composition of GnomAD and clinical trial subjects with *LMNA* mutations were significantly different (p=0.024) with the clinical trial cohort being more enriched for white patients (78 vs 21%) and less enriched for Latino patients (7 vs. 21%). There were no differences for other genes. The rates of synonymous mutations were different among patients of different ethnicities, p<0.001 for all genes.

Discussion: Partial lipodystrophy due to *LMNA* mutations may be underdiagnosed in Latinos, leading to reduced participation in clinical trials. The lack of differences in other genes suggests there is no overall cohort bias. Different rates of synonymous mutations suggest there may be evolutionary drivers to racial differences in inherited forms of lipodystrophy, such as founder mutations or heterozygote advantage. Future work will determine prevalence of pLOF variants in lipodystrophy-associated genes in other genetic data sets enriched for minority subjects.

Diabetes Mellitus and Glucose Metabolism

BENCH TO BEDSIDE: NOVEL MECHANISMS IN DIABETES AND METABOLISM

Renal GLUT2 is Essential in Regulating Systemic Glucose Homeostasis by Glycosuria

Leticia M. de Souza Cordeiro, PhD[1], Nagavardhini Devisetty, PharmD[1], David McDougal, PhD[2], Dorien J.M. Peters, PhD[3], Kavaljit H. Chhabra, MPharm, PhD[1].

[1]University of Rochester, Rochester, NY, USA, [2]Pennington Biomedical Research Center, Baton Rouge, LA, USA, [3]Leiden University Medical Center, Leiden, Netherlands.

Diabetes increases renal GLUT2 levels and consequently, worsens hyperglycemia by enhancing glucose reabsorption. We recently demonstrated that renal GLUT2 is a primary effector of the central melanocortin system in regulating glucose homeostasis. Therefore, we hypothesized that renal GLUT2 is essential for maintaining systemic glucose homeostasis by regulating glycosuria. To test the hypothesis, we generated kidney-specific inducible *Glut2* knockout (KO) mice [$Glut2^{LoxP/LoxP}$ x $KspCad^{CreERT2}$ (inducible by tamoxifen)]. These mice exhibited 90% reduction in *Glut2* expression selectively in the kidneys, without affecting the expressions of other renal glucose transporters, such as *Glut1*, *Sglt1*, and *Sglt2*. To evaluate the physiological contribution of renal GLUT2 in systemic glucose homeostasis, we performed oral glucose tolerance tests (OGTT) in kidney-specific *Glut2* KO mice and their control littermates (Ctrl). We observed that the kidney-specific GLUT2 deficient mice exhibited improved glucose tolerance compared to their Ctrls (AUC for OGTT, 41,950 ±2,014 vs. 52,165 ±1,686 mg/dL.min). To measure glycosuria in the kidney-specific *Glut2* KO mice, we placed the mice in metabolic cages and collected 24h urine after acclimating the mice in the new cages. Indeed, the GLUT2 deficient mice had ~1,800-fold increase in urine glucose levels (53.5 ±11 vs. 0.03 ±0.005 mg/24h) and exhibited an increased urine volume (2.5 ±0.3 vs. 0.9 ±0.3 mL/24h) and water intake (7.6 ±0.7 vs. 4.9 ±0.7 mL/24h) compared to their Ctrl littermates. The improvement in glucose tolerance in the kidney-specific *Glut2* KO mice was independent of the insulin signaling because we did not observe any changes in insulin tolerance tests (ITT) (AUC for ITT, 10,982 ±414 vs. 11,275 ±583 mg/dL.min) and serum insulin levels (1.07 ±0.14 vs. 1.05 ±0.13 ng/mL) between the groups. Importantly, the kidney-specific GLUT2 deficient mice had normal serum creatinine (0.42 ±0.02 vs. 0.41 ±0.03 mg/dL), free fatty acid (0.43 ±0.14 vs. 0.53±0.14 nmol/μL), β-hydroxybutyrate (0.29 ±0.01 vs. 0.27 ±0.02 mM) and glucagon (14 ±4 vs. 10 ±1 pg/mL) levels. Moreover, the kidney-specific *Glut2* KO mice had normal glomerular area (4,190 ±119 vs. 4,219 ±186 μm²) as measured by kidney histology and normal glomerular filtration rate (153 ±9 vs. 173 ±10 [μL/min/b.w.]/100) compared with their Ctrl littermates, indicating the absence of any known renal injury. Altogether, we have developed a new mouse model in which we can knockout *Glut2* selectively in the kidneys in adult mice. We show that loss-of-function of kidney-specific GLUT2 improves glucose tolerance due to elevated glycosuria without producing any known side effects. In conclusion, blocking kidney-specific GLUT2 has the potential to treat diabetes.

