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Recent advances in glucokinase activators for the treatment of type 2 diabetes

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Glucokinase is a member of the hexokinase family of enzymes that are responsible for the phosphorylation of glucose to glucose-6-phosphate for further utilization in cells. The enzyme plays a key role in glucose homeostasis. Phosphorylation of glucose by glucokinase in the liver promotes glycogen synthesis, while in the β-cell it results in insulin release. Activators of glucokinase increase the sensitivity of the enzyme to glucose, leading to increased insulin secretion and liver glycogen synthesis and a decrease in liver glucose output. Thus, small molecule glucokinase activators have been demonstrated to be effective glucose-lowering agents in animal models of type 2 diabetes and have advanced into clinical studies.

Introduction

Type 2 diabetes mellitus (non-insulin-dependent diabetes mellitus; often abbreviated to NIDDM or T2D), which comprises approximately 90–95% of all diabetes cases, is a chronic metabolic disorder involving the dysregulation of glucose metabolism, β-cell dysfunction and impaired insulin sensitivity. It is becoming more prevalent because of the recent dramatic rise in obesity levels [1,2]. T2D is associated with several complications, for example (i) macrovascular complications [3,4] resulting from hyperlipidemia and hypertension, which can lead to end-stage renal disease, limb amputation and accelerated atherosclerosis (cardiovascular disease) [5,6] and (ii) chronic microvascular complications [4] such as retinopathy (blindness), nephropathy and neuropathy. While several options are currently available for the treatment of T2D, no single marketed drug is capable of achieving long-lasting blood glucose control in the majority of T2D patients [7]. Therefore, the use of initial monotherapy may have to be expanded to complex combination therapies as the disease progresses [8]. The glucose-phosphorylating enzyme, glucokinase (GK), represents an attractive target for T2D therapies and has been reviewed previously [9–12]. The present article will provide a brief overview of GK activation by small molecules along with the status of lead and clinical candidates reported over the past couple of years.

Glucokinase: its role in glucose homeostasis

Glucokinase (GK or GLK, also known as hexokinase IV or hexokinase D (ATP: D-glucose-6-phosphotransferase, EC 2.7.1.2)), is a 50-kDa cytoplasmic enzyme and one of the four hexokinases found in mammals that catalyze the conversion of glucose to glucose-6-phosphate (G-6-P), the first step of glucose metabolism [13,14]. In addition to neuronal/neuroendocrine cells GK is selectively expressed in pancreatic β-cells and liver parenchymal cells (hepatocytes), both of which are known to play crucial roles in whole-body blood glucose homeostasis [15,16]. Indeed, studies in transgenic animals have confirmed that GK plays a crucial role in whole-body glucose homeostasis. Animals that do not express GK die within days of birth with severe diabetes while animals overexpressing GK have improved glucose tolerance. More elaborately, mice that are deficient in pancreatic β-cells die because of profound hyperglycemia, whereas mice lacking hepatic GK suffer from impaired insulin secretion. By contrast, overexpression of GK in the livers of diabetic or non-diabetic mice resulted in improved glucose tolerance [17]. GK is often referred to as a 'glucose sensor' in β-cells [18]. The concentration of glucose at which GK demonstrates half-maximal activity is approximately 8.0 mM. The other three hexokinases experience glucose saturation at much lower concentrations (<1.0 mM). Therefore, the

