ADENOSINE A_{2A} RECEPTOR AGONIST ATL146e IMPROVES ISLET TRANSPLANTATION OUTCOMES IN MICE.

Davidson AB, Montoya JT, Zeng Q, Johnson A, Banerjee K, and Thompson GS
Department of Surgery, Burrell College of Osteopathic Medicine, Las Cruces, NM.

Introduction: Peri-transplant inflammatory events limit the success of clinical islet transplantation. The effect of the A_{2A} adenosine receptor (A_{2A}AR) agonist ATL146e on *in vitro* islet function and functional islet graft survival was investigated.

Methods: Islets were isolated from C57BL/6 recipient mice and underwent: 1) glucose-stimulated insulin secretion (GSIS), with or without ATL146e (100nM) in 2hr static incubations using 50 islets/well; or 2) syngeneic islet transplantation (50 islets, renal subcapsular site) in diabetic (IP streptozotocin, 18mg/kg) C57BL/6 recipient mice. In transplant recipients ATL146e (60 ng/kg/min) was administered by mini-osmotic pump beginning 2 days pre-transplant.

Results: Glucose-stimulated insulin secretion from ATL-treated islets demonstrated a stimulation index (SI) significantly less than that observed for vehicle-treated islets (3.75±0.45 for islets alone vs. 2.36±0.22 for islets + ATL, $p<0.01$**)**. Decreased SIs in ATL146e-treated islets resulted from increased (1.79±0.20, n=9, $p<0.05$) basal insulin secretion. Islets co-cultured with neutrophils demonstrated a significant reduction in glucose-stimulated insulin secretion, however, culture with ATL146e abrogated this effect (SI=1.41±0.17 for islets + neutrophils vs. 2.57±0.18 for + with neutrophils + ATL146e, $p<0.05$). Addition of an ATL146e antagonist (SCH58261) reverse ATL146e's effect on islet/neutrophil co-cultures (SI=2.59±0.31 for islets + neutrophils + ATL146e vs. 1.11±0.42 for islets + neutrophils + ATL146e + SCH58261, $p<0.05$). Further, islets co-cultured with A_{2A}AR knock-out neutrophils showed no improvement in GSIS upon addition of ATL146e (SI=1.24±0.40 for islets + KO neutrophils vs. 0.90±0.10 for islets + KO neutrophils + ATL146e, $p<0.05$). *In vivo*, diabetic mice receiving vehicle or ATL146e alone did not achieve normoglycemia (n=5,8; >30days). Recipients grafted with 50 syngeneic islets (n=10) achieved normoglycemia within 30 days (20.6±3.5 days). ATL146e therapy enhanced the return to normoglycemia (9.9±2.9 days, n=7, $p<0.031$). Transplantation of syngeneic islets to C57BL/6 A_{2A}AR-KO recipients treated with ATL146e showed no improvement in functional graft success over controls.

Conclusions: *In vitro* studies show that ATL146e abrogates the deleterious effects of neutrophils on GSIS, and that this effect is mediated by specific activation of A_{2A}ARs on neutrophils. Additionally, ATL146e administration *in vivo* improves islet transplant outcome, with reduced grafted islet mass requirement, and shorter time to hyperglycemic reversal.