Antimicrobial Activity of New 2,4-Disubstituted Thiazolidinone Derivatives

Wael A. El-Sayed[a], Yasser K. Abdel-Monem[b], Nabil M. Yousif[a], Nashwa Tawfek[a], Mohamed T. Shaaban[c], and Adel A.-H. Abdel-Rahman[b,*]

[a] Photochemistry Department, National Research Center, El Dokki, Cairo, Egypt
[b] Chemistry Department, Faculty of Science, Menoufia University, Shebin El-Koam, Egypt. E-mail: adelnassar63@hotmail.com
[c] Botany Department, Faculty of Science, Menoufia University, Shebin El-Koam, Egypt

* Author for correspondence and reprint requests

Z. Naturforsch. **64c**, 785–789 (2009); received May 7/June 11, 2009

A number of new disubstituted 2,5-thiazolidinone derivatives were synthesized and tested for their antimicrobial activity against *Bacillus subtilis* (Gram-positive), *Pseudomonas aeruginosa* (Gram-negative), and *Streptomyces* species (Actinomycetes). They displayed different degrees of antimicrobial activities or inhibitory actions.

Key words: Thiazolidinones, Diaza-1,3-thiazole, Antimicrobial Activity

Introduction

Thiazole derivatives are considered as one of the most important classes of heterocyclic compounds; their derivatives are characterized by high biological activity in pharmaceutical fields and have shown antibacterial (Tsuji and Ishikawa, 1994), antifungal (López-García *et al.,* 2003), antitumour (Srivastava *et al.*, 1977), antiviral (Srivastava *et al.*, 1977; Kirsi *et al.*, 1983), anti-inflammatory (Mgonzo *et al.*, 1995; Geronikaki *et al.*, 2003) and antineoplastic (Milne, 2000) activities as well as growth inhibitory activity of gastrointestinal (Sato *et al.*, 2000; Takahashi *et al.*, 1999), biliary, and pancreatic adenocarcinoma cells (Motomura *et al.*, 2000). The aminothiazole ring system has found application in drug development for the treatment of HIV infection, hypertension and inflammation (Kearney *et al.*, 1988). On the other hand, 1,3-thiazolidines are considered as an important class of antimicrobial agents with activity against a broad spectrum of Gram-positive pathogens including *Staphylococci*, *Streptococci*, and *Enterococci* (Singh *et al.*, 1981). It is known that the entrance of arylidene moieties at different positions of the thiazolidine ring enhances the antimicrobial activity (Brown, 1961; Nasr *et al.*, 2003). Dioxotetrahydrothiazole derivatives with a carbonyl group at positions 2 and 4 are an important group of heterocyclic compounds with diverse biological activities, *i.e.*, they are known as antineoplastics (DeLima *et al.*, 1992). These derivatives have been extensively studied, chemically as well as biologically (Labouta *et al.*, 1987), in an effort to generate new translation initiation inhibitors for cancer therapy (Chen *et al.*, 2004). In addition, 2,4-dioxotetrahydro-1,3-thiazoles inhibit the growth of gastrointestinal (Sato *et al.*, 2000; Takahashi *et al.*, 1999), biliary, and pancreatic adenocarcinoma cells (Motomura *et al.*, 2000). Moreover, thiazolidine derivatives are reported to show a variety of pharmacological properties, such as antibacterial, antifungal (DeLima *et al.*, 1992), anthelmintic (Vagdevi *et al.*, 2006), cardiotonic (Andreani *et al.*, 1993), anticonvulsant (El-Feky, 1993), cyclooxygenase and lipoxygenase inhibitory (Unangst *et al.*, 1993) activities.

In view of the above-mentioned findings and as continuation of our effort to identify new candidates that may be suitable in designing new, potent, selective, and less toxic antimicrobial agents (Abdel-Rahman *et al.*, 2008; El-Sayed *et al.*, 2008, 2009) we report in the present work the synthesis and antimicrobial activity of new substituted thiazole derivatives.

