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ORIGINAL ARTICLE

Synthesis and antidiabetic activity of some new chromonyl-2,4-thiazolidinediones

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Abstract

A series of chromonyl-2,4-thiazolidinediones/imidazolidinediones/2-thioxo-imidazolidine-4-ones (**IIIa–i**, **IVa–i**) was prepared by Knoevenagel reaction of 2,4-thiazolidinedione/2,4-imidazolidinedione/2-thioxo-imidazolidine-4-one (**IIa–c**) with 2/3-formyl chromone (**Ia–b**) and then alkylation with methyl/ethyl iodide. The prepared compounds were tested for their insulinotropic activities in INS-1 cells. Compounds ıV**b** and ıV**c** (at lower concentration, 1 µg/mL) were able to increase insulin release in the presence of 5.6 mmol/L glucose." should be written as "Compounds IVb and IVc (at lower concentration, 1 µg/mL) and also IIId and IIIg (at higher concentration) were able to increase insulin release in the presence of 5.6 mmol/L glucose. Compounds ıV**b** and ıV**c** (at lower concentration, 1 µg/mL) were able to increase insulin release in the presence of 5.6 mmol/L glucose.

Keywords: *Chromone; 2,4-thiazolidinediones; chromonyl-2,4-thiazolidinediones; antidiabetic activity; synthesis*

Introduction

Diabetes mellitus (DM) is a common chronic metabolic disease characterized by high levels of glucose in the blood and associated with serious long-term complications, such as neuropathy, nephropathy, retinopathy, cataracts, accelerated atherosclerosis and increased risk of myocardial infarction, stroke, and amputation[1]. 2,4-Thiazolidinediones (2,4-TZDs) are a new class of antidiabetic agents that increase insulin sensitivity, enhance glucose control, improve the lipid profile, suppress inflammatory cytokines, and improve vascular structure and function[2]. Chromones, occurring widely throughout the plant kingdom, are one of the most representative families of plant secondary metabolites and display a remarkable spectrum of biological activities such as anticancer, antimicrobial, and antidiabetic activities[3–6].

In our previous studies, we synthesized novel furochromonyl-TZDs[5] and chromonyl-TZDs[6] and tested for their antidiabetic activity. In this study, we describe further modifications of the 2,4-TZD derivatives containing the chromone ring. The synthesized compounds were tested for their insulinotropic activities in INS-1 cells.

Materials and methods

Chemical analysis

Melting points were measured on an electrothermal 9100 type apparatus (electrothermal Engineering Ltd., Essex, UK) and are uncorrected. All instrumental analyses were performed in the Central Laboratory of the Faculty of Pharmacy of Ankara University. Infrared (IR) spectra were recorded as potassium bromide disks with a Jasco FT/IR-420 spectrometer (Easton, MD, USA). ^{1}H nuclear magnetic resonance (NMR) spectra were measured with a Varian Mercury 400 MHz FT-NMR spectrometer (Varian Inc., Palo Alto, CA, USA) in dimethylsulfoxide (DMSO)-d_6 and $CDCl_3$; all chemical shifts are reported as δ (ppm) values. Mass spectra were recorded on a Waters ZQ Micromass LC-MS spectrometer (Waters Corporation, Milford, MA, USA) using the electrospray ionization (ESI) (+) method. Elementary analyses were performed on a Leco 932 CHNS analyzer (Leco-932, St. Joseph, MI, USA), and satisfactory results within ±0.4% of calculated values (C, H, N) were obtained. For the chromatographic analysis, Merck silica gel 60 (230–400 mesh ASTM (American Society for Testing and Materials)) was used. The chemical reagents used in the synthesis were purchased from E. Merck (Darmstadt, Germany),

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Fluka (Buchs, Switzerland), and Aldrich (Milwaukee, MI, USA). 2,4-TZD (**II**)[7], *o*-acetoxy acetophenone[8], α-acetyl-2-hydroxy acetophenone[9], 2-methyl chromone[10], and 2-formyl chromone[11] were synthesized according to the literature.

Chemical synthesis

General synthesis of compounds IIIa–c, IVa–c

A mixture of 2/3-formyl chromone (**Ia–b**) (0.001 mol) and thiazolidine-2,4-dione/imidazolidine-2,4-dione/2-thioxo-imidazolidine-4-one (**IIa–c**) (0.001 mol) was refluxed in the presence of 1 mL glacial acetic acid and sodium acetate (0.001 mol). The crude product was crystallized from dimethylformamide (DMF).

