Regulating Cancerous Hyperactive Wnt Signalling Through a Novel Gid8 Mechanism in Drosophila and Human Embryonic Kidney Cells

Shah, Ashka (School: Jericho High School)

Hyperactive Wnt signalling causes over 90% of colorectal and 50% of breast cancers. It is driven by excessive nuclear translocation of mutant ß-catenin (ArmS10 in Drosophila), yet the exact translocation mechanism is unknown. Gid8, a ubiquitin ligase known to promote normal Wnt signalling, has never been tested in hyperactive, cancerous Wnt signalling. This study aimed to investigate Gid8's role in hyperactive Wnt signalling and pinpoint functional domains that mediate its involvement. GAL4/UAS-edited Drosophila expressing ArmS10 with/without Gid8 knockdown were analyzed. Gid8 knockdown suppressed hyperactive Wnt activity, rescuing cancerous phenotypes ($p<0.0001$), revealing Gid8 as a novel regulator of oncogenic Wnt signalling. Fluorescent imaging revealed that Gid8 colocalized with Kinesin-2/IFT140 ($p<0.0001$), transport proteins that guide wild-type ß-catenin, suggesting Gid8 is involved in nuclear transport. This uncovers a previously unrecognized mechanism that may explain how ArmS10's nuclear entry works. Supporting this, co-immunoprecipitation assays confirmed Gid8/ArmS10 binding ($p<0.01$), providing the first biochemical evidence that Gid8 facilitates nuclear translocation in hyperactive Wnt signalling. To make these findings more specific, human embryonic kidney cells were transfected with wild-type or mutated Gid8. Only wild-type Gid8 localized to the nucleus, while a C-terminal mutation prevented localization ($p<0.0001$). This confirms Gid8's C-terminal's essential role in Wnt activity and highlights it as a target to disrupt hyperactive signalling. This research uncovers Gid8 and its C-terminal as novel and highly specific regulators of hyperactive Wnt signalling, offering a targeted entry point to disrupt Wnt-driven cancers where current therapies have failed.

Awards Won: