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REVIEW ARTICLE

The current state of GPCR-based drug discovery to treat metabolic disease

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One approach of modern drug discovery is to identify agents that enhance or diminish signal transduction cascades in various cell types and tissues by modulating the activity of GPCRs. This strategy has resulted in the development of new medicines to treat many conditions, including cardiovascular disease, psychiatric disorders, HIV/AIDS, certain forms of cancer and Type 2 diabetes mellitus (T2DM). These successes justify further pursuit of GPCRs as disease targets and provide key learning that should help guide identifying future therapeutic agents. This report reviews the current landscape of GPCR drug discovery with emphasis on efforts aimed at developing new molecules for treating T2DM and obesity. We analyse historical efforts to generate GPCR-based drugs to treat metabolic disease in terms of causal factors leading to success and failure in this endeavour.

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Abbreviations

DPP4, dipeptidyl peptidase-4; α-MSH, α melanocyte stimulating hormone; PAM, positive allosteric modulator; POMC, pro-opiomelanocortin peptide; SSTR5, somatostatin receptor 5; T2DM, Type 2 diabetes mellitus.

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Introduction

The **GPCRs** are a large family of heptahelical plasma membrane-spanning proteins that have evolved to couple extracellular signals into intracellular responses (Lefkowitz, 2007). It is often claimed that 20–50% of approved drugs act in some way *via* GPCRs (Santos *et al.*, 2017). GPCRs typically couple to two main families of intracellular effector proteins to transduce signals: heterotrimeric G-proteins and β-arrestins (Lefkowitz, 2007). Heterotrimeric G-proteins regulate well-described biochemical pathways such as cAMP synthesis, phospholipase C activation, ion channel activity and small GTPase signalling (Lefkowitz, 2007). In contrast, β-arrestins desensitize GPCRs and activate alternative signal transduction pathways such as kinase cascades (Lefkowitz, 2007). A detailed coverage of the intricacies of canonical GPCR biology and signal transduction can be found in the literature and as such is outside the scope of this review. The GPCR family has members that selectively respond to an exceptionally diverse palette of ligands, including photons, protons, small organic molecules, carbohydrates, lipids, peptides and large proteins (Alexander *et al.*, 2017a). Moreover, many GPCRs exhibit selective distribution in organs, tissues and cell types. Not surprisingly, many individual GPCRs have been investigated as potential therapeutic targets for Type 2 diabetes mellitus (T2DM), obesity and cardiovascular disease. In this review, we retrospectively evaluate the outcomes of targeting single GPCRs for T2DM and metabolic disease, with the thought that this might provide insights for future efforts. This review is intended to sample areas we feel are worthy of illumination and is not meant to be exhaustive.

Overview

It is generally asserted that GPCRs are highly amenable to drug discovery and represent the best target-class for identifying novel therapeutic agents (Roth and Kroeze, 2015; Santos *et al.*, 2017). The GPCR phylogenetic tree represents five families of transmembrane receptors as designated by the GRAFS (or Class) nomenclature Glutamate (Class C), Rhodopsin (Class A), Adhesion (Class B2), Frizzled (Class F), Secretin (Class B1) (Alexander *et al.*, 2017a). The GPCR-ome consists of more than 400 olfactory receptors and approximately 300 potential drug-target receptors (Roth and Kroeze, 2015). Within this selection of 300 drug-target GPCRs, approximately 150 are orphan receptors (no endogenous ligand has been described for them). Among the GPCR drug-target set, there is a diversity of natural ligand, protein structure, biological function and consequently druggability of the individual GPCRs.

Many pharmacological modalities exist by which to target GPCRs. Although these are well-elucidated concepts, they are worth listing (see Table 1). Briefly, agents to target GPCR biology can act indirectly, such as an injectable antibody to

Table 1

Different pharmacological approaches to target GPCRs

Pharmacology	Mechanisms of action	Representative targets	Examples: drug, disease
Indirect orthosteric agonism or antagonism by changing natural ligand concentration	Enzyme inhibitors, antibodies, transporter inhibitors	DPP4 (GLP-1, GIP receptors)	**Sitagliptin**, T2DM
		ACE (AT_1, AT_2 receptors)	**Captopril**, hypertension
		Cycloxygenase (many PG, lipoxin and Tx receptors)	Aspirin, pain and inflammation
Indirect antagonism by reducing receptor expression	siRNA, CRISPR, ASO	Glucagon receptor	ISIS 325568 (Phase II), T2DM
Negative allosteric modulation	Small molecules, peptides, antibodies. Alter the affinity and/or efficacy of the natural ligand	CCR5	**Maraviroc**, HIV/AIDS
Positive allosteric modulation	Small molecules, peptides, antibodies. Alter the affinity and/or efficacy of the natural ligand	CaS receptors	Cinacalcet, secondary hyperparathyroidism
Orthosteric antagonist	Small molecules, peptides, antibodies.	β_1- and β_2-adrenoceptors	**Propranolol**, cardiac arrythmia and heart failure
	Block the binding of the natural ligand in a competitive manner	H_2 receptors	**Ranitidine**, dyspepsia
		GnRH receptors	Degarlix, prostate cancer
Orthosteric agonist	Small molecules, peptides,antibodies. Mimic the binding of the natural ligand to activate the receptor.	β_2-adrenoceptors	**Salbutamol**, asthma
		GLP-1 receptors	Exenatide, T2DM
		SST_2 and SST_5 receptors	**Octreotide**, GH secreting tumours and acromegaly

Examples of the key pharmacological methods to drug GPCRs and some examples from approved medicines.

sequester the natural agonist ligand (such as anti-CGRP antibodies for migraine) (Dodick *et al.*, 2014) or a small molecule enzyme inhibitor to prevent degradation of the endogenous agonist, such as dipeptidyl peptidase-4 (**DPP4**) inhibitors that block the degradation of glucagon-like peptide-1 (**GLP-1**) and glucose-dependent insulinotropic peptide (**GIP**) and thereby enhance the effects of **GLP-1** receptor and **GIP receptor** activation respectively (Ahren, 2007). Canonical gene therapy approaches such as siRNA and antisense as well as emerging approaches such as CRISPR have shown some promise, but drug delivery has been a limiting factor with these methods.