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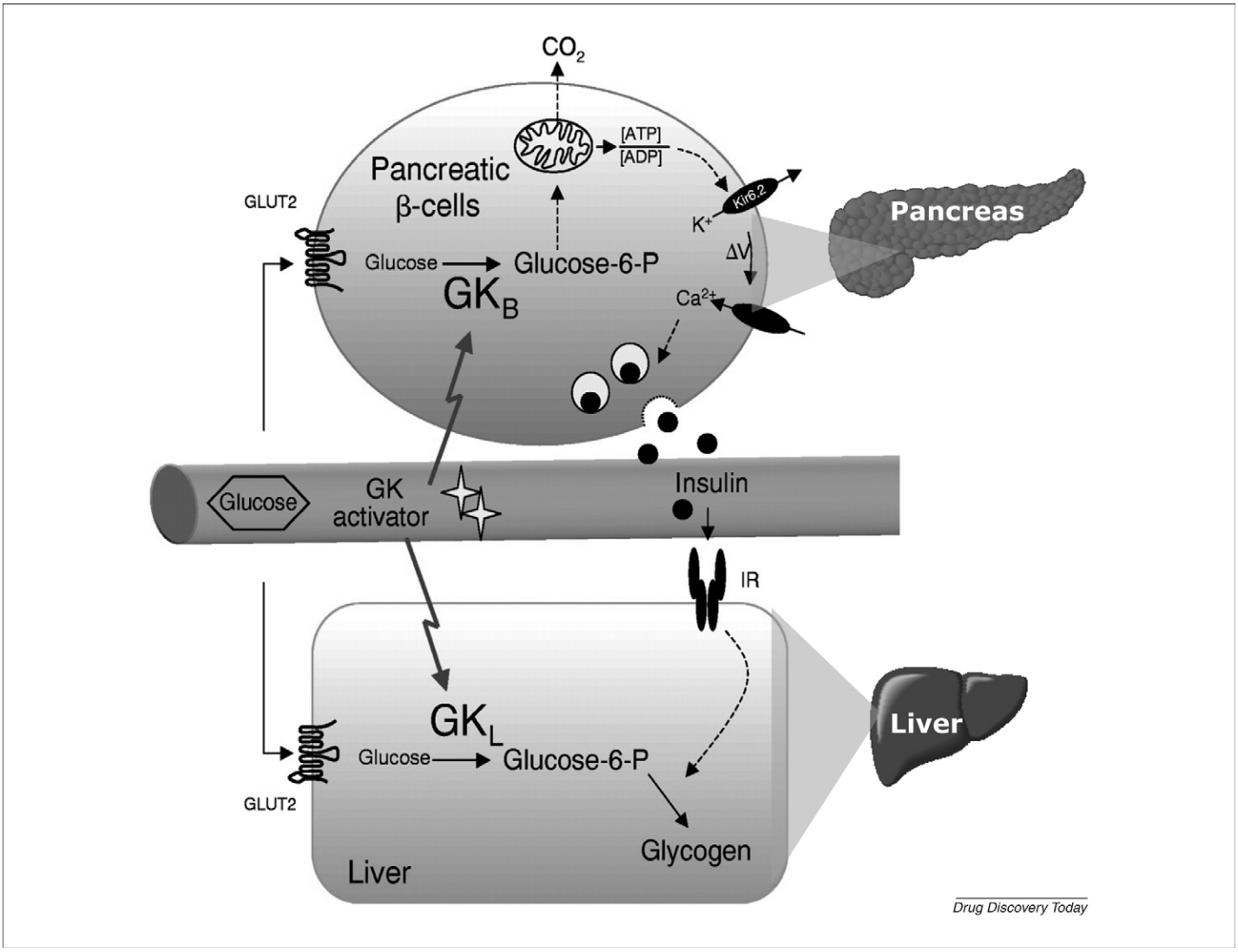


FIGURE 1

The role of GK in glucose homeostasis [22].

metabolism of glucose by GK increases as the concentration of glucose in the blood increases from fasting (5 mM) to postprandial (10–15 mM) levels following a meal containing carbohydrate [19].

Glucokinase in pancreatic β-cells and liver parenchymal cells: a target for T2D

GLUT2, the main plasma membrane glucose transporter in the pancreatic β-cell and liver parenchymal cells transports glucose across the cell membrane [20]. Under physiological glucose concentrations, this process is not rate limiting to the overall rate of glucose uptake in these cells. The rate of glucose uptake is limited by the rate of phosphorylation of glucose to G-6-P, which is catalyzed by GK (GK_B in pancreatic β-cells and GK_L in liver, Fig. 1) [21,22]. Under normal physiological conditions and concentrations, GK can only phosphorylate glucose if the concentration of glucose is above 1–2 mM. GK has a low affinity for glucose as indicated by its $K_{0.5}$ of 6–10 mM. Also GK is not inhibited by physiological concentrations of G-6-P [13]. Phosphorylation of glucose leads to glycogen synthesis in liver and glycolysis, the metabolic pathway that converts glucose into pyruvate, in pancreatic β-cells (Fig. 1). The increase of the citric acid cycle and electron transport results in an increased ATP/ADP ratio leading to the closure of ATP-sensitive K^+ channels, followed by membrane depolarization and subsequently Ca^{2+} influx. This causes the conversion of a reserve pool of insulin granules into a 'readily released' pool and, finally, the release of insulin from β islets into the circulation (Fig. 1).

It is worth mentioning that hepatic GK activity and the intracellular location of GK are controlled by a protein produced in hepatocytes called glucokinase regulatory protein (GKRP) [23]. Small molecules may activate GK either directly or by destabilizing the GK–GKRP complex. The former class of compounds would be predicted to stimulate glucose utilization in both liver and pancreas, whereas the latter class would be likely to act exclusively in the liver. Because T2D is characterized by the defective glucose utilization in both tissues, compounds with either profile may be useful for the treatment of T2D.

