ELSEVIER

Il Farmaco 57 (2002) 747–751

IL FARMACO

www.elsevier.com/locate/farmac

Synthesis and anti-HIV activity of 2,3-diaryl-1,3-thiazolidin-4-(thi)one derivatives

Angela Rao [a], Anna Carbone [a], Alba Chimirri [a], Erik De Clercq [b], Anna Maria Monforte [a], Pietro Monforte [a,*], Christophe Pannecouque [b], Maria Zappalà [a]

[a] *Dipartimento Farmaco-Chimico, Università di Messina, viale Annunziata, 98168 Messina, Italy*
[b] *Rega Institute for Medical Research, Katholieke Universiteit Leuwen, 10 Minderbroedersstraat, B-3000 Leuwen, Belgium*

Received 9 March 2002; received in revised form 9 April 2002; accepted 20 April 2002

Abstract

Several 2,3-diaryl-1,3-thiazolidine-4-thione derivatives and 2,3-diaryl-1,3-thiazolidin-4-ones bearing a methyl group at C-5 position have been synthesized and tested as anti-HIV agents. The results of the in vitro tests showed that some of them proved to be effective inhibitors of HIV-1 replication.

Keywords: 2,3-Diaryl-1,3-thiazolidin-4-ones; 2,3-Diaryl-1,3-thiazolidine-4-thiones; Anti-HIV activity; NNRTIs

1. Introduction

To date, 15 anti-retroviral agents have been licensed for clinical use in the treatment of AIDS or HIV infection [1]. All these compounds belong to three distinct classes: nucleoside and nonnucleoside reverse transcriptase inhibitors (NRTIs and NNRTIs) and protease inhibitors (PIs). In recent years the combination therapy has been found to decrease the viral load and the AIDS-related death rate. Despite these results, there is an urgent need for new anti-retroviral drugs due to resistance development, cross-resistance within classes, toxicity, adverse drug–drug interactions and other limitations, which may lead to failure of long-term treatment [2].

A successful strategy in the development of anti-retroviral drugs has been and is still today the targeting of the virally-encoded reverse transcriptase (RT) enzyme [3].

Two classes of compounds, NRTIs and NNRTIs potently and selectively inhibit this enzyme and, as above reported, play a key role in the combination therapy for HIV infections.

In the last decade, our research group has been developing different series of 1*H*,3*H*-thiazolo[3,4-*a*]benzimidazole derivatives (TBZs) as anti-HIV agents [4–8], some of which proved to be potent NNRTIs.

Recently, we carried out a 3D-QSAR and flexible docking study [9] and demonstrated that, similarly to other NNRTIs, the TBZ biological activity was associated with the ability to assume a butterfly-like conformation by means of a suitable spatial location of lipophilic groups important for the RT site binding. In particular, we observed that plausible pharmacophoric elements were the benzene ring of the benzimidazole moiety, the dihalosubstituted aryl group at C-1 and the nitrogen atom of the thiazole nucleus.

In view of this, we designed new anti-HIV agents keeping all key structural requirements for a potent RT inhibition. The results of our research culminated in the discovery of 2,3-diaryl-1,3-thiazolidin-4-ones (**1**) a new class of highly potent NNRTIs. Some of them proved to be effective in HIV-1 replication at nanomolar concentrations and minimally toxic to MT-4 cells [10,11].

In order to find new anti-HIV-1 agents, to further explore the salient features controlling the activity, especially lipophilicity, and to shed more light on the SAR of this series of compounds, we report here the synthesis of new derivatives in which some structural modifications have been introduced on 1,3-thiazolidin-

* Corresponding author
E-mail address: monforte@pharma.unime.it (P. Monforte).

0014-827X/02/$ - see front matter
PII: S0014-827X(02)01268-5

4-one nucleus, that is the replacement of the carbonyl moiety with the isostere thiocarbonyl functionality (**2**) and the introduction of a methyl group at the C-5 atom (**3**–**4**). These structural modifications have been carried out only on derivatives of type-**1** that had proved to be particularly active as anti-HIV agents [10,11].