Molecules that act by directly binding GPCRs are classified as either orthosteric (bind in the same pocket as the natural ligand) or allosteric (bind in a different pocket to the natural ligand). The pharmacology of orthosteric GPCR ligands is well understood. Blocking the action of the natural ligand to stimulate a GPCR is the effect of an antagonist (e.g. histamine H_1 receptor antagonists for allergy) (Sadek and Stark, 2016). Agonists, such as the **β_2-adrenoceptor** agonists for asthma, mimic the cellular pharmacology of the endogenous agonist (adrenaline for the β_2-adrenoceptor) and typically have an enhanced duration of action. Recently, attention has focused on the discovery and characterization of allosteric modulators of GPCRs. Such molecules cooperatively interact with the natural orthosteric ligand to potentiate receptor signalling (positive allosteric modulator, PAM) or attenuate receptor signalling (negative allosteric modulator). Typically, these molecules work by stabilizing unique conformations of the receptor and modulating either the affinity or efficacy of an orthosteric ligand. For example, **cinacalcet** is a PAM of the **calcium sensing receptor** used to treat hypoparathyroidism (Jensen and Brauner-Osborne, 2007). Allosteric modulators provide several potential advantages over orthosteric molecules with respect to selectivity, mechanism and kinetics of action and the ability to modulate receptors for which the orthosteric site appears undruggable with conventional approaches. To date, unfortunately, limited success has been demonstrated with GPCR allosteric modulators in the clinic. Other pharmacological ligand categorizations exist including partial agonists, inverse agonists, bitopic ligands, kinetically selective and biased ligands. Discussion of these is beyond the scope of this article but they are well reviewed in the literature.

The medical need in metabolic disease: an opportunity for GPCRs

One of the major medical needs of the 21st century is the prevention and treatment of the epidemic of T2DM, obesity and related co-morbidities (Zimmet *et al.*, 2014). It is projected that by 2035, over half a billion individuals will have T2DM. A sizeable component of this epidemic is likely resultant from dietary and societal changes over the last century. Given the magnitude and seriousness of the oncoming problem, it is probable that in concert with preventative strategies, an enhanced armamentarium for pharmacological treatment of these conditions will be needed.

Design of new therapeutic approaches must take into account that glucose homeostasis and energy metabolism are regulated by highly coordinated signalling networks linking the enteric nervous system and the CNS. Normal interplay among the processes controlling glucose disposal and energy expenditure can, however, become dysregulated when excessive caloric intake leads to profound lipid accumulation and development of obesity. This condition is characterized by an increase in intra-abdominal visceral fat and ectopic deposition of lipid in non-adipose tissues such as liver and skeletal muscle. Adiposity raises intracellular lipid content and intermediate metabolites such as DAG, acetyl-CoA, and various ceramides, which are associated with the development of insulin resistance (Samuel and Shulman, 2016). Reduced insulin sensitivity in muscle is particularly detrimental to overall glucose control because this tissue accounts for the majority of whole-body glucose utilization. Furthermore, impaired insulin action in adipocytes promotes lipolysis, which increases the availability of fatty acids for uptake by the liver. This heightened flux of pre-formed fatty acids increases hepatic triglyceride synthesis *via* re-esterification and promotes gluconeogenesis by elevating acetyl-CoA from fatty acid oxidation, thereby contributing to overall hyperglycaemia (Samuel and Shulman, 2016). If an increase in insulin secretion by pancreatic beta cells fails to sufficiently compensate, impaired glucose tolerance and T2DM eventually ensue (DeFronzo and Tripathy, 2009).

The underlying genetic basis that predisposes susceptibility and/or influences the onset of T2DM is still not well understood. However, consistent with the critical physiological impairments that characterize progression of the disease, most genetic loci linked to T2DM risk are associated with insulin secretion and pathways controlling pancreatic islet development (Shungin *et al.*, 2015). It is also becoming clear that genetic variance points to a role of the brain in obesity susceptibility as adiposity is associated with genes involved in hypothalamic pathways that control food intake and energy expenditure (Shungin *et al.*, 2015). These genetic findings support other emerging evidence of a 'brain-centred glucoregulatory system' that works in concert with islets to integrate the complex control of glucose homeostasis (Schwartz *et al.*, 2013). Defining the neuronal circuity that helps control glucose homeostasis should provide new therapeutic targets, many of which may be GPCRs.

GPCR-targeted drug discovery for T2DM and obesity: historical approaches

Academia and the pharmaceutical industry have invested substantial resources into basic science, target validation, drug discovery and drug development supporting GPCR-targeted medications. The genomic era of GPCR discovery was presaged by the molecular cloning of the β-adrenoceptor in 1986 (Lefkowitz, 2007). This led to the identification of the genes encoding numerous GPCRs for hormones, neurotransmitters and regulatory peptides many of which had been hypothesized decades earlier. The current molecular biology toolkit for GPCRs has powered drug discovery research toward the generation of several investigational molecules for treating metabolic diseases, and there is a wealth of publications, patents and clinical trials that exemplify this. To

simplify our analysis, we have created a top 20 list of what we regard as the most extensively studied and historically promising GPCRs for T2DM and obesity that have been tested in humans (Table 2). Of these, we further highlight some of the most intensely investigated GPCRs in short vignettes on the **glucagon receptor**, the **β_3-adrenoceptor**, the melanocortin **MC_4 receptor**, the cannabinoid **CB_1 receptor**, the orphan receptor **GPR119**, the **free fatty acid FFA1 receptor**/(GPR40) and the glucagon-like peptide-1 (GLP-1) receptor.

Why so challenging: lost in translation from mice to men?

The receptor for glucagon, the principal counter-regulatory hormone to insulin, was pharmacologically characterized in the early 1970s (Rodbell, 1973), but it was not until the cloning of the glucagon receptor in 1993 (Jelinek *et al*., 1993) that heterologous expression of this receptor became possible. This enabled structure–function studies to be undertaken and initiated the identification of small molecule and antibody antagonists (Pearson *et al*., 2016). Glucagon receptor-null mice and studies using glucagon receptor antisense oligonucleotides in diabetic rodent models confirmed the glucose-lowering potential of antagonizing glucagon receptors but also demonstrated that strong interference in this pathway results in compensatory alpha-cell proliferation in the pancreas (Sloop *et al*., 2004). Ultimately, several antibody and small molecule glucagon receptor antagonists were identified as putative anti-diabetic agents, with some showing indications of efficacy in humans (Pearson *et al*., 2016). However, based on the overall benefit–risk analysis, it remains unclear whether glucagon receptor antagonists will ultimately be submitted to, and approved by, regulatory agencies.

