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Synthesis and SAR of Thiazolylmethylphenyl Glucoside as Novel *C*-Aryl Glucoside SGLT2 Inhibitors

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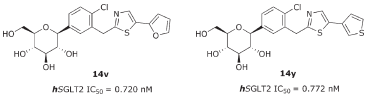
ABSTRACT Novel *C*-aryl glucoside SGLT2 inhibitors containing the thiazole motif were designed and synthesized for biological evaluation. Among the compounds assayed, thiazole containing furanyl moiety **14v** and thiophenyl moiety **14y** demonstrated the best *in vitro* inhibitory activity against SGLT2 in this series to date (IC_{50} = 0.720 nM for **14v** and IC_{50} = 0.772 nM for **14y**). Both of these compounds have been further evaluated on a urinary glucose excretion test and the urine volumes excreted.



KEYWORDS Thiazole, SAR, SGLT2, glucoside, diabetes

Diabetes has become an increasing concern worldwide. In 2010, approximately 285 million people around the world will have diabetes, corresponding to 6.4% of the world's adult population, with a prediction that by 2030 the number of people with diabetes will have grown to 438 million.[1] Type 2 diabetes is the most common disorder of glucose homeostasis, accounting for nearly 90−95% of all cases of diabetes.[2]

Sodium-dependent glucose cotransporters (SGLTs) couple the transport of glucose against a concentration gradient with the simultaneous transport of Na^+ down a concentration gradient.[3] It is estimated that 90% of renal glucose reabsorption is facilitated by SGLT2.[4] SGLT2 inhibitors have attracted considerable attention because there is need for novel antidiabetic agents with a unique mode of action to compensate for existing therapies and avert potential side effects.[5]

Bristol-Myers Squibb and Johnson & Johnson have identified, respectively, dapagliflozin **1** and canagliflozin **2**, potent, selective SGLT2 inhibitors for the treatment of type 2 diabetes.[6−9] At present, dapagliflozin is the most advanced SGLT2 inhibitor in clinical trials (Phase 3), and canagliflozin is the second most advanced clinical candidate. In addition, Boehringer Ingelheim, Lexicon, Astellas, and Pfizer are reported to be in various phases of clinical trials.[10−13]

We have explored metabolically more stable *C*-glucosides bearing a heteroaromatic ring in order to develop novel SGLT2 targeting antidiabetic agents since *C*-glucosides are more stable than *O*-glucoside derivatives. We envisioned that replacement of the distal ring or proximal ring of dapagliflozin **1** with a heterocyclic ring might improve the physicochemical properties while maintaining a similar level of antidiabetic activity, thereby providing a high possibility for clinical development.[14−17] For instance, C log *P* values of this series of compounds would routinely show 2.7−3.0, while that of dapagliflozin (**1**) is shown to be 3.4. Along this line, the structure of dapagliflozin (**1**) was modified into **5**, bearing a thiazole ring at the distal ring position as shown in Figure 1. Herein, we report the synthesis and structure−activity relationship (SAR) evaluation of thiazolylmethylphenyl glucoside congeners.

Preparation of the thiazole compound is described in Scheme 1. Thus, previously reported carboxylic acid **6**[14,15] was coupled with 2-amino-1-(furan-2-yl)ethanone hydrochloride in the presence of EDCI, HOBt, and NMM to provide the corresponding amide **7** in 82% yield. Amide **7** smoothly underwent thionation and subsequent cyclization by the action of Lawesson reagent in refluxing THF to lead to thiazole **8**. Finally, total removal of benzyl protection was affected with TMSI at mild heating for 15 h to produce the target compound **14v** in 73% yield.

