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## Synthesis and glycogen phosphorylase inhibitor activity of functionalized 1,4-benzodioxanes

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Dedicated to Professor Károly Lempert on the occasion of his 85<sup>th</sup> birthday.

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A series of novel 1,4-benzodioxanes **11a–e** and *rac*-**12a–d** carrying thiazolidine-2,4-dione moiety was synthesized and their glycogen phosphorylase inhibitor activity was also evaluated.

### 1. Introduction

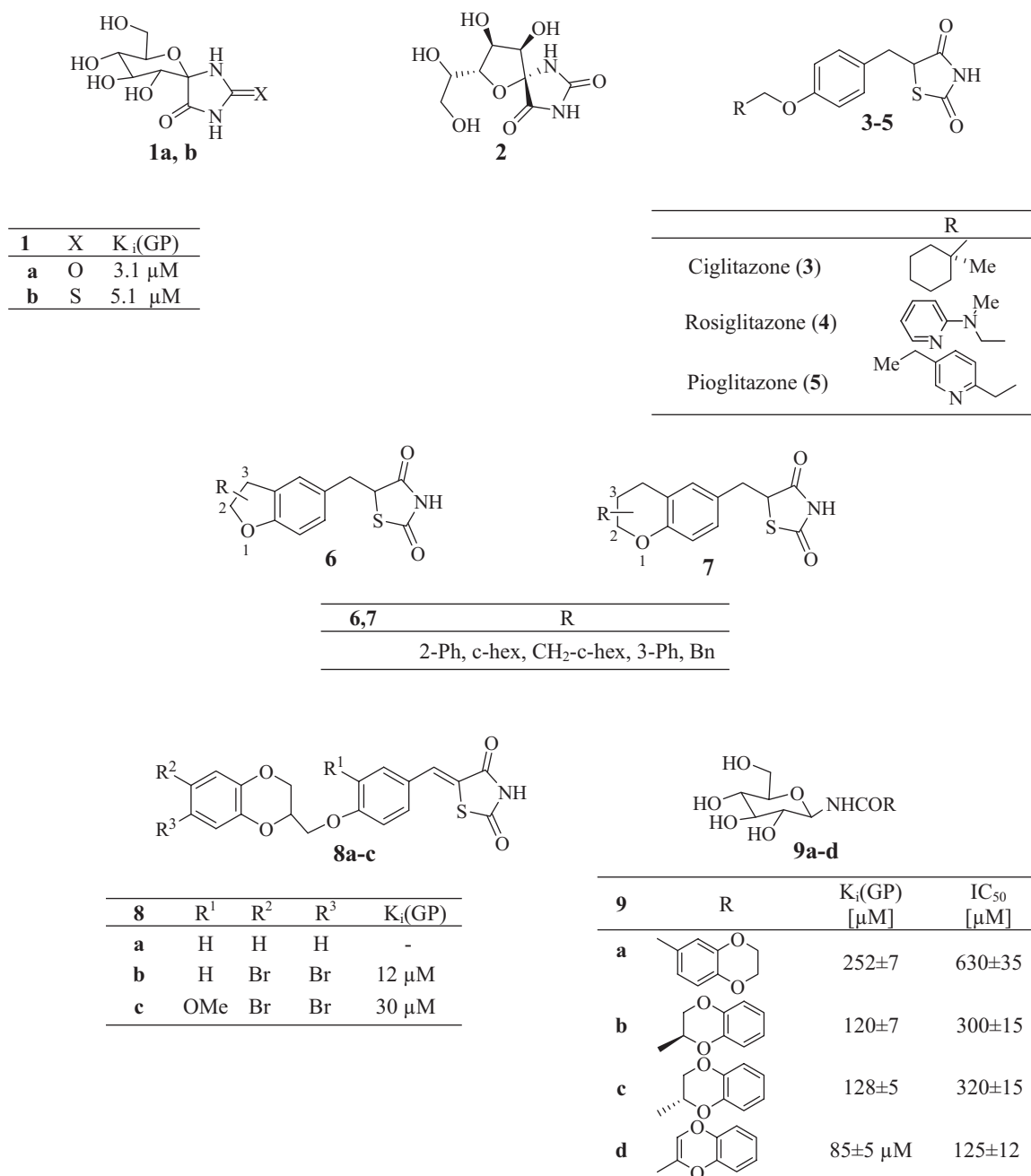
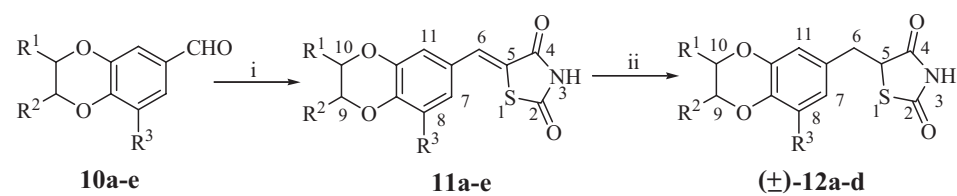
Type II diabetes is caused by abnormal insulin secretion and/or insulin resistance. Insufficient action of insulin in the liver results in increased blood sugar levels. Although the molecular etiology of this disease remains unknown, several classes of oral hypoglycaemic drugs [sulfonylureas, biguanides, thiazolidinediones (Cheng and Fantus 2005; Krentz and Balley 2005)] and  $\alpha$ -glycosidase inhibitors [acarbose, miglitol (Laar et al. 2003)] have been used as symptomatic treatments for type II diabetes. The numerous side effects observed in a large proportion of patients led to the investigation of the mechanism of hepatic glucose output (Agius 2007) which can be regulated by inhibition of glycogen phosphorylase (GP). Since this is the rate-limiting enzyme in glycogen degradation, it seemed reasonable to suppose that liver-specific GP inhibitors could be a powerful tool for controlling glucose levels in patients suffering from type II diabetes (Agius 2007; Morral 2003; Oikonomakos 2002; Oikonomakos and Somsák 2008; Somsák et al. 2008). Since several GP inhibitors carry a hydantoin (**1a**, **2**) or thiohydantoin (**1b**) moiety (Agasimundin et al. 1998; Ósz et al. 1999), one can assume that related heterocycles, such as glitazone-type molecules [citglitazone (**3**) (Shoda et al. 1982), rosiglitazone (**4**) (Cantello et al. 1994), and pioglitazone (**5**) (Ikeda et al. 1990), employed as oral hypoglycemics] and their dihydrobenzofuran (**6**), dihydrobenzopyran (**7**) and 1,4-benzodioxane (**8a**) analogues (Clark et al. 1991; de Nanteuil et al. 1995) may also possess a GP inhibitory effect (Fig. 1). Indeed, we have found recently that 1,4-benzodioxanes carrying a 5-arylidene-thiazolidine-2,4-dione (**8b**, **c**) or *N*- $\beta$ -D-glucopyranosylcarbamoyl (**9a–d**) moiety possess a significant GP inhibitory effect (Czakó et al. 2009; Juhász et al. 2007) (Fig. 1). Therefore, as a continuation of our investigation into the synthesis of 1,4-benzodioxane derivatives of potential hypoglycaemic agents, we prepared novel analogues of **8b** and **8c** in order to obtain further information about their structure-activity relationship (SAR).