A presumed drawback of GCGR antagonists is that these agents may only affect hyperglycaemia and have little effect on body weight. In this light, one of the first GPCR targets investigated to reduce adiposity was the β_3-adrenoceptor. These receptors are expressed in brown (thermogenic) adipocytes in rodents and humans (Lowell and Bachman, 2003), and receptor activation was proposed to stimulate energy expenditure and promote weight loss and improve insulin action. Agonists at the β_3-adrenoceptor, such as **CL-316243**, reduced food intake, and stimulated lipolysis and thermogenesis consequently inducing weight loss and improved insulin sensitivity in rodents (Susulic *et al*., 1995). Genetic evidence from β_3-adrenoceptor knockout mice (Susulic *et al*., 1995) and obese T2DM humans (Walston *et al*., 1995) supported the *in vitro* and preclinical pharmacological studies bringing much excitement to the development of novel orally available β_3-adrenoceptor agonists. However, clinical studies with several β_3-adrenoceptor agonists demonstrated little effect on lipolysis, energy expenditure and body weight in humans (Larsen *et al*., 2002). An explanation for this lack of efficacy is that β_3-adrenoceptors in humans are expressed only in brown adipocytes, which are relatively sparse in humans, while in rodents, β_3-adrenoceptors are found in both white and brown adipocytes (Ito *et al*., 1998). Moreover, activation of β_3-adrenoceptors in rodent white adipose tissue stimulates the lipolysis necessary to fuel β-oxidation of fatty acids in brown adipose tissue, an effect lacking in humans (Lowell and Bachman, 2003).

Derived from the *proopiomelanocortin* (POMC) gene, **α-melanocyte stimulating hormone (α-MSH)** analogues offered similar promise in preclinical studies, inhibiting food intake and stimulating energy expenditure in both dietary (DIO) and genetic, leptin-deficient rodent obesity/diabetes models (such as the *ob*/*ob*, *agouti* and *db*/*db* mouse models) (Fisher *et al*., 1999). Indeed, the adipokine **leptin** stimulates POMC signalling in a specific population of hypothalamic arcuate nucleus-containing and brainstem POMC-containing neurons (Berglund *et al*., 2012). The anti-obesity effects of **α-MSH** analogues are attributed to activation of MC_4 receptors in the CNS. Consistent with the effects of administration of α-MSH analogues, genetic null mutations of either POMC or MC_4 receptors result in severe obesity in both mice and humans (Tao, 2009). Of particular interest is the weight loss promoting effects of α-MSH administration in human POMC-deficient patients (Kuhnen *et al*., 2016). However, the effect of α-MSH analogues on body weight in the general obese patient population is not clear (Krishna *et al*., 2009). It may be that cardiovascular side effects have been dose-limiting in human studies. Future work will need to examine if cardiovascular side effects exhibit tachyphylaxis following repeated dosing, whether α-MSH analogues with differing pharmacokinetic properties, partial agonist pharmacology or possibly PAMs can safely and efficaciously be used to modulate MC_4 receptors in obese and T2DM patients.

The endocannabinoid system is involved in the regulation of food intake, body weight and peripheral metabolism. Genetic and pharmacological evidence in rodents and humans support that blockade of CB_1 receptors produces reductions in food intake and body weight and improves insulin sensitivity (Kang, 2013). The prototypical CB_1 receptor antagonist SR141716 (rimonabant) was the first to advance to the clinic and new drug registration in Europe for the treatment of obesity and metabolic disease (Lafontan *et al*., 2007). However, the excitement over this new obesity medication was short-lived as significant neuropsychiatric side effects were produced in clinical settings by rimonabant, causing withdrawal of the drug from the market. Thus, although the genetic and pharmacological validation of antagonists of CB_1 receptors in the CNS for obesity was successful, rimonabant failed clinically because of safety concerns that were not evident from preclinical studies. Recently, peripherally-restricted, non-brain penetrant, CB_1 receptor antagonists have demonstrated efficacy in obese diabetic preclinical models (Hsiao *et al*., 2015). Thus, there may yet be a path forward for clinical development of a CB_1 receptor antagonist for metabolic disease.

The lysophospholipid receptor GPR119 received much attention as a novel anti-diabetic target because it is expressed both in pancreatic beta cells and in L and K cells of the gut. The therapeutic potential of this receptor was supported by key data showing GPR119 agonists stimulate insulin and incretin secretion and also reduce body weight in rodents (Ritter *et al*., 2016). These findings are in line with studies of *Gpr119* null mice that show a reduction in post-prandial GLP-1 secretion (Lan *et al*., 2009). Numerous companies advanced GPR119 agonists into clinical trials. However, to date, none have progressed beyond Phase II development (Kang,

Table 2

Top 20 GPCRs for Type 2 diabetes mellitus, metabolic syndrome, and obesity that have been tested in humans

		Receptor	Representative ligands	Pharmacology	Proven or proposed site of action	Therapeutic approach	Route of administration	Preclinical Validation	Outcome of the most advanced phase of testing	Reference
Approved Drugs	1	GLP-1 receptor	Exenatide	Agonist	CNS, GI Tract, Islet, PNS	Peptide, protein	Parenteral	Glucose lowering in rodent and primate models	Approved drug/Byetta and others	Graaf *et al.* (2016)
	2	$5HT_{2C}$ receptor	**Lorcaserin**, **fenfluramine**	Agonist, combination (fenfluramine + phentermine)	CNS	Small molecule	oral	Fen-Phen	Approved drug/Belviq; approved drug (Fen-Phen) subsequently withdrawn	Weintraub *et al.* (1992)
	3	**Calcitonin receptors**	Pramlintide	Agonist	CNS/GI tract/PNS/ Islet	Peptide	Parenteral	Inhibition of gastric emptying and glucagon secretion in rodents	Approved drug/Symlin	Mietlicki-Baase (2016)
	4	μ-opioid receptor	**Naltrexone**	Antagonist (combination with **bupropion**)	CNS	Small molecule	oral	Inhibition of food intake in rodents	Approved drug/ Contrave	Greenway *et al.* (2009)
	5	CB_1 receptor	Rimonabant	Antagonist/ inverse agonist	CNS	Small molecule	oral	Appetite suppression and weight loss in rodent and primate models	Approved drug subsequently withdrawn/ Accomplia	Colombo *et al.* (1998)
Phase III	6	FFA1 receptor (GPR40)	TAK875	Agonist	Islet, L-cell	Small molecule	oral	Glucose lowering in rodent and primate models	Glucose lowering in Phase III	Kaku *et al.* (2015)

continues

Table 2

(Continued)

