Principal Investigator: Hye-Won Shin

Grant Title: Roles of glucosylceramide-flippase in glycosphingolipid metabolism

Abstract

Lipid transport is an essential cellular process with importance to human health, disease development, and therapeutic strategies. Type IV P-type ATPases (P4-ATPases) have been identified as membrane lipid flippases by utilizing nitrobenzoxadiazole (NBD)-labeled lipids as substrates. Among the 14 human P4-ATPases, ATP10D was shown to flip NBD-glucosylceramide (GlcCer) across the plasma membrane. Here, we found that conversion of incorporated GlcCer (d18:1/12:0) to other sphingolipids is accelerated in cells exogenously expressing ATP10D but not its ATPase-deficient mutant. These findings suggest that 1) ATP10D flips unmodified GlcCer as well as NBD-GlcCer at the plasma membrane and 2) ATP10D can translocate extracellular GlcCer which is subsequently converted to other metabolites. Notably, exogenous expression of ATP10D led to the reduction in cellular hexosylceramide (HexCer) levels. Moreover, the expression of GlcCer flippases also reduced cellular HexCer levels in fibroblasts derived from patients with Gaucher disease (GD), which is a lysosomal storage disorder with excess GlcCer accumulation. It is noteworthy that the cellular distribution of lysosomes and lysosomal proteolytic activity remained largely unaffected in GD fibroblasts. However, we found that lysosomal membranes of GD fibroblasts were susceptible to damage when exposed to a lysosomotropic agent. Our study highlights the contribution of ATP10D to the regulation of cellular GlcCer levels and maintaining lipid homeostasis.

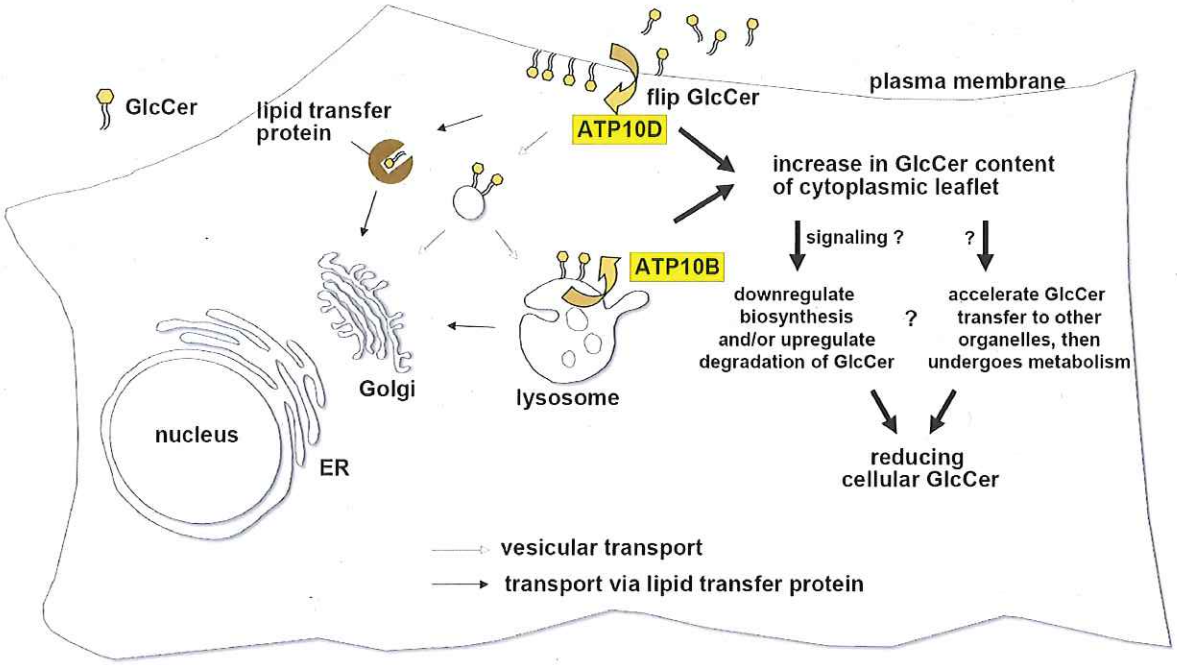

Figure. Schematic model of a decrease in the cellular GlcCer levels in cells expressing GlcCer flippases

1. Kita, N., Hamamoto, A., B.Gowda, S.G., Takatsu, H., Nakayama, K., Arita, M., Hui, S.P., and *Shin, H.-W. (2024) Glucosylceramide flippases contribute to cellular glucosylceramide homeostasis. ***J. Lipid Res. 65,*** 100508. DOI: 10.1016/j.jlr.2024.100508.

2. Hamamoto, A., Kita, N., B.Gowda, S.G., Takatsu, H., Nakayama, K., Arita, M., Hui, S.P., and *Shin, H.-W. (2024) Lysosomal membrane integrity in fibroblasts derived from patients with Gaucher disease. ***Cell Struct. Funct.*** **49,** 1-10. DOI: 10.1247/csf.23066.