Journal of Research in Pharmacy

Research Article

www.jrespharm.com

Synthesis, characterization and antimicrobial evaluation of new 2-(2-thienylcarbonyl)hydrazono-3-alkyl/aryl-4-thiazolidinone and 2-aryl-3-(2-thienylcarbonyl)amino-4-thiazolidinone derivatives

Efe Doğukan DİNCEL[1,2] *, Dilek ŞATANA[3], Süheyla ÖZBEY[4], Nuray ULUSOY-GÜZELDEMİRCİ[1]

1 İstanbul University, Faculty of Pharmacy, Department of Pharmaceutical Chemistry, 34116 Beyazıt, İstanbul, Turkey
2 İstanbul University, Institute of Graduate Studies in Health Sciences, 34126 İstanbul, Turkey
3 İstanbul University, İstanbul Faculty of Medicine, Department of Microbiology, 34390 İstanbul, Turkey
4 Hacettepe University, Faculty of Engineering, Department of Engineering Physics, 06800 Ankara, Turkey
* Corresponding Author. E-mail: efe.dincel@istanbul.edu.tr (E.D.D.); Tel. +90-536-483 45 13.

Received: 2 December 2021 / Revised: 7 January 2022/ Accepted: 12 January 2022

ABSTRACT: A series of novel 2-(2-thienylcarbonyl)hydrazono-3-alkyl/aryl-4-thiazolidinone (**2a-f**), 2-(2-thienyl)-5-[(4-bromophenyl)amino]-1,3,4-oxadiazole (**3**), 2-aryl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (**5a-h**) and 2-aryl-3-(2-thienylcarbonyl)amino-5-methyl-4-thiazolidinone (**6a-h**) were designed and synthesized. The structural elucidations of the novel compounds were performed by IR, ^{1}H-NMR, mass and elemental analysis. The compounds were evaluated for their antimycobacterial and antifungal activities. According to the biological activity studies activities of the compounds, **2f** (MIC: >12.5 µg/ml, 76% inhibition), **3** (MIC: >12.5 µg/ml, 43% inhibition), and **5c** (MIC: >12.5 µg/ml, 38% inhibition), displayed antimycobacterial activity. **2b (**MIC: 25 µg/ml) displayed antifungal activity against *T. rubrum*. Besides, x-ray crystallography studies were performed to illuminate the structure of **2e**. Consequently, the obtained results relvealed that **2f**, **3** and **5c** present a leading structure for future drug development due to its straightforward synthesis and relevant bioactivity.

KEYWORDS: 4-Thiazolidinones; oxadiazole; crystal structure; antimycobacterial activity; antifungal activity.

1. INTRODUCTION

4-Thiazolidinones are thiazolidine derivatives which possess a sulfur atom at position one, a nitrogen atom at position three, and a carbonyl group at position four. 4-Thiazolidinones have been considered as a pharmacologically active scaffold due to their wide range of reported biological activities in literature. The compounds bearing 4-thiazolidinone moiety have been reported in literature by their antibacterial [1,2], antifungal [2,3], antitubercular [4], anticancer [5], anti-inflammatory [6], analgesic [7], anticonvulsant [8], antiviral [9], cytotoxic [10] and antidiabetic [11] activities. In the recent studies, Türe and collaborators (2021) reported the design, synthesis, and anticancer activity of novel 4-thiazolidinone-phenylaminopyrimidine hybrids. Some of these compounds displayed superior anticancer activity on K562 (chronic myeloid leukemia) cell lines [12]. Çakır and collaborators (2015) described novel 4-thiazolidinones as non-nucleoside inhibitors of Hepatitis C virus NS5B RNA-Dependent RNA Polymerase [13]. In another study, Küçükgüzel and co-workers (2013) reported 2-heteroarylimino-5-arylidene-4-thiazolidinones as a new class of non-nucleoside inhibitors of HCV NS5B polymerase [14]. Herein twenty novel 4-thiazolidinone derivatives were synthesized and eleven of them inhibited NS5B higher than 50% at 100 µM. Tatar and co-workers (2008) reported the synthesis, characterization and screening of antimicrobial, antituberculosis, antiviral and anticancer activity of novel 1,3-thiazolidine-4-ones [15]. Some of these compounds displayed marginal activity against *Staphylococcus aureus* ATCC 29213, *Bacillus subtilis* A57 and *Candida albicans* A177. Besides some compounds displayed varying degree of antituberculosis activity against *Mycobacterium tuberculosis* H37Rv strain. The most potent compound displayed 90% inhibition of mycobacterial growth at 6.25 µg/ml. In another study, Tatar et al. (2010) described the design, synthesis, anti-tuberculosis and antiviral activity of novel 2-isonicotinoylhydrazono-5-arylidene-4-thiazolidinones [16]. Within the synthesized compounds, the most potent one displayed antimycobacterial activity with an IC_{90} value of 2.039 µg/ml and an IC_{50} value of 1.267

Dinçel ED, Şatana D, Özbey S, Ulusoy-Güzeldemirci N. Synthesis, characterization and antimicrobial evaluation of new 2-(2-thienylcarbonyl)hydrazono-3-alkyl/aryl-4-thiazolidinone and 2-aryl-3-(2-thienylcarbonyl)amino-4-thiazolidinone derivatives. J Res Pharm. 2022; 26(3): 641-654.

ISSN: 2630-6344

http://dx.doi.org/10.29228/jrp.161

µg/ml against *Mycobacterium tuberculosis* H37Rv strain. Compounds such as ralitoline (anticonvulsant), pioglitazone (hypoglycemic) and thiazolidomycin (activity against Streptomyces species), based on 4-thiazolidinone pharmacophore are already in clinical use as commercial products (Figure 1).

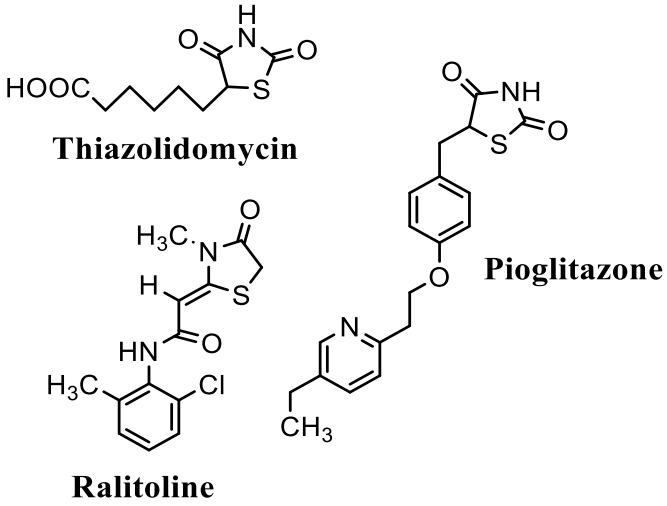

Figure 1. Chemical structures of some drugs bearing 4-thiazolidinone moiety.