		Receptor	Representative ligands	Pharmacology	Proven or proposed site of action	Therapeutic approach	Route of administration	Preclinical Validation	Outcome of the most advanced phase of testing	Reference
Phase I and Phase II, other human studies	7	β_3-adrenoceptor	CL-316243, L-796568, TAK-677	Agonist	Adipose	Small molecule	oral	Lipolytic, hypophagic and insulin sensitizing effects in rodents	Phase II	Larsen *et al.* (2002)
	8	**CCK_1 receptor**	GI181771X	Agonist	PNS	Small molecule	oral	Hypophagic effects in rodents	Not efficacious, Phase II	Jordan *et al.* (2008)
	9	Glucagon receptor	TT-401	Agonist (oxyntomodulin, triagonists)	Liver, islet	Peptide	Parenteral	Glucose lowering in rodents	Phase II	Muller *et al.* (2017)
			LY2409241	Antagonist	Liver	Small molecule, antibody, antisense oligonucleotides	oral, parenteral	Glucose lowering in rodents	Glucose lowering in Phase II	Pearson *et al.* (2016)
	10	**GHS (ghrelin) receptor**	**PF-5190457**	Antagonist, inverse agonist	CNS	Small molecule	oral	Hypophagic and weight loss effects in rodents	Phase II diabetes – discontinued for strategic reasons	Asakawa *et al.* (2003)
	11	GIP receptor	SAR438335/ RG7697	Agonist (twincretins, triagonists)	Adipose, islet	Peptide	Parenteral	Glucose lowering, reduced insulin resistance in rodents and primates	Phase II, efficacy in Phase I	Finan *et al.* (2013)
	12	GPBA receptor	SB-756050	Agonist	L-cell	Small molecule	oral	Glucose lowering in rodents	Phase I	Hodge *et al.* (2013)
	13	GPR119	PSN821	Agonist	L-Cell, islet	Small molecule	oral	Glucose lowering and incretin elevation in rodents	Limited efficacy in Phase II	Ritter *et al.* (2016)
	14	H_3 receptor	Betahistine (H_3 antagonist, H_1 agonist)	Antagonist/ inverse agonist	CNS	Small molecule	oral	Prevention of weight gain in rodents	Not efficacious, Phase II	Provensi *et al.* (2016)

continues

Table 2

(Continued)

	Receptor	Representative ligands	Pharmacology	Proven or proposed site of action	Therapeutic approach	Route of administration	Preclinical Validation	Outcome of the most advanced phase of testing	Reference
15	**HCA_2 receptor**	**GSK256073, acipimox**	Agonist	Adipose	Small molecule	oral	Reduction of lipolysis and plasma NEFA, glucose lowering in rodents	Glucose lowering and reduced NEFA in Phase II. Response not durable (GSK256073)	Dobbins *et al.* (2015)
16	MC_4 receptor	**RM-493 (setmelanotide)**	Agonist	CNS	Small molecule and peptide	oral, parenteral	Weight loss, increased energy expenditure and improved glucose tolerance in rodents and primates	Phase II, increased energy expenditure in obese individuals	Chen *et al.* (2015)
17	**MCH_1** receptor	AMG076	Small molecule	CNS	Small molecule	oral	Weight loss, increased energy expenditure and improved glucohomeostasis in rodents.	Phase I	Motani *et al.* (2013)
18, 19	**Y_2 /Y_4 receptors**	**PYY-(3-36)**, TM30388	Agonist	CNS	Peptide	Parenteral	Inhibition of food intake in rodents	Short-term efficacy in humans, inhibition of food intake	Batterham *et al.* (2002)
20	**Y_5 receptor**	MK-0557, velneperit	Antagonist	CNS	Small molecule	oral	Numerous genetic and pharmacological rodent models of obesity	Phase II, limited efficacy for weight loss	Erondu *et al.* (2006)

This table summarizes GPCRs that have been investigated for the treatment of diabetes and obesity in humans. Data are obtained from publications and www.clinicaltrials.gov. It represents a summary of information known to the authors and is not intended to be exhaustive.

2013). It is generally understood that these compounds are well tolerated, but development has been discontinued primarily due to a lack of efficacy with respect to glucose control (Katz *et al.*, 2012). These results represent another example of the difficulty in translating efficacy signals from rodent models to glucose lowering in man.

Small molecule agonists of a related GPCR, the long-chain fatty acid FFA1 receptor (formerly GPR40), were shown to stimulate insulin secretion and lower blood glucose in diabetic rodents (Yashiro *et al.*, 2012) and humans with T2DM (Mancini and Poitout, 2015). Consistent with these results, the stimulatory effects of fatty acids on insulin secretion are lost in FFA1 receptor knockout mice (Lan *et al.*, 2008). Interestingly, a new report shows the ability of FFA1 receptor agonists or potentiators to increase GLP-1 to levels sufficient for reducing food intake and body weight in obese rodents (Gorski *et al.*, 2017). Several FFA1 receptor agonists have entered clinical testing with mixed results. An early molecule **AMG-837** did not lower glucose in Phase I trials (Luo *et al.*, 2012). However, another agonist, **TAK-875** demonstrated sustained glucose lowering as a monotherapy in Phase III trials (Mancini and Poitout, 2015). Unfortunately, due to a possible hepatotoxicity risk, TAK-875 is no longer being advanced in the clinic. As with the glucagon receptor antagonists, despite the extensive efforts of the academic and industrial scientific community, it is unclear at this time if FFA1 receptor agonists will be submitted for regulatory review.

In summary, there are many different factors that can prevent the translation of preclinical projects into fully fledged drugs that could be administered to patients. These can range from non-translatable animal models, safety signals and commercial concerns to unexplainable lack of efficacy in the clinic. As a counter to this list of failures, it is useful to consider the GLP-1 receptor agonists as an example of successful GPCR drug discovery.

A key success: GLP-1 receptor agonist therapy

Discovery and characterization of the physiological actions of the incretin hormones, GIP and GLP-1, and their respective receptors (both secretin family members) during the late 1980s and mid-1990s provided a fundamental understanding of the glucose-regulatory roles of these peptides (Drucker, 2006). This work set the stage for clinical investigations of several GLP-1 receptor agonists during the 2000s. It is noteworthy that the main impetus for pursuing these agents came from preclinical and clinical studies characterizing the ability of GLP-1 to enhance glucose-dependent insulin secretion, regulate gastric transit and reduce food intake, not from genetic studies. Further, analyses of the reported *Glp-1r* null mouse lines have not revealed profound physiological effects resulting from loss of the receptor (Hansotia *et al.*, 2004). In spite of a lack of 'genetic validation' that is often demanded as critical evidence, GLP-1 receptor agonists were successfully developed and are now entrenched in the modern algorithm for treating T2DM because these agents improve glucose homeostasis and reduce body weight. In light of the aetiology and progressive nature of T2DM, the ability to both enhance peripheral glucose disposal and decrease energy intake is clearly a desirable attribute. A key component of the efficacy of GLP-1 receptor agonists is the pleiotropic effects they induce by acting on many different cell types and target tissues (Drucker, 2016). Similarly, there are a wide range of receptors and signal transduction systems that can regulate endogenous GLP-1 secretion making it a critical nexus for pharmaceutical intervention. Small molecule inhibitors of DPP4, the enzyme that inactivates incretins, are also a successful class of drugs. They are extensively reviewed in the literature; thus, we will not discuss them in detail here.