Small molecules as glucokinase activators

The allosteric pocket of GK, which is the binding site for GK activators (GKAs), is about 20 Å remote from the glucose binding.

It has been shown that the glucose-lowering effect of recently discovered GKAs is due to binding to this pocket. Many of these GK activators have been shown [24–28] to have potent antihyperglycemic actions in rodents, by increasing pancreatic insulin secretion and by enhancing hepatic glucose metabolism. It is worthy of mention that a bifunctional enzyme, 6-phosphofructo-2-kinase/fructose-2,6-bisphosphatase (6PF2K/F26P_2ase), was found to be an activator of GK [29], although it remains to be clarified whether this is an endogenous activator of GK that acts by a similar mechanism to GKAs.

Gain-of-function mutations of GK

Mutations of the GK gene can produce severe diabetic syndromes. For example, loss of function mutations cause maturity-onset diabetes of the young type 2 (MODY-2) and permanent neonatal diabetes mellitus (PNDM) [30,31]. Hyperglycemia in MODY-2 patients results from defective glucose utilization in both the pancreas and liver. Defective glucose utilization in the pancreas of MODY-2 patients results in a raised threshold for glucose-stimulated insulin secretion. By contrast, gain-of-function, or activating, mutations of GK in humans produce hypoglycemia and hyperinsulinemia. Thus, the activation of GK, which acts as a glucose sensor, thereby upregulates insulin (from the pancreas) and promotes glucose storage as glycogen in the liver under elevated blood glucose conditions. This may prove to be a promising approach for the development of treatments for T2D. Several small molecule GKAs have been discovered and characterized that are capable of eliciting glucose-stimulated insulin secretion (GSIS) in β-cells and pancreatic islets [12,31,32]. Notably, unlike sulfonylureas, GKAs have no effect on GSIS at low glucose concentrations, thereby reducing the possibility of hypoglycemia. Conversely, at low glucose concentrations, GKAs are able to stimulate those cellular processes that would normally take place only at significantly higher glucose levels.

The activator–glucokinase complex

GK is a monomeric enzyme with a single active site and displays kinetic cooperativity for glucose with a half-maximal velocity reached at a substrate concentration ($S_{0.5}$) of <7.5 mM. This suggests that a conformational change enhances the catalytic action of GK. In other words, the enzyme exists in distinct forms that interconvert slowly as a result of substrate binding. Owing to the slow rate of interconversion compared to the catalytic cycle, one form is predominant at steady state and leads to positive kinetic cooperativity. This traditional enzyme kinetics characterization of GK cooperativity has recently been supported by the cocrystallization of a GKA–GK complex and the demonstration of superopen, open and closed conformations of the enzyme (Fig. 2a) [33]. While the crystal structure of GK in its open form has yet to be described, each of these conformations has different kinetic parameters for glucose. Moreover, GK possesses two catalytic cycles (although the present consensus is in favor of the existence of a single catalytic cycle), the ratio of which is responsible for the sigmoidal response to glucose. At low glucose concentrations the 'superopen' low affinity conformation is favored and, hence, results in a slow catalytic cycle. Binding of the glucose substrate to GK, however, induces the formation of 'open' and 'closed' structures that is activated catalytic forms of GK [26]. In other words, high glucose