In this study two novel series of 4-thiazolidinones were prepared in an attempt to obtain new compounds with antimicrobial properties. Since the structure of 4-thiazolidinones obtained from asymmetric thiourea derivatives has been frequently discussed due to the possibility of structural isomers involving 2- and 3- positions of the thiazolidinone ring depending upon the nitrogen involved in ene thiolization, X-ray diffraction studies were carried out to determine the position of the 2-thienylcarbonylamino residue [17]. The antimycobacterial activity of the compounds against *Mycobacterium tuberculosis* H37Rv was evaluated employing the BACTEC 460 radiometric system. The highest inhibition observed at 12.5 µg/ml was 76%. The antifungal activity of the title compounds against representative fungi was also investigated.

2. RESULTS AND DISCUSSION

2.1. Chemistry

The effects of modification of the ring substituents of the 4-thiazolidinone ring system on biological activity were examined through a series of compounds prepared as described in Figure 2 and Figure 3. 4-Alkyl/aryl-1-(2-thienylcarbonyl)-3-thiosemicarbazides (**1a-f**) were synthesized from thiophene-2-carboxylic acid hydrazide and alkyl/arylisothiocyanates. Cyclization of **1a-f** with $BrCH_2COOC_2H_5$ in the presence of anhydrous CH_3COONa afforded the target 4-thiazolidinones (**2a-f**). An oxadiazole derivative (**3**) was also isolated from the reaction mixture of **2f** by fractional crystallization from C_2H_5OH. Diverse crystals were precipitated at different times during crystallization. The spectral data indicated the difference of these compounds from eachother. 4-Thiazolidinone and 1,3,4-oxadiazole derivatives can be formed from hydrazinecarbothioamides under same conditions. This situation was reported in literature too [18]. Herein, *N*-(4-bromophenyl)-2-(thiophene-2-carbonyl)hydrazine-1-carbothioamide (**1f**) undergo desulfuration to afford 2-(2-thienyl)-5-[(4-bromophenyl)amino]-1,3,4-oxadiazole (**3**) with ethylbromoacetate in the presence of anhydrous sodium acetate. *N*-aryl groups are likely to be less nucleophilic than *N*-alkyl ones. It seems most likely that after the formation of the *S*-alkyl intermediate the carbonyl group attacks the carbon bearing the S atom and makes the *S*-R group leaving group affording ring closure. The absence of C=O bands in the IR spectrum also supports the 1,3,4-oxadiazole structure. *N*-benzylidene-2-thiophenecarboxylic acid hydrazides (**4a-h**) were prepared by condensation of thiophene-2-carboxylic acid hydrazide with appropriate aromatic aldehydes employing a literature method [19]. Compounds **4a-h** furnished 4-thiazolidinones (**5a-h** and **6a-h**) on cyclodehydration with thioglycolic (**5a-h**) or thiolactic (**6a-h**) acid. The structures of **2a-f**, **3**, **5a-h** and **6a-h** were determined by spectral (IR, ^{1}H-NMR, EIMS) and analytical methods (Table 1).

http://dx.doi.org/10.29228/jrp.161
J Res Pharm 2022; 26(3): 641-654

Journal of Research in Pharmacy
Research Article

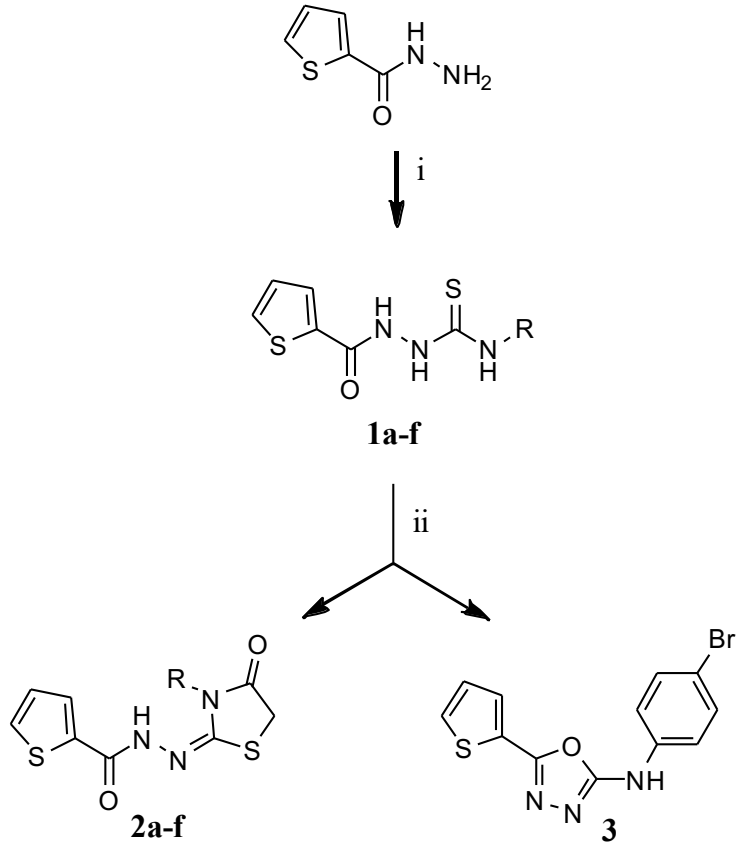

Figure 2. Synthesis of the title compounds (**2a-f** and **3**). (i: RNCS, EtOH, reflux; ii: Ethylbromoacetate, fused sodium acetate, EtOH, reflux).

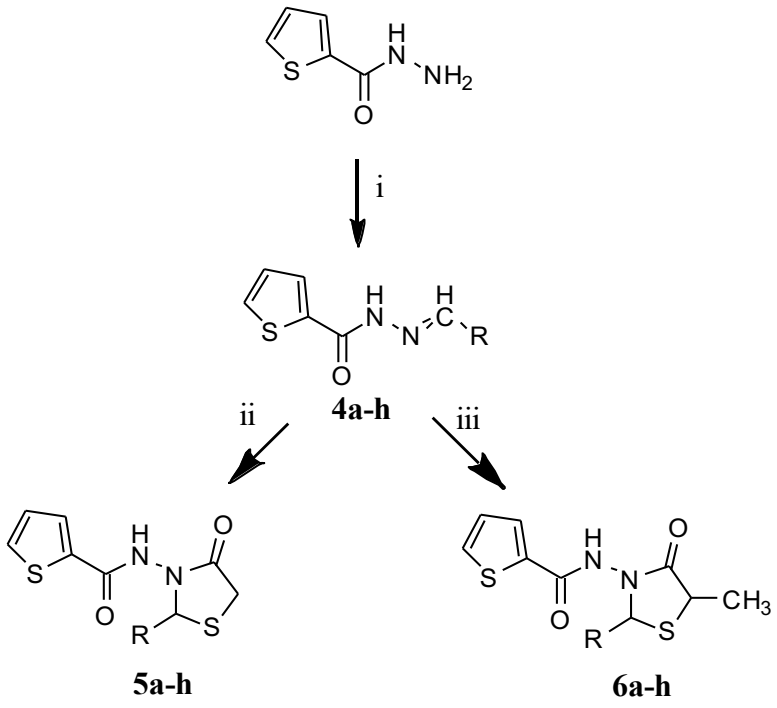

Figure 3. Synthesis of the title compounds (**5a-h** and **6a-h**). (i: RCHO, EtOH, reflux; ii: thioglycolic acid, benzene, reflux; iii: thiolactic acid, benzene, reflux).

http://dx.doi.org/10.29228/jrp.161
J Res Pharm 2022; 26(3): 641-654

The IR spectra exhibited N-H and C=O bands in the 3254-3142 and 1664-1615 cm^{-1} regions attributed to the common CONHN functions of **2a-f**, **5a-h** and **6a-h**. The characteristic lactam C=O absorptions of **2a-f**, **5a-h** and **6a-h** were observed in the 1716-1694 cm^{-1} region.

The ^{1}H-NMR spectra **2a-f** and **5a-h** displayed resonances attributed to the methylene ring protons at about δ 4.18-3.75 ppm. Due to magnetic nonequivalence caused by the chiral center produced by the nucleophilic addition of thioglycolic acid to the N=CH function of **4**, the methylene ring protons of **5a-h** displayed geminal coupling and absorbed as two doublets. Additional support for the structure of **5a-h** and **6a-h** was obtained from the absorption positions of the N-CH-S protons which showed upfield shifts and absorbed at about δ 5.91-5.85 ppm due to the loss of the sp^2 character of the involved C atom. The CH-CH_3 protons of **6a-h** showed a quartet and a doublet at δ 4.11-4.06 and 1.55-1.54 ppm as anticipated. The molecular ions observed in the EIMS confirmed molecular weights of the compounds. The IR, ^{1}H NMR, and MS spectra of the novel compounds are in agreement with the assigned structures. No unacceptable side reactions were observed, and products were obtained in moderate to good yields [20-22].

2.2. X-Ray Diffraction Studies

To definitely determine the position of the thienylcarbonyl residue and thus propose the correct mode of ene thiolization in at least N_4 alkyl substituted thiosemicarbazides which may be regarded as asymmetric thiourea derivatives, X-ray diffraction studies were carried out on **2e** (Tables 3, 4). A perspective view as shown in Figure 4 reveals that the studies compound exhibits nearly planar conformation. The thiazole moiety slightly deviates from planarity [maximum deviation -0.039(5)Å for C_1] while the thiophene ring is planar. The S1-C6-N2-N3-H3 ring, P, completed by the hydrogen bond of N3-H3…S1 is essentially planar, with a maximum deviation from the least-squares plane P, defined by all atoms of 0.007(3)Å for N3. The C9 and O1 atoms deviate from P by only 0.023(4) and 0.013(3) Å, and so lie nearly in this plane. The torsion angles around the N2-N3 and N3-C9 bonds are 179.2(4) and 179.1(3)° respectively, for the C6-N2-N3-C9 and N2-N3-C9-O1 atoms.

The dihedral angles between the best planes of the thiazole and the thiophene ring, the thiazole and P ring are respectively 7.3(7) and 3.1(6)°. The allyl group attached to the thiazole ring is oriented with torsion angle C2-N1-C3-C4 of -101.4(5)°. Crystallographic and refinement parameters are summarized in Table 2,3 and 4.

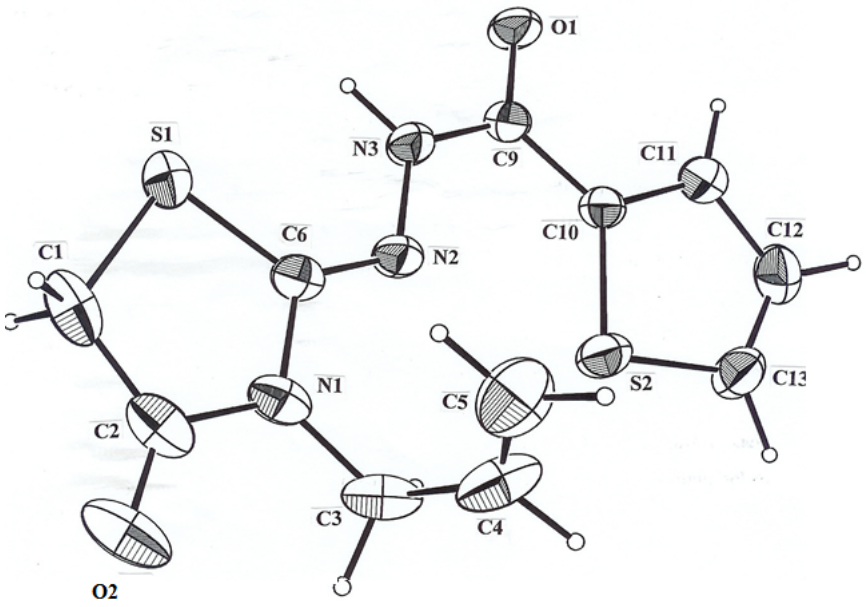

Figure 4. A perspective view of **2e** from X-ray diffraction studies.

Table 1. Physical and analytical data of the synthesized compounds (**2a-f**, **3**, **5a-h** and **6a-h**).

Comp.	R	Formula (M.W.)	M.p. (°C)	Yield (%)	Analysis(calcd./found) C	H	N
2a	CH_3	$C_9H_9N_3O_2S_2$ (255.30)	218-219	76	42.33 42.77	3.55 3.53	16.45 16.65
2b	C_2H_5	$C_{10}H_{11}N_3O_2S_2$ (269.34)	153-154	84	44.59 44.13	4.11 3.93	15.60 15.56
2c	C_3H_7	$C_{11}H_{13}N_3O_2S_2$ (283.36)	153	77	46.62 46.33	4.62 4.78	14.82 14.75
2d	C_4H_9	$C_{12}H_{15}N_3O_2S_2$ (297.39)	148-149	69	48.46 48.17	5.08 5.03	14.12 14.10
2e	CH_2-CH=CH_2	$C_{11}H_{11}N_3O_2S_2$ (281.35)	141-142	66	46.95 46.29	3.94 3.81	14.93 14.95
2f	4-BrC_6H_4	$C_{14}H_{10}BrN_3O_2S_2$ (396.28)	200-201	30	42.43 42.72	2.54 2.34	10.60 10.95
3	4-BrC_6H_4	$C_{12}H_8BrN_3OS$ (322.18)	240-241	28	44.73 44.34	2.50 2.33	13.04 12.83
5a	C_6H_5	$C_{14}H_{12}N_2O_2S_2$ (304.38)	190-191	99	55.24 55.64	3.97 4.03	9.20 8.77
5b	3-$CH_3C_6H_4$	$C_{15}H_{14}N_2O_2S_2.0.5H_2O$ (327.41)	205-207	87	55.03 54.31	4.62 4.20	8.56 7.92
5c	4-$CH_3C_6H_4$	$C_{15}H_{14}N_2O_2S_2$ (318.41)	204-205	91	56.58 56.49	4.43 4.34	8.79 8.64
5d	4-$CH_3OC_6H_4$	$C_{15}H_{14}N_2O_3S_2$ (334.41)	219-220	41	53.87 54.15	4.21 4.16	8.37 8.23
5e	4-FC_6H_4	$C_{14}H_{11}FN_2O_2S_2$ (322.37)	172	98	52.16 52.10	3.44 3.33	8.68 8.61
5f	3-ClC_6H_4	$C_{14}H_{11}ClN_2O_2S_2.H_2O$ (356.83)	158	71	47.12 47.60	3.67 3.17	7.85 7.97
5g	4-ClC_6H_4	$C_{14}H_{11}ClN_2O_2S_2$ (338.82)	197	96	49.62 49.02	3.27 2.92	8.26 7.97
5h	4-BrC_6H_4	$C_{14}H_{11}BrN_2O_2S_2$ (383.28)	213-214	99	43.87 44.14	2.89 2.67	7.30 7.19
6a	C_6H_5	$C_{15}H_{14}N_2O_2S_2$ (318.41)	182-184	95	56.58 56.00	4.43 4.29	8.79 8.46
6b	3-$CH_3C_6H_4$	$C_{16}H_{16}N_2O_2S_2.1.5H_2O$ (359.45)	149-153	65	53.46 53.95	5.33 4.67	7.79 7.39
6c	4-$CH_3C_6H_4$	$C_{16}H_{16}N_2O_2S_2.0.5H_2O$ (341.44)	189-191	95	56.28 56.40	5.01 4.72	8.20 8.07
6d	4-$CH_3OC_6H_4$	$C_{16}H_{16}N_2O_3S_2$ (348.44)	185-186	93	55.15 55.00	4.62 4.69	8.03 7.92
6e	4-FC_6H_4	$C_{15}H_{13}FN_2O_2S_2$ (336.40)	183-184	91	53.55 53.00	3.89 3.85	8.32 8.08
6f	3-ClC_6H_4	$C_{15}H_{13}ClN_2O_2S_2.1.5H_2O$ (379.87)	154-155	80	47.43 47.42	4.25 3.25	7.37 6.91
6g	4-ClC_6H_4	$C_{15}H_{13}ClN_2O_2S_2.0.5H_2O$ (361.85)	190-191	98	49.79 49.59	3.89 3.53	7.74 7.46
6h	4-BrC_6H_4	$C_{15}H_{13}BrN_2O_2S_2.H_2O$ (415.33)	198-199	94	43.37 43.65	3.64 3.43	6.74 6.76

http://dx.doi.org/10.29228/jrp.161
J Res Pharm 2022; 26(3): 641-654

Table 2. Experimental data for the crystallographic analysis [a].

Formula: $C_{11}H_{11}N_3O_2S_2$
Formula weight = 281.36
Crystal system: monoclinic
Space group: $P2_1/c$ $Z = 4$
a =5.655(1) Å
b = 14.387(2) Å β = 96.02(1)°
c = 15.428(2) Å
V = 1248.3(4) Å ^{3}
D_x = 1.50 g/cm^3
μ(Mo K_α)=0.41 mm^{-1}
$2\theta_{max}$= 52.6° with Mo K_α
No. of reflections measured = 2900
No. of reflections used = 1280 (I>2σ(I))
R = 0.045
wR = 0.049
$(\Delta/\sigma)_{max}$ =0.001
$(\Delta/\rho)_{max}$ =0.26
$(\Delta/\rho)_{min}$ =–0.24

[a] Measurement: Enraf Nonius CAD-4 diffractometer.
Program system: CAD-4 EXPRESS Software.
Structure determination: SIR in MolEN.
Refinement: full matrix least-squares.

Table 3. Fractional atomic coordinates and equivalent isotropic displacement parameters (Å^2) for **2e**.