2. Experimental

2.1. Chemistry

Amines, aldehydes and mercaptoacids employed in this study are commercially available. M.p.s were determined on a Kofler hot stage apparatus and are uncorrected. Elemental analyses (C, H, N) were carried out on a Carlo Erba Model 1106 Elemental Analyzer and the results are within $\pm 0.4\%$ of the theoretical values. Merck silica gel 60 F_{254} plates were used for analytical TLC; column chromatography was performed on Merck silica gel 60 (230–400 mesh). ^{1}H NMR spectra were recorded in $CDCl_3$ on a Varian Gemini-300 spectrometer. Chemical shifts were expressed in δ (ppm) relative to TMS as internal standard and coupling constants (J) in Hz. The log P values were calculated by using the XLOGP v. 2.0 program [12].

2.1.1. General procedure for the synthesis of 2,3-diaryl-1,3-thiazolidine-4-thiones (**2a**–**i**)

A $C_6H_5CH_3$ solution (50 ml) of the suitable 2,3-diaryl-1,3-thiazolidin-4-one (1 mmol), obtained according to a previously reported procedure [10], was heated under stirring with Lawesson's reagent (0.6 mmol) for 2 h. After removal of the solvent under reduced pressure, the oily residue was treated with Et_2O to afford derivatives **2b** and **2e** as solids, or chromatographed on silica gel column using $CHCl_3$ as eluant to give compounds **2a**, **2c**–**d** and **2f**–**i**. All compounds were recrystallized from EtOH.

*2.1.1.1. 2-(2,6-Difluorophenyl)-3-phenyl-1,3-thiazolidine-4-thione (**2a**).* Yield: 82%, m.p. 146–148 °C; ^{1}H NMR: δ 4.54 (d, 1H, $J = 16.4$, $5H_A$), 4.77 (dd, 1H, $J = 2.4$ and 16.4, $5H_B$), 6.79–7.39 (m, 8H, Ar and H-2). *Anal*. $C_{15}H_{11}F_2NS_2$ (C, H, N).

*2.1.1.2. 2-(2,6-Dichlorophenyl)-3-phenyl-1,3-thiazolidine-4-thione (**2b**).* Yield: 95%, m.p. 152–154 °C; ^{1}H NMR: δ 4.63 (d, 1H, $J = 16.7$, $5H_A$), 4.74 (dd, 1H, $J = 3.0$ and 16.7, $5H_B$), 7.10–7.47 (m, 8H, Ar), 7.50 (d, 1H, $J = 3.0$ H-2). *Anal*. $C_{15}H_{11}Cl_2NS_2$ (C, H, N).

*2.1.1.3. 2-(2,6-Difluorophenyl)-3-(6-methyl-pyridin-2-yl)-1,3-thiazolidine-4-thione (**2c**).* Yield: 12%, m.p. 115 °C; ^{1}H NMR: δ 2.45 (s, 3H, CH_3), 4.52 (d, 1H, $J = 16.7$, $5H_A$), 4.77 (dd, 1H, $J = 2.4$ and 16.7, $5H_B$), 6.77–7.71 (m, 7H, Ar and H-2). *Anal*. $C_{15}H_{12}F_2N_2S_2$ (C, H, N).

*2.1.1.4. 2-(2,6-Dichlorophenyl)-3-(6-methyl-pyridin-2-yl)-1,3-thiazolidine-4-thione (**2d**).* Yield: 40%, m.p. 130 °C; ^{1}H NMR: δ 2.45 (s, 3H, CH_3), 4.59 (d, 1H, $J = 16.7$, $5H_A$), 4.74 (dd, 1H, $J = 3.0$ and 16.7, $5H_B$), 6.95–7.73 (m, 6H, Ar), 8.03 (d, 1H, $J = 3.0$ H-2). *Anal*. $C_{15}H_{12}Cl_2N_2S_2$ (C, H, N).

*2.1.1.5. 2-(2,6-Difluorophenyl)-3-(4-methyl-pyridin-2-yl)-1,3-thiazolidine-4-thione (**2e**).* Yield: 89%, m.p. 155–158 °C; ^{1}H NMR: δ 2.33 (s, 3H, CH_3), 4.51 (d, 1H, $J = 16.7$, $5H_A$), 4.76 (dd, 1H, $J = 2.2$ and 16.7, $5H_B$), 6.78–8.30 (m, 7H, Ar and H-2). *Anal*. $C_{15}H_{12}F_2N_2S_2$ (C, H, N).

*2.1.1.6. 2-(2,6-Dichlorophenyl)-3-(4-methyl-pyridin-2-yl)-1,3-thiazolidine-4-thione (**2f**).* Yield: 50%, m.p. (dec.) 185 °C; ^{1}H NMR: δ 2.32 (s, 3H, CH_3), 4.59 (d, 1H, $J = 17.0$, $5H_A$), 4.73 (dd, 1H, $J = 3.0$ and 17.0, $5H_B$), 6.94–8.27 (m, 7H, Ar and H-2). *Anal*. $C_{15}H_{12}Cl_2N_2S_2$ (C, H, N).

*2.1.1.7. 2-(2,6-Dichlorophenyl)-3-(4-methyl-pyrimidin-2-yl)-1,3-thiazolidine-4-thione (**2g**).* Yield: 81%, m.p. (dec.) 146 °C; ^{1}H NMR: δ 2.47 (s, 3H, CH_3), 4.56 (d, 1H, $J = 17.0$, $5H_A$), 4.74 (dd, 1H, $J = 2.7$ and 17.0, $5H_B$), 6.99–8.57 (m, 6H, Ar and H-2). *Anal*. $C_{14}H_{11}Cl_2N_3S_2$ (C, H, N).

*2.1.1.8. 2-(2,6-Difluorophenyl)-3-(4,6-dimethyl-pyridin-2-yl)-1,3-thiazolidine-4-thione (**2h**).* Yield: 8%, m.p. (dec.) 95 °C; ^{1}H NMR: δ 2.45 (s, 3H, CH_3), 4.45 (d, 1H, $J = 16.7$, $5H_A$), 4.66 (dd, 1H, $J = 3.0$ and 16.7, $5H_B$), 6.62–7.49 (m, 6H, Ar and H-2). *Anal*. $C_{16}H_{14}F_2N_2S_2$ (C, H, N).

*2.1.1.9. 3-(6-Bromo-pyridin-2-yl)-2-(2,6-difluorophenyl)-1,3-thiazolidine-4-thione (**2i**).* Yield: 28%, m.p. 86–89 °C; ^{1}H NMR: δ 4.49 (d, 1H, $J = 17.0$, $5H_A$), 4.71 (dd, 1H, $J = 2.4$ and 17.0, $5H_B$), 6.80–8.10 (m, 7H, Ar and H-2). *Anal*. $C_{14}H_9BrF_2N_2S_2$ (C, H, N).

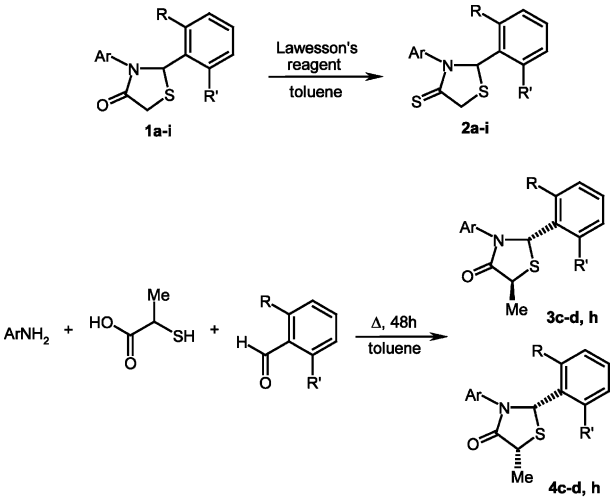
2.1.2. Synthesis of 5-methyl-2,3-diaryl-1,3-thiazolidin-4-ones (**3c**–**d**, **h**, **4c**–**d**, **h**)

To a stirred solution of heteroaromatic amine (8 mmol) in dry $C_6H_5CH_3$ (50 ml), racemic 2-mercaptopropionic acid (16 mmol) and the appropriate aromatic aldehyde (8 mmol) were added. The reaction mixture was refluxed for 48 h and then neutralized by a solution of $NaHCO_3$. After removal of the solvent under reduced pressure, the oily residue was powdered by treatment with a mixture of EtOH and Et_2O to afford the mixture of the two diastereoisomers **3** and **4**, which were

separated by silica gel column chromatography eluting with $CHCl_3$. Compounds **3** eluted before the corresponding isomers **4**. All compounds were recrystallized from EtOH.

*2.1.2.1. trans-2-(2,6-Difluorophenyl)-3-(6-methyl-pyridin-2-yl)-5-methyl-1,3-thiazolidin-4-one (**3c**).* Yield: 26%, m.p. 72–74 °C; ^{1}H NMR: δ 1.65 (d, 3H, J = 6.8, CH–CH_3), 2.32 (s, 3H, CH_3), 4.42 (q, 1H, J = 6.8, H_5), 6.77–8.09 (m, 7H, Ar and H-2). *Anal*. $C_{16}H_{14}F_2N_2OS$ (C, H, N).

*2.1.2.2. cis-2-(2,6-Difluorophenyl)-3-(6-methyl-pyridin-2-yl)-5-methyl-1,3-thiazolidin-4-one (**4c**).* Yield: 11%, m.p. 96–99 °C; ^{1}H NMR: δ 1.76 (d, 3H, J = 6.8, CH–CH_3), 2.29 (s, 3H, CH_3), 4.21 (q, 1H, J = 6.8, H_5), 6.74–7.95 (m, 7H, Ar and H-2). *Anal*. $C_{16}H_{14}F_2N_2OS$ (C, H, N).



Compds	Ar	R	R'
a		F	F
b		Cl	Cl
c		F	F
d		Cl	Cl
e		F	F
f		Cl	Cl
g		Cl	Cl
h		F	F
i		F	F

Scheme 1.

*2.1.2.3. trans-2-(2,6-dichlorophenyl)-3-(6-methyl-pyridin-2-yl)-5-methyl-1,3-thiazolidin-4-one (**3d**).* Yield: 12%, m.p. 135–138 °C; ^{1}H NMR: δ 1.69 (d, 3H, J = 7.1, CH–CH_3), 2.29 (s, 3H, CH_3), 4.39 (q, 1H, J = 7.1, H_5), 6.81–8.01 (m, 7H, Ar and H-2). *Anal*. $C_{16}H_{14}Cl_2N_2OS$ (C, H, N).

*2.1.2.4. cis-2-(2,6-Dichlorophenyl)-3-(6-methyl-pyridin-2-yl)-5-methyl-1,3-thiazolidin-4-one (**4d**).* Yield: 6%, m.p. 137–140 °C; ^{1}H NMR: δ 1.76 (d, 3H, J = 6.8, CH–CH_3), 2.25 (s, 3H, CH_3), 4.19 (q, 1H, J = 6.8, H_5), 6.80–7.96 (m, 7H, Ar and H-2). *Anal*. $C_{16}H_{14}Cl_2N_2OS$ (C, H, N).

*2.1.2.5. trans-2-(2,6-Difluorophenyl)-3-(4,6-dimethyl-pyridin-2-yl)-5-methyl-1,3-thiazolidin-4-one (**3h**).* Yield: 16%, m.p. 105–107 °C; ^{1}H NMR: δ 1.65 (d, 3H, J = 7.1, CH–CH_3), 2.27 (s, 6H, CH_3), 4.41 (q, 1H, J = 7.1, H_5), 6.68–7.90 (m, 6H, Ar and H-2). *Anal*. $C_{17}H_{16}F_2N_2OS$ (C, H, N).

*2.1.2.6. cis-2-(2,6-Difluorophenyl)-3-(4,6-dimethyl-pyridin-2-yl)-5-methyl-1,3-thiazolidin-4-one (**4h**).* Yield: 11%, m.p. 115–117 °C; ^{1}H NMR: δ 1.76 (d, 3H, J = 6.8, CH–CH_3), 2.26 (s, 6H, CH_3), 4.18 (q, 1H, J = 6.8, H_5), 6.66–7.74 (m, 6H, Ar and H-2). *Anal*. $C_{17}H_{16}F_2N_2OS$ (C, H, N).

2.2. Pharmacology

The antiviral experiments using MT-4 cells and HIV-1 (III_B) and -2 (ROD) strains were performed following procedures that have already been described [13,14].

3. Results and discussion

The synthesis of 2,3-diaryl-1,3-thiazolidine-4-thiones (**2**) was carried out by reacting compounds **1** with Lawesson's reagent under reflux in toluene as reported in Scheme 1. The 5-methyl-2,3-diaryl-1,3-thiazolidin-4-ones (**3** and **4**) were obtained by reacting a suitable 2,6-dihalobenzaldehyde with an equimolar amount of a heteroaromatic amine in the presence of an excess of 2-mercaptopropionic acid in refluxing toluene. Owing to the asymmetry of 2-mercaptopropionic acid, the reaction furnished a ca. 2:1 mixture of racemic diastereoisomers **3** and **4** characterized by a different disposition of the substituents at C-2 and C-5. The *trans*-**3** and *cis*-**4** isomers were separated by column chromatography and their structure was assigned on the basis of ^{1}H NMR spectroscopy assisted by NOE measurements. In derivatives **3**, irradiation of 5-methyl resonance gave a positive NOE peak at the H-2 proton so proving the *trans* relationship of the methyl group and the aryl substituent at C-2. On the contrary, in derivatives **4** a

Table 1
Anti-HIV-1 activity, cytotoxicity, selectivity index and log *P* values for compounds **1**–**4**

Comp.	EC_{50} (μM) [a]	CC_{50} (μM) [b]	SI [c]	Log *P* [d]
1a	2.30±0.75	>429	>186	3.43
2a	NA	35.8±1.2		4.48
1b	0.401±0.09	38.1±4.5	95	4.35
2b	NA	31.1±4.4		5.40
1c	0.082±0.03	126.0±34.9	1536	3.22
2c	6.51±0.6	39.4±0.0	6	4.27
3c	3.43±1.7	26.34±0.8	8	4.32
4c	1.90±0.2	20.0±8.6	11	4.32
1d	0.044±0.01	284.7±33.6	6470	4.14
2d	11.48±1.9	21.9±1.2	2	5.19
3d	3.79±1.4	89.23±30.0	24	5.24
4d	1.33±0.2	139.9±40.5	106	5.24
1e	0.248±0.03	242.2±0.3	976	3.34
2e	4.34±1.2	332.8±81.9	77	4.39
1f	0.147±0.05	>368.5	>2500	4.26
2f	NA	179.3±62.8		5.32
1g	0.044±0.02	180±7.5	2454	3.10
2g	0.28±0.02	24.4±15.4	86	4.15
1h	0.059±0.012	43.4±2.9	735	3.66
2h	6.42±0.4	44.3±3.6	7	4.71
3h	3.11±1.4	35.89±1.2	12	4.76
4h	2.06±0.6	36.8±3.0	18	4.76
1i	0.030±0.01	32.0±0.54	1066	3.79
2i	0.58±0.2	31.0±2.8	54	4.84

NA, not active, at subtoxic concentrations.
[a] Concentration required to reduce HIV-1-(III_B) induced cytopathic effect by 50% in MT-4 cells.
[b] Concentration required to reduce MT-4 cell viability by 50%.
[c] Selectivity index: ratio CC_{50}/EC_{50}.
[d] Calculated with the XLOGP program.

positive NOE peak at the H-2 resonance is observed on irradiation of the H-5 signal.

