

P028 The Role of Kinesin Light Chains in Insulin Granule Movement

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Background. The recruitment of insulin-containing large dense core vesicles (LDCVs) along microtubules to the plasma membrane is essential for glucose-stimulated insulin secretion from pancreatic beta cells. Previous studies have suggested this process is dependent on kinesin-1 (Kif5B). Kinesin consists of two heavy (KHC) and two light chains (KLC), and uses energy derived from ATP hydrolysis to transport cargoes. Earlier studies revealed that AMPK activation inhibits both basal and glucose-stimulated insulin vesicle movement. Both KHC and KLC1 have consensus AMPK phosphorylation sites. Results. Eight KLC splice variants, all containing a consensus AMPK phosphorylation site at Ser-521 have been described. Conventional PCR of MIN6 cell- or isolated mouse islet-derived RNA revealed the presence of KLC isoforms 1a, 1b, 1c and 1f and Real Time PCR (Sybergreen™) showed that the level of KLC1b and 1c expression was <2%, and KLC1f ~50%, of that of KLC1a. A peptide corresponding to the sequence around Ser-521 was efficiently phosphorylated by AMPK *in vitro*. Conclusion. LDCV trafficking in beta cells is likely to involve KLC-1a or -1f, and may be regulated by AMPK-mediated phosphorylation. A KLC1 phospho-Ser520 specific antibody is being used to determine whether KLC1 isoforms are phosphorylated in living beta cells in response to glucose deprivation and AMPK activation.