Selective nonpeptide somatostatin 5 (sst5) agonist inhibits glucose-stimulated insulin secretion in human and rat islets

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Congenital hyperinsulinism (CHI) is a rare heterogenous genetic disease characterized by dysregulated insulin secretion resulting in persistent hypoglycemia. The most common forms are associated with mutations within the SUR1 and Kir6.2 genes of the β-cell K_{ATP} channel. CHI is typically diagnosed in the newborn period and if undiagnosed or untreated, the resulting severe hypoglycemia can cause seizures, permanent developmental delays, coma, and even death. Unfortunately, there are very limited options to treat CHI patients: the K_{ATP} channel opener diazoxide, the injectable sst2-selective peptide agonists octreotide and lanreotide, chronic glucose infusions, or near-total pancreatectomy. Diazoxide is ineffective in CHI patients with mutations in the K_{ATP} channel (~50% of CHI patients). Although many patients initially respond to sst2-selective peptide agonists, the risk of impairing glucagon secretion through activation of the sst2 receptor in the α-cells, as well as the development of tachyphylaxis, often render these therapies inadequate for long-term use. The activation of sst5 also potently suppresses insulin secretion from β-cells, but does not suppress glucagon, and is less prone to agonist driven desensitization compared to sst2. We hypothesize that an orally available, sst5-selective agonist may be a beneficial new approach to managing hyperinsulinemia.

We launched an iterative medicinal chemistry program that led to the discovery of selective nonpeptide sst5 agonists, possessing EC_{50} values < 1 nM in cell-based assays of receptor activation. These sst5 agonists potently suppress insulin and raise plasma glucose in multiple studies of glycemic control in rats. To unravel the mechanism of selective sst agonism and its translation from rat studies to humans, we have evaluated the effect of these sst5 agonists on static glucose-stimulated insulin secretion (GSIS) assays in pancreatic islets isolated from human donors and from naïve Sprague Dawley rats. As comparators, we used the somatostatin 14 (SS14) peptide to suppress insulin from islets.

In both human and rat islets, we found that selective sst5 agonists suppressed insulin secretion more effectively than selective sst2 agonists, demonstrating their potential efficacy in human conditions. These studies support our program to identify and develop potent nonpeptide selective sst5 agonists with pharmaceutical and safety characteristics suitable for evaluation in human clinical trials.

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Hypothesis: An Oral Drug Selectively Targeting sst5 is the Optimal Strategy for Treating HI

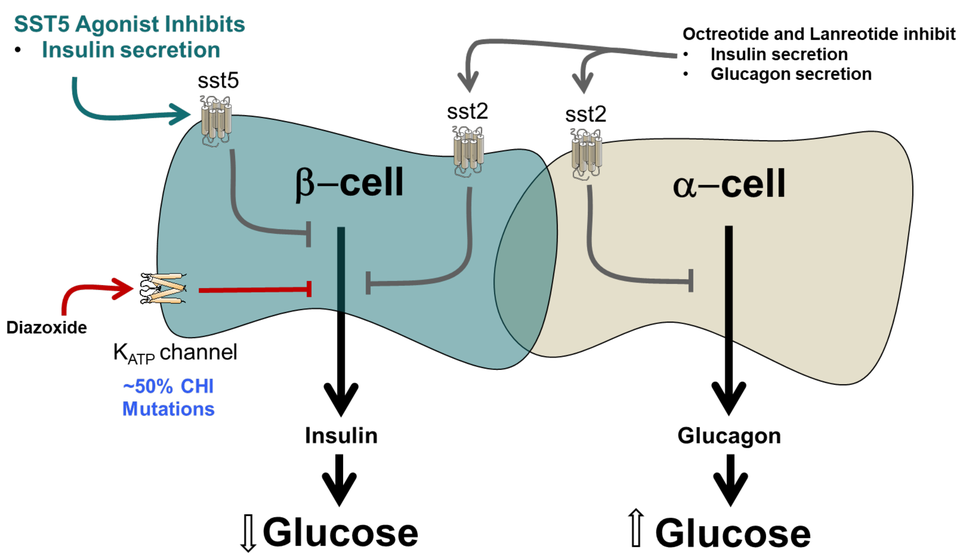

Figure 1. Glucose regulation in HI pancreatic islets by somatostatin agonists and diazoxide.

Diazoxide inhibits GSIS in rat and human islets.

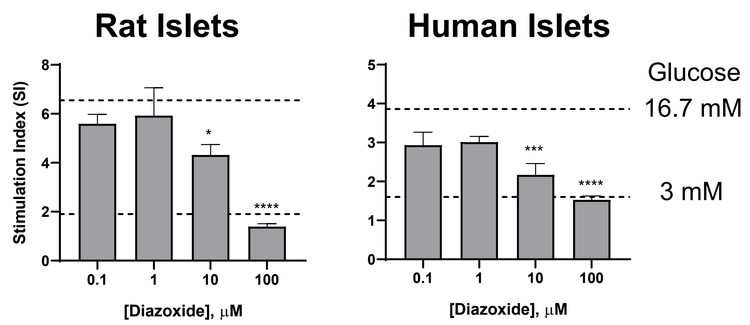

Figure 2. Rat and human islets were treated with 16.7 mM glucose in the presence of increasing concentrations of diazoxide for 90 min. Stimulation Index (SI) was calculated as stimulated insulin levels/basal insulin levels. Dotted lines represent SI at 3 mM and 16.7 mM glucose with no diazoxide. Mean SI±SEM (n = 7 independent experiments). ***, p< 0.001; ****, p<0.0001 (One way ANOVA compared to 16.7 mM glucose)