The cell-based SGLT2 AMG (methyl-α-D-glucopyranoside) inhibition assay was performed to evaluate the inhibitory effects of all prepared compounds on *h*SGLT2 activities.[18,19] Exploration of the SAR began by replacing the phenyl moiety at the distal ring position of dapagliflozin **1** with the 2-thiazolyl moiety. Table 1 shows the structure−activity relationship upon alteration of the substituent at the distal thiazole ring employing only the β-anomer. Initially, ethyl on the C-5 thiazole ring showed decent activity (**14a**, IC_{50} = 16.7 nM). However, as the number of carbons increases, the inhibitory activity against *h*SGLT2 fluctuates. For example, a 7-fold drop in activity is observed by varying the length of the alkyl chain from ethyl (**14a**, IC_{50} = 16.7 nM) to *n*-butyl (**14b**, IC_{50} = 119 nM), whereas a 9-fold increase in activity is observed from *n*-butyl (**14b**, IC_{50} = 119 nM) to *n*-pentyl (**14c**, IC_{50} = 13.3 nM). Additional 3-fold increase in activity is observed from *n*-pentyl (**14c**) to *n*-hexyl (**14d**, IC_{50} = 5.0 nM). A branched alkyl

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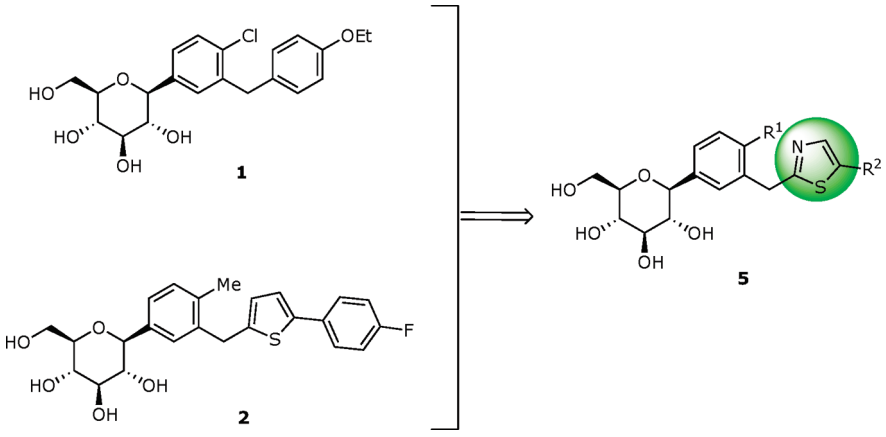
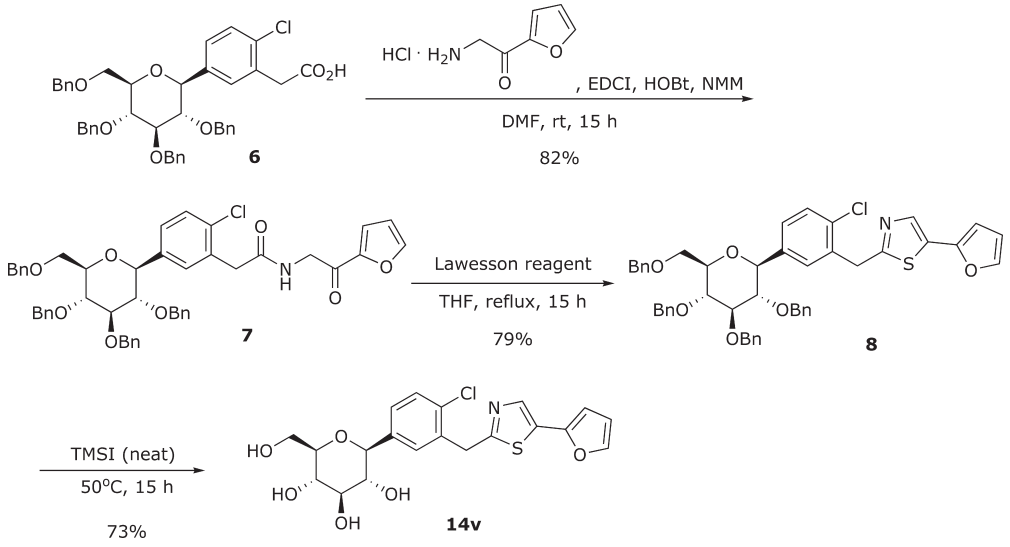


Figure 1. Exploration of C-glucoside bearing thiazole at the distal ring.