All compounds obtained were tested for anti-HIV activity by determining their ability to inhibit the replication of HIV-1 (III_B) or -2 (ROD) in MT-4 cells and the results compared with that of the analogous 2,3-diaryl-1,3-thiazolidin-4-ones (**1**). As shown in Table 1, some of them proved to be effective inhibitors of HIV-1 replication, whereas, as observed for other classes of NNRTIs, none of the tested compounds inhibited the replication of HIV-2 (ROD) in MT-4 cells at subtoxic concentrations (data not shown).

Analogously to derivatives **1** [10,11], the anti-HIV-1 activity of the 2,3-diaryl-1,3-thiazolidine-4-thiones (**2**) increases by substituting the phenyl ring at N-3 of the thiazolidinone ring with a 2-pyridinyl or 2-pyrimidinyl nucleus bearing an appropriate substituent in a suitable position. In particular, the best results have been obtained with derivatives **2g** and **2i** in which the substituent is a methyl group or a bromine atom present on C adjacent to the nitrogen atom, respectively.

The presence of chlorine atoms at 2 and 6 position on the phenyl ring at C-2 generally increases the anti-HIV activity of compounds **1**. On the contrary, in compounds **2**, the 2,6-difluorophenyl substituted derivatives are more active than the corresponding chlorine ones (**2c** compared to **2d** and **2e** to **2f**), whereas in 5-methyl-1,3-thiazolidin-4-ones (**3** and **4**) the nature of the halogen atoms does not seem to influence the anti-HIV activity (**3c** vs. **3d** and **4c** vs. **4d**). Moreover, not even the stereochemistry seems to play a key role in anti-HIV activity, the *cis* derivatives **4** being only slightly more active than the *trans*-**3** ones.

The replacement of the carbonyl with the thiocarbonyl group negatively influences the activity which appreciably decreases also when a methyl group is present at C-5 atom. In fact, compounds **2**–**4** are all less active than the corresponding derivatives **1**.

These results point out that, as suggested by our molecular modelling studies [11], the antiviral efficacy of 2,3-diaryl-1,3-thiazolidin-4-(thi)ones is modulated by the nature of the aryl substituent at N-3 and in particular by the presence of lipophilic groups in proper positions of the heterocycle. However, a further increase in the lipophilicity (log *P* values, Table 1) resulting from structural modifications on the thiazolidinone nucleus negatively influences anti-HIV activity.

Acknowledgements

This work was supported in part by the Minister dell Istruzione dell Università e della Ricerca (COFIN 2000), Roma, Italy.

References

[1] E. De Clercq, Antiviral drugs: current state of the art, J. Clin. Virol. 22 (2001) 73–89.
[2] J.A. Tavel, K.D. Miller, H. Masur, Guide to major clinical trials of antiretroviral therapy in human immunodeficiency virus-infected patients: protease inhibitors, non-nucleoside reverse transcriptase inhibitors, and nucleotide reverse transcriptase inhibitors, Clin. Infect. Dis. 28 (1999) 643–676.
[3] H. Jonckheere, J. Anné, E. De Clercq, The HIV-1 reverse transcription (RT) process as target for RT inhibitors, Med. Res. Rev. 20 (2000) 129–154.
[4] A. Chimirri, S. Grasso, A.M. Monforte, P. Monforte, M. Zappalà, Anti-HIV agents II. Synthesis and *in vitro* anti-HIV activity of novel 1*H*,3*H*-thiazolo[3,4-*a*]benzimidazoles, II Farmaco 46 (1991) 925–933.
[5] A. Chimirri, S. Grasso, C. Molica, A.M. Monforte, P. Monforte, M. Zappalà, Anti-HIV agents. IV. Synthesis and *in vitro* anti-HIV activity of novel 1-(2,6-difluorophenyl)-1*H*,3*H*-thiazolo[3,4-*a*]benzimidazoles, II Farmaco 51 (1996) 279–282.
[6] A. Chimirri, S. Grasso, C. Molica, A.M. Monforte, P. Monforte, M. Zappalà, G. Bruno, F. Nicolò, M. Witvrouw, H. Jonckeere, J. Balzarini, E. De Clercq, Structural features and anti-human immunodeficiency virus (HIV) activity of the isomers of 1-(2′,6′-difluorophenyl)-1*H*,3*H*-thiazolo[3,4-*a*]benzimidazole, a potent non-nucleoside HIV-1 reverse transcriptase inhibitor, Antiviral Chem. Chemother. 8 (1997) 363–370.