### 2. Investigations, results and discussion

Thiazolidine-2,4-diones **11a–e** and **12a–d** were prepared from the corresponding 1,4-benzodioxane aldehydes **10a–e** in one and two steps respectively, as shown in Scheme 1. Condensation of the neat aldehydes **10a–e** with thiazolidine-2,4-dione and anhydrous sodium acetate at 110 °C for 10 min in a CEM-Discover MW reactor afforded **11a–e** 5-arylidene-2,4-thiazolidinediones in excellent yield (88–92%), whose catalytic hydrogenation in acetic acid at high pressure (12 atm) in the presence of 10% palladium/carbon resulted in the corresponding racemic dihydro-(**12a**, **b**) and diastereomeric tetrahydro-(**12c**, **d**) derivatives respectively. Since the latter compounds could not be separated by either crystallization or chromatography, they were characterized and tested as a 1:1 mixture of the diastereomers. The 1,4-benzodioxane aldehydes **10a** and **10e** were prepared as described in the literature (Kashima et al. 1987; Vallejos et al. 2005). 5-Methoxyprotochatechualdehyde (**13**) was alkylated with 1,2-dibromoethane in acetone in the presence of potassium carbonate resulting in 3-methoxy-4,5-ethylendioxybenzaldehyde (**10b**) in 81% yield as shown in Scheme 2. The 6- and 7-formyl-2-benzylidene-1,4-benzodioxanes (**10c**, **d**) of (*Z*)-geometry were prepared from 3- and 4-propargylprotochatechualdehydes (**14a**, **b**) and iodobenzene by Sonogashira coupling as published by Chowdhury et al. (Chowdhury et al. 1998) in moderate yields (42% and 35%, respectively) (Scheme 3).

