mucosal injury using human induced pluripotent stem cell-derived 2D monolayer crypt-villus structural small intestine (2D-hiPSC-SI).

Materials and Methods: Human iPS cell-derived intestinal organoids were seeded and cultured in Air-liquid interface. The permeability of 2D-hiPSC-SI was evaluated using Lucifer yellow. Changes in structure and mucosal permeability of 2D-hiPSC-SI after addition of aspirin were confirmed over time, and changes in intestinal epithelium-related markers were evaluated by real-time qPCR and Immunofluorescence staining. The effect of Irsogladine on prevention of aspirin mucosal injury was examined by adding Irsogladine to the culture medium.

Results: Cultured 2D-hiPSC-SI showed multi-lineage differentiation into small intestinal epithelium comprised of absorptive cells, goblet cells, enteroendocrine cells, and Paneth cells, which express CD10, MUC2, chromogranin A, and lysozyme, respectively. RNA in situ hybridization revealed intestinal stem cells that express Lgr5. ASA administration induced an increase in the mucosal permeability of 2D-hiPSC-SI. ASA-injured 2D-hiPSC-SI showed decreased mRNA expression of multi-lineage small intestinal cell markers as well as intestinal stem cell marker Lgr5. Administration of Irsogladine on the basal side of the 2D-hiPSC-SI resulted in significant increases in Mki67 and Muc2 mRNA expression by 2D-hiPSCs at 48 h compared with the control group. Administration of 400 μg/mL Irsogladine to the ASA-induced small intestinal injury model resulting in significantly decreased mucosal permeability of 2D-hiPSC-SI. In immunofluorescence staining, Irsogladine significantly increased the fluorescence intensity of MUC2 under normal conditions and administration of 400 μg/mL ASA.

Conclusions: we established a novel ASA-induced small intestinal injury model using human iPSC-derived small intestine. Irsogladine maintains mucosal permeability and goblet cell differentiation against ASA-induced small intestinal injury.