5-(4-Oxo-4H-chromen-2-yl methylene)-thiazolidine-2,4-dione) (IIIa) Yield: 57.36%, mp: 316°C, ^{1}H NMR (DMSO-d_6): δ = 6.93 (s, 1H, =C**H**), 7.53 (t, 1H, Jo = 7.60, 6-**H**), 7.64 (s, 1H, 3-**H**), 7.75 (d, 1H, $J_{8,7}$ = 8.80 Hz, 8-**H**), 7.86 (t, 1H, Jo = 7.20, 7-**H**), 8.03 (d, 1H, $J_{5,6}$ = 7.60 Hz, 5-**H**), 12.82 (s, 1H, N**H**), MS (ESI+) *m/z* (rel intensity): 274 (M + 1, 100), Anal. for $C_{13}H_7NO_4S{\cdot}1.5H_2O$: Calc. C, 52.00; H, 3.33; N, 4.67; S, 10.67%. Found C, 52.03; H, 3.23; N, 5.09; S, 10.04%.

5-(4-Oxo-4H-chromen-2-yl methylene)-imidazolidine-2,4-dione) (IIIb) Yield: 30.81%, mp: 333°C, ^{1}H NMR (DMSO-d_6): δ = 6.26 (s, 1H, =C**H**), 6.66 (s, 1H, 3-**H**), 7.47 (ψt, 1H, Jo = 7.20 and 7.60 Hz, 6-**H**), 7.81 (ψt, 1H, Jo = 8.00 and 7.60 Hz, 7-**H**), 7.99 (d, 1H, $J_{8,7}$ = 7.60 Hz, 8-**H**), 8.24 (d, 1H, $J_{5,6}$ = 8.80 Hz, 5-**H**), 10.99 (s, 1H, N**H**), 11.60 (s, 1H, N**H**), MS (ESI+) *m/z* (rel intensity): 257 (M + 1, 100), Anal. for $C_{13}H_8N_2O_4{\cdot}0.9H_2O$: Calc. C, 57.31; H, 3.60; N, 10.29%. Found C, 57.60; H, 3.51; N, 9.95%.

5-(4-Oxo-4H-chromen-2-yl methylene)-2-thioxo-imidazolidine-4-one) (IIIc) Yield: 56.50%, mp: 339°C, ^{1}H NMR (DMSO-d_6): δ = 6.29 (s, 1H, =C**H**), 6.74 (s, 1H, 3-**H**), 7.45 (ψt, 1H, Jo = 7.20 and 7.60 Hz, 6-**H**), 7.80 (td, 1H, Jo = 6.80 and 8.80 Hz, Jm = 2.00 Hz, 7-**H**), 7.96 (dd, 1H, $J_{8,7}$ = 7.60 Hz, $J_{8,6}$ = 1.60 Hz, 8-**H**), 8.18 (d, 1H, $J_{5,6}$ = 8.40 Hz, 5-**H**), 12.42 (s, 1H, N**H**), 12.69 (s, 1H, N**H**), MS (ESI+) *m/z* (rel intensity): 273 (M + 1, 100), Anal. for $C_{13}H_8N_2O_3S{\cdot}H_2O$: Calc. C, 53.79; H, 3.45; N, 9.66; S, 11.03%. Found C, 53.80; H, 3.45; N, 9.83; S, 11.21%.

*5-(4-Oxo-4H-chromen-3-yl methylene)-thiazolidine-2,4-dione) (**IVa**)* Yield: 87.19%, mp: 311°C (Ref. 12 mp: 271°C), ^{1}H NMR (DMSO-d_6): δ = 7.56 (ψt, 1H, Jo = 7.20 and 7.80 Hz, 6-**H**), 7.61 (s, 1H, =C**H**), 7.73 (d, 1H, $J_{8,7}$ = 8.40 Hz, 8-**H**), 7.88 (td, 1H, Jo = 7.80 and 8.00 Hz, Jm = 1.60 Hz, 7-**H**), 8.13 (dd, 1H, $J_{5,6}$ = 8.40 Hz, $J_{5,7}$ = 1.60 Hz, 5-**H**), 8.85 (s, 1H, 2-**H**), 12.45 (s, 1H, N**H**), MS (ESI+) *m/z* (rel intensity): 274 (M + 1, 100), Anal. for $C_{13}H_7NO_4S$: Calc. C, 57.14; H, 2.58; N, 5.13; S, 11.73%. Found C, 57.30; H, 2.08; N, 5.19; S, 11.61%.

5-(4-Oxo-4H-chromen-3-yl methylene)-imidazolidine-2,4-dione) (IVb) Yield: 86.17%, mp: 363°C (Ref. 13 mp: 345–348°C), ^{1}H NMR (DMSO-d_6): δ = 6.35 (s, 1H, =C**H**), 7.55 (ψt, 1H, Jo = 7.20 and 7.80 Hz, 6-**H**), 7.74 (d, 1H, $J_{8,7}$ = 7.60 Hz, 8-**H**), 7.87 (td, 1H, Jo = 7.60 and 8.00 Hz, Jm = 1.60 Hz, 7-**H**), 8.14 (dd, 1H, $J_{5,6}$ = 8.00, $J_{5,7}$ = 1.60 Hz, 5-**H**), 8.76 (s, 1H, 2-**H**), 10.40 (s, 1H, N**H**), 11.30 (s, 1H, N**H**), MS (ESI+) *m/z* (rel intensity): 257 (M + 1, 100), Anal. for $C_{13}H_8N_2O_4{\cdot}0.1H_2O$: Calc. C, 60.51; H, 3.18; N, 10.86%. Found C, 60.46; H, 3.11; N, 11.05%.