The 1,4-benzodioxanes **11a–e** and **12a–d** were tested against unphosphorylated rabbit muscle GP (RMGPb) as described previously (Juhász et al. 2007; Ósz et al. 1999) and their inhibitory activities are given in Table 1.

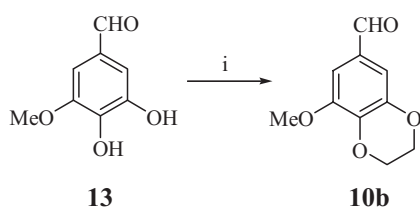
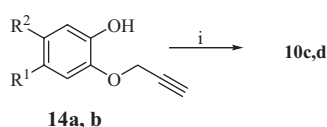
The IC<sub>50</sub> value of **11a**, possessing a 1,4-benzodioxane nucleus of half-chair conformation (entry 1), has clearly indicated that its inhibition action on RMGPb is significantly weaker than those of *rac*-**8b** and -**8c** 1,4-benzodioxanes possessing an aryl linker between their thiazolidine-2,4-dione and 1,4-benzodioxane moieties. It is also noteworthy that introduction of a methoxy or bromine group at C-8 of **11a** (**11b**, **11e**) decreased the GP inhibitor activity (entries 2 and 5), while a

Fig. 1: Structure of some hydantoin, thiohydantoin, thiazolidine-2,4-dione and *N*-acyl-β-D-glucopyranosylamine derivatives

10, 11	R <sup>1</sup>	R <sup>2</sup>	R <sup>3</sup>
a	H	H	H
b	H	H	OMe
c	(Z)-PhCH=	H	H
d	H	(Z)-PhCH=	H
e	H	H	Br

12	R <sup>1</sup>	R <sup>2</sup>	R <sup>3</sup>
a	H	H	H
b	H	H	OMe
c	Bn	H	H
d	H	Bn	H

Scheme 1: (i) thiazolidine-2,4-dione, NaOAc, MW, 110 °C; (ii) AcOH, Pd(C)/H<sub>2</sub>, 12 atm

Scheme 2: (i) BrCH<sub>2</sub>CH<sub>2</sub>Br, K<sub>2</sub>CO<sub>3</sub>, acetone, Δ

14	R <sup>1</sup>	R <sup>2</sup>
a	CHO	H
b	H	CHO

Scheme 3: (i) PhI, (PPh<sub>3</sub>)<sub>2</sub>PdCl<sub>2</sub>, CuCl<sub>2</sub>, Et<sub>3</sub>N, 110 °C

benzylidene group of (Z)-geometry at its C-10 or C-9 (**11c**, **11d**) significantly increased it (entries 3 and 4). Interestingly, the latter modification (**11a** → **11d**) makes the inhibition one order of magnitude stronger than that of **11c** and it possess a GP inhibitor activity comparable with those of *rac*-**8c** and **8c**. Finally, saturation of the exocyclic double bond of **11c** and **11d** by catalytic hydrogenation (**11c** → **12c**, **11d** → **12d**) also caused a significant decrease of their activities.

In good agreement with our earlier results observed in the series of *N*-(β-D-glucopyranosyl)amides **9a-d** (Czakó et al. 2009), these data also underline the importance of the π-donor ability of the aryl moiety of these compounds (**11c**, **d** > **12c**, **d** > **11a**, **b**, **e** ≅ **12a**, **b**) and the presence of an aryl linker between the thiazolidine-2,4-dione and 1,4-benzodioxane moieties of **8b** and **8c** (*K<sub>i</sub>*(GP): 12, 25, 30, 100 μM for **8b**, **11c**, **8c**, **11d**, resp.) which

**Table 1: GP inhibitor activities of functionalized 1,4-benzodioxanes 11a-e and 12a-d**

Compd.	<i>K<sub>i</sub></i> (μM)*	IC <sub>50</sub> (μM)
11a	233 ± 11.0	700 ± 6.5
11b	243 ± 11.0	730 ± 6.0
11c	25 ± 3.0	75 ± 4.2
11d	100 ± 8.5	300 ± 5.0
11e	283 ± 12.2	850 ± 8.2
12a	200 ± 9.5	600 ± 5.8
12b	257 ± 11.8	770 ± 7.6
12c	217 ± 10.4	650 ± 6.0
12d	217 ± 10.4	650 ± 6.0

\* Calculated by the Cheng-Prusoff equation: *K<sub>i</sub>* = IC<sub>50</sub>/(1+[S]/*K<sub>m</sub>*) (Cheng and Prusoff 1973)

seems to be an important prerequisite for glycogen phosphorylase inhibitor activity.