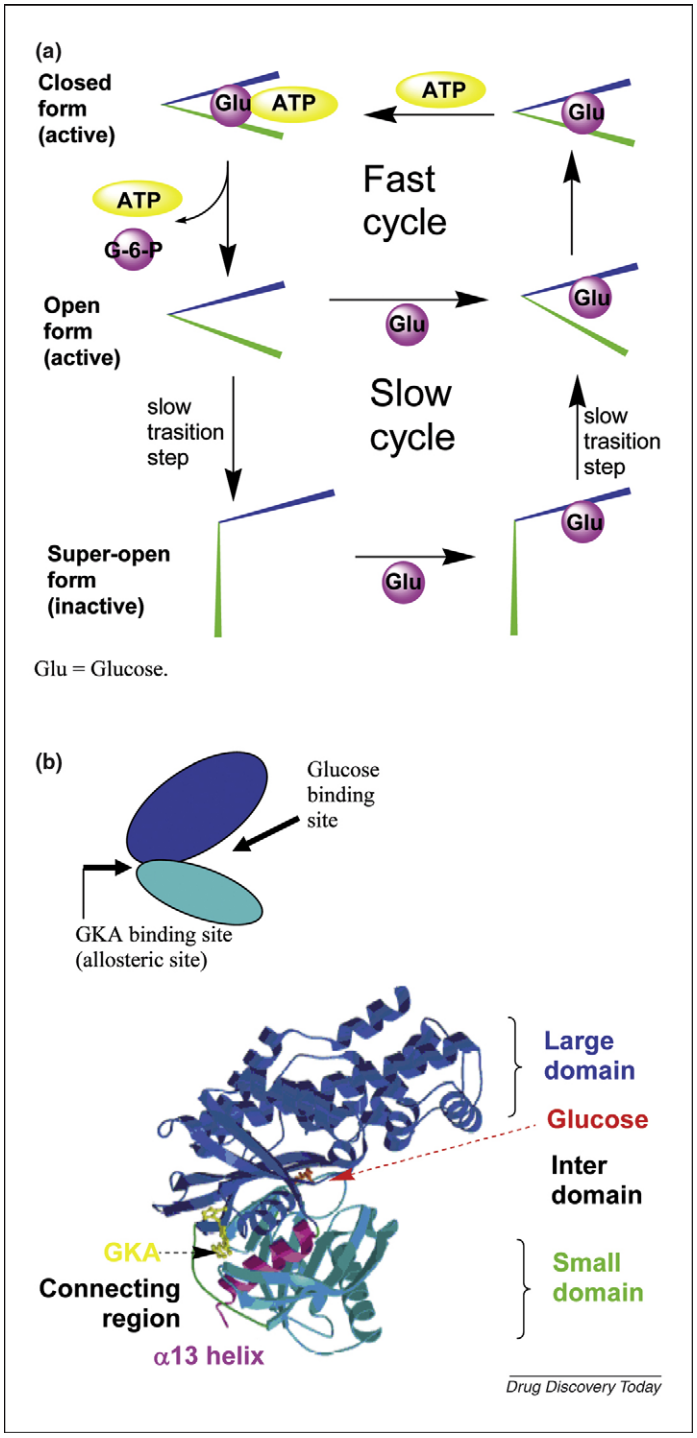


FIGURE 2

(a) Superopen, open and closed conformations of GK and its allosteric activation. **(b)** Binding sites of GK and ribbon drawing of a GKA–GK–glucose cocrystal structure [33].

concentration favors the 'open' and 'closed' high affinity conformations that are associated with a fast catalytic cycle. GKAs appear to bind to an allosteric site at the hinge region formed between the two lobes of the enzyme (Fig. 2b). Thus, GKAs stabilize the 'open' and 'closed' high affinity conformations, thereby activating GK for catalysis. The allosteric pocket at the hinge region is not formed in the 'superopen' conformation and is not accessible to GKAs for binding. This is the basis for the selectivity displayed by GKAs that

increases the affinity of GK for glucose through selectively stabilizing the high affinity conformations of the enzyme. Nevertheless, GKAs also elicit GSIS via indirect (i.e. noncatalytic) GK-dependent mechanisms, although the possibility of insulinotropic effects of GKAs independent of their kinetic effects on GK is believed to be entirely conjectural.

In hepatocytes, the GKRP binds to the superopen form of GK [34] thereby inhibiting GK activity. Binding of a GKA to the 'open'

