00455 A PDZ Domain-containing Protein, GIPC3, Positively Regulates Hedgehog Signalling and Shows Potential as a Therapeutic Target for Melanoma

Sathya Narayanan Patmanathan, Woo Wei Meng, Ting Yong Zheng Jonathan

Nanyang Technological University

Aims: Hedgehog (Hh) signalling activation is evident in many cancers. In this study, we investigated the role of a putative adaptor protein GIPC3 in Hh signalling and explored the potential of targeting this protein in Hh signaling-associated cancers.

Methodology: We used lentivirus-based shRNA technique for gene knockdown and QRT-PCR to measure gene expression. Pathways association of GIPC3-interacting proteins was determined using Ingenuity® Pathway Analysis (IPA). The Cancer Genome Atlas (TCGA) was utilized to identify expression and association of GIPC3 and Hh pathway components in cancers.

Result: Using shRNA knockdown in primary and established mouse fibroblasts, we showed that Gipc3 is required for Hh signaling activation, suggesting Gipc3 is a novel positive regulator of the Hh pathway. We then identified Gipc3-interacting proteins and analyzed these proteins using IPA. We found that over 90% of GIPC3's interactors are associated with cancer pathogenesis. To explore association of GIPC3 with cancers, we assessed TCGA and revealed upregulation of GIPC3 mRNA in several Hh pathway-associated cancers. We thus examined GIPC3 expresion in a number of Hh pathwayassociated cancer cell lines and demonstrated that GIPC3 expression was increased in the majority of melanoma (N=7) and head and neck (HNSCC) cancer (N=15) lines. We further revealed strong positive correlations (Spearman $r>0.8$; $p<0.01$) between the expression of GIPC3 and Hh pathway components GLI1, GLI2 and GPR161, in the melanoma lines, but not in HNSCCs. Gipc3 knockdown in mouse melanoma line, B16F10, resulted in a decrease of Gli1 expression, suggesting Gipc3 positively regulates Hh signaling in melanoma.

Conclusion: Our findings suggest a novel role of GIPC3 in melanoma pathogenesis via regulating the Hh pathway. Given the reported contribution of both canonical and non-canonical Hh signalling to melanoma progression, unraveling the mechanism of how GIPC3 regulates the Hh pathway during melanoma progression may shed light on new therapeutics for melanoma.