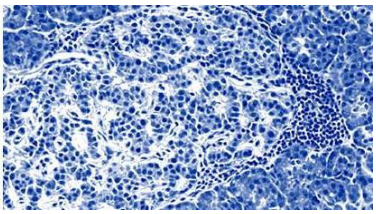
nPOD
Network for Pancreatic Organ Donors with Diabetes



Beta Cell Autophagy Is Reduced in Type 1 Diabetes

AUTHORS

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PURPOSE

Autophagy is a dynamic recycling mechanism that maintains cellular homeostasis, alleviates cellular stress, and generates energy by recycling damaged macromolecules and organelles. Crinophagy is a specific type of autophagy where secretory granules directly fuse with lysosomes for degradation. Impairment of autophagy and crinophagy have been implicated in beta cell death and dysfunction and this has been demonstrated in the context of type 2 diabetes. However, there is currently no literature describing beta cell autophagy in type 1 diabetes. Our study is therefore aimed at analyzing autophagy as a function of type 1 diabetes pathogenesis.

METHODS

We utilized immunofluorescent (IF) staining of LC3 (autophagosome marker), Lamp1 (lysosome marker), proinsulin (beta cell marker), and DAPI (nuclear marker) to analyze islet autophagy. We first analyzed paraffin embedded pancreas tissue from NOR (non-obese resistant- doesn't develop diabetes) and diabetic NOD (non-obese diabetic- develops spontaneous diabetes). We then performed IF on paraffin embedded pancreas tissue obtained from nPOD, collected from non-diabetic, autoantibody positive and type 1 diabetic human organ donors.

SUMMARY OF RESULTS

In the diabetic NOD mouse islets, we observe a significant decrease in the total number of lysosomes ($p=0.0129$), with no change in autophagosome numbers when compared to NOR control pancreas. We also observe a trend of decreased colocalization of autophagosomes with lysosomes, suggesting modestly reduced macroautophagy in islets of diabetic NOD mice. However, we observe significantly reduced colocalization of proinsulin with lysosomes in the islets of diabetic NOD mice compared to the NOR controls ($p=0.0121$), indicating reduction in a specific type of autophagy known as crinophagy. Interestingly, qualitative analysis of islets of diabetic NOD mice shows less granulation and more diffused proinsulin staining when compared to the NOR mice controls. Analysis of human pancreatic tissue sections showed no difference in autophagosome or lysosome numbers. Interestingly, however, both macroautophagy ($p=0.044$) and crinophagy ($p=0.0018$) were significantly

reduced in residual proinsulin positive cells of pancreas from type 1 diabetic individuals compared to both the nondiabetic and autoantibody positive individuals.

CONCLUSIONS

This study provides evidence that macroautophagy and crinophagy are both significantly reduced in the context of type 1 diabetes. These data are the first demonstration of reduced autophagy in human islets from type 1 diabetic organ donors. Given the static nature of our observations, further studies should be performed to address how and when autophagy is reduced during the pathogenesis of type 1 diabetes, and to determine the functional effects of autophagy reduction in the context of disease initiation.