5-(4-Oxo-4H-chromen-3-yl methylene)-2-thioxo-imidazolidine-4-one) (IVc) Yield: 91.78%, mp: 326°C (Ref. 14 mp: 292–295°C), ^{1}H NMR (DMSO-d_6): δ = 6.37 (s, 1H, =C**H**), 7.57 (ψt, 1H, Jo = 7.40 and 7.60 Hz, 6-**H**), 7.75 (d, 1H, $J_{8,7}$ = 8.00 Hz, 8-**H**), 7.88 (td, 1H, Jo = 7.60 and 8.00 Hz, Jm = 1.60 Hz, 7-**H**), 8.17 (dd, 1H, $J_{5,6}$ = 8.00 Hz, $J_{5,7}$ = 1.60 Hz, 5-**H**), 8.92 (s, 1H, 2-**H**), 11.81 (s, 1H, N**H**), 12.43 (s, 1H, N**H**), MS (ESI+) *m/z* (rel intensity): 273 (M + 1, 100), Anal. for $C_{13}H_8N_2O_3S$: Calc. C, 57.35; H, 2.96; N, 10.29; S, 11.78%. Found C, 57.22; H, 2.93; N, 10.43; S, 11.90%.

General synthesis of compounds IIId, IIIf, IIIh, IVd, IVf, IVh

Compounds **IIIa–c**/**IVa–c** (0.0004 mmol) and anhydrous sodium carbonate (0.0004 mmol) were dissolved in 3 mL DMF. Methyl iodide (0.0008 mmol) was added to this mixture and stirred at 40°C. The reaction mixture was poured on to ice. The residue was filtered off. The filtrate was purified by column chromatography using silica gel 60 (230–400 mesh ASTM) as the adsorbent and petroleum ether:chloroform (1:1) as the eluent.

3-Methyl-5-(4-oxo-4H-chromen-2-yl methylene)-thiazolidine-2,4-dione) (IIId) Yield: 41.85%, mp: 275°C, ^{1}H NMR (DMSO-d_6): δ = 3.12 (s, 3H, -C**H$_3$**), 6.97 (s, 1H, =C**H**), 7.54 (ψt, 1H, Jo = 7.20 and 7.60 Hz, 6-**H**), 7.77 (s, 1H, 3-**H**), 7.78 (d, 1H, $J_{8,7}$ = 8.40 Hz, 8-**H**), 7.87 (ψt, 1H, Jo = 8.40 and 8.80 Hz, 7-**H**), 8.04 (d, 1H, $J_{5,6}$ = 8.00 Hz, 5-**H**), MS (ESI+) *m/z* (rel intensity): 288 (M + 1, 100), Anal. for $C_{14}H_9NO_4S$: Calc. C, 58.53; H, 3.16; N, 4.88; S, 11.16%. Found C, 58.59; H, 3.13; N, 4.92; S, 10.85%.

1,3-Dimethyl-5-(4-oxo-4H-chromen-2-yl methylene)-imidazolidine-2,4-dione) (IIIf) Yield: 16.23%, mp: 208°C, ^{1}H NMR ($CDCl_3$): δ = 3.19 (s, 3H, -C**H$_3$**), 3.56 (s, 3H, -C**H$_3$**), 6.42 (s, 1H, =C**H**), 6.45 (s, 1H, 3-**H**), 7.45 (m, 2H, 6-**H** and 8-**H**), 7.72 (td, 1H, Jo = 6.80 and 8.80 Hz, Jm = 1.60 Hz, 7-**H**), 8.22 (dd, 1H, $J_{5,6}$ = 8.00 Hz, $J_{5,7}$ = 1.20 Hz, 5-**H**), MS (ESI+) *m/z* (rel intensity): 285 (M + 1, 100), Anal. for $C_{15}H_{12}N_2O_4$: Calc. C, 63.38; H, 4.25; N, 9.85%. Found C, 63.06; H, 4.06; N, 9.67%.

3-Methyl-2-(methylthio)-5-(4-Oxo-4H-chromen-2-yl methylene)-imidazolidine-4-one) (IIIh) Yield: 72.53%, mp: 292°C, ^{1}H NMR ($CDCl_3$): δ = 2.80 (s, 3H, -C**H$_3$**), 3.19 (s, 3H, -C**H$_3$**), 6.62 (s, 1H, =C**H**), 7.39 (ψt, 1H, Jo = 7.20 and 8.00 Hz, 6-**H**), 7.47 (d, 1H, $J_{8,7}$ = 8.00 Hz, 8-**H**), 7.62 (s, 1H, 3-**H**), 7.67 (td, 1H, Jo = 7.20 and 7.60 Hz, Jm = 1.60 Hz, 7-**H**), 8.20 (dd, 1H, $J_{5,6}$ = 8.40 Hz, $J_{5,7}$ = 1.60 Hz, 5-**H**), MS (ESI+) *m/z* (rel intensity): 301 (M + 1, 100), Anal. for $C_{15}H_{12}N_2O_3S$: Calc. C, 59.99; H, 4.03; N, 9.33; S, 10.68%. Found C, 60.30; H, 4.22; N, 9.16; S, 10.35%.