Pharmaceutical development of agonists targeting the GLP-1 receptor has largely focused on engineering strategies to prolong the pharmacokinetic properties of native GLP-1 or **exendin-4**, a potent GLP-1 receptor agonist isolated from the salivary gland of the Gila monster lizard (39 amino acids, 53% similarity with GLP-1) (Eng *et al.*, 1992). Analogues of GLP-1 include **liraglutide**, **dulaglutide** and **albiglutide**. Although there are subtle differences in the efficacy responses for these compounds reported from various clinical trials, each of these agents improves glycaemic control and leads to some degree of weight loss. Overall, this therapeutic class is well tolerated, but the incidence of nausea often limits aggressive dose escalation (Harris and McCarty, 2015). The benefit of GLP-1 receptor agonist therapy beyond improving glycaemic control and body weight has been investigated in large cardiovascular outcome trials for liraglutide. Consistent with improving overall cardiometabolic health, these studies showed that liraglutide therapy reduced the rate of myocardial infarction, stroke and death from cardiovascular causes (Marso *et al.*, 2016). Furthermore, high-risk patients had lower rates of cardiovascular events if they were undergoing treatment with liraglutide (Marso *et al.*, 2016). In other trials, liraglutide 3.0 mg (a higher dose than the standard 1.8 mg) has been shown to be an effective adjunctive therapy to diet and exercise for weight management itself and was approved for this by the FDA. Clearly, the demonstration that GLP-1 receptor agonism improves cardiovascular outcomes and has anti-obesity effects in humans is a major breakthrough in the field of metabolic disease. While it has been determined that activation of GLP-1 receptors lowers body weight by decreasing energy intake, as distinct from increasing fat oxidation, the mechanism(s) by which this occurs continues to be a significant area of investigation.

Exploring GPCR-based mechanisms to increase circulating GLP-1

Based on the success of direct acting GLP-1 receptor agonists and DPP4 inhibitors that stabilize endogenously released GLP-1, more recent efforts have focused on enhancing the secretion of GLP-1 from intestinal L cells as well as other incretins, for example, GIP from K cells. Following bariatric surgery, GLP-1 levels are significantly increased, and these enhanced levels are thought to be partly responsible for the observed improvement in metabolic disease (Jorgensen *et al.*, 2013). Can compounds acting directly on the L cell raise GLP-1 levels to those seen in bariatric surgery or to levels that exceed those observed with currently available therapeutics?

Initial efforts targeting GPCRs expressed on the L cell yielded candidate molecules for GPR119 and the bile acid **GPBA receptor** (TGR5). The GPBA receptor agonist SB-756050 and the dual **FXR** and GPBA receptor agonist INT-767 have progressed to the clinic (Hodge *et al.*, 2013); however, positive data have not been forthcoming. Although GPBA receptor agonists have shown robust GLP-1 secretagogue activity and can improve glucose control in rodent models, these compounds appear to also induce gallbladder filling, which may limit their use due to on-target adverse pharmacology (Li *et al.*, 2011). Intestinally restricted GPBA receptor agonists have been suggested as a means of mitigating unwanted on-target pharmacology (Duan *et al.*, 2015). Efficacy of the GPR119 agonist GSK1292263 has been examined in humans with T2DM (Nunez *et al.*, 2014) but this compound had no effect on either GLP-1 or GIP levels nor did it improve glucose control in patients.

As mentioned previously, some agonists targeting the FFA1 receptor stimulate release of GLP-1 and thus present a novel way of targeting the incretin system. First generation FFA1 receptor agonists (e.g. TAK-875) were found to selectively act on islet beta cells and increase insulin secretion with little effect on GLP-1 secretion possibly through selective coupling to $G\alpha_q$ signal transduction pathways (Hauge *et al.*, 2016). However, newer classes of FFA1 receptor agonists molecules increase GLP-1 secretion by apparently coupling to both G_s and G_q pathways (Hauge *et al.*, 2016), making these molecules significantly more attractive as candidate therapeutic agents. The potential for FFA1 receptor agonists to stimulate sufficient GLP-1 secretion for reducing body weight may enhance the attraction of this type of molecule. A related receptor, **FFA4**/GPR120, is also activated by long-chain fatty acids and ω-3 fatty acids. Activation of FFA4 receptors induces GLP-1 secretion from the L-cell in rodents in addition to inducing anti-lipolytic, insulin sensitizing and anti-inflammatory effects by action on adipose tissue and macrophages (Oh *et al.*, 2014). Much effort has been put into the generation of FFA4 receptor agonists, however, it appears that none have yet reached clinical testing. An interesting concept has been advanced from demonstrations that simultaneous activation of FFA1 and FFA4 receptors has synergistic anti-diabetic effects in animal models by virtue of dual glucose-lowering and insulin-sensitizing effects (Oh *et al.*, 2014; Satapati *et al.*, 2017). It has been proposed that a dual FFA1/FFA4 receptor agonist could be a highly efficacious anti-diabetic agent as it is acting on several key tissues (i.e. L-cell, beta cell, adipocyte and macrophage) and physiological systems thought to be necessary for an effective antidiabetic agent.

Another potential mechanism to increase endogenous GLP-1 concentrations is to inhibit signalling pathways that negatively regulate GLP-1 release. The hormone **somatostatin** plays such a role in the gut as it exerts an inhibitory tone on GLP-1 secretion. In support of this concept, blockade of the somatostatin **SST_5 receptor** has been shown to enhance GLP-1 release (Farb *et al.*, 2017). Although different groups have reported SST_5 receptor antagonists, data from clinical trials evaluating this mechanism in humans have not been disclosed. Further efforts aimed at deep mining of GPCRs expressed in most entero-endocrine cell types have been reported; this may lead to a host of potential new targets to explore either alone or in combination, ultimately with the goal of eliciting a therapeutically relevant increase in GLP-1 and other incretins that can mimic those seen in bariatric surgery.

A look to the future of GPCR drug discovery in metabolic disease

Drug discovery is beset by failure at all stages. This is exemplified by the estimate of >99% of projects initiated in preclinical industrial labs failing to deliver new medicines (Paul *et al.*, 2010). The success rate of developing T2DM therapies for all target classes (not just GPCRs) typifies this. Within the last 30 years, there have only been four classes of medications launched for T2DM that engage novel targets: the thiazolidinediones (oral **PPARγ** agonists), GLP-1-based drugs (injectable GLP-1 analogues and oral DPP4 inhibitors), **pramlintide** (the injectable amylin receptor agonist) and glucose reabsorption inhibitors (oral **SGLT2** inhibitors). Our opinion is that there is no obvious panacea or magic solution that can be applied to revolutionize GPCR-based drug discovery for metabolic disease.

We conclude this review article by offering 10 questions that frame the modern drug discovery paradigm and summarise some of the key factors to consider when initiating and progressing a GPCR drug discovery campaign.