Atom	X	Y	Z	Beq
S1	0.9912(2)	0.38208(9)	0.85381(8)	4.37(3)
S2	0.9800(2)	0.75999(8)	0.90976(9)	4.35(3)
O1	1.5216(5)	0.6148(2)	1.0247(2)	4.36(7)
O2	0.4136(6)	0.4351(3)	0.7050(3)	7.4(1)
N1	0.7010(6)	0.5128(3)	0.7910(2)	4.26(9)
N2	1.0145(6)	0.5699(3)	0.8823(2)	3.39(7)
N3	1.2222(6)	0.5555(2)	0.9367(2)	3.45(8)
C1	0.7209(9)	0.3488(4)	0.7880(3)	5.5(1)
C2	0.5921(8)	0.4350(4)	0.7557(3)	5.0(1)
C3	0.6101(8)	0.6052(4)	0.7695(3)	5.4(1)
C4	0.743(1)	0.6547(4)	0.7050(3)	6.2(1)
C5	0.907(1)	0.6235(4)	0.6628(3)	6.2(1)
C6	0.9118(7)	0.4993(3)	0.8461(3)	3.36(9)
C9	1.3381(7)	0.6284(3)	0.9755(3)	3.24(9)
C10	1.2510(7)	0.7240(3)	0.9610(3)	3.24(9)
C11	1.3830(8)	0.7997(3)	0.9916(3)	4.3(1)
C12	1.2620(9)	0.8841(4)	0.9731(4)	5.3(1)
C13	1.0474(9)	0.8731(3)	0.9303(3)	4.7(1)
H1	0.639	0.315	0.823	5.8
H2	0.762	0.312	0.743	8.2
H3	1.282	0.494	0.947	4.4
H4	0.700	0.718	0.693	8.0
H11	1.538	0.795	1.022	5.3
H12	1.333	0.946	0.984	7.1
H13	0.919	0.918	0.911	5.1
H31	0.440	0.602	0.750	5.7
H32	0.629	0.640	0.820	4.1
H51	0.9774	0.6625	0.6231	8.2
H52	0.9588	0.5610	0.6710	8.2

Table 4. Selected geometric parameters of **2e**.

S1 - C1	1.808(5)	N2 - C6	1.270(6)
S1 - C6	1.746(5)	N3 - C9	1.344(5)
S2 - C10	1.729(4)	C1 - C2	1.497(7)
S2 - C13	1.694(5)	C3 - C4	1.489(8)
O1 - C9	1.236(5)	C4 - C5	1.270(8)
O2 - C2	1.210(6)	C9 - C10	1.470(6)
N1 - C2	1.363(7)	C10 - C11	1.375(6)
N1 - C3	1.451(7)	C11 - C12	1.408(7)
N1 - C6	1.404(5)	C12 - C13	1.329(7)
N2 - N3	1.386(4)	-	-
-	-	-	-
C1 — S1 — C6	91.2(2)	O2 — C2 — N1	124.6(5)
C10 — S2 — C13	91.8(2)	N1 — C2 — C1	111.3(4)
C2 — N1 — C6	116.6(4)	N1 — C3 — C4	113.6(4)
N3 — N2 — C6	117.8(3)	S2 — C10— C11	110.0(3)
N2 — N3 — C9	119.7(3)	S2 — C13— C12	112.5(4)
S1 — C6 — N1	111.8(3)	C3 — C4 — C5	128.3(5)
S1 — C1 — C2	108.7(4)	C10 —C11— C12	112.3(4)
O1 — C9 — N3	119.2(4)	C11 —C12 —C13	113.4(4)
N3 — C9 — C10	121.5(3)	-	-
-	-	-	-
C2 — N1 — C3 — C4	-101.4(5)	-	-
N3 — N2 — C6 — S1	0.2(6)	-	-
C6 — N2 — N3 — C9	179.2(4)	-	-
N2 — N3 — C9 — C10	-0.7(6)	-	-
N1 — C3 — C4 — C5	6.3(8)	-	-
N3 — C9 — C10 — S2	10.6(6)	-	-

2.3. Biological Activity Evaluation

The antimycobacterial activity of the title compounds were investigated employing the BACTEC 460 radiometric system. Compounds showing inhibition against *Mycobacterium tuberculosis* H37R**v** at 12.5 µg/ml are presented in Table 5. 4-BrC_6H_4 substitution increased the antimycobacterial activity for **2a-f**. As can be seen in Table 6, the compounds showed only marginal activity against representative dermatophyte strains. Compound **2b** being the most active showed inhibition at 25 µg/ml against *T. rubrum*.

Table 5. Antimycobacterial activity of **2f**, **3** and **5c**

Compound	R	MIC (µg/ml)	% Inhibition[a]
2f	4-BrC_6H_4	>12.5	76
3	4-BrC_6H_4	>12.5	43
5c	4-$CH_3C_6H_4$	>12.5	38

[a]MIC Rifampicin = 0.25 µg/ml, 97-99% inhibition vs. *M. tuberculosis* H37Rv.

4. CONCLUSION

In this study, in an attempt to find novel antifungal and antimycobacterial agents, 23 diverse 2-(2-thienylcarbonyl)hydrazono-3-alkyl/aryl-4-thiazolidinone, 1,3,4-oxadiazole and 2-aryl-3-(2-thienylcarbonyl)amino-4-thiazolidinone derivatives were designed and synthesized. X-Ray crstallographic structure of **2e** was illuminated. The final compounds were screened for their *in vitro* antifungal activity against *T. rubrum, T. mentagrophytes, M. canis* and antituberculosis activity against *Mycobacterium tuberculosis* H37Rv strain. The compounds displayed only marginal activity against the representative dermatophyte strains. The activity results revealed that **2f**, **3** and **5c** was the most active antimycobacterial compounds by 76%, 43% and 38% inhibition values respectively. In this context, these compounds could be an interesting starting point for further structural optimization to obtain promising and more potent antitubercular agents.

Table 6. Antifungal activity of **2a-e**, **3**, **5a-h** and **6a-h**.

Compound	A MIC (µg/ml)	B MIC (µg/ml)	C MIC (µg/ml)
2a	50	50	50
2b	25	50	50
2c	50	50	50
2d	>50	50	50
2e	50	50	50
3	50	50	50
5a	50	50	50
5b	50	50	50
5c	50	50	50
5d	>50	50	50
5e	>50	50	>50
5f	>50	50	50
5g	50	50	50
5h	50	50	50
6a	50	50	50
6b	50	50	50
6c	50	50	50
6d	50	50	50
6e	50	>50	50
6f	50	50	50
6g	50	50	>50
6h	50	50	50
Ketoconazole	1.6	6	6

[a] A = *T. rubrum*, B = *T. mentagrophytes*, C = *M. canis*.

5. MATERIALS AND METHODS

5.1. Chemistry

All chemicals were purchased locally from Aldrich, Fluka and Merck. Melting points were determined with a Büchi 530 melting point apparatus in open capillaries and uncorrected. IR (KBr) and ^{1}H-NMR (DMSO-d_6) spectra were recorded on Perkin-Elmer 1600 FT-IR, Bruker AC 200 (200 MHz) and Oxford Pulsar (60 MHz) instruments, respectively. EIMS were recorded on a VG Zab Spec (70 eV) instrument. Elemental analyses were performed on a Carlo Erba 1106 elemental analyzer.

