ABSTRACT

Principal Investigator: Taiho Kambe

Grant Title: Inhibition of cancer cell growth by suppressing Golgi mannosidase II activation

Purpose

Golgi mannosidase II (GMII) plays a crucial role in distinguishing hybrid-type from complex-type N-linked glycans, which are implicated in cancer development and abnormal cell proliferation. As a result, GM II has been a long-standing target for cancer therapy. We recently found that GMII is a zinc-dependent enzyme activated by zinc transported via zinc transporters, ZNT5 and ZNT7, which localize in the early secretory compartments. GMII remains inactive unless both ZNTs function properly.

In this study, we aim to demonstrate that the reduction of GMII activity due to the loss of ZNT5 and ZNT7 functions is effective in suppressing cancer cell growth using a nude mouse xenograft model. Furthermore, considering the current situation where GMII inhibitors have not been clinically applied due to their side effects impairing lysosomal mannosidase (LAMAN) functions, we aim to prove that the loss of ZNT5 and ZNT7 functions selectively decreases GMII activity without impairing LAMAN activity. These results suggest the potential of targeting ZNT5 and ZNT7 as a novel strategy for cancer therapy.

Methods

MIA-*Z5Z7*-DKO cells were generated using the CRISPR/Cas9 system. After confirming that their growth in culture was comparable to WT cells, both cell types were subcutaneously inoculated into the right or left shoulder areas of nude mice. For GMII activity measurement, the substrate solution was prepared at pH 7.5 (10 mM Tris-HCl, 0.5 mM $MgCl_2$, and 0.1% Triton X-100). For LAMAN activity measurement, a separate substrate solution was prepared at pH 4.0 (100 mM CH_3COOH).

Results

To assess the effects of loss of ZNT5 and ZNT7 on pancreatic cancer cell growth, we used a nude mouse xenograft model. MIA-*Z5Z7*-DKO cells exhibited significantly reduced growth over four weeks compared to that of WT cells. Consistently, tumors excised after four weeks from mice inoculated with MIA-*Z5Z7*-DKO cells weighed significantly less than those from WT MIA PaCa-2-injected mice. Furthermore, we confirmed that GMII activity was substantially decreased in *Z5Z7*-DKO cells, while the activity of its homolog, lysosomal mannosidase (LAMAN), remained unchanged. These findings indicate that functional impairment of ZNT5 and ZNT7 may contribute to the suppression of pancreatic cancer progression.