Submitted by: Dr. Rebecca Ng Mei Li
Date: 29 September 2017

Role of Sphingosine Kinase in Insulin Signalling and Its Action in Hepatocytes

Mei Li NG1, 2, Pu Xia2
1 Advanced Medical and Dental Institute (AMDI), University of Science (USM), Malaysia
2 Centenary Institute, Sydney Medical School, University of Sydney, Australia

Abstract (300 word limit)

Background and aims: Sphingosine kinase (SK) is a key enzyme involves in the homeostasis of sphingolipids metabolism by catalyzing phosphorylation of sphingosine to sphingosine 1-phosphate (S1P). Increased level of sphingolipids are associated with insulin resistance. In this study, we aimed to examine the potential role of SK in insulin signalling and its metabolic actions in hepatocytes.

Methodology: A human hepatocarcinoma cell line (Huh7), a non-neoplastic human hepatic cell line (PH5CH8) and freshly-isolated mouse primary hepatocytes from wild-type (WT) or SK1 knockout (SK1-/-) were used as experimental models. SK expression and activity were manipulated by gene overexpression of WT or dominant-negative SK, siRNA-mediated knockdown, and using a specific chemical inhibitor, SKI-II.

Findings: Treatment of hepatocytes with insulin resulted in two-folds increase in SK enzyme activity. Inhibition of SK activity by overexpressing a dominant-negative mutant (SKG82D) or the enzyme inhibitor SKI-II significantly attenuated insulin-induced Akt activation, while overexpression of WT-SK markedly enhanced Akt activation. Inhibition of SK activity markedly inhibited the association of IRS-1 with p85 regulatory subunit of PI3-K, suggesting a molecular target for the action of SK on insulin signaling. Interestingly, we revealed that SK2 was predominantly insoform for insulin-induced Akt activation in hepatocytes. Insulin-induced Akt activation and phosphorylation of two important Akt downstream targets, GSK3α and 70S6K were significantly attenuated in the hepatocytes where SK2 was knocked-down by its specific siRNA. Insulin-induced suppression of phosphoenolpyruvate carboxykinase (PEPCK) gene, a rate-limiting enzyme in gluconeogenesis, was abolished by knockdown of SK2 expression. In contrast, the siRNA-mediated knockdown of SK1 expression had no significant effects on insulin-induced Akt activation and PEPCK suppression, indicating an isoform-specific effect of SK2 on insulin action in hepatocytes.

Conclusion: Our data suggest a potential role of SK2 in regulation of insulin signaling and glucose metabolic action in hepatocytes.

(291 words)

Image

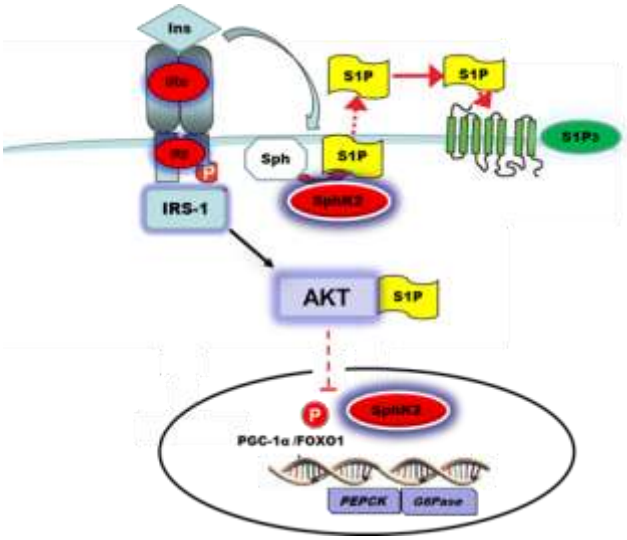


Figure 1. Proposed model molecular mechanism of SphK2 action to enhance insulin-mediated glucose metabolic actions in the hepatocytes.

Recent Publications (minimum 5)

Peer-reviewed journals

1. **M. L. Ng**, Wadham, C., & Sukocheva, O. A. (2017). The role of sphingolipid signalling in diabetes-associated pathologies (Review). International Journal of Molecular Medicine, 39(2), 243–252.
2. Lovelace, Michael D., Varney, Bianca,Sundaram, Gayathri, Franco, Nunzio F. **M. L. Ng**, Pai, Saparna, Lim, Chai K., Guillemin, Gilles J., Brew, Bruce J. (2016). Current evidence for a role of the Kynurenine pathway of tryptophan metabolism in Multiple Sclerosis. Frontiers in Immunology. 7: p. 246.
3. **M. L. Ng**, Lim SC, Liu JJ, Tai ES, Dalan R and Boehm BO (2015). Latent autoimmune diabetesof the adult (LADA) in Singapore: Study on the islet cell autoimmunity. J Clin Cell Immunol, 6:5.
4. Ong, Y. H., W. C. A. Koh, **M. L. Ng**, Z. Y. Tam, S. C. Lim and B. O. Boehm (2017). Glutamic acid decarboxylase and islet antigen 2 antibody profiles in people with adult-onset diabetes mellitus: a comparison between mixed ethnic populations in Singapore and Germany." Diabet Med 34(8): 1145-1153.
5. **M. L. Ng**, Lim SC, Liu JJ, Tai ES, Boehm BO. (2014). Epitope mapping of polyclonal autoantibodies to zinc transporter 8 (znt8) and glutamic acid decarboxylase (gad65) by peptide-based microarray. Diabetes Research and Clinical Practice 106S1 (2014) S47–S267. Poster presented at the 10th IDF-WPR Congress and 6th AASD Scientific Meeting, Singapore. (conference proceedings)

Dr. Ng Mei Li has keen interested in finding novel therapeutic target for diabetes mellitus. This work revealed for the first time, the potential role of SphK as novel therapeutic target for hyperglycemia in diabetes mellitus. She is looking for research collaboration with scientist from multidisciplinary background in the area of diabetes and metabolic disease research. Email: nmeili@usm.my