5.1.2. Synthesis of 2-(2-thienylcarbonyl)hydrazono-3-alkyl/aryl-4-thiazolidinones ***(2a-f)***

A mixture of an appropriate thiosemicarbazide **1** (0.005 mol), ethyl bromoacetate (0.0055 mol) and fused sodium acetate (0.02 mol) were refluxed in 30 ml absolute C_2H_5OH for 3-4 h. The reaction mixture was cooled, diluted with water and allowed to stand overnight. The precipitate thus obtained was filtered, dried and recrystallized from C_2H_5OH.

3-Methyl-2-(2-thienylcarbonyl)hydrazono-4-thiazolidinone (2a)

White solid, mp, 218-219 °C, yield 76%. Anal. Calcd. for $C_9H_9N_3O_2S_2$: C, 42.33; H, 3.55; N, 16.45%. Found: C, 42.77; H, 3.53; N, 16.65%. IR υ_{max} (KBr, cm^{-1}): 3146 (N-H stretching), 1707 (C=O stretching, ring), 1617 (C=O stretching). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 3.18 (s, N-CH_3 and solvent H_2O), 4.05 (s, 2H, CH_2-S), 7.18 (distorted s, 1H, C_4-H thiophene), 7.80 (s, 2H, C_3-H and C_5-H thiophene), 10.78 (s, 1H, NH).

3-Ethyl-2-(2-thienylcarbonyl)hydrazono-4-thiazolidinone (2b)

White solid, mp, 153-154 °C, yield 84%. Anal. Calcd. for $C_{10}H_{11}N_3O_2S_2$: C, 44.59; H, 4.11; N, 15.60%. Found: C, 44.13; H, 3.93; N, 15.56%. IR υ_{max} (KBr, cm^{-1}): 3148 (N-H stretching), 1706 (C=O stretching, ring), 1615 (C=O stretching). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 1.19 (t, *J* = 7.1 Hz, 3H, CH_2-C**H$_3$**), 3.77 (q, *J* = 6.9 Hz, 2H, C**H$_2$**-CH_3), 4.04 (s, 2H, CH_2-S), 7.17 (t, *J* = 4.3 Hz, 1H, C_4-H thiophene), 7.80 (d, *J* = 4.7 Hz, 2H, C_3-H and C_5-H thiophene), 10.78 (s, 1H, NH). EIMS m/z (%): 269 (M^+, 67), 111 (100).

3-Propyl-2-(2-thienylcarbonyl)hydrazono-4-thiazolidinone (2c)

White solid, mp, 153 °C, yield 77%. Anal. Calcd. for $C_{11}H_{13}N_3O_2S_2$: C, 46.62; H, 4.62; N, 14.82%. Found: C, 46.33; H, 4.78; N, 14.75%. IR υ_{max} (KBr, cm^{-1}): 3143 (N-H stretching), 1715 (C=O stretching, ring), 1616 (C=O stretching). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 0.88 (t, J = 7.01 Hz, 3H, -CH_2-C**H**$_3$), 1.44-1.85 (m, 2H, N-CH_2-C**H**$_2$-), 3.70 (distorted t, *J* = 7.0 Hz, 2H, N-C**H**$_2$-), 4.06 (s, 2H, CH_2-S), 7.17 (distorted t, *J* = 7.0 Hz, 1H, C_4-H thiophene), 7.80 (s, 2H, C_3-H and C_5-H thiophene), 10.75 (s, 1H, NH).

3-Butyl-2-(2-thienylcarbonyl)hydrazono-4-thiazolidinone (2d)

White solid, mp, 148-149 °C, yield 69%. Anal. Calcd. for $C_{12}H_{15}N_3O_2S_2$: C, 48,46; H, 5.08; N, 14.12%. Found: C, 48.17; H, 5.03; N, 14.10%. IR υ_{max} (KBr, cm^{-1}): 3150 (N-H stretching), 1710 (C=O stretching, ring), 1619 (C=O stretching). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 0.90 (t, *J* = 7.2 Hz, 3H, N-$CH_2CH_2CH_2C$**H**$_3$), 1.31 (sextet, *J* = 7.3 Hz, 2H, N-CH_2CH_2C**H**$_2CH_3$), 1.62 (quin, *J* = 7.3 Hz, 2H, N-CH_2C**H**$_2CH_2CH_3$), 3.72 (t, *J* = 7.1 Hz, 2H, N-C**H**$_2CH_2CH_2CH_3$), 4.04 (s, 2H, CH_2-S), 7.16 (t, *J* = 4.3 Hz, 1H, C_4-H thiophene), 7.79 (d, *J* = 4.5 Hz, 2H, C_3-H and C_5-H thiophene), 10.75 (s, 1H, NH).

3-Allyl-2-(2-thienylcarbonyl)hydrazono-4-thiazolidinone (2e)

White solid, mp, 141-142 °C, yield 66%. Anal. Calcd. for $C_{11}H_{11}N_3O_2S_2$: C, 46.95; H, 3.94; N, 14.93%. Found: C, 46.29; H, 3.81; N, 14.95%. IR υ_{max} (KBr, cm^{-1}): 3142 (N-H stretching), 1716 (C=O stretching, ring), 1624 (C=O stretching). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 4.08 (s, 2H, CH_2-S), 4.33 (d, *J* = 5.0 Hz, 2H, N-CH_2), 5.18 (dd, *J* = 10.1; 1.0 Hz, cis, 1H, CH=**H**CH), 5.19 (dd, *J* = 17.2; 0.9 Hz, trans, 1H, CH=**H**CH), 5.78-5.97 (m, 1H, =CH), 7.15 (t, *J* = 4.2 Hz, 1H, C_4-H thiophene), 7.79 (d, *J* = 4.9 Hz, 2H, C_3-H and C_5-H thiophene), 10.76 (s, 1H, NH). EIMS m/z (%): 281 (M^+, 100).