Experimental

General

Melting points were determined using a Büchi apparatus. IR spectra (KBr) were recorded with a Bruker-Vector22 instrument (Bruker, Bremen,

Germany). ^{1}H NMR spectra were recorded with a Varian Gemini spectrometer at 300 MHz and 200 MHz with TMS as internal standard. Chemical shifts were reported in δ scale (ppm) relative to TMS as a standard, and the coupling constants (J values) are given in Hz. The progress of the reactions was monitored by TLC using aluminium silica gel plates 60 F_{245}. EI-mass spectra were recorded with a HP D5988 A 1000 MHz instrument (Hewlett-Packard, Palo Alto, CA, USA). Antiviral activity against hepatitis B virus (HBV) was tested at the Liver Institute, Menoufia University, Shebin El-Koam, Egypt.

Sample preparation

Each of the test compounds and standards was dissolved in 12.5% DMSO, at concentrations of 500 μg/mL. Further dilutions of the compounds and standards in the test medium were prepared at the required quantities.

Culture of microorganisms

Bacteria strains were supplied from Botany Department, Faculty of Science, Menoufia University, Shebin El-Koam, Egypt, namely *Bacillus subtilis* (ATCC 6633, Gram-positive), *Pseudomonas aeruginosa* (ATCC 27853, Gram-negative) and *Streptomyces* species (Actinomycetes). The bacterial strains were maintained on Mueller-Hinton agar (MHA) medium (Oxoid, Chemical Co., UK) for 24 h at 37 °C. The medium was molten on a water bath, inoculated with 0.5 mL of the culture of the specific microorganism and poured into sterile Petri dishes to form a layer of about 3–4 mm thickness. The layer was allowed to cool and harden. With the aid of a cork-borer, cups of about 10 mm diameter were produced (Jorgensen *et al.*, 1999).