Diabetes Mellitus and Glucose Metabolism

BENCH TO BEDSIDE: NOVEL MECHANISMS IN DIABETES AND METABOLISM

Scientific and Regulatory Considerations for the Approval of the First Generic Glucagon

Eric Pang, PhD[1], William Chong, MD[2], Markham C. Luke, MD PhD[1].

[1]FDA CDER OGD Division of Therapeutic Performance, Silver Spring, MD, USA, [2]FDA CDER OGD, Silver Spring, MD, USA.

Glucagon for Injection (NDA 020928) is a polypeptide hormone identical to human glucagon approved 20 years ago for severe hypoglycemia in patients with diabetes mellitus. On Dec 28, 2020, the U.S. FDA approved the first generic version of glucagon for injection USP, 1 mg/vial packaged in an emergency kit. The generic and the reference listed drug (RLD) version, i.e., the innovator version, of glucagon were each produced through different manufacturing processes. The RLD version of glucagon is produced via recombinant DNA in yeast while the generic version of glucagon is produced by peptide synthesis. The FDA published its current thinking on how to ensure sameness between the generic and innovator peptide products prepared with different manufacturing processes in a Draft Guidance for Industry: Submission of Abbreviated New Drug Applications for Certain Highly Purified Synthetic Peptide Drug Products, which applies to five peptide drug products, including glucagon. In this presentation, we aim to provide an overview of the regulatory recommendations for submitting generic glucagon drug products for approval, as outlined in the aforementioned draft guidance. Although glucagon may be produced using different manufacturing processes, the sameness in glucagon can be adequately demonstrated using analytical methods, which involve demonstrating physicochemical properties, as well as primary and secondary structures, oligomers and aggregation states. Biological assays may also be used as part of the demonstration of active pharmaceutical ingredient sameness. Synthetic glucagon may have different impurity

profiles when compared to the RLD recombinant product. As part of the ANDA review, impurities in the synthetic drugs are analyzed and controlled, in addition, the potential immunogenicity of new impurities, which are not in the RLD products, are assessed and compared using non-clinical assays. In this work, we will discuss non-clinical assays for assessing the immunogenicity risk of these impurities, for both adaptive and innate immune responses. In conclusion, the sameness of an approved generic synthetic glucagon to an RLD can be adequately established through various analytical methods and biological assays.

Diabetes Mellitus and Glucose Metabolism

BENCH TO BEDSIDE: NOVEL MECHANISMS IN DIABETES AND METABOLISM

Selective Somatostatin 5 (SST5) and Somatostatin 2 (SST2) Nonpeptide Agonists Potently Suppress Glucose- and Tolbutamide-Stimulated Dynamic Insulin Secretion From Isolated Human Islets

Elizabeth Rico, PhD, Jian Zhao, PhD, Mi Chen, BS, Ana Karin Kusnetzow, PhD, Yun Fei Zhu, PhD, Stephen F. Betz, PhD.
Crinetics Pharmaceuticals, San Diego, CA, USA.

Congenital hyperinsulinism (HI) is the most common cause of persistent hypoglycemia in newborns and infants and arises from dysregulated insulin secretion. Rapid recognition and treatment are vital to prevent seizures, permanent developmental delays, coma, or even death. Very few medical options exist to treat congenital HI patients: the K_{ATP} channel activator diazoxide, the injectable somatostatin receptor peptide agonists octreotide and lanreotide, or chronic glucose infusions. However, side effects and/or limited efficacy render these therapies inadequate for many patients.

Somatostatin is a 14-amino acid peptide hormone with a broad spectrum of biological actions, which are regulated through five somatostatin receptor subtypes (SST1-SST5). Somatostatin's common physiological role is to down-regulate secretion of other hormones in various tissues. Its role in the maintenance of euglycemia is to regulate insulin and glucagon secretion from pancreatic β- and α-cells, respectively. Somatostatin regulates insulin secretion by decreasing the intracellular levels of cAMP, inhibition of voltage-gated calcium channels (VGCC), activation of the G protein-activated inward rectifier K^+ channel (GIRK), and direct inhibition of insulin exocytosis.