TABLE 1

Structures of key GKAs reported by various pharmaceutical companies

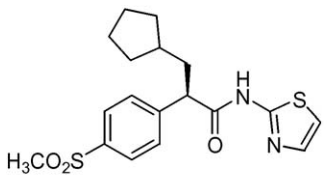
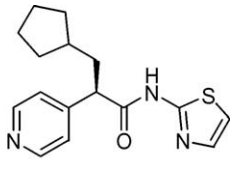
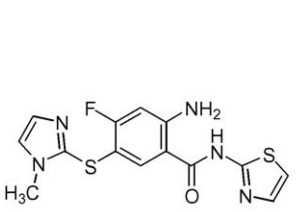
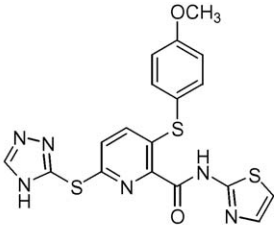
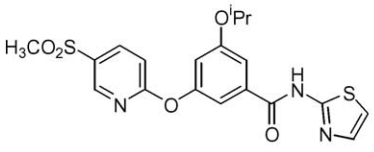
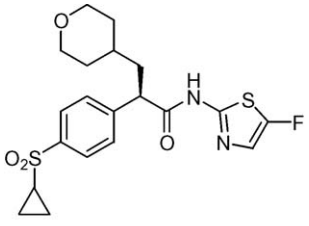
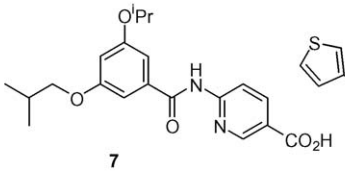
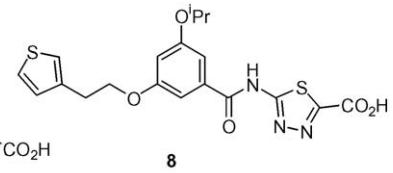
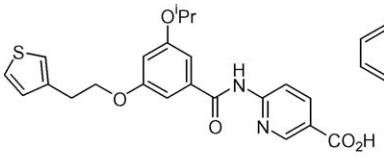
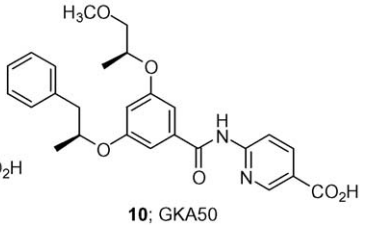
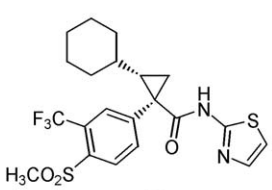
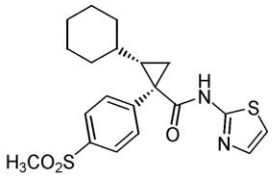
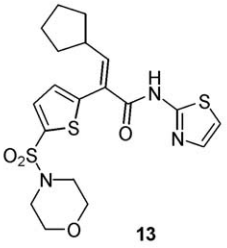
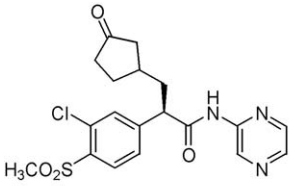
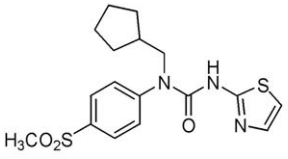
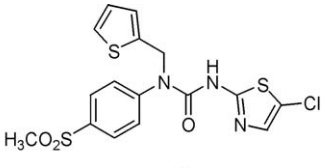
Entry	GKAS reported by	Structures
1.	Roche	 **1**; Ro-28-1675  **2**; Ro-0275145
2.	Banyu	 **3**  **4**  **5**
3.	Prosidon/OSI	 **6**; PSN010 (LY2599506)
4.	AstraZeneca	 **7**  **8**  **9**; GKA22  **10**; GKA50

TABLE 1 (*Continued*)

Entry	GKAS reported by	Structures
5.	Eli Lilly	 **11**  **12**; LY-2121260  **13**
6.	Hoffmann-La-Roche	 **14**; piragliatin (Ro-4389620 or GK2 or R1440)
7.	Prosidion/OSI	 **15**  **16**

and 'closed' conformations of GK, however, prevents relaxation to the superopen form and activates GK for catalysis. Recently, it has been proposed that the GKRP–GK complex dissociates as GKAs bind to the allosteric site [32]. Notably, although in theory GKAs could activate GK either directly or by destabilizing the GKRP–GK complex, as discussed in 'Small molecules as glucokinase activators' section, all the GKAs reported, however, have been shown to have dual actions.

The work on cocrystal structures [33] of GK with its activators has revealed a palm-shaped structure consisting of a small and a large domain separated by an interdomain space (Fig. 2b). The residues Glu256 and Glu290 of the large domain and Thr168 and Lys169 of the small domain along with Asn204 and Asp205 of the interdomain space play a key role in glucose binding. The allosteric site at the hinge region formed between the two domains is surrounded by the linking region of the large domain (β1 strand and α5 helix) and the small domain [α13 helix (magenta), Fig. 2b]. This site is composed of a top created by residues 65–68, which are part of the first connection linking the two domains of GK, and a floor created by the hydrophobic residues Met235, Met210, Ile211, Val62, Val452 and Ile159.

