Effects of MSDC-0602 and GLP1 on Pancreatic Islets in ZDSD/Pco Rats Challenged with a High Fat Diet

Jerry R. Colca[1], Heitor F. Ferrari[2], Charles D. Mackenzie[2], Rob R.Eversole[3], Troy Gobbett[4], and Richard Peterson[4].
[1]Metabolic Solutions Development Company, Kalamazoo, MI; [2]Michigan State University; [3]Western Michigan University; [4]PreClinOmics, Indianapolis, IN, USA.

ABSTRACT:

MSDC-0602 is a new insulin sensitizing agent designed to act on the mitochondrial target, mTOT. We have previously shown MSDC-0602 lowered circulating glucose and insulin in a phase 2 clinical trial. Here we asked whether MSDC-0602 would have beneficial effects on the pancreatic islets in the ZDSD/Pco rat, a model selected for beta cell dysfunction in response to a high fat diet. Since this model responds to GLP1, we also evaluated the combination of GLP1 treatment with this mTOT modulator. Five matched groups of ZDSD/Pco rats were selected at 14 weeks of age and then treated for 6 weeks. Group 1 was maintained on the standard chow diet while the other 4 groups were switched to the 5SCA (Purina) high fat diet and then given either oral vehicle and saline injection (group 2), oral MSDC-0602 (30 mg/kg) and saline injection (group 3), oral vehicle and exenatide (s.q. 1 μg) (group 4), or oral MSDC-0602 (30 mg/kg) and exenatide (s.q. 1 μg) (group 5). The change in diet caused glucose levels to progressively rise to over 500 mg/dl in the control treated rats (group 2). Either treatment alone significantly reduced glucose levels over 4 week treatment as compared to the group 2 (control treatment on the high fat diet), but the combination of MSDC-0602 and exenatide prevented any rise in glucose. At sacrifice, the pancreas was removed, fixed, and stained for insulin, PDX1, and Ki67, as an index of proliferation. Both treatments increased the number of islet cells and insulin staining, but the largest increase was group 5 where both treatments were combined. Exenatide increased the number of Ki67 positive cells and the combination with MSDC-0602 increased this further. Interestingly, however, individual cells were either positive for Ki67 or insulin and/or PDX1 but not both, indicating that the individual islet cells were at different stages of differentiation.. These data suggest that the combination of an mTOT modulator with GLP1 therapy may be useful for the restoration of pancreatic islets in diabetes..

INTRODUCTION:

Recently, a new class of insulin sensitizers has been identified , which modify a newly identified mitochondrial target thus avoiding the side effects associated with direct activation of nuclear receptors (1-3). This mechanism appears to involve alteration in nutrient sensing pathways that favor the differentiated state of the beta cells, a least in vitro (4).

The ZDSD/Pco rat is a new rat model of type 2 diabetes that undergoes beta cell dysfunction in response to a high fat diet (5). As such, these rats are a model for a key feature of type 2 diabetes that involves a progressive loss of beta cell function that appears to involve both beta cell death and loss of differentiated phenotype.

Recent clinical evidence suggests a potential powerful synergy between first generation insulin sensitizers and GLP1 in the clinic when given as an early treatment (6). Here we have asked whether the combination of GLP1 treatment with an mTOT modulator currently in clinical trials might function to prevent loss of functional beta cells in the ZDSD/Pco rat.

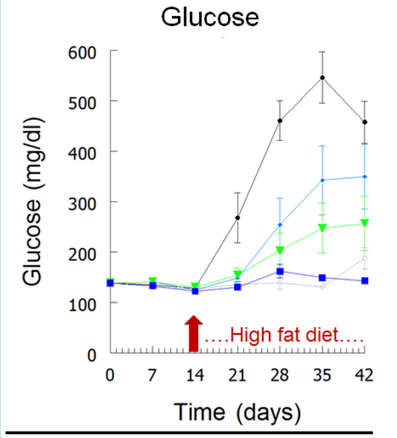
METHODS:

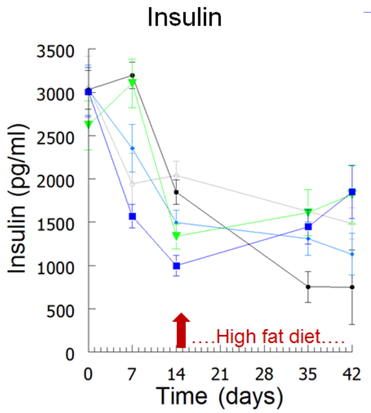
Male ZDSD/Pco rats were maintained on standard chow 5008 (Purina) until treatment groups were switched to the high fat diet (Purina 5SCA). Treated animals were given a single oral dose of vehicle (1% SCMC/0.1% Tween 80) or MSDC-0602 (15 mg/kg) and a 0.1 ml subcutaneous injection of vehicle or 1 μg exenatide. Control rats (given both vehicles) were maintained on the normal diet and the treatment groups were switched to the high fat diet as indicated. Glucose and insulin were measured weekly from tail vein samples. On day 36 an oral glucose tolerance test (OGTT); 2 g/kg) was conducted after a 16 hour fast. Glucose samples were taken at 0,30,60, 90, and 120 minutes; insulin was measured at 0 and 30 minutes and the change at 30 minutes is shown. Data are mean and SE; N=10. At sacrifice, the pancreas was fixed in formalin and shipped to MSU for histology and IHC. A range of anti-bodies were used to identify and quantify the presence of insulin, Ki67, collagen, CD3 lymphocytes, PDX-1 and Bcl2. An Image –Pro system was used with Image J to quantify the degree of positive staining.

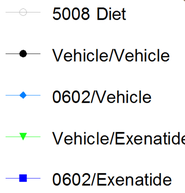
REFERENCES

1. Colca, et al., (2013) Identification of a Mitochondrial Target of Thiazolidinedione Insulin Sensitizers (mTOT)-relationship to newly identified mitochondrial pyruvate carrier proteins. PLOS ONE 8:e61551.
2. Colca, et al., Clinical proof of concept with MSDC-0160, a prototype mTOT modulating insulin sensitizer. Clinical Pharmacology and Therapeutics. 93 352-359
3. Chen, Z. (2012) Insulin Resistance and Metabolic Derangements in Obese Mice Are Ameliorated by a Novel Peroxisome Proliferator-activated Receptor -sparing Thiazolidinedione J. Biol. Chem., 287: 23537-23548.
4. Rohatgi, et al., (2013) Novel Insulin Sensitizer Modulates Nutrient Sensitizing Pathways and Maintains b-cell Phenotype in Human Islets. PLOS ONE :8 e62012-1-13.
5. Davis, et al., (2013) Age-Related Differences in Response to High-Fat Feeding on Adipose Tissue and Metabolic Profile in ZDSD Rats. Obesity 2013, http://dx.doi.org/10.1155/2013/584547.
6. Abdul-Ghani, et al., this meeting 72-OR and 384-OR.