Scheme 1



group at the position appears to decrease activity (**14f**, IC_{50} = 51.4 nM), whereas the conjugated alkenyl group exemplified in **14g** restores inhibitory activity against *h*SGLT2 (IC_{50} = 4.69 nM). Carbocycle at the position also appears to be tolerant (**14i**, IC_{50} = 9.71 nM). Alkoxy groups on the thiazole were also explored. These alkoxides (**14k**−**14n**) appear to be tolerated across the board (IC_{50} = 8.2 nM−19.6 nM). Meanwhile, isosteric replacement of alkoxy groups with alkylthio groups for this region appears to diminish inhibitory activity in approximately 3-fold, primarily implying that the bulky sulfur atom is less favored at this location. Replacement of this moiety with phenyl group **14s** improved the inhibitory activity against *h*SGLT2 (IC_{50} = 13.3 nM). Interestingly, the somewhat big biphenyl group **14u** showed good inhibitory activity (IC_{50} = 9.31 nM). Meanwhile, substitution of phenyl with 2-furyl (**14v**) or 3-thiophenyl (**14y**) dramatically improved the inhibitory activity against *h*SGLT2 (**14v**, IC_{50} = 0.720 nM vs **14y**, IC_{50} = 0.772 nM). Substituted thiophene **14z** decreases activity 10-fold. However, thiazole **14aa** and thiadiazole **14ab** demonstrated lower *in vitro* activity (**14aa**, IC_{50} = 3.13 nM; **14ab**, IC_{50} = 14.0 nM) than furan **14v** or thiophene **14y**, likely suggesting that increased polarity is not favorable in the region. This phenomenon is unequivocally observed in carboxylic acid **14ad**, which is completely devoid of any activity against *h*SGLT2 (IC_{50} > 10,000 nM).

It was encouraging to note that considerable inhibitory activity against *h*SGLT2 was already evident in compounds **14v** and **14y**. At this juncture, we proceeded to explore the C-4 region of the proximal phenyl ring by using 5-(furan-2-yl)thiazolyl and 5-(thiophen-3-yl)thiazolyl as lead scaffolds. The inhibitory activity data is shown in Table 2. The small methyl group (**15a** and **15b**) or other halogen atoms (**15c**−**15f**) appear to maintain a similar level of activity, albeit in slightly decreased potency. On the contrary, hydrogen **15i**, cyano **15j**, **15k**, or cyclopropyl **15l** demonstrated a loss of activity in an order of magnitude, suggesting that merely small lipophilic groups are favored at the C-4 position of the proximal phenyl ring.

Next, an isomeric thiazole series of compounds was explored by using 2-furanyl and 3-thiophenyl as lead scaffolds

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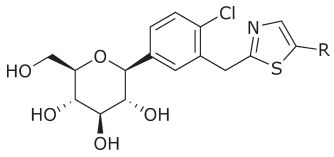
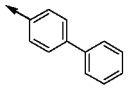
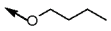
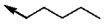
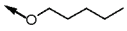
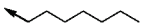
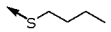
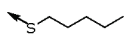
Table 1. *In Vitro* Screening Data for *h*SGLT2 Inhibitory Activity



compound	R	*h*SGLT2 IC_{50} (nM) [a]	compound	R	*h*SGLT2 IC_{50} (nM) [a]	compound	R	*h*SGLT2 IC_{50} (nM) [a]
1	-	0.49 ± 0.04 [b]	**14k**		18.8	**14u**		9.31
14a		16.7	**14l**		12.4	**14v**		0.720
14b		119	**14m**		19.6	**14w**		1.57
14c		13.3	**14n**		8.2	**14x**		1.14
14d		5.0	**14o**		30.9	**14y**		0.772
14e		12.4	**14p**		56.4	**14z**		13.3
14f		51.4	**14q**		58.7	**14aa**		3.13
14g		4.69	**14r**		18.9	**14ab**		14.0
14h		11.5	**14s**[c]		13.3	**14ac**		50.3
14i		9.71	**14t**		21.2	**14ad**		>10,000
14j		25.4						