1. *What is the target validation?* Will engaging the GPCR be efficacious for the pathophysiology being targeted? This can encompass a wide variety of information including human genetic data, an efficacious compound in the clinic, compelling data from preclinical models using tool compounds and knockout mice phenotypes. A good example is the GLP-1 receptor, as it does not show particularly strong validation attributes in mouse knockout models or from human genetics – yet this receptor is the pre-eminent T2DM GPCR drug target.
2. *Are preclinical models predictive of clinical efficacy?* It will be difficult to discover and develop a drug if they are not. Clearly, some GPCRs, such as the β_3-adrenoceptor and the GPR119, display robust efficacy in rodents, but this does not translate to treating human disease.
3. *Will the approach be safe?* There is enhanced scrutiny on the safety of metabolic medications because they are typically used chronically. Natural biological processes do not usually have target engagement to the extent that synthetic molecules do. This supra-pharmacological action of synthetic molecules can lead to undesirable side-effects or toxicology in some cases. Ligands of both CB_1 and GPBA receptors demonstrated robust clinical and preclinical-efficacy respectively. However, safety concerns led to termination of those efforts.
4. *Is it technically feasible?* What is the probability of technical success? Are there basic science problems such as selectivity, potency, adsorption, metabolism, excretion and toxicology that cannot be solved in a reasonable timeframe?
5. *Are there preclinical and clinical biomarkers for target engagement and translational pharmacology?* If the compound progresses into clinical development, it is important to

have a method to show the compound is actually engaging the target.

These first five questions are predominantly science rooted and occur at the front end of projects. Often, groups may focus on these first questions and conduct experiments to obtain answers.

6. *How will efficacy of the medicine compare to the existing standard of care (that may be a generic or biosimilar molecule)?*
7. *Where will the medicine fit in the treatment paradigm and what will the therapeutic landscape look like in >10 years time?*
8. *Can a solid intellectual property position be established?*
9. *Is the new medicine commercially viable?* What is the overall probability of success given cost of goods, patent life, projected market, *etc*.
10. *Can efficacy data be generated to warrant reimbursement of the product by payers?* Statistical significance is the historical benchmark to get a medicine approved. In the modern era, drugs need to show substantial benefit to the healthcare system to engender payment.

These remaining questions have more of a commercial and medical bent and are often out of the control of a GPCR-focused scientist, but awareness of these issues is important in resource-constrained environments.

Nomenclature of targets and ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in http://www.guidetopharmacology.org, the common portal for data from the IUPHAR/BPS Guide to PHARMACOLOGY (Harding *et al*., 2018) and are permanently archived in the Concise Guide to PHARMACOLOGY 2017/2018 (Alexander *et al*., 2017a,b,c,d).

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Conflict of interest

All authors are employees of Eli Lilly and Company and may own company stock or possess stock options.

References

Ahren B (2007). Dipeptidyl peptidase-4 inhibitors: clinical data and clinical implications. Diabetes Care 30: 1344–1350.

Alexander SPH, Christopoulos A, Davenport AP, Kelly E, Marrion NV, Peters JA *et al*. (2017a). The Concise Guide to PHARMACOLOGY 2017/18: G protein-coupled receptors. Br J Pharmacol 174 (Suppl 1): S17–S129.

Alexander SPH, Fabbro D, Kelly E, Marrion NV, Peters JA, Faccenda E *et al*. (2017b). The Concise Guide to PHARMACOLOGY 2017/18: Enzymes. Br J Pharmacol 174: S272–S359.

Alexander SPH, Cidlowski JA, Kelly E, Marrion NV, Peters JA, Faccenda E *et al*. (2017c). The Concise Guide to PHARMACOLOGY 2017/18: Nuclear hormone receptors. Br J Pharmacol 174: S208–S224.

Alexander SPH, Kelly E, Marrion NV, Peters JA, Faccenda E, Harding SD *et al*. (2017d). The Concise Guide to PHARMACOLOGY 2017/18: Transporters. Br J Pharmacol 174: S360–S446.

Asakawa A, Inui A, Kaga T, Katsuura G, Fujimiya M, Fujino MA *et al*. (2003). Antagonism of ghrelin receptor reduces food intake and body weight gain in mice. Gut 52: 947–952.

Batterham RL, Cowley MA, Small CJ, Herzog H, Cohen MA, Dakin CL *et al*. (2002). Gut hormone PYY(3-36) physiologically inhibits food intake. Nature 418: 650–654.

Berglund ED, Vianna CR, Donato J Jr, Kim MH, Chuang JC, Lee CE *et al*. (2012). Direct leptin action on POMC neurons regulates glucose homeostasis and hepatic insulin sensitivity in mice. J Clin Invest 122: 1000–1009.

Chen KY, Muniyappa R, Abel BS, Mullins KP, Staker P, Brychta RJ *et al*. (2015). RM-493, a melanocortin-4 receptor (MC4R) agonist, increases resting energy expenditure in obese individuals. J Clin Endocrinol Metab 100: 1639–1645.

Colombo G, Agabio R, Diaz G, Lobina C, Reali R, Gessa GL (1998). Appetite suppression and weight loss after the cannabinoid antagonist SR 141716. Life Sci 63: PL113–PL117.

DeFronzo RA, Tripathy D (2009). Skeletal muscle insulin resistance is the primary defect in type 2 diabetes. Diabetes Care 32 (Suppl 2): S157–S163.

Dobbins R, Byerly R, Gaddy R, Gao F, Mahar K, Napolitano A *et al*. (2015). GSK256073 acutely regulates NEFA levels via HCA2 agonism but does not achieve durable glycaemic control in type 2 diabetes. A randomised trial. Eur J Pharmacol 755: 95–101.

Dodick DW, Goadsby PJ, Spierings EL, Scherer JC, Sweeney SP, Grayzel DS (2014). Safety and efficacy of LY2951742, a monoclonal antibody to calcitonin gene-related peptide, for the prevention of migraine: a phase 2, randomised, double-blind, placebo-controlled study. Lancet Neurol 13: 885–892.

Drucker DJ (2006). The biology of incretin hormones. Cell Metab 3: 153–165.

Drucker DJ (2016). The cardiovascular biology of glucagon-like peptide-1. Cell Metab 24: 15–30.

Duan H, Ning M, Zou Q, Ye Y, Feng Y, Zhang L *et al*. (2015). Discovery of intestinal targeted TGR5 agonists for the treatment of type 2 diabetes. J Med Chem 58: 3315–3328.

Eng J, Kleinman WA, Singh L, Singh G, Raufman JP (1992). Isolation and characterization of exendin-4, an exendin-3 analogue, from Heloderma suspectum venom. Further evidence for an exendin receptor on dispersed acini from guinea pig pancreas. J Biol Chem 267: 7402–7405.