3-Methyl-5-(4-oxo-4H-chromen-3-yl methylene)-thiazolidine-2,4-dione) (IVd) Yield: 78.95%, mp: 249°C, ^{1}H NMR (DMSO-d_6): δ = 3.09 (s, 3H, -C**H$_3$**), 7.58 (ψt, 1H, Jo = 7.20 and 8.00 Hz, 6-**H**), 7.72 (s, 1H, =C**H**), 7.75 (d, 1H, $J_{8,7}$ = 8.80 Hz, 8-**H**), 7.89 (ψt, 1H, Jo = 7.60 and 8.00 Hz, 7-**H**), 8.14 (dd, 1H, $J_{5,6}$ = 7.60 Hz, $J_{5,7}$ = 1.60 Hz, 5-**H**), 8.93 (s, 1H, 2-**H**), MS (ESI+) *m/z* (rel intensity): 288 (M + 1, 25), 310 (100), Anal. for $C_{14}H_9NO_4S$: Calc. C, 58.53; H, 3.16; N, 4.88; S, 11.16%. Found C, 58.63; H, 3.39; N, 4.87; S, 11.06%.

1,3-Dimethyl-5-(4-oxo-4H-chromen-3-yl methylene)-imidazolidine-2,4-dione) (IVf) Yield: 49.58%, mp: 162°C, ^{1}H NMR (DMSO-d_6): δ = 2.96 (s, 3H, -C**H**$_3$), 2.99 (s, 3H, -C**H**$_3$), 6.35 (s, 1H, =C**H**), 7.51 (ψt, 1H, Jo = 7.20 and 7.40 Hz, 6-**H**), 7.68 (d, 1H, $J_{8,7}$ = 8.00 Hz, 8-**H**), 7.82 (td, 1H, Jo = 7.80 and 7.80 Hz, Jm = 1.20 Hz, 7-**H**), 8.08 (dd, 1H, $J_{5,6}$ = 8.40 Hz, $J_{5,7}$ = 1.20 Hz, 5-**H**), 8.56 (s, 1H, 2-**H**), MS (ESI+) *m/z* (rel intensity): 285 (M + 1, 35), 307 (100), Anal. for $C_{15}H_{12}N_2O_4$: Calc. C, 63.38; H, 4.25; N, 9.85%. Found C, 63.24; H, 4.56; N, 9.75%.

3-Methyl-2-(methylthio)-5-(4-oxo-4H-chromen-3-yl methylene)-imidazolidine-4-one) (IVh) Yield: 58.48%, mp: 277°C, ^{1}H NMR (DMSO-d_6): δ = 2.74 (s, 3H, -C**H**$_3$), 3.09 (s, 3H, -C**H**$_3$), 7.01 (s, 1H, =C**H**), 7.56 (ψt, 1H, Jo = 6.80 and 8.00 Hz, 6-**H**), 7.73 (d, 1H, $J_{8,7}$ = 8.40 Hz, 8-**H**), 7.87 (ψt, 1H, Jo = 7.20 and 7.60 Hz, 7-**H**), 8.15 (d, 1H, $J_{5,6}$ = 7.60 Hz, 5-**H**), 9.69 (s, 1H, 2-**H**), MS (ESI+) *m/z* (rel intensity): 301 (M + 1, 35), 323 (100), Anal. for $C_{15}H_{12}N_20_3S$: Calc. C, 59.99; H, 4.03; N, 9.33; S, 10.68%. Found C, 60.01; H, 4.19; N, 9.15; S, 10.49%.

General synthesis of compounds IIIe, IIIg, IIIi, IVe, IVg, IVi

Compounds **IIIa–c/IVa–c** (0.0008 mmol) and anhydrous sodium carbonate (0.0016 mmol) were dissolved in 5 mL DMF. Ethyl iodide (0.0032 mmol) was added to this mixture and stirred at 40°C. The reaction mixture was poured on to ice. The residue was filtered off. The filtrate was purified by column chromatography using silica gel 60 (230–400 mesh ASTM) as the adsorbent and petroleum ether:chloroform (1:1) as the eluent.