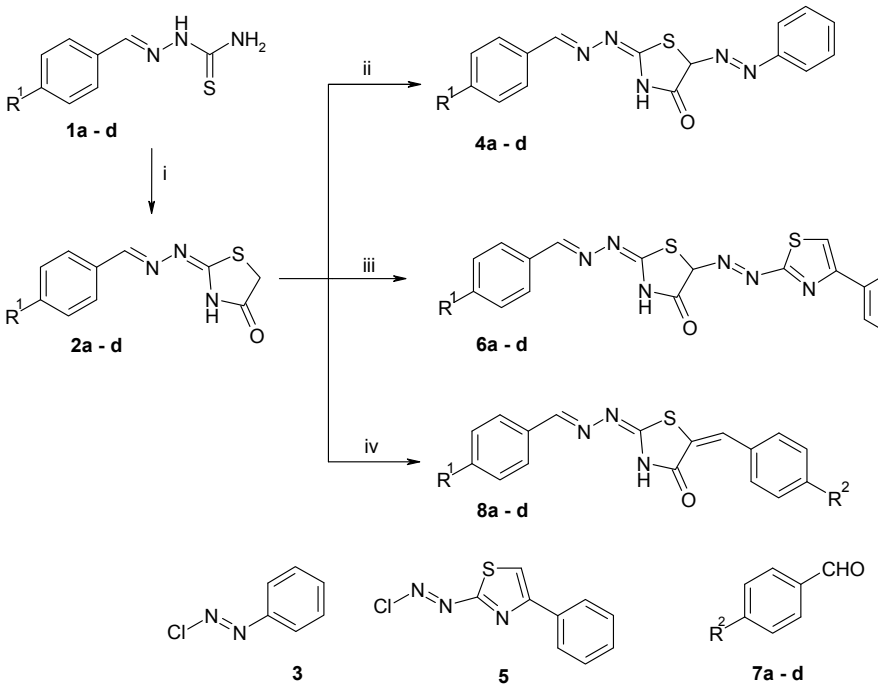
Agar diffusion technique

The antibacterial activities of the synthesized compounds were tested against *Bacillus subtilis*, *Pseudomonas aeruginosa*, and *Streptomyces* species using MH medium (17.5 g casein hydrolysate, 1.5 g soluble starch, 1000 mL beef extract). A stock solution of each synthesized compound (500 μg/mL) in DMSO was prepared and graded quantities of the test compounds were incorporated in a specified quantity of sterilized liquid MH medium. Different concentrations of the test compounds in DMF were placed separately in cups in the agar medium. All plates were incubated at 37 ºC overnight. The inhibition zones were measured after 24 h. The minimum inhibitory concentration (MIC) was defined as the intercept of the grave of logarithm concentrations versus diameter of the inhibition zones (Janssen *et al.*, 1987; Greenwood, 2000).

Results and Discussion

Chemistry

The (*E*)-2-(*E*)-4-substituted arylidenehydrazonothiazolidin-4-one derivatives **2a–d** were synthesized by the reaction of the corresponding substituted arylidinehydrazinothioamide derivatives **1a–d** with ethyl chloroacetate in ethanol at reflux temperature (Fig. 1). The chemical structures of the thiazolidine derivatives **2a–d** were confirmed by their spectral and analytical data. Thus, their IR spectra showed a characteristic absorption band in the carbonyl frequency region at 1644–1692 cm^{-1} corresponding to the C=O groups. The ^{1}H NMR spectrum of **2b**, as a representative example, showed a singlet peak at δ 3.91 ppm for the CH_2 group, signals of the aromatic protons at δ 7.55–8.44 ppm, and an NH signal as singlet at δ 12.1 ppm (which disappeared in D_2O exchange). Furthermore, the mass spectrum of **2b** showed the molecular ion peak at *m/z* 253 ([MH]$^+$, 69%), which is in accordance with its molecular formula. The coupling reaction of chlorodiazonium compounds at the active methylene group in compounds **2a–b** was the convenient route to synthesize the substituted diazothiazolidinone derivatives. Thus, reaction of the (*E*)-2-(*E*)-4-substituted arylidenehydrazonothiazolidin-4-one derivatives **2a–d** with (*E*)-1-chloro-2-phenyldiazene (**3**) or (*E*)-2-(chlorodiazenyl)-4-phenylthiazole (**5**) afforded the (*E*)-2-(*E*)-4-substituted arylidenehydrazono-5-[(*E*)-phenyldiazenyl]thiazolidin-4-ones **4a–d** in 70–78% yields or the (*E*)-2-(*E*)-4-substituted arylidenehydrazono-5-[(*E*)-4-phenylthiazol-2-yl]diazenylthiazolidin-4-ones **6a–d** in 77–80% yields, respectively. The chemical structures of compounds **4a–d** and **6a–d** were proved on the basis of their IR, ^{1}H NMR and mass spectra which all agreed with the assigned structures. Thus, the ^{1}H NMR spectrum of compound **4b**, as a representative example, revealed the presence of a singlet peak corresponding to H-5 in the thiazolidine ring at δ 3.37 ppm, C*H*=N as singlet at δ 8.06 ppm, signals of the aromatic protons at δ 7.43–8.05 ppm,



Compound	R^1	R^2
1a	H	–
1b	Cl	–
1c	Br	–
1d	$N(CH_3)_2$	–
2a	H	–
2b	Cl	–
2c	Br	–
2d	$N(CH_3)_2$	–
4a	H	–
4b	Cl	–
4c	Br	–
4d	$N(CH_3)_2$	–

Compound	R^1	R^2
6a	H	–
6b	Cl	–
6c	Br	–
6d	$N(CH_3)_2$	–
7a	–	H
7b	–	Cl
7c	–	Br
7d	–	$N(CH_3)_2$
8a	Br	H
8b	Br	Cl
8c	Br	Br
8d	Br	$N(CH_3)_2$

Fig. 1. Synthesis of 2,4-disubstituted thiazolidinone derivatives. (i) $ClCH_2COOEt$, EtOH, reflux; (ii) **3**, KOH/H_2O; (iii) **5**, KOH/H_2O; (iv) **7a–d**, EtOH/reflux

and an NH signal as broad singlet at δ 11.33 ppm (which disappeared in D_2O exchange) in addition to the disappearance of the signal corresponding to the CH_2 group. Moreover, the mass spectrum of this compound showed the molecular ion peak at *m/z* 357 ($[MH]^+$, 66%), which is in accordance with its molecular formula. On the other hand, when the thiazolidine derivatives **2a–d** were allowed to react with the aromatic aldehydes **7a–d** in the presence of acetic acid in ethanol at reflux temperature, the corresponding (2*E*,5*E*)-4-substituted arylidene-2-[(*E*)-(4-arylidene)hydrazono] thiazolidin-4-ones **8a–d** were obtained in 78–80% yields. The IR spectra of the disubstituted 2,5-thiazolidinone derivatives showed the presence of characteristic absorption bands at 3325, 1682 and 1605 cm^{-1} corresponding to NH, C=O, and C=N, respectively. The ^1H NMR spectrum of compound **8a** showed signals of the aromatic protons at δ 6.88–8.05 ppm in addition to an NH signal at δ 10.55 ppm. The disappearance of the singlet peak of the CH_2 group also confirmed the assigned structure.

Antimicrobial activity

The antimicrobial activity of the synthesized compounds was evaluated against three microorganisms; *Bacillus subtilis* (ATCC 6633, Gram-positive), *Pseudomonas aeruginosa* (ATCC 27853) (Gram-negative) and *Streptomyces* species (Actinomycetes). The values of minimal inhibitory concentration (MIC) of the tested compounds are presented in Table I. The results of the antimicrobial activity test revealed that **6b** and **6c** showed the highest activity against *B. subtilis* with MIC values of 75 μg/mL followed by **1c**, **2a**, **2c**, and **4a**. Compounds **1d** and **2d** showed the highest inhibition activity against *P. aeruginosa*, whereas **6c** and **6d** were the most active among the series of tested compounds against *Streptomyces* species with MIC values of 75 μg/mL. Some compounds did not show any activity against the three microorganisms.

From the structure-activity relationship it is clear that substitution at the *p*-position in the phenyl ring of the thiazolidinone derivatives with a chlorine or bromine atom increases the antibacterial activity against *B. subtilis* (ATCC 6633). This was not the case for *P. aeruginosa* since substitution with *N,N*-dimethylamine resulted in higher activity. Additionally, the *p*-substituted bromo- and *N,N*-dimethylaminophenyl groups in the thiazolidinone derivatives revealed the highest activity against *Streptomyces* species. On the other hand, **6c** containing an 1,3-thiazole ring showed higher activity against the three microorganisms compared to its substituted phenyl analogue **4c**. Furthermore, substitution of the active methylene group in the thiazolidine ring of compounds **8a–d** resulted in the loss of antimicrobial activity except for **8a** which showed relatively higher activity against *Streptomyces* species.

Conclusion

From the results of the antimicrobial activity tests and structure-activity relationship, it can be concluded that the antimicrobial activity against *B. subtilis*, *P. aeruginosa*, and *Streptomyces* species (Actinomycetes) depends, to some extent, on the substituent of the aryl group placed at position 4 as well as substitution at position 5 of the thiazolidine ring. Thus, **6c** containing 4-bromophenyl and 2-diazo-1,3-thiazol groups at position 4 of the thiazolidine ring showed high activity.

Table I. Minimum inhibitory concentrations (MIC in μg/mL) of the title compounds. The negative control DMSO showed no activity.