Key glucokinase activators

Since the emergence of the initial report on the pivotal role of GK in glucose homeostasis, the search for orally active small molecule GKAs has become a major focus area of numerous pharmaceutical companies. As a result of drug discovery research initiated by various companies, several GKAs have been reported during the past few years [35]. A phenylacetamide derivative [24,25] **1** (the *R*-enantiomer, Ro-28-1675 of Roche, Entry 1, Table 1) was reported as an orally active GKA (EC_{50} = 0.75 μM) that potently activated recombinant human GK in a dose-dependent manner without affecting other hexokinases of brain and muscle. It showed significant reduction in the elevation of glucose in an oral glucose tolerance test (OGTT) in several rodent models of diabetes [11]. Because of its potential cardiovascular risk (hERG IC_{50} = 2.8 μM; Purkinje fiber ΔAPD_{90} = 20%), however, further development of this compound was abandoned. The crystal structure of a pyridine variant of compound **1** that is compound **2** of Roche (Entry 1, Table 1) complexed with GK [24,25,36] indicated that the activator binds to an allosteric site located approximately 20 Å distant from the glucose binding site (supporting the traditional mechanism of action). This study characterized the allosteric site as a relatively

small and well-defined pocket, which in some way helps to explain the structural homology of GKAs discovered thus far.

A structural elaboration of an activator (**3**, Entry 2, Table 1) reported by Banyu that belongs to a novel class of substituted amino benzamides [37] yielded substituted pyridine carboxamides [38] and heteroaryl carbonylbenzenes [39] represented by compounds **4** and **5**, respectively (Entry 2, Table 1). On the basis of *in vitro* results in MIN6 cells (pancreatic β-cells) and *in vivo* results in rodents, compound **6** (Entry 3, Table 1) was identified as a promising activator of GK [28]. This compound, reported by OSI increased GK activity by 4.3-fold (median effective concentration (EC_{50}) 130 nmol/l) and improved insulin secretion *in vitro*. Its effect on glucose metabolism was studied in primary rat hepatocytes using a 2-deoxy-D-[^{3}H]glucose (2-DG) uptake assay where, in liver, it increased 2-DG incorporation into glycogen 6-fold. It reduced blood glucose by both pancreatic and hepatic mechanisms and increased insulin levels when tested in C57B1/6 mice @ 1 and 10 mg/kg (p.o, *q.d*). Compound **6** reduced blood glucose from 20.5 ± 1.5 mM to 5.8 ± 0.5 nM (versus 17.9 ± 1.4 mM to 13.0 ± 1.5 mM for vehicle-treated mice) in *db/db* mice and showed robust effects in both fed and fasted states over an 11-hour treatment period when administered to female Zucker Diabetic Fatty (ZDF) rats. It also showed improved OGTT in ob/ob mice. On the basis of these results in the advanced models of T2D compound **6** has been advanced into clinical trials.

Several novel 1,3,5-trisubstituted benzenes represented by compounds **7**–**10** from AstraZeneca (Entry 4, Table 1) have been reported as benzamide-based activators of GK. Compounds **7** and **8** enhanced free GK levels, glucose phosphorylation, glycolysis and glycogen synthesis in a concentration-dependent manner when administered to rat hepatocytes [40]. Compound **7** improved the affinity of GK for glucose 2- and 4-fold at 1.0 and 10 μM, respectively. Similarly, compound **8** gave a 4- and 11-fold increase at the same concentrations. These compounds caused translocation of GK from nucleus into the cytoplasm in a manner that is similar to a 'glucose-like' effect. Following a single oral dose to female mice that had been fasted overnight, compound **7** showed a dose-dependent reduction in blood glucose levels. The thiophene derivative **9** [41] showed good *in vitro* potency (GK $EC_{50} = 0.09$ μM) and favorable physical properties. It also exhibited good PK properties ($F = 100\%$ in female Han-Wister rats) and desirable antihyperglycemic effects *in vivo*. Although it improved glucose tolerance in female Zucker rats following a single oral dose it required 30 mg/kg to achieve significantly reduced OGTT. To improve the potency of **9**, further SAR work was carried out. Compound **10** was obtained by introducing α-branching with (*S*)-stereochemistry in the isopropoxy group and varying the alkoxy side chain with desired stereochemistry [42]. This compound showed good *in vitro* potency (enzyme $EC_{50} = 0.03$ μM) and PK properties both in rats ($F = 99\%$) and dogs ($F = 100\%$) and showed antihyperglycemic effects at doses as low as 1.0 mg/kg in acute OGTT in high fat diet fed female Zucker rats.