[a] These data were obtained by single determinations. [b] The IC_{50} value was obtained by in-house multiple determinations. [c] Compound **14s** had been disclosed in WO2005012326.[22]

(**16a**−**16d**, Table 3). This series exhibited a slight decrease in inhibitory activities against *h*SGLT2 compared with that of the previous thiazole series. For instance, 2-(furan-2-yl)thiazol-5-yl **16a** (IC_{50} = 1.42 nM) showed a 2-fold decrease in activity than the counterpart **14v** (IC_{50} = 0.720 nM). As a more clear demonstration, two pairs of examples are presented, including furans **14w** (IC_{50} = 1.57 nM), **16b** (IC_{50} = 4.47 nM), and thiophenes **14x** (IC_{50} = 1.14 nM) and **16c** (IC_{50} = 2.79 nM), demonstrating close to a 3-fold decrease of activity, respectively.

Selected promising compounds (**14v** and **14y**) were tested on animal models for *in vivo* efficacy. Urinary glucose excretion was evaluated in normal SD rats. As shown in Figure 2, a single oral dose of thiazole **14y** (1 mg/kg or 10 mg/kg) increased urinary glucose excretion in normal SD rats, resulting in 160-fold (1 mg/kg) or 730-fold (10 mg/kg) elevation in glucose disposal relative to that of vehicle controls. However, a single oral dose of thiazole **14v** (1 mg/kg or 10 mg/kg) increased urinary glucose excretion in normal SD rats, resulting in 230-fold (1 mg/kg) or 700-fold (10 mg/kg) elevation in glucose disposal relative to that of vehicle controls. Urine volumes excreted in normal SD rats are also shown in Figure 2. Dapagliflozin **1** (1 mg/kg) caused increased urine volume over that of the vehicle by 5.7-fold, while thiazole **14y** (1 mg/kg or 10 mg/kg) increased urine volume by 2.5-fold (1 mg/kg) or by 4.2-fold, respectively. Thiazole **14v** increased urine volume to a lesser degree at a dose of 10 mg/kg (3.7-fold increase).

In order to further evaluate this series, the PK properties of **14y** and **14v** were measured in male SD rats. After oral administration of 5 mg/kg of **14y** to rats, a C_{max} of 1.54 μg/mL was obtained at 0.38 h. The elimination half-life of **14y**

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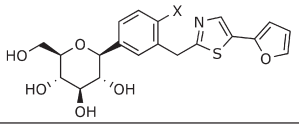
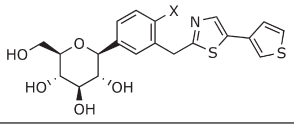
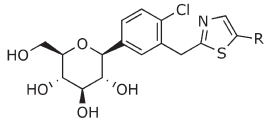
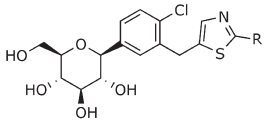
Table 2. *In Vitro* Inhibitory Activity against *h*SGLT2

X				
	compound	*h*SGLT2 IC_{50} (nM) [a]	compound	*h*SGLT2 IC_{50} (nM) [a]
Cl	**14v**	0.720	**14y**	0.772
Me	**15a**	1.43	**15b**	1.28
Br	**15c**	1.89	**15d**	6.36
F	**15e**	6.11	**15f**	7.36
CF_3	**15g**	9.73	**15h**	14.6
H	**15i**	22.6	—	—
CN	**15j**	36.7	**15k**	140
cyclopropyl	**15l**	54.8	—	—

[a] These data were obtained by single determinations.