Erondu N, Gantz I, Musser B, Suryawanshi S, Mallick M, Addy C *et al*. (2006). Neuropeptide Y5 receptor antagonism does not induce clinically meaningful weight loss in overweight and obese adults. Cell Metab 4: 275–282.

Farb TB, Adeva M, Beauchamp TJ, Cabrera O, Coates DA, DeShea Meredith T*et al*. (2017). Regulation of endogenous (male) rodent GLP-1 secretion and human islet insulin secretion by antagonism of somatostatin receptor 5. Endocrinology 158: 3859–3873.

Finan B, Ma T, Ottaway N, Muller TD, Habegger KM, Heppner KM *et al*. (2013). Unimolecular dual incretins maximize metabolic benefits in rodents, monkeys, and humans. Sci Transl Med 5: 209ra151.

Fisher SL, Yagaloff KA, Burn P (1999). Melanocortin-4 receptor: a novel signalling pathway involved in body weight regulation. Int J Obes Relat Metab Disord 23 (Suppl 1): 54–58.

Gorski JN, Pachanski MJ, Mane J, Plummer CW, Souza S, Thomas-Fowlkes BS *et al*. (2017). GPR40 reduces food intake and body weight through GLP-1. Am J Physiol Endocrinol Metab 313: E37–E47.

Graaf C, Donnelly D, Wootten D, Lau J, Sexton PM, Miller LJ *et al*. (2016). Glucagon-like peptide-1 and its class B G protein-coupled receptors: a long march to therapeutic successes. Pharmacol Rev 68: 954–1013.

Greenway FL, Whitehouse MJ, Guttadauria M, Anderson JW, Atkinson RL, Fujioka K *et al*. (2009). Rational design of a combination medication for the treatment of obesity. Obesity (Silver Spring) 17: 30–39.

Hansotia T, Baggio LL, Delmeire D, Hinke SA, Yamada Y, Tsukiyama K *et al*. (2004). Double incretin receptor knockout (DIRKO) mice reveal an essential role for the enteroinsular axis in transducing the glucoregulatory actions of DPP-IV inhibitors. Diabetes 53: 1326–1335.

Harding SD, Sharman JL, Faccenda E, Southan C, Pawson AJ, Ireland S *et al*. (2018). The IUPHAR/BPS Guide to PHARMACOLOGY in 2018: updates and expansion to encompass the new guide to IMMUNOPHARMACOLOGY. Nucl Acids Res 46 (D1): D1091–D1106.

Harris KB, McCarty DJ (2015). Efficacy and tolerability of glucagon-like peptide-1 receptor agonists in patients with type 2 diabetes mellitus. Ther Adv Endocrinol Metab 6: 3–18.

Hauge M, Ekberg JP, Engelstoft MS, Timshel P, Madsen AN, Schwartz TW (2016). Gq and Gs signaling acting in synergy to control GLP-1 secretion. Mol Cell Endocrinol 449: 64–73.

Hodge RJ, Lin J, Vasist Johnson LS, Gould EP, Bowers GD, Nunez DJ (2013). Safety, pharmacokinetics, and pharmacodynamic effects of a selective TGR5 agonist, SB-756050, in type 2 diabetes. Clin Pharmacol Drug Dev 2: 213–222.

Hsiao WC, Shia KS, Wang YT, Yeh YN, Chang CP, Lin Y *et al*. (2015). A novel peripheral cannabinoid receptor 1 antagonist, BPR0912, reduces weight independently of food intake and modulates thermogenesis. Diabetes Obes Metab 17 (5): 495–504.

Ito M, Grujic D, Abel ED, Vidal-Puig A, Susulic VS, Lawitts J *et al*. (1998). Mice expressing human but not murine beta3-adrenergic receptors under the control of human gene regulatory elements. Diabetes 47: 1464–1471.

Jelinek LJ, Lok S, Rosenberg GB, Smith RA, Grant FJ, Biggs S *et al*. (1993). Expression cloning and signaling properties of the rat glucagon receptor. Science 259: 1614–1616.

Jensen AA, Brauner-Osborne H (2007). Allosteric modulation of the calcium-sensing receptor. Curr Neuropharmacol 5: 180–186.

Jordan J, Greenway FL, Leiter LA, Li Z, Jacobson P, Murphy K *et al*. (2008). Stimulation of cholecystokinin-A receptors with GI181771X does not cause weight loss in overweight or obese patients. Clin Pharmacol Ther 83: 281–287.

Jorgensen NB, Dirksen C, Bojsen-Moller KN, Jacobsen SH, Worm D, Hansen DL *et al*. (2013). Exaggerated glucagon-like peptide 1 response is important for improved beta-cell function and glucose tolerance after Roux-en-Y gastric bypass in patients with type 2 diabetes. Diabetes 62: 3044–3052.

Kaku K, Enya K, Nakaya R, Ohira T, Matsuno R (2015). Efficacy and safety of fasiglifam (TAK-875), a G protein-coupled receptor 40 agonist, in Japanese patients with type 2 diabetes inadequately controlled by diet and exercise: a randomized, double-blind, placebo-controlled, phase III trial. Diabetes Obes Metab 17: 675–681.

Kang SU (2013). GPR119 agonists: a promising approach for T2DM treatment? A SWOT analysis of GPR119. Drug Discov Today 18: 1309–1315.

Katz LB, Gambale JJ, Rothenberg PL, Vanapalli SR, Vaccaro N, Xi L *et al*. (2012). Effects of JNJ-38431055, a novel GPR119 receptor agonist, in randomized, double-blind, placebo-controlled studies in subjects with type 2 diabetes. Diabetes Obes Metab 14: 709–716.

Krishna R, Gumbiner B, Stevens C, Musser B, Mallick M, Suryawanshi S *et al*. (2009). Potent and selective agonism of the melanocortin receptor 4 with MK-0493 does not induce weight loss in obese human subjects: energy intake predicts lack of weight loss efficacy. Clin Pharmacol Ther 86: 659–666.

Kuhnen P, Clement K, Wiegand S, Blankenstein O, Gottesdiener K, Martini LL *et al*. (2016). Proopiomelanocortin deficiency treated with a melanocortin-4 receptor agonist. N Engl J Med 375: 240–246.

Lafontan M, Piazza PV, Girard J (2007). Effects of CB1 antagonist on the control of metabolic functions in obese type 2 diabetic patients. Diabetes Metab 33: 85–95.

Lan H, Hoos LM, Liu L, Tetzloff G, Hu W, Abbondanzo SJ *et al*. (2008). Lack of FFAR1/GPR40 does not protect mice from high-fat diet-induced metabolic disease. Diabetes 57: 2999–3006.

Lan H, Vassileva G, Corona A, Liu L, Baker H, Golovko A *et al*. (2009). GPR119 is required for physiological regulation of glucagon-like peptide-1 secretion but not for metabolic homeostasis. J Endocrinol 201: 219–230.