3-Ethyl-5-(4-oxo-4H-chromen-2-yl methylene)-thiazolidine-2,4-dione) (IIIe) Yield: 49.88%, mp: 268°C, ^{1}H NMR (DMSO-d_6): δ = 1.18 (ψt, 3H, J = 6.80 and 7.20 Hz, -C**H**$_3$), 3.70 (q, 2H, J = 7.40 Hz and 7.60 Hz, -C**H**$_2$-), 6.97 (s, 1H, =C**H**) 7.54 (ψt, 1H, Jo = 7.20 and 7.60 Hz, 6-**H**), 7.78 (m, 2H, 3-**H** and 8-**H**), 7.87 (ψt, 1H, Jo = 7.40 and 8.20 Hz, 7-**H**), 8.04 (d, 1H, $J_{5,6}$ = 8.00 Hz, 5-**H**), MS (ESI+) *m/z* (rel intensity): 302 (M + 1, 100), Anal. for $C_{15}H_{11}NO_4S$: Calc. C, 59.79; H, 3.68; N, 4.65; S, 10.64%. Found C, 59.93; H, 3.66; N, 4.72; S, 10.16%.

3-Ethyl-5-(4-oxo-4H-chromen-2-yl methylene)-imidazolidine-2,4-dione) (IIIg) Yield: 18.03%, mp: 115°C, ^{1}H NMR ($CDCl_3$): δ = 1.26-1.31 (6H, m, -C**H**$_3$), 3.72 (q, 2H, J = 7.20 and 7.60 Hz, -C**H**$_2$-), 4.19 (q, 2H, J = 7.20 and 7.60 Hz, -C**H**$_2$-), 6.40 (s, 1H, =C**H**), 6.47 (s, 1H, 3-**H**), 7.45 (m, 1H, 6-**H** and 8-**H**), 7.72 (ψt, 1H, Jo = 7.20 and 8.00 Hz, 7-**H**), 8.23 (d, 1H, $J_{5,6}$ = 7.20 Hz, 5-**H**), MS (ESI+) *m/z* (rel intensity): 313 (M + 1, 100), Anal. for $C_{17}H_{16}N_2O_4$: Calc. C, 65.38; H, 5.13; N, 8.97%. Found C, 65.60; H, 5.15; N, 8.55%.

3-Ethyl-2-(ethylthio)-5-(4-oxo-4H-chromen-3-yl methylene)-imidazolidine-4-one) (IIIi) Yield: 71.87%, mp: 160°C, ^{1}H NMR (DMSO-d_6): δ = 1.18 (ψt, 3H, J = 6.80 and 7.20 Hz, -C**H**$_3$), 1.50 (ψt, 3H, J = 7.20 and 7.60 Hz, -C**H**$_3$), 3.43 (q, 2H, J = 6.80 and 7.60 Hz, -C**H**$_2$-), 3.59 (q, 2H, J = 6.80 and 7.60 Hz, -C**H**$_2$-), 6.49 (s, 1H, =C**H**), 7.36 (s, 1H, 3-**H**), 7.50 (ψt, 1H, Jo = 7.20 and 7.60 Hz, 6-**H**), 7.62 (d, 1H, $J_{8,7}$ = 8.40 Hz, 8-**H**), 7.83 (t, 1H, Jo = 7.80, 7-**H**), 8.03 (d, 1H, $J_{5,6}$ = 7.60 Hz, 5-**H**), MS (ESI+) *m/z* (rel intensity): 329 (M + 1, 100), Anal. for $C_{17}H_{16}N_2O_3S$. $0.1H_2O$: Calc. C, 61.86; H, 4.91; N, 8.49; S, 9.70%. Found C, 61.62; H, 5.06; N, 8.48; S, 9.56%.

3-Ethyl-5-(4-oxo-4H-chromen-3-yl methylene)-thiazolidine-2,4-dione) (IVe) Yield: 77.10%, mp: 233°C, ^{1}H NMR (DMSO-d_6): δ = 1.12 (t, 3H, J = 7.20 Hz, -C**H**$_3$), 3.63 (q, 2H, J = 7.20 Hz, -C**H**$_2$-), 7.55 (td, 1H, Jo = 7.60 and 8.00 Hz, Jm = 1.20 Hz, 6-**H**), 7.69 (s, 1H, =C**H**), 7.72 (d, 1H, $J_{8,7}$ = 8.40 Hz, 8-**H**), 7.86 (td, 1H, Jo = 7.80 and 8.00 Hz, Jm = 1.60 Hz, 7-**H**), 8.11 (dd, 1H, $J_{5,6}$ = 8.40 Hz, $J_{5,7}$ = 1.60 Hz, 5-**H**), 8.90 (s, 1H, 2-**H**), MS (ESI+) *m/z* (rel intensity): 302 (M + 1, 65), 86 (100), Anal. for $C_{15}H_{11}NO_4S$: Calc. C, 59.79; H, 3.68; N, 4.65; S, 10.64%. Found C, 59.83; H, 3.38; N, 4.81; S, 10.80%.