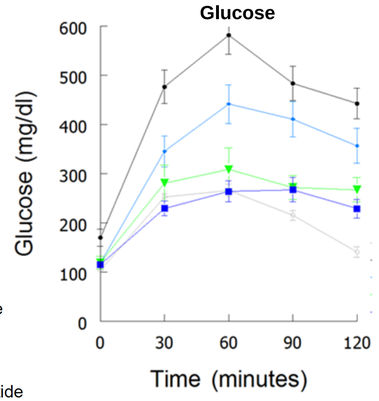
Effect on Glucose and Insulin

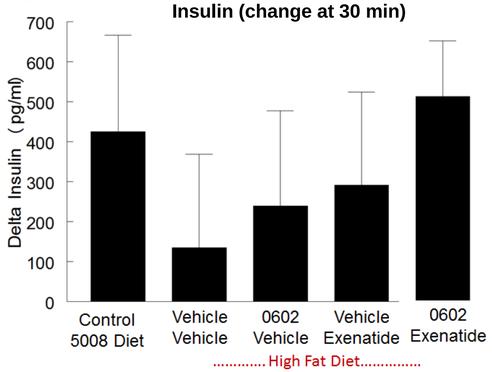






Oral Glucose Tolerance

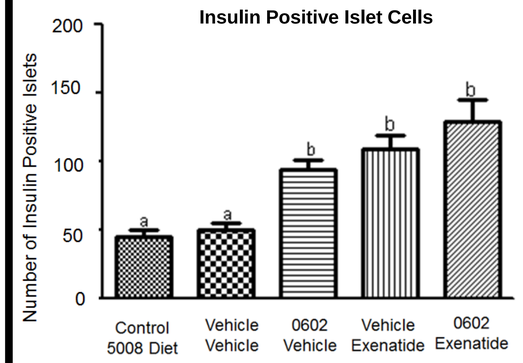


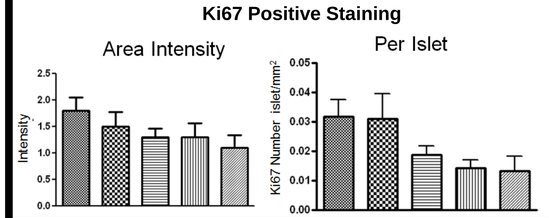


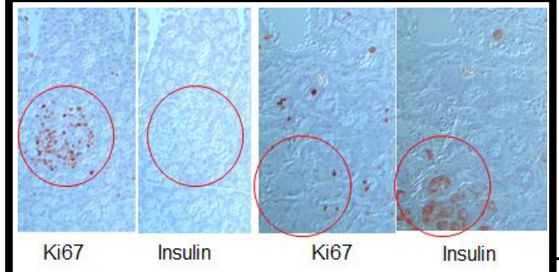
There is an apparent synergism between GLP1 and MSDC-0602 on

- Circulating glucose and insulin levels
- Oral glucose tolerance test (OGTT)

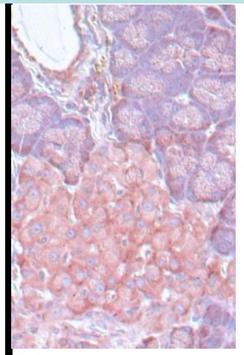
Effects on Pancreatic Islets



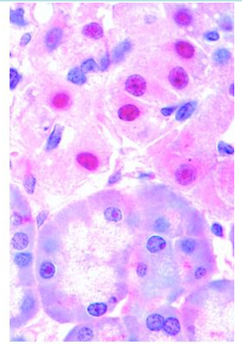




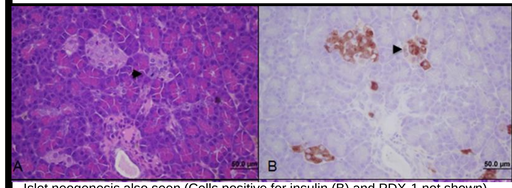
- Islets positive for Ki67 are usually negative for insulin
- Supports work in human islets showing similar phenomenon (4)
- Synergistic increase with MSDC-0602 and GLP1



Bcl2 staining in an islet and ductal epithelium of 0602+exenatide group

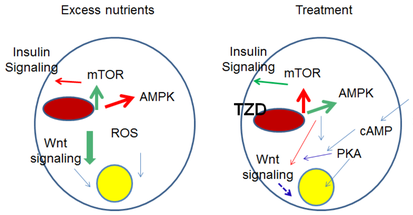


PDX-1 staining in an islet of 0602+exenatide group



Islet neogenesis also seen (Cells positive for insulin (B) and PDX-1 not shown)

Developing Hypothesis



Excess nutrients inhibit AMPK and activate mTOR favoring loss of beta cell phenotype/functional beta-cells. Metabolic inflammatory signals also may trigger cell death pathways.

mTOT modulation by 0602 together with GLP1 action elicit signals favoring maintenance of beta cell phenotype. [see also Rohatghi, N., et al. (4); Aly, H., et al., PLOS ONE, *in press*, 6/12/13.]

Conclusion

These results suggest that there could be a therapeutic value to combining mTOT modulators with GLP1 agonists to preserve functional beta cells.