3-(4-Bromophenyl)-2-(2-thienylcarbonyl)hydrazono-4-thiazolidinone (2f)

White solid, mp, 200-201 °C, yield 30%. Anal. Calcd. for $C_{14}H_{10}BrN_3O_2S_2$: C, 42.43; H, 2.54; N, 10.60%. Found: C, 42.72; H, 2.34; N, 10.95%. IR υ_{max} (KBr, cm^{-1}): 3254 (N-H stretching), (1715 C=O stretching, ring), 1626 (C=O stretching). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 4.18 (s, 2H, CH_2-S), 7.10 (s, 1H, C_4-H thiophene), 7.23-7.75 (m, 4H, Ar-H), 7.84 (d, *J* = 5.0 Hz, 2H, C_3-H and C_5-H thiophene), 10.77 (s, 1H, NH). EIMS m/z (%): 397 [$(M+2)^+$, 30], 395 (M^+, 29), 183 (100).

5.1.3. Synthesis of 2-(2-thienyl)-5-[(4-bromophenyl)amino]-1,3,4-oxadiazole (3)

Obtained as described under compounds 2 and isolated from the reaction mixture of 2f by fractional crystallization from C_2H_5OH.

White solid, mp, 240-241 °C, yield 28%. Anal. Calcd. For $C_{12}H_8BrN_3OS$: C, 44.73; H, 2.50; N, 13.04%. Found: C, 44.34; H, 2.33; N, 12.83%. IR υ_{max} (KBr, cm^{-1}): 3252 (N-H stretching band). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 7.26 (t, *J* = 4.3 Hz, 1H, C_4 thiophene), 7.54 (d, *J* = 9.0 Hz, 2H, Ar-H), 7.58 (d, *J* = 9.0 Hz, 2H, Ar-H), 7.65 (d, *J* = 3.5 Hz, 1H, C_3-thiophene), 7.85 (d, *J* = 4.8 Hz, 1H, C_5-thiophene), 10.90 (S, 1H, NH). EIMS m/z (%): 323 ($(M+2)^+$, 9), 321 (M^+, 27), 151 (100).

5.1.4. Synthesis of 2-aryl-3-(2-thienylcarbonyl)amino-4-thiazolidinones and 2-aryl-3-(2-thienylcarbonyl)amino-5-methyl-4-thiazolidinones (5a-h, 6a-h)

A mixture of **4** (0.005 mol) and thioglycolic/thiolactic acid (0.005 mol) was refluxed in 30 ml dry benzene for 5-6 h using a Dean-Stark water separator. Excess benzene was evaporated in vacuo. The resulting residue was triturated with saturated $NaHCO_3$ solution until CO_2 evolution ceased and was allowed to stand overnight. The solid thus obtained was washed with water, dried, and recrystallized from C_2H_5OH-H_2O.

2-Phenyl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (5a)

White solid, mp, 190-191 °C, yield 99%. Anal. Calcd. for $C_{14}H_{12}N_2O_2S_2$: C, 55.24; H, 3.97; N, 9.20%. Found: C, 55.64; H, 4.03; N, 8.77%. IR υ_{max} (KBr, cm^{-1}): 3220 (N-H stretching), 1701 (C=O stretching, ring), 1653 (C=O stretching). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 3.85 (s, 2H, CH_2-S), 5.91 (s, 1H, N-CH-S), 7.05-7.42 (m, 6H, C_4-H thiophene and Ar.-H), 7.70-7.87 (m, 2H, C_3-H and C_5-H thiophene), 10.69 (s, 1H, NH).

2-(3-Methylphenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (5b)

White solid, mp, 205-207 °C, yield 87%. Anal. Calcd. For $C_{15}H_{14}N_2O_2S_2.0.5H_2O$: C, 53.03; H, 4.62; N, 8.56%. Found: C, 54.31; H, 4.20; N, 7.92%. IR υ_{max} (KBr, cm^{-1}): 3228 (N-H/O-H stretching), 1705 (C=O stretching), 1654 (C=O) stretching). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 2.29 (s, 3H, Ar CH_3), 3.75, 3.89 (2d, *J* = 16.0 Hz, 2H, CH_2-S), 5.86 (s, 1H, N-CH-S), 7.09-7.20 (m, 3H, Ar and C_4-H thiophene), 7.35 (d, *J* = 8.0 Hz, 2H, Ar-H), 7.71 (d, J = 3.6 Hz, 1H, C_3-thiophene), 7.82 (d, *J* = 4.8 Hz, 1H, C_5-H thiophene), 10.62 (s, 1H, NH).

2-(4-Methylphenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (5c)

White solid, mp, 204-205 °C, yield 91%. Anal. Calcd. For $C_{15}H_{14}N_2O_2S_2$: C, 56.58; H, 4.43; N, 8.79%. Found: C, 56.49; H, 4.34; N, 8.64%. IR υ_{max} (KBr, cm^{-1}): 3237 (N-H stretching), 1701 (C=O ring), 1653 (C=O). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 2.29 (s, 3H, Ar-CH_3), 3.75; 3.89 (2d, *J* = 16.0 Hz, 2H, CH_2-S), 5.86 (s, 1H, N-CH-S), 7.09-7.20 (m, 3H, Ar and C_4-H thiophene), 7.35 (d, *J* = 8.0 Hz, 2H, Ar), 7.71 (d, *J* = 3.6 Hz, 1H, C_3-H thiophene), 7.82 (d, J = 4.8 Hz, 1H, C_5-H), 10.62 (s, 1H, NH). EIMS m/z (%): 318 (M^+, 100).

2-(4-Methoxyphenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (5d)

White solid, mp, 219-220 °C, yield 41%. Anal. Calcd. For $C_{15}H_{14}N_2O_3S_2$: C, 53.84; H, 4.21; N, 8.37%. Found: C, 54.15; H, 4.16; N, 8.23%. IR υ_{max} (KBr, cm^{-1}): 3232 (N-H stretching), 1702 (C=O ring), 1654 (C=O). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 3.75 (s, 3H, OCH_3), 3.83 (s, 2H, CH_2-S), 5.87 (s, 1H, N-CH-S), 6.85-7.35 (m, 4H, C_4-H thiophene and ar), 7.70-7.87 (m, 2H, C_3-H and C_5-thiophene), 10.64 (s, 1H, NH).