Several studies have evaluated the effect of somatostatin, somatostatin peptide analogs, and a limited number of nonpeptide somatostatin receptor agonists on insulin secretion in static assays using isolated human islets. However, the lack of highly selective agonists has made the interpretation of the contribution of SST receptor subtypes difficult to discern. Our programs for the treatment of hyperinsulinism, acromegaly, and other indications have led to the development of selective nonpeptide SST2, SST3, SST4, and SST5 agonists, possessing $EC_{50}s < 1$ nM in cell-based assays of receptor activation and selectivity > 130 times over the other members of the family. The ability of these selective nonpeptide agonists to regulate glucose- and tolbutamide-stimulated dynamic insulin secretion

from human islets was evaluated using a perifusion system (Biorep, FL).

We found that selective SST2 and SST5 agonists potently suppressed dynamic insulin secretion in contrast to SST3 or SST4 selective agonists. Importantly, SST5 agonists were shown to have a greater effect than selective SST2 agonists or diazoxide, demonstrating their potential utility in human conditions such as congenital HI. In addition, SST5 activation is also known to have a smaller effect on glucagon secretion and is also less prone to agonist-driven desensitization than SST2 activation. Taken together, these studies support our program to identify, characterize, and develop potent, nonpeptide, orally-bioavailable, selective SST5 agonists with appropriate pharmaceutical and safety characteristics for the treatment of congenital HI.

Diabetes Mellitus and Glucose Metabolism

BENCH TO BEDSIDE: NOVEL MECHANISMS IN DIABETES AND METABOLISM

Serum Sclerostin Is Associated With Central Fat Distribution in Patients With Type 2 Diabetes and Peripheral Neuropathy

BRUNA COSTI, DOCTOR[1], CAROLINA CORREIA, DOCTOR[2], KAROLINE MEDEIROS, DOCTOR[1], PAULO BARROS FILHO, DOCTOR[1], FRANCISCO BANDEIRA, PhD[1].
[1]Division of Endocrinology and Diabetes, Agamenon Magalhaes Hospital, University of Pernambuco Medical School, Recife, Brazil, [2]Department of Neurology, Oswaldo Cruz University Hospital, University of Pernambuco Medical School, Recife, Brazil.

Background: Peripheral neuropathy (PN) is the most common chronic diabetic complication occurring in both type 1 and type 2 (T2DM) patients as well as in prediabetes states. Metabolic syndrome seems to be as important as glycemic control in determining the onset and course of DPN, but the mechanisms underlying this association is far from conclusive. Sclerostin (SCL) is a glycoprotein secreted by osteocytes that has an antagonistic effect on the Wnt/beta-catenin pathway which is related to bone formation as well as to increased ectopic fat including marrow fat. Besides to its well-documented role in bone metabolism, the relationship between SCL and adiposity and metabolic syndrome is poorly understood. **Objective:** To determine SCL levels in patients with T2DM and DPN and evaluate their relationship with metabolic and body composition parameters. **Design:** Cross-sectional study including 56 patients with T2DM and DPN. Serum SCL levels, glycemic and lipid profile, anthropometric measurements, and percent body fat (PBF) were determined. **Results:** Mean age was 61.80 ± 10.67 years, duration of T2DM 13.30 ± 8.13 years, 57.1% men, 78.6% hypertensive, body mass index 28.74 ± 5.04 kg/m^2, abdominal circumference 99.86 ± 13.37 cm, waist-to-hip ratio (WHR) 0.97 ± 0.08, fasting plasma glucose 169.73 ± 83.13 mg/dL, HbA1c 8.88 ± 2.09%, Triglycerides (TG) 163.54 ± 75.93 mg/dL, SCL 207.41 ± 215.13 pg/mL, and PBF 34.45 ± 8.42%. There were significant correlations between SCL and TG (r=0.407, p=0.003) and significant differences in TG according to quartiles (< p25 vs >p75) of SCL: 125.15 ± 47.45 mg/dL vs 223.50 ± 88.77 mg/dL,