[7] A. Chimirri, S. Grasso, A.M. Monforte, P. Monforte, A. Rao, M. Zappalà, G. Bruno, F. Nicolò, C. Pannecouque, M. Witvrouw, E. De Clercq, Synthesis, structure and *in vitro* anti-human immunodeficiency virus activity of novel 3-methyl-1*H*,3*H*-thiazolo[3,4-*a*]benzimidazoles, Antiviral Chem. Chemother. 9 (1998) 431–438.
[8] A. Chimirri, S. Grasso, P. Monforte, A. Rao, M. Zappalà, A.M. Monforte, C. Pannecouque, M. Witvrouw, J. Balzarini, E. De Clercq, Synthesis and biological activity of novel 1*H*,3*H*-thiazolo[3,4-*a*]benzimidazoles: non-nucleoside human immunodeficiency virus type 1 reverse transcriptase inhibitors, Antiviral Chem. Chemother. 10 (1999) 211–217.
[9] M.L. Barreca, A. Chimirri, A. Carotti, A. Carrieri, A.M. Monforte, M. Pellegrini Calace, A. Rao, Comparative Molecular Field Analysis (CoMFA) and docking studies of non-nucleoside HIV-1 RT inhibitors (NNRTIs), Bioorg. Med. Chem. 7 (1999) 2283–2292.
[10] M.L. Barreca, A. Chimirri, L. De Luca, A.M. Monforte, P. Monforte, A. Rao, M. Zappalà, J. Balzarini, E. De Clercq, C. Pannecouque, M. Witvrouw, Discovery of 2,3-diaryl-1,3-thiazolidin-4-ones as potent anti-HIV-1 agents, Bioorg. Med. Chem. Lett. 11 (2001) 1793–1796.
[11] M.L. Barreca, J. Balzarini, A. Chimirri, E. De Clercq, L. De Luca, H.D. Höltje, M. Höltje, A.M. Monforte, P. Monforte, C. Pannecouque, A. Rao, M. Zappalà, Design, synthesis, structure–activity relationships and molecular modelling studies of 2,3-diaryl-1,3-thiazolidin-4-ones potent anti-HIV agents, J. Med. Chem. in press.
[12] R. Wang, Y. Gao, L. Lai, Calculating partition coefficient by atom-additive method, Perspect. Drug Discov. 19 (2000) 47–66.
[13] R. Pauwels, J. Balzarini, M. Baba, R. Snoeck, D. Schols, P. Herdewijin, J. Desmyter, E. De Clercq, Rapid and automated tetrazolium-based colorimetric assay for the detection of anti-HIV compounds, J. Virol. Methods 20 (1988) 309–321.
[14] M. Witvrouw, D. Schols, G. Andrei, R. Snoeck, M. Hosoya, R. Pauwels, J. Balzarini, E. De Clercq, Antiviral activity of low-MW dextran sulfate (derived from dextran MW 1000) compared to dextran sulfate samples of higher MW, Antiviral Chem. Chemother. 2 (1991) 171–179.