3-Ethyl-5-(4-oxo-4H-chromen-3-yl methylene)-imidazolidine-2,4-dione) (IVg) Yield: 49.58%, mp: 223°C, ^{1}H NMR ($CDCl_3$): δ = 1.11 (ψt, 3H, J = 6.80 and 7.60 Hz, -C**H**$_3$), 3.48 (q, 2H, J = 7.20 Hz, -C**H**$_2$-), 6.42 (s, 1H, =C**H**), 7.52 (td, 1H, Jo = 7.20 and 7.80 Hz, Jm = 1.20 Hz, 6-**H**), 7.70 (d, 1H, $J_{8,7}$ = 8.80 Hz, 8-**H**), 7.84 (td, 1H, Jo = 7.60 and 8.00 Hz, Jm = 1.60 Hz, 7-**H**), 8.11 (dd, 1H, $J_{5,6}$ = 8.40 Hz, $J_{5,7}$ = 1.60 Hz, 5-**H**), 8.74 (s, 1H, 2-**H**), 10.53 (s, 1H, N**H**), MS (ESI+) *m/z* (rel intensity): 285 (M + 1, 65), 307 (100), Anal. for $C_{15}H_{12}N_2O_4$: Calc. C, 63.38; H, 4.25; N, 9.85%. Found C, 63.42; H, 4.48; N, 9.79%.

3-Ethyl-2-(ethylthio)-5-(4-oxo-4H-chromen-3-yl methylene)-imidazolidine-4-one) (IVi) Yield: 70.49%, mp: 195°C, ^{1}H NMR (DMSO-d_6): δ = 1.13 (t, 3H, J = 7.20 Hz, -C**H**$_3$), 1.42 (ψt, 3H, J = 6.80 and 7.60 Hz, -C**H**$_3$), 3.36 (q, 2H, J = 7.20 and 7.60 Hz, -C**H**$_2$-), 3.53 (q, 2H, J = 7.20 Hz, -C**H**$_2$-), 6.97 (s, 1H, =C**H**), 7.52 (t, 1H, Jo = 8.00 Hz, 6-**H**), 7.69 (d, 1H, $J_{8,7}$ = 8.80 Hz, 8-**H**), 7.84 (td, 1H, Jo = 7.20 Hz, Jm=1.20 Hz, 7-**H**), 8.12 (dd, 1H, $J_{5,6}$ = 8.00 Hz, $J_{5,7}$ = 1.60 Hz, 5-**H**), 9.61 (s, 1H, 2-**H**), MS (ESI+) *m/z* (rel intensity): 329 (M + 1, 90), 351 (100), Anal. for $C_{17}H_{16}N_2O_3S$: Calc. C, 62.18; H, 4.91; N, 8.53; S, 9.76%. Found C, 62.16; H, 4.95; N, 8.35; S, 9.40%.

Insulin releasing activity studies

Cell culture of INS-1 cells

INS-1 cells, generously provided by Dr. C. Wollheim, Geneva, Switzerland[15], were grown in plastic culture bottles or microwells for 4–6 days (half confluence: $1–2 \times 10^6$ cells per mL) in RPMI medium supplemented with 10% (v/v) fetal calf serum, 100 U of penicillin per mL, and 0.1 mg of streptomycin per mL. Cells were seeded at a density of 5×10^5 cells/mL. The medium was changed every 5 days, and the cells were detached from the culture flask with trypsin 1 week after seeding, centrifuged, and reseeded as described above. Prior to the experiment cells were washed twice and then incubated in Krebs–Ringer buffer containing 10 mM HEPES and 0.5 % bovine serum albumin (KRBH).

Insulin release

To measure insulin secretion, half-confluent cells in microwells were incubated for 90 min at 37°C in the aforementioned KRBH buffer. Insulin released into the medium was assayed with a radioimmunoassay using rat insulin (Novo Nordisk, Bagsvaerd, Denmark) as a standard, (mono ^{125}I–Tyr A^{14})-insulin as the labeled compound (Sanofi-Aventis, Germany), and anti-insulin antibodies from Linco (St. Louis, MO, USA). Each compound had been checked for non-interference with the insulin radioimmunoassay. The data

were corrected for the effects of solubilizing compounds (ethanol, DMSO).