A series of acylcyclopropyl acetamide derivatives, for example **11** and **12** (Entry 5, Table 1) have been reported by Eli Lilly to be activators of GK [26,43,44]. Compound **11** showed an enzyme EC_{50} of 0.16 μM. Compound **12** [26,44] increases insulin release in studies conducted in freshly isolated rodent pancreatic islets. X-ray crystallographic studies indicated that, like other GKAs, compound **12** also binds to the allosteric site of GK. When tested in primary rat hepatocytes, using a 2-DG uptake assay, compound **12** stimulated 2-DG uptake by 140% ($EC_{50} = 1.7 \pm 0.4$ μM) in the presence of ~5 mM glucose. Also, when dosed orally at 50 mg/kg, compound **12** produced a 28% decrease in the area under the glucose curve following an OGTT in overnight-fasted Wister rats. Notably, **12** induced a 2-fold increase in GK protein levels in pancreatic β-cells. An olefin derivative **13** has also been reported by Eli Lilly to be an activator of GK.

Compound **14** [45] (Entry 6, Table 1) of Hoffmann-La-Roche, which showed an effect on increasing hepatic glucose metabolism in a pancreatic clamp study, was identified as a promising activator of GK [46]. In phase 2 clinical studies this compound was tested, with each being used either in monotherapy or in combination with metformin. Although compound **14** showed a dose-dependent reduction of fasting glucose and glucose excursions following an OGTT in healthy subjects and T2D patients, it has recently been replaced by another activator, R1511, because of an as-yet undisclosed reason. The chemical structure of R1511, which is presently in phase 1 clinical trials, has not yet been disclosed.

A series of urea derivatives have been reported by Prosidion/OSI as GK activators in which the chiral α-carbon of **1** (Entry 1, Table 1) has been replaced by an achiral nitrogen atom [47]. For example, compound **15** (Entry 7, Table 1) was found to be a good GKA, *in vitro* (GK $EC_{50} = 16.0$ and 9.7 μM @ 5 mM and 15 mM glucose, respectively). Compound **15** did not, however, show blood glucose-lowering effects when dosed orally at 100 mg/kg to overnight-fasted C57BL/6J mice. Further SAR work yielded compound **16** (Entry 7, Table 1) (GK $EC_{50} = 6.6$ μM @ 5 mM glucose) that lowered blood glucose levels by up to 34% compared to vehicle control, when administered orally at 100 mg/kg to overnight-fasted C57BL/6J mice.

Glucokinase activators under development

Several GKAs [9–12,35,48] are presently at various stages of clinical development. Although chemical structures of many of these activators are not available a list summarizing their development status and related information is given in Table 2. The present section is organized based on the molecules that are in (a) the most advanced stage followed by (b) phase 1 clinical trials and (c) preclinical studies. Compound **14** was the most advanced candidate, entering phase 2 trials; however, it has been withdrawn for an as-yet unknown reason. The other most advanced candidate AZD6370 is presently undergoing phase 2 trials and is being developed for the potential treatment of obesity and T2D [49]. Apart from these two candidates several compounds are presently in phase 1 clinical trials (Table 2). For example, compound R1511 has been presented as 'prioritized backup' and replaced the earlier lead **14**. Compound **6** entered phase 1 trial, based on its strong *in vitro* and *in vivo* preclinical data, whereas phase 1 clinical trials of AZD1656 have been initiated in Europe. Another compound, ID1101 [50], presently in phase 1 clinical studies has shown a good safety profile. In phase 1 clinical trials, ID1101 enhanced glucose-stimulated insulin release from pancreatic islet cells and glucose disposition by the liver, thereby reducing blood glucose and body weight.

As shown in Table 2, a couple of compounds, for example NN9101 (or TTP355), TTP399 and ARRY403 are presently undergoing preclinical studies. Preclinical results of compound NN9101

TABLE 2

Status summary of selected GKAs

Compound	Company	Status	Refs
14	Hoffmann-La-Roche	Phase 2 (study on hold)	[45]
AZD6370	AstraZeneca	Phase 2	[48]
R1511 or GK3	Hoffmann-La-Roche	Phase 1	[12,48]
6	Prosidon/Lilly	Phase 1	[26,48]
AZD1656	AstraZeneca	Phase 1	[48]
ID1101	Innodia	Phase 1	[49,50]
NN9101 or TTP355	Novo-Nordisk/Trans Tech Pharma	Clinical development	[12,48]
TTP399	Novo-Nordisk/Trans Tech Pharma	Preclinical	[12]
ARRY403	Array Biopharma	Preclinical	[51]
ARRY588	Array Biopharma	Discovery	[51,52]
PSN105	Prosidion/OSI	Halted in preclinical	[48]
1	Roche	Halted in preclinical	[48]