Compound	*Bacillus subtilis* (Gram-positive)	*Pseudomonas aeruginosa* (Gram-negative)	*Streptomyces* species (Actinomycetes)
1a	–[a]	–	–
1b	250	–	–
1c	100	–	500
1d	–	75	–
2a	100	–	125
2b	–	–	–
2c	100	–	125
2d	–	75	–
4a	100	500	–
4b	75	–	100
4c	250	250	–
4d	100	250	100
6a	100	250	–
6b	75	–	125
6c	75	250	75
6d	125	250	75
8a	125	250	100
8b	–	–	500
8c	250	–	125
8c	250	–	250
Penicillin	31	46	33

[a] Totally inactive (MIC > 500 μg/mL).

Abdel-Rahman A. A.-H., El-Sayed W. A., Abdel-Bary H. M., Abdel-Megied A. E.-S., and Morcy E. M. (2008), Amino acid derivatives, VIII [1]: synthesis and antimicrobial evaluation of α-amino acid esters bearing an indole side chain. Monatsh. Chem. **139**, 1095–1101.

Andreani A., Rambaldi M., Locatelli A., Leoni R., Bossa M., Chiericozzi I., Galatulas G., and Salvatore A. (1993), Synthesis of lactams with potential cardiotonic activity. Eur. J. Med. Chem. **28**, 825–829.

Brown F. G. (1961), 4-Thiazolidinones. Chem. Rev. **61**, 463–521.

Chen H., Fan Y.-H., Natarajan A., Guo Y., Iyasere J., Harbinski F., Luis L., Christ W., Aktas H., and Halperin J. (2004), Synthesis and biological evaluation of thiazolidine-2,4-dione and 2,4-thione derivatives as inhibitors of translation initiation. Bioorg. Med. Chem. Lett. **14**, 5401–5405.

DeLima M. C. A., Costa D. L. B., Goes A. J. S., Galdino S. L., Pitta I. R., and Luu-Duc C. (1992), Synthèse et activité antimicrobienne de dérivés chlorobenzyl benzylidéne imidazolidinediones et thiazolidinediones substituées. Pharmazie **47**, 182–184.

El-Feky S. A. H. (1993), Synthesis and anticonvulsant properties of some quinazolinone thiazolidine and 4-thiazolidinone derivatives. Pharmazie **48**, 894–896.

El-Sayed W. A., Ramiz M. M. M., and Abdel-Rahman A. A.-H. (2008) *C*-Furyl glycosides, I: Synthesis and antimicrobial evaluation of *C*-furyl glycosides and chalcones derived therefrom. Monatsh. Chem. **139**, 1499–1505.

El-Sayed W. A., Nassar I. F., and Abdel-Rahman A. A.-H. (2009), *C*-Furyl glycosides, II: Synthesis and antimicrobial evaluation of *C*-furyl glycosides bearing pyrazolines, isoxazolines, and 5,6-dihydropyrimidine-2(1*H*)-thiones. Monatsh. Chem. **140**, 365–370.

Geronikaki A., Hadjipavlou-Litina D., Chatziopoulos C., and Soloupis G. (2003), Synthesis and biological evaluation of new 4,5-disubstituted-thiazolyl amides, derivatives of 4-hydroxy-piperidine or of 4-*N*-methyl piperazine. Molecules **8**, 472–479.

Greenwood D. (2000), Antimicrobial Chemotherapy, 4th ed. Oxford University Press, New York, p. 114.

Janssen A. M., Scheffer J. J., and Svendsen A. B. (1987), Antimicrobial activity of essential oils; a 1976–1986 literature review. Planta Med. **53**, 395–400.

Jorgensen J. H., Jurnide J. D., and Washington J. A. (1999), Antimicrobial susceptibility tests. In: Manual of Clinical Microbiology, 7th ed. (Muarry P. R., Baron E. J., and Yolken R. C., eds.). American Society for Microbiology, Washington, DC, USA, pp. 1526–1543.

Kearney P. C., Fernandz M., and Flygare J. A. (1988), Solid-phase synthesis of 2-aminothiazoles. J. Org. Chem. **63**, 196–200.