Results and discussion

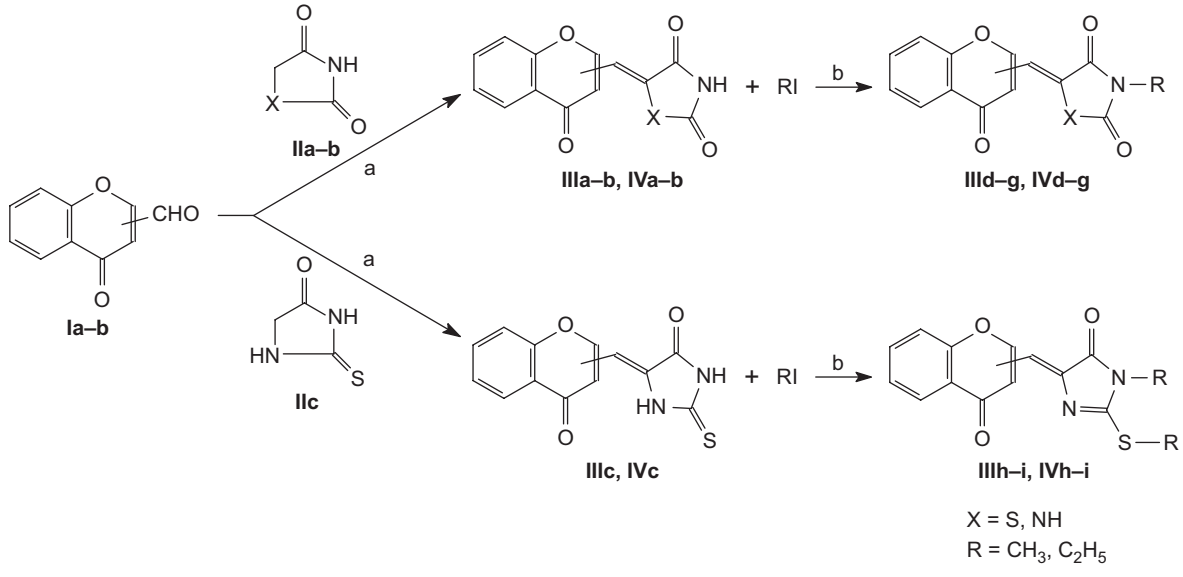
The most common method involves the Baker–Venkataraman rearrangement wherein an *o*-hydroxyacetophenone is acetylated[8] and the ester is treated with base (pyridine/KOH) to effect an acyl group migration, forming a 1,3-diketone[9]. The ensuing diketone is then cyclized under strongly acidic conditions using sulfuric acid to furnish 2-methyl chromone[10]. 2,4-TZD (**II**) was synthesized with $ClCH_2COOH$ and thiourea in hot water[7]. Compounds **IIIa–c**, **IVa–c** were prepared by Knoevenagel reaction of 2,4-thiazolidinedione/2,4-imidazolidinedione/2-thioxo-imidazolidine-4-one (**IIa–c**) with 2/3-formyl chromone (**Ia–b**). Compounds **IIId–i**, **IVd–i** were synthesized by alkylation of compounds **IIIa–c**, **IVa–c** with methyl/ethyl iodide in the presence of anhydrous sodium carbonate/DMF (Scheme 1).

The structure of the synthesized chromonyl compounds was elucidated by IR, ^1H NMR, mass spectral data, and elementary analysis findings. All spectral data were in accordance with assumed structures. In ^1H NMR spectra, chromone protons were observed between 6.45 and 9.69 ppm; methylene protons for chromonyl-2,4-TZDs, chromonyl-2,4-imidiazolidinediones, and chromonyl-2-thioxo-imidazolidine-4-ones were seen at 6.93–7.72 ppm, 6.26–6.42 ppm, and 6.29–7.01 ppm as a singlet, respectively. In mass spectra, the compounds had an M + H ion peak. In the literature it was reported that in reactions using unsubstituted imidazolidinediones and benzaldehydes or 3-substituted thiazolidinedione and 3-formyl chromone in an acidic medium, the main product was the Z isomer[16,17]. The coupled ^{13}C NMR study of arylidene thiazolidinediones and imidazolidinediones also showed that only the Z isomer was formed[18,19]. In this study, only one isomer of the compounds was obtained. After the appropriate crystals of compounds **IVd**, **IVg**, **IVi** were obtained, X-ray analysis was performed and the compounds **IVd** (supplementary

Table 1. Effects of chromon-2-yl-2,4-thiazolidinediones/imidazolidinediones/2-thioxo-imidazolidine-4-ones **IIIa–i** on glucose-mediated insulin release from INS-1 cells.

Compound	Insulin release (%)
Glucose (3.0 mM)	59.47 ± 7.70
Glucose (5.6 mM)	100.0
Plus **IIIa** (1 μg/mL)	58.51 ± 5.03
Plus **IIIa** (10 μg/mL)	29.37 ± 4.28
Plus **IIIb** (1 μg/mL)	80.46 ± 6.42
Plus **IIIb** (10 μg/mL)	71.94 ± 6.85
Plus **IIIc** (1 μg/mL)	103.3 ± 2.62
Plus **IIIc** (10 μg/mL)	50.89 ± 6.38
Plus **IIId** (1 μg/mL)	96.85 ± 2.78
Plus **IIId** (10 μg/mL)	125.8 ± 11.24
Plus **IIIe** (1 μg/mL)	96.58 ± 6.43
Plus **IIIe** (10 μg/mL)	106.9 ± 3.02
Plus **IIIf** (1 μg/mL)	88.88 ± 10.81
Plus **IIIf** (10 μg/mL)	88.07 ± 2.22
Plus **IIIg** (1 μg/mL)	103.9 ± 3.13
Plus **IIIg** (10 μg/mL)	160.1 ± 14.34
Plus **IIIh** (1 μg/mL)	Not tested
Plus **IIIh** (10 μg/mL)	Not tested
Plus **IIIi** (1 μg/mL)	67.10 ± 4.55
Plus **IIIi** (10 μg/mL)	72.27 ± 8.79
Plus glibenclamide (1 μg/mL)	172.14 ± 7.98
Plus DMSO (1 μg/mL)	100.5 ± 4.87
Plus DMSO (10 μg/mL)	94.12 ± 5.32

Note. INS-1 cells in multiwells were washed three times and incubated in KRBH buffer for 90 min at 5.6 mM glucose. The results are expressed as percent insulin release at 5.6 mM glucose alone. Values obtained in the presence of 3.0 mM glucose (substimulatory concentration) and glibenclamide (1 μg/mL) served as negative and positive controls. The final concentration of the solvent DMSO was either 0.01 or 0.1%; a DMSO control (even at 1%) had no effect as shown in the table data. Each value represents the mean ± SEM of six independent experiments.