showed good glucose regulation, no weight gain and increased insulin content in pancreas. Its glucose-lowering effect was also observed in first human data, including reduction in both fasting and postprandial glucose. Another activator that is TTP399, which is in preclinical development, belongs to a chemical class different from that of NN9101. A potent GK activator [51], ARRY403 (GK EC_{50} = 79 nM @ 5 mM glucose) showed good PK across the species (F = 55% in mouse; 39% in rat; 44% in dog and 33% in monkey) and significant blunting of the postprandial glucose excursion at 3 and 10 mg/kg in acute OGTT dose–response study in C57BL/6J normal mice. It also reduced both nonfasted and postprandial glucose in *ob/ob* mice, in ZDF female rats and NONcNZO10/LtJ mice, without causing hypoglycemia or abnormal weight gain. It is now ready to enter human clinical trials. Another potent activator of human GK *in vitro*, ARRY588 (GK EC_{50} = 42 ± 25 nM @ 5 mM glucose) showed good PK properties (F = 59% and 24% in CD1 mice and rats, respectively) elicited glucose-lowering activity in a dose–response OGTT study in C57BI/6J mice (AUC reductions of 23% and 40% @ 10 and 30 mg/kg p.o.) [51,52]. In a 14-day study in ob/ob mice it showed dose-dependent reduction of glucose excursion in OGTT performed on days 1 and 14, normalized nonfasting glucose by day 3 (10 and 30 mg/kg dose), and comparable reduction in fasting glucose to that seen in C57BI/6J mice.

Earlier, the GK activator PSN105, which was identified along with compound **6** but with a different profile, had been undergoing preclinical studies. Although this compound showed promising antihyperglycemic effects in diabetic animals [53], its further development has been halted recently. Similarly, the progress of compound **1** the most extensively studied GKA at the discovery stage has also been halted because of the reasons discussed earlier.

Potential benefits and possible issues with GKAs

Although GKAs have been proposed as a promising therapy for the potential treatment of 'diabesity' (diabetes and obesity), it has yet to be confirmed that the same small molecule activators could modulate GK activity simultaneously in liver, pancreas and brain. Because of their dual pancreatic and hepatic actions it was anticipated, however, that GKAs are capable of eliciting their desired glucose-lowering effects when used as monotherapy. Indeed, compound **14** was tested successfully as a monotherapy in addition to its combination with metformin during clinical studies. GKAs also have great potential in combination therapy. For example, combining with an insulin sensitizer, such as a thiazolidinedione (TZD) may allow GKAs to deliver enhanced glucose-lowering efficacy owing to the third action elicited by the TZD. While the combination of GKAs with a sulfonylurea may not be complicated by potential incidence of hypoglycemic effects (although not proven by preclinical or clinical data), a combo therapy using GKA/GLP1 (glucagon-like peptide 1) analogs or GKA/DPP4 (dipeptidyl peptidase 4) inhibitors could be a way forward. Indeed, promising preclinical data have been generated for ARRY403 in combination with either metformin, pioglitazone (a TZD) or sitagliptin (a DPP-4 inhibitor) [51]. While hypoglycemia, β-cell hyperplasia, steatosis and weight gain have not been major issues in these preclinical studies, the excessive accumulation of liver glycogen or increased conversion of glucose into fatty acids and triacylglycerols (as observed with massive overexpression of GK activity in rat liver) could be potential concerns [54]. None of these observations has been reported in humans with activating mutations of GK or in rodents treated chronically with a GKA.

Finally, apart from their therapeutic benefits GKAs have great potential as molecular tools for studying the function and physiology of insulin secreting cells such as β-cells and other glucose-sensitive cells [55].

Conclusions

GK activators hold promise as oral antihyperglycemic agents

The fact that GKAs can influence glucose homeostasis in the liver and pancreatic β-cells, two sites of action crucial to diabetes, makes GK activation an attractive target for the potential treatment of T2D. Moreover, as combination therapy is already widely practiced, GKAs dual pancreatic and hepatic actions could be more effective than present oral antidiabetic agents in controlling blood glucose. Despite their potential, GKAs are still in the early stages of clinical development. Clinical studies have been halted at phase 1 for two candidates and at phase 2 for one candidate. Nevertheless,

because of establishing its vital role in glucose homeostasis, considerable progress has been made in understanding the biology of GK. Cocrystal structures of GK with its activators have radically improved the knowledge of GK structure and function, and helped medicinal chemists to design and develop novel and promising activators. Thus, several small molecule GK activators have been discovered that have shown promising glucose-lowering effects in animal models of T2D, and have advanced into human clinical studies. With the promising preclinical data in hand, GK activators have the potential to become useful antidiabetic agents and could be a new addition to the physician's arsenal in the fight against T2D.

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