### 3. Experimental

Analytical TLC was performed on Kieselgel 60 plates F<sub>254</sub> (Merck). For workup, the solutions were dried (MgSO<sub>4</sub>) and concentrated in vacuum. <sup>1</sup>H and <sup>13</sup>C NMR spectra were recorded on Bruker WP-200 and Bruker WP-360 spectrometers. The chemical shifts are given in δ (ppm) and spin-spin coupling constants (*J*) in Hz. Microanalyses were performed on a Carlo-Erba Tpy 1106 analyser. Compounds listed below gave satisfactory C, H, N analysis (±0.4%). The reagents were purchased from Sigma-Aldrich. The arylaldehydes **10a**, **10e**, **14a** and **14b** were prepared according to the respective procedures reported in the literature (Kashima et al. 1987; Plourde and Spaetzel 2002; Vallejos et al. 2005; Wei et al. 2006).

#### 3.1. General procedure for the preparation of 11a-e

A mixture of aldehyde **10a-e** (0.5 mmol), thiazolidine-2,4-dione (2 mmol) and anhydrous sodium acetate (0.2 mmol) was heated in a CEM-Discover MW reactor at 110 °C for 10 min. Then water was added to the reaction mixture and the solid product was filtered off, washed with chloroform and dried to give **11a-e** (Table 2).

**Table 2: Physical and selected NMR data, and yields for functionalized 1,4-benzodioxanes 11a-e and 12a-d**

Compd.	Yield %	m.p. °C	Molecular Form.	Selected <sup>1</sup> H-NMR data	Selected <sup>13</sup> C-NMR data
11a	88	>250	C <sub>12</sub> H <sub>9</sub> NO <sub>4</sub> S	7.67 (1H, s, H-6)	119.53 C-5; 130.23 C-6
11b	91	192–198	C <sub>13</sub> H <sub>11</sub> NO <sub>5</sub> S	7.63 (1H, s, H-6)	121.03 C-5; 131.99 C-6
11c	89	207–212	C <sub>19</sub> H <sub>13</sub> NO <sub>4</sub> S	4.18 (1H, s, PhCH=); 7.52 (1H, s, H-6)	107.43 PhCH=; 122.61 C-5; 142.67 C-10; 146.27 C-6
11d	90	216–221	C <sub>19</sub> H <sub>13</sub> NO <sub>4</sub> S	5.88 (1H, s, PhCH=); 7.60 (1H, s, H-6)	107.79 PhCH=; 124.61 C-5; 143.56 C-9; 144.29 C-6
11e	92	>250	C <sub>12</sub> H <sub>8</sub> BrNO <sub>4</sub> S	7.24 (1H, s, H-6)	121.98 C-5; 128.18 C-6
12a	88	158–161	C <sub>12</sub> H <sub>11</sub> NO <sub>4</sub> S	2.86 (1H, dd, <i>J</i> = 10.0 and <i>J</i> = 14.4, H-6 <sub>A</sub> ); 3.27 (1H, dd, <i>J</i> = 4.0 and <i>J</i> = 14.4, H-6 <sub>B</sub> ); 4.73 (1H, dd, <i>J</i> = 4.0 and <i>J</i> = 10.0, H-5)	35.37 C-6; 52.20 C-5
12b	79	129–131	C <sub>13</sub> H <sub>13</sub> NO <sub>5</sub> S	2.87 (1H, dd, <i>J</i> = 9.8 and <i>J</i> = 14.2, H-6 <sub>A</sub> ); 3.27 (1H, dd, <i>J</i> = 4.2 and <i>J</i> = 14.0, H-6 <sub>B</sub> ); 4.76 (1H, dd, <i>J</i> = 4.0 and <i>J</i> = 9.8, H-5)	37.46 C-6; 53.65 C-5
12c	35	217–226	C <sub>19</sub> H <sub>17</sub> NO <sub>4</sub> S	2.78–2.94 (3H, m, H-6 <sub>A</sub> and PhCH <sub>2</sub> -); 3.17–3.29 (1H, m, H-6 <sub>B</sub> ); 3.83–3.92 (1H, m, H-10); 4.58–4.61 (1H, m, H-5)	36.34 PhCH <sub>2</sub> -; 37.38 C-6; 54.77 C-5; 73.21 C-10
12d	39	231–239	C <sub>19</sub> H <sub>17</sub> NO <sub>4</sub> S	2.80 (1H, dd, <i>J</i> = 10.1 and <i>J</i> = 14.0, H-6 <sub>A</sub> ); 2.95–2.97 (2H, m, PhCH <sub>2</sub> -); 3.31 (1H, dd, <i>J</i> = 3.6 and <i>J</i> = 14.0, H-6 <sub>B</sub> ); 3.89–3.94 (1H, m, H-9); 4.49 (1H, dd, <i>J</i> = 3.9 and <i>J</i> = 10.1, H-5)	36.38 PhCH <sub>2</sub> -; 37.99 C-6; 55.96 C-5; 73.18 C-9

