Figure 9. VEGF-A expression is significantly reduced in PdxPBcreER;VEGFfl/fl islets after administration of 3 doses of 8mg tamoxifen. Islets were isolated from adult PdxPBcreER;VEGFfl/fl and VEGFfl/fl mice (n=4 mice) at the indicated time points before and after tamoxifen treatment. Isolated islets were cultured for 48 hours at 37°C in aliquots of 70 islets per 500ul of media. Islets and culture media (450 ul) were subsequently collected and stored at -80°C. VEGF-A levels in the culture media samples were then measured using an VEGF-A ELISA (n=3 wells). ** p-value < 0.0001 by t-test
Figure 10. Decreased VEGF-A expression reduces vascular density in Pdx^{PB}\text{Cre}^{ER};\text{VEGF}^{fl/fl} islets. (A) Mice were infused with tomato-lectin to label the functional vasculature. Sections of the pancreas from Pdx^{PB}\text{Cre}^{ER};\text{VEGF}^{fl/fl} mice were imaged to assess the lectin labeled vessels at the indicated time points. Phase contrast was used to determine the islet perimeters. All images are at 20X. (B) The vascular density of VEGF^{fl/fl} islets (n=15) was unchanged 1 week (wk) and 1 month (mo) after tamoxifen. However, the vascular density of tm treated Pdx^{PB}\text{Cre}^{ER};\text{VEGF}^{fl/fl} islets was decreased 1mo post tam (n=20) when compared to –tm islets (n=12), there was no significant change in islets 1 wk after tm (n=31). (C) Measurements of area per vessel of Pdx^{PB}\text{Cre}^{ER};\text{VEGF}^{fl/fl} islets showed a significant decrease at 1wk (n=32) and 1mo (n=25) post tm when compared to islets from mice not treated with tamoxifen (n=20). No change was observed in VEGF^{fl/fl} islets (n=30). **p-value= 0.0031 and ***p-value<0.0001
Figure 11. Glucose Tolerance changes in Pdx<sup>PB</sup>Cre<sup>ER</sup>;VEGF<sup>fl/fl</sup> mice post tamoxifen treatment. (A) All Pdx<sup>PB</sup>Cre<sup>ER</sup>;VEGF<sup>fl/fl</sup> (○) and VEGF<sup>fl/fl</sup> (■) mice underwent glucose tolerance testing (GTT) prior to tamoxifen injection. (B) One week after 3X8mg of tamoxifen, both Pdx<sup>PB</sup>Cre<sup>ER</sup>;VEGF<sup>fl/fl</sup> (○) and VEGF<sup>fl/fl</sup> (■) mice, underwent another GTT. (C) There was a significant decrease in glucose clearance in the Pdx<sup>PB</sup>Cre<sup>ER</sup>;VEGF<sup>fl/fl</sup> mice after receiving tamoxifen (red) when compared to their GTTs before tamoxifen (black). (D) This was still observed with the GTTs of male (▲) and female (□) mice were separated and compared. **p-value= 0.003 by unpaired t-test.
Figure 12. Summary of Glucose Tolerance tests in Pdx$^{PB\text{Cre}}_{ER}\text{;VEGF}^{fl/fl}$. Glucose Tolerance Tests in Pdx$^{PB\text{Cre}}_{ER}\text{;VEGF}^{fl/fl}$ Mice Return to ‘Normal’ 1 Month Post Tamoxifen Treatment This graph shows the GTT results for three Pdx$^{PB\text{Cre}}_{ER}\text{;VEGF}^{fl/fl}$ mice that were tested at all three time points. Prior to tamoxifen treatment these mice had normal glucose clearance (-■-). However, at 1 week post tamoxifen treatment their GTTs results showed a delay in glucose clearance (-▲-). This delay in glucose clearance was no longer observed in the tamoxifen treated Pdx$^{PB\text{Cre}}_{ER}\text{;VEGF}^{fl/fl}$ mice at 1 month (-▼-) ***p-value= 0.003 by unpaired t-test.