Kirsi J. J., North J. A., McKernan P. A., Murray B. K., Canonico P. G., Huggins J. W., Srivastava P. C., and Robins R. K. (1983), Broad-spectrum antiviral activity of 2-β-D-ribofuranosylselenazole-4-carboxamide, a new antiviral agent. Antimicrob. Agents Chemother. **24**, 353–361.

Labouta I. M., Salama H. M., Eshba N. H., Kader O., and El-Chrbini E. (1987), Potential anti-microbial: syntheses and *in vitro* anti-microbial evaluation of some 5-arylazo-thiazolidones and related compounds. Eur. J. Med. Chem. **22**, 485–489.

López-García B., Veyrat A., Pérez-Payá E., González-Candelas L., and Marcos J. F. (2003), Comparison of the activity of antifungal hexapeptides and the fungicides thiabendazole and imazalil against postharvest fungal pathogens. Int. J. Food Microbiol. **89**, 163–170.

Mgonzo R., Geronikaki A., and Kourounakis P. N. (1995), Synthesis and antiinflammatory activity of some new thiazole derivatives. Pharmazie **50**, 505–506.

Milne G. W. A. (ed.) (2000), Ashgate Handbook of Antineoplastic Agents. Gower, London, UK.

Motomura W., Okumura T., Takahashi N., Obara T., and Kohgo Y. (2000), Activation of peroxisome poliferator-activated receptor γ by troglitazone inhibits cell growth through the increase of p27Kip1 in human pancreatic carcinoma cells. Cancer Res. **60**, 5558–5564.

Nasr M. N., Gineinah M. M., and El-Bendary E. R. (2003), Synthesis and *in vitro* antibacterial evaluation of novel imidazo[2',1':5,1]-1,2,4-triazolo[4,3-*c*]-quinazoline derivatives of 5-thioxo-1,2,4-triazole, 4-oxothiazolidine, and their open-chain counterparts. Arch. Pharm. **336**, 560–566.

Sato H., Ishihara S., Kawashima K., Moriyama N., Suetsugu H., Kazumori H., Okuyama T., Rumi M. A. K., Fukuda R., Nagasue N., and Kinoshita Y. (2000), Expression of peroxisome proliferator-activated receptor (PPAR) γ in gastric cancer and inhibitory effects of PPAR γ agonists. Br. J. Cancer **83**, 1394–1400.

Singh S. P., Parmar S. S., Raman K., and Stenberg V. I. (1981), Chemistry and biological activity of thiazolidinones. Chem. Rev. **81**, 175–203.

Srivastava P. C., Pickering M. V., Allen L. B., Streeter D. G., Campbell M. T., Witkowski J. T., Sidwell R. W., and Robins R. K. (1977), Synthesis and antiviral activity of certain thiazole *C*-nucleosides. J. Med. Chem. **20**, 256–261.

Takahashi N., Okumura T., Motomura W., Fujimoto Y., Kawabata I., and Kohgo Y. (1999), Activation of PPAR γ inhibits cell growth and induces apoptosis in human gastric cancer cells. FEBS Lett. **455**, 135–139.

Tsuji K. and Ishikawa H. (1994), Synthesis and antipseudomonal activity of new 2-isocephems with a dihydroxypyridone moiety at C-7. Bioorg. Med. Chem. Lett. **4**, 1601–1606.

Unangst P. C., Connor D. T., Cetenko W. A., Sorenson R. J., Sircar J. C., Wright C. D., Schrier D. J., and Dyer R. D. (1993), Oxazole, thiazole, and imidazole derivatives of 2,6-di-*tert*-butylphenol as dual 5-lipoxygenase and cyclooxygenase inhibitors. Bioorg. Med. Chem. Lett. **3**, 1729–1734.

Vagdevi H. M., Vaidya V. P., Latha K. P., and Padmashali B. (2006), Synthesis and pharmacological examination of some thiazolidinone derivatives of naphtho[2,1-*b*]-furan. Indian J. Pharm. Sci. **68**, 719–725.