Scheme 1. General synthesis of IIIa–i/IVa–i. (a) CH_3COONa/CH_3COOH, (b) Na_2CO_3/DMF.

Table 2. Effects of chromon-3-yl-2,4-thiazolidinediones/imidazolidinediones/2-thioxo-imidazolidine-4-ones **IVa–i** on glucose-mediated insulin release from INS-1 cells.

Compound	Insulin release (%)
Glucose (3.0 mM)	82.18 ± 7.49
Glucose (5.6 mM)	100
Plus **IVa** (1 μg/mL)	58.28 ± 7.99
Plus **IVa** (10 μg/mL)	40.73 ± 3.96
Plus **IVb** (1 μg/mL)	142.7 ± 17.60
Plus **IVb** (10 μg/mL)	138.3 ± 17.84
Plus **IVc** (1 μg/mL)	155.4 ± 35.16
Plus **IVc** (10 μg/mL)	116.7 ± 11.84
Plus **IVd** (1 μg/mL)	98.68 ± 6.43
Plus **IVd** (10 μg/mL)	107.3 ± 8.48
Plus **IVe** (1 μg/mL)	103.2 ± 9.56
Plus **IVe** (10 μg/mL)	122.5 ± 1.49
Plus **IVf** (1 μg/mL)	104.4 ± 5.55
Plus**IVf** (10 μg/mL)	100.8 ± 6.40
Plus **IVg** (1 μg/mL)	89.29 ± 6.16
Plus **IVg** (10 μg/mL)	99.94 ± 8.52
Plus **IVh** (1 μg/mL)	125.4 ± 5.10
Plus **IVh** (10 μg/mL)	134.3 ± 2.92
Plus **IVi** (1 μg/mL)	117.5 ± 10.12
Plus **IVi** (10 μg/mL)	137.6 ± 7.05
Plus glibenclamide (1 μg/mL)	138.0 ± 13.99
Plus DMSO (10 μg/mL)	96.27 ± 14.75

Note. INS-1 cells in multiwells were washed three times and incubated in KRBH buffer for 90 min at 5.6 mM glucose. The results are expressed as percent insulin release at 5.6 mM glucose alone. Values obtained in the presence of 3.0 mM glucose (substimulatory concentration) and glibenclamide (1 μg/mL) served as negative and positive controls. The final concentration of the solvent DMSO was either 0.01 or 0.1%; a DMSO control (even at 1%) had no effect as shown in the table data. Each value represents the mean ± SEM of 4–6 independent experiments.

material reference number: FL2008), **IVg** (supplementary material reference number: at2183), and **IVi** (CCDC deposition number: CCDC-604172) were found to be Z isomers[20–22].

Derivatives of chromone were synthesized and their biological activity with respect to insulin release was tested. All the biological results of the tested compounds are given in Tables 1 and 2. According to their insulinotropic activities in INS-1 cells, compounds **IIIg**, **IVh**, **IVi** (at higher concentration; 10 μg/mL) were able to increase insulin release in the presence of 5.6 mmol/L glucose, but to a lower extent than the reference compound glibenclamide. Compounds **IVb**, **IVc** (at lower concentration; 1 μg/mL) showed more potent insulinotropic effect than glibenclamide (1 μg/mL). The effects of compounds **IVb** and **IVc** were more pronounced at the lower than at the higher concentrations, which is well known from other insulinotropic compounds. But compounds **IIIg**, **IVh**, and **IVi** had an insulinotropic effect at higher concentrations. Compared to compounds derived from 2-formyl chromone, compounds derived from 3-formyl chromone showed a better insulinotropic effect. Introducing methyl or ethyl groups to 2,4-thiazolidinedione, 2,4-imidazolidinedione, and 2-thioxo-imidazolidine-4-one rings of the synthesized compounds did not improve the insulinotropic effect, except for **IIIg**.

It was shown that introducing a 4-oxo-4*H*-chromen-3-yl methylene group at the fifth position of 2,4-imidazolidinedione and 2-thioxo-imidazolidine-4-one rings increased the insulinotropic effect. Also, it should be pointed out that compounds **IVb** and **IVc** are promising molecules, and this kind of structure could be a guide for further investigations on insulinotropic activity studies in INS-1 cells. The effect of these synthesized compounds structurally related to thiazolidinediones may also improve insulin sensitivity, which was not experimentally proven in our experiments.

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Declaration of interest

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