Table 3. *In Vitro* Inhibitory Activity against *h*SGLT2

R				
	compound	*h*SGLT2 IC_{50} (nM) [a]	compound	*h*SGLT2 IC_{50} (nM) [a]
	**14v**	0.720	**16a**	1.42
	**14w**	1.57	**16b**	4.47
	**14x**	1.14	**16c**	2.79
	**14y**	0.772	**16d**	1.11

[a] These data were obtained by single determinations.

following oral administration was 2.51 h. Compound **14y** showed moderate oral bioavailability (F = 15%). Furanyl compound **14v** also demonstrated PK profiles similar to those of compound **14y**, showing slightly better oral bioavailability (F = 20.6%). For comparison, in-house oral administration of 5 mg/kg of dapagliflozin **1** to rats showed a C_{max} of 2.50 μg/mL at 0.67 h. The elimination half-life of **1** following oral administration was 4.01 h. Dapagliflozin **1** showed superior oral bioavailability (F = 88%). Therefore, it is concluded that the decreased *in vivo* efficacy of the current SGLT2 inhibitors **14v** and **14y**, compared with that of dapagliflozin **1**, should be attributed to the 1.5-fold lower *in vitro* potencies as well as their unfavorable PK properties.

In summary, the potential of metabolically more stable *C*-glucosides bearing a thiazole ring as antidiabetic agents was exploited. Among the compounds tested, thiazole containing furanyl moiety **14v** or thiophenyl moiety **14y** showed the best *in vitro* inhibitory activity against *h*SGLT2 in this series to date (IC_{50} = 0.720 nM for **14v**; IC_{50} = 0.772 nM for **14y**). These compounds demonstrated reasonable glucose excretion

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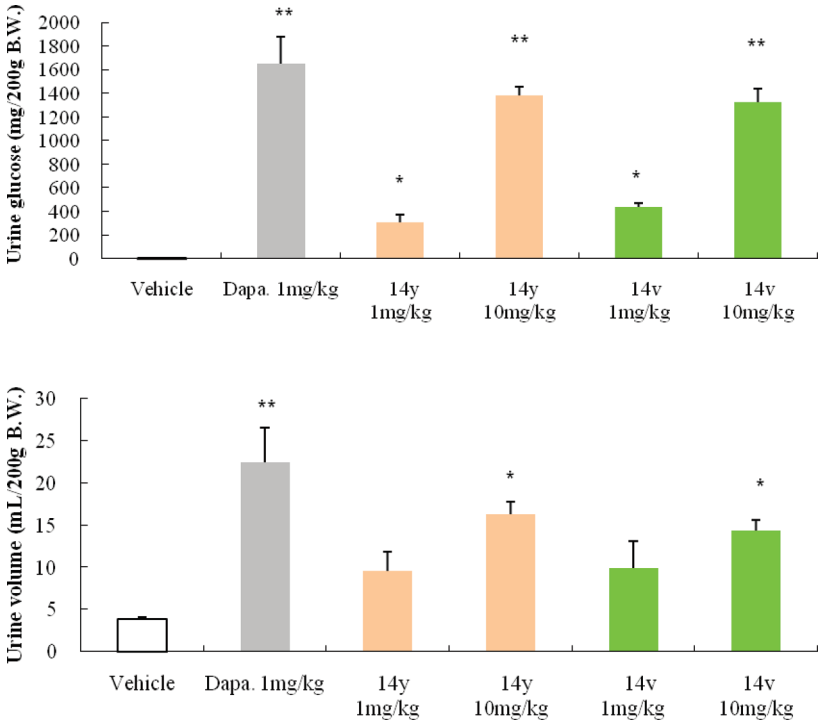

Figure 2. Effects of dapagliflozin, compound **14y** and **14v** on urine glucose excretion and urine volume in normal Sprague−Dawley rats. All results are expressed as means ± SEM. The statistical analysis was performed by one-way ANOVA followed by Dunnett's posthoc test. $*P < 0.05$ vs vehicle. $**P < 0.01$ vs vehicle.

and glucosuria in normal SD rats along with moderate PK profiles. Further work concerning optimization of SGLT2 inhibitors suitable for development is ongoing on the basis of this series.

SUPPORTING INFORMATION AVAILABLE

Experimental details. This material is available free of charge via the Internet at http://pubs.acs.org.

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