Larsen TM, Toubro S, van Baak MA, Gottesdiener KM, Larson P, Saris WH *et al*. (2002). Effect of a 28-d treatment with L-796568, a novel beta(3)-adrenergic receptor agonist, on energy expenditure and body composition in obese men. Am J Clin Nutr 76: 780–788.

Lefkowitz RJ (2007). Seven transmembrane receptors: something old, something new. Acta Physiol (Oxf) 190: 9–19.

Li T, Holmstrom SR, Kir S, Umetani M, Schmidt DR, Kliewer SA *et al*. (2011). The G protein-coupled bile acid receptor, TGR5, stimulates gallbladder filling. Mol Endocrinol 25: 1066–1071.

Lowell BB, Bachman ES (2003). β-Adrenergic receptors, diet-induced thermogenesis, and obesity. J Biol Chem 278: 29385–29388.

Luo J, Swaminath G, Brown SP, Zhang J, Guo Q, Chen M *et al*. (2012). A potent class of GPR40 full agonists engages the enteroinsular axis to promote glucose control in rodents. PloS one 7: e46300.

Mancini AD, Poitout V (2015). GPR40 agonists for the treatment of type 2 diabetes: life after 'TAKing' a hit. Diabetes Obes Metab 17: 622–629.

Marso SP, Bain SC, Consoli A, Eliaschewitz FG, Jodar E, Leiter LA *et al*. (2016). Semaglutide and cardiovascular outcomes in patients with type 2 diabetes. N Engl J Med 375: 1834–1844.

Mietlicki-Baase EG (2016). Amylin-mediated control of glycemia, energy balance, and cognition. Physiol Behav 162: 130–140.

Motani AS, Luo J, Liang L, Mihalic JT, Chen X, Tang L *et al*. (2013). Evaluation of AMG 076, a potent and selective MCHR1 antagonist, in rodent and primate obesity models. Pharmacol Res Perspect 1: e00003.

Muller TD, Finan B, Clemmensen C, DiMarchi RD, Tschop MH (2017). The new biology and pharmacology of glucagon. Physiol Rev 97: 721–766.

Nunez DJ, Bush MA, Collins DA, McMullen SL, Gillmor D, Apseloff G *et al*. (2014). Gut hormone pharmacology of a novel GPR119 agonist (GSK1292263), metformin, and sitagliptin in type 2 diabetes mellitus: results from two randomized studies. PloS one 9: e92494.

Oh DY, Walenta E, Akiyama TE, Lagakos WS, Lackey D, Pessentheiner AR *et al*. (2014). A Gpr120-selective agonist improves insulin resistance and chronic inflammation in obese mice. Nat Med 20: 942–947.

Paul SM, Mytelka DS, Dunwiddie CT, Persinger CC, Munos BH, Lindborg SR *et al*. (2010). How to improve R&D productivity: the pharmaceutical industry's grand challenge. Nat Rev Drug Discov 9: 203–214.

Pearson MJ, Unger RH, Holland WL (2016). Clinical trials, triumphs, and tribulations of glucagon receptor antagonists. Diabetes Care 39: 1075–1077.

Provensi G, Blandina P, Passani MB (2016). The histaminergic system as a target for the prevention of obesity and metabolic syndrome. Neuropharmacology 106: 3–12.

Ritter K, Buning C, Halland N, Poverlein C, Schwink L (2016). G protein-coupled receptor 119 (GPR119) agonists for the treatment of diabetes: recent progress and prevailing challenges. J Med Chem 59: 3579–3592.

Rodbell M (1973). The problem of identifying the glucagon receptor. Fed Proc 32: 1854–1858.

Roth BL, Kroeze WK (2015). Integrated approaches for genome-wide interrogation of the druggable non-olfactory G protein-coupled receptor superfamily. J Biol Chem 290: 19471–19477.

Sadek B, Stark H (2016). Cherry-picked ligands at histamine receptor subtypes. Neuropharmacology 106: 56–73.

Samuel VT, Shulman GI (2016). The pathogenesis of insulin resistance: integrating signaling pathways and substrate flux. J Clin Invest 126: 12–22.

Santos R, Ursu O, Gaulton A, Bento AP, Donadi RS, Bologa CG *et al*. (2017). A comprehensive map of molecular drug targets. Nat Rev Drug Discov 16: 19–34.

Satapati S, Qian Y, Wu MS, Petrov A, Dai G, Wang SP *et al*. (2017). GPR120 suppresses adipose tissue lipolysis and synergizes with GPR40 in antidiabetic efficacy. J Lipid Res 58: 1561–1578.

Schwartz MW, Seeley RJ, Tschop MH, Woods SC, Morton GJ, Myers MG *et al*. (2013). Cooperation between brain and islet in glucose homeostasis and diabetes. Nature 503: 59–66.

Shungin D, Winkler TW, Croteau-Chonka DC, Ferreira T, Locke AE, Magi R *et al*. (2015). New genetic loci link adipose and insulin biology to body fat distribution. Nature 518: 187–196.

Sloop KW, Cao JX, Siesky AM, Zhang HY, Bodenmiller DM, Cox AL *et al*. (2004). Hepatic and glucagon-like peptide-1-mediated reversal of diabetes by glucagon receptor antisense oligonucleotide inhibitors. J Clin Invest 113: 1571–1581.

Susulic VS, Frederich RC, Lawitts J, Tozzo E, Kahn BB, Harper ME *et al*. (1995). Targeted disruption of the beta 3-adrenergic receptor gene. J Biol Chem 270: 29483–29492.

Tao YX (2009). Mutations in melanocortin-4 receptor and human obesity. Prog Mol Biol Transl Sci 88: 173–204.

Walston J, Silver K, Bogardus C, Knowler WC, Celi FS, Austin S *et al*. (1995). Time of onset of non-insulin-dependent diabetes mellitus and genetic variation in the beta 3-adrenergic-receptor gene. N Engl J Med 333: 343–347.

Weintraub M, Sundaresan PR, Madan M, Schuster B, Balder A, Lasagna L *et al*. (1992). Long-term weight control study. I (weeks 0 to 34). The enhancement of behavior modification, caloric restriction, and exercise by fenfluramine plus phentermine versus placebo. Clin Pharmacol Ther 51: 586–594.

Yashiro H, Tsujihata Y, Takeuchi K, Hazama M, Johnson PR, Rorsman P (2012). The effects of TAK-875, a selective G protein-coupled receptor 40/free fatty acid 1 agonist, on insulin and glucagon secretion in isolated rat and human islets. J Pharmacol Exp Ther 340: 483–489.

Zimmet PZ, Magliano DJ, Herman WH, Shaw JE (2014). Diabetes: a 21st century challenge. The Lancet Diabetes & endocrinology 2: 56–64.