Peptide and nonpeptide sst agonists inhibit GSIS in rat and human islets more potently than diazoxide

EC_{50} (nM)	sst1		sst2		sst3		sst4		sst5	
	human	rat	human	rat	human	rat	human	rat	human	rat
SS14	0.8	ND	0.13	0.18	0.16	0.13	0.07	0.063	0.063	1.3
Agonist 1	>10000	ND	440	2300	39	22	5.7	0.62	0.39	0.36
Agonist 2	89	ND	0.040	0.30	4.1	6.4	0.34	0.071	2800	24

Table 1. Agonist activity at human and rat sst receptors. G_i activation was measured using the CisBio cAMP assay in CHO-K cells stably expressing each human or rat sst receptor. ND: Not Determined

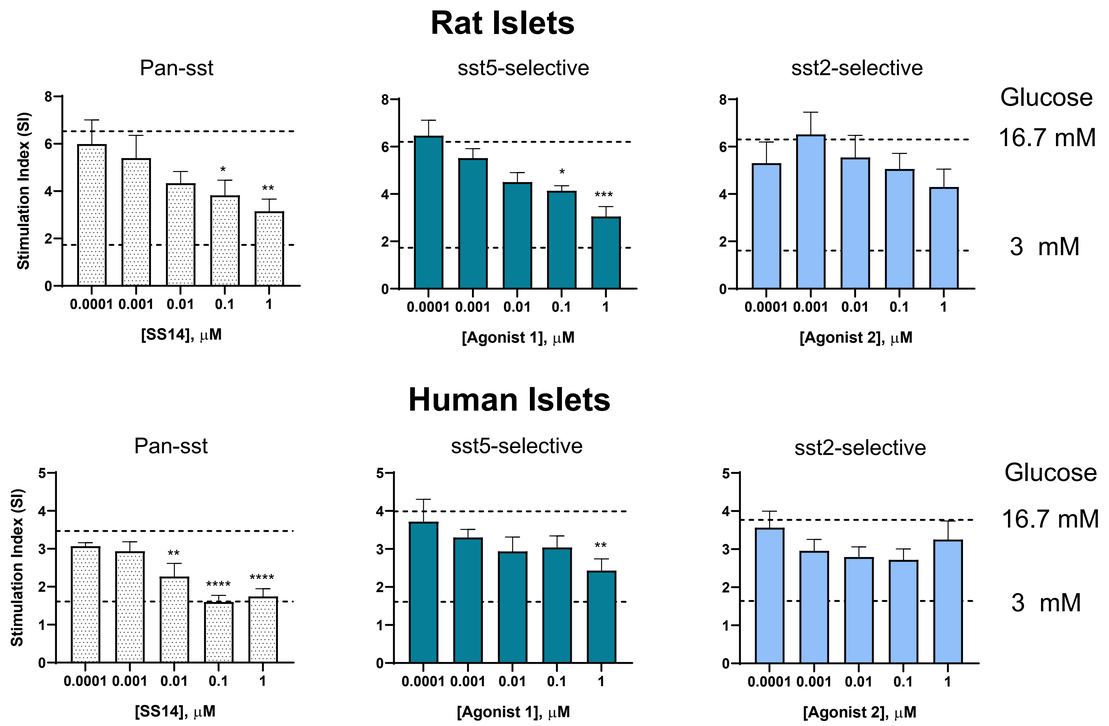

Figure 3. Rat and human islets were treated with 16.7 mM glucose in the presence of increasing concentrations of somatostatin 14 (SS14), sst5-specific small molecule (agonist 1) or sst2-specific small molecule (agonist 2) for 90 min. Stimulation Index (SI) was calculated as stimulated insulin levels/basal insulin levels. Dotted lines represent SI at 3 mM and 16.7 mM glucose with no SS14 or sst agonist. Mean SI±SEM (n = 7 independent experiments). *, p<0.05; **, p < 0.01; ***, p< 0.001; ****, p<0.0001 (One way ANOVA compared to 16.7 mM glucose).

Conclusions: We have developed several potent and selective nonpeptide sst5 and sst2 agonists and evaluated GSIS in human and rat pancreatic islets. Selective sst5 agonists appear to suppress insulin secretion more effectively than selective sst2 agonists, suggesting their potential utility in treating human diseases of hyperinsulinism.

- Static GSIS is inhibited by SS14, a potent agonist of all sst receptors, in human and rat islets (100% and 75% suppression at 1 μM in human and rat respectively).
- The sst5-selective nonpeptide suppressed > 60% insulin secretion at 1 μM in both species.
- The sst2-selective nonpeptide suppressed < 50% insulin secretion at 1 μM in both species.

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