### 3.2. General procedure for the preparation of rac-12a-d

The arylidene derivatives (**11a-d**) (0.5 mmol) were hydrogenated in acetic acid (25 mL) in the presence of 10% Pd(C) (200 mg) at 12 atm pressure until the pressure was stabilized. The catalyst was filtered off on celite, the solvent was evaporated, and the residue was purified by column chromatography (CC) on silica gel using a mixture of hexane and ethyl-acetate (1:1) as eluent (Table 2).

### 3.3. 3-Benzylidene-6-formyl-1,4-benzodioxane (10c) and 2-benzylidene-6-formyl-1,4-benzodioxane (10d)

A mixture of iodobenzene (7.35 mmol), (Ph<sub>3</sub>P)<sub>2</sub>PdCl<sub>2</sub> (0.34 mmol) and CuI (0.63 mmol) in anhydrous Et<sub>3</sub>N was stirred under nitrogen at room temperature for 30 min, and then propargylprotocatechualdehyde (**14a** or **14b**) (10.2 mmol) in anhydrous Et<sub>3</sub>N (50 mL) and THF (15 mL) was added dropwise. The reaction mixture was stirred at room temperature for 1 h and then at 100 °C for 20 h. The precipitation was filtered off and the solvent was evaporated. The residue was dissolved in ethyl acetate (150 mL) and washed with water (3 × 100 mL). The organic phase was dried, evaporated and then purified by CC on silica gel using a mixture of hexane and ethyl acetate (4:1) as eluent.

**10c**: Yield 35%; m.p. 88–91 °C; <sup>1</sup>H NMR: δ (ppm): 9.87 (1H, s, CHO), 7.72–7.26 (8H, m, Ar-H), 5.96 (1H, s, CH), 4.82 (2H, s, CH<sub>2</sub>).

**10d**: Yield 42%; m.p. 84–85 °C; <sup>1</sup>H NMR: δ (ppm): 9.83 (1H, s, CHO), 7.67–7.19 (8H, m, Ar-H), 5.65 (1H, s, CH), 4.61 (2H, s, CH<sub>2</sub>).

### 3.4. 6-Formyl-8-methoxy-1,4-benzodioxane (10b)

To a solution of **13** (7.14 mmol) in dry acetone (60 mL) anhydrous K<sub>2</sub>CO<sub>3</sub> (24 mmol) was added and stirred for 10 min at room temperature. Then 1,2-dibromoethane (17.4 mmol) was added to the solution, and the reaction mixture was refluxed with stirring and the progress of the reaction was followed by TLC. After 3 days, K<sub>2</sub>CO<sub>3</sub> was filtered off and the solvent was evaporated. The residue was purified by CC, using hexane and ethyl-acetate (3:1) as eluent to give **10b** as white crystals (81% yield, m.p.: 69–72 °C). <sup>1</sup>H-NMR: δ (ppm): 9.79 (1H, s, CHO), 7.08 (2H, s, Ar-H), 4.43–4.39 (2H, m, CH<sub>2</sub>), 4.33–4.29 (2H, m, CH<sub>2</sub>), 3.95 (1H, s, OMe).

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