2-(4-Fluorophenyl)-3-(2-thienylcarbontl)amino-4-thiazolidinone (5e)

White solid, mp, 172 °C, yield: 98%. Anal. Calcd. For $C_{14}H_{11}FN_2O_2S_2$: C, 52.16; H, 3.44; N, 8.68%. Found: C, 52.10; H, 3.33; N, 8.61%. IR υ_{max} (KBr, cm^{-1}): 3226 (N-H stretching), 1694 (C=O ring), 1652 (C=O). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 3.77, 3.93 (2d, *J* = 15.9 Hz, 2H, CH_2-S), 5.91 (s, 1H, N-CH-S), 7.11-7.24 (m, 3H, ar and C_4-H thiophene), 7.49-7.58 (m, 2H, Ar-H), 7.71 (d, *J* = 3.6 Hz, 1H, C_3-H thiophene), 7.83 (d, *J* = 4.8 Hz, 1H, C_5-H thiophene), 10.63 (s, 1H, NH). EIMS m/z (%): 322 (M^+, 100).

2-(3-Chlorophenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (5f)

White solid, mp, 158 °C, yield: 71%. Anal. Calcd. For $C_{14}H_{11}ClN_2O_2S_2.H_2O$: C, 47.12; H, 3.67; N, 7.85%. Found: C, 47.60; H, 3.17; N, 7.97%. IR υ_{max} (KBr, cm^{-1}): 3223 (N-H/O-H), 1708 (C=O ring), 1655 (C=O). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 3.78, 3.99 (2d, *J* = 15.8 Hz, 2H, CH_2-S), 5.91 (s, 1H, N-CH-S), 7.14 (t, *J* = 4.3 Hz, 1H, C_4-H thiophene), 7.41 (s, 3H, Ar), 7.59 (s, 1H, Ar), 7.73 (d, *J* = 4.4 Hz, 1H, C_3-H thiophene), 7.85 (d, *J* = 5.0Hz, 1H, C_5-H thiophene), 10.71 (s, 1H, NH).

2-(4-Chlorophenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (5g)

White solid, mp, 187 °C, yield: 96%. Anal. Calcd. For $C_{14}H_{11}ClN_2O_2S_2$: C, 49.62; H, 3.27; N, 8.26%. Found: C, 49.02; H, 2.92; N, 7.97%. IR υ_{max} (KBr, cm^{-1}): 3232 (N-H), 1702 (C=O ring), 1649 (C=O). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 2.29 (s, 3H, Ar-CH_3), 3.84-3.90 (m, 2H, CH_2S), 5.90 (s, 1H, NCHS), 7.14 (t, *J* = 4.3 Hz, 1H, C_4 thiophene), 7.50, 7.54 (2s, 4H, Ar), 7.69-7.89 (m, 2H, C_3-H and C_5-H thiophene), 10.69 (s, 1H, NH).

2-(4-Bromophenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (5h)

White solid, mp, 213-214 °C, yield: 99%. Anal. Calcd. For $C_{14}H_{11}BrN_2O_2S_2$: C, 43.87; H, 2.89; N, 7.30%. Found: C, 44.14; H, 2.67; N, 7.19%. IR υ_{max} (KBr, cm^{-1}): 3236 (N-H stretching), 1701 (C=O ring), 1648 (C=O). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 3.84-3.88 (m, 2H, CH_2-S), 5.91 (s, 1H, NCHS), 7.13 (dd, *J* = 3.9, 1.1 Hz, 1H, C_4-H thiophene), 7.48 (s, 4H, Ar-H), 7.69-7.88 (m, 2H, C_3-H and C_5-H thiophene), 10.68 (s, 1H, NH).

5-Methyl-2-phenyl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6a)

White solid, mp, 182-184 °C, yield 95%. Anal. Calcd. For $C_{15}H_{14}N_2O_2S_2$: C, 56.58; H, 4.43; N, 8.79%. Found: C, 56.00; H, 4.29; N, 8.46%. IR υ_{max} (KBr, cm^{-1}): 3228 (N-H stretching), 1697 (C=O ring), 1661 (C=O ring).). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 1.55 (d, *J* = 6.9 Hz, 3H, C**H**$_3$CHS), 4.10 (q, *J* = 6.2 Hz, 1H, CH_3C**H**S), 5.90 (s, 1H, NCHS), 7.05-7.42 (m, 6H, C_4-H thiophene and ar), 7.69-7.87 (m, 2H, C_3-H and C_5-H thiophene), 10.75 (s, 1H,, NH).

5-Methyl-2-(3-methylphenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6b)

White solid, mp, 149-153 °C, yield 65%. Anal. Calcd. For $C_{16}H_{16}N_2O_2S_2.1.5H_2O$: C, 53.46; H, 5.33; N, 7.79%. Found: C, 53.95; H, 4.67; N, 7.39%. IR υ_{max} (KBr, cm^{-1}): 3245 (NH/OH), 1698 (C=O ring), 1661 (C=O).). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 1.54 (d, *J* = 6.8 Hz, 3H, C**H**$_3$CHS), 2.29 (s, 3H, Ar CH_3), 4.06 (q, *J* = 7.0 Hz, 1H, CH_3C**H**S), 5.85 (s, 1H, NCHS), 7.09-7.20 (m, 3H, ar and C_4-H thiophene), 7.35 (d, *J* = 8.0 Hz, 2H, Ar), 7.71 (d, *J* = 3.8 Hz, 1H, C_3-H thiophene), 7.82 (d, *J* = 4.8 Hz, 1H, C_5-H thiophene), 10.66 (s, 1H, NH).

5-Methyl-2-(4-methylphenyl)-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6c)

White solid, mp, 189-191 °C, yield 95%. Anal. Calcd. For $C_{16}H_{16}N_2O_2S_2.0.5H_2O$: C, 56.28; H, 5.01; N, 8.20%. Found: C, 56.40; H, 4.72; N, 8.07%. IR υ_{max} (KBr, cm^{-1}): 3230 (NH/OH stretching), 1700 (C=O ring), 1658 (C=O). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 1.54 (d, *J* = 6.8 Hz, 3H, C**H**$_3$CHS), 2.29 (s, 3H, Ar CH_3), 4.06 (q, *J* = 7.0 Hz, 1H, CH_3C**H**S), 5.85 (s, 1H, NCHS), 7.09-7.20 (m, 3H, ar and C_4-H thiophene), 7.35 (d, *J* = 8.0 Hz, 2H, Ar), 7.71 (d, *J* = 3.8 Hz, 1H, C_3-H thiophene), 7.82 (d, *J* = 4.8 Hz, 1H, C_5-H thiophene), 10.66 (s, 1H, NH). EIMS m/z (%): 332 (M^+,3), 245 (100).

2-(4-Methoxyphenyl)-5-methyl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6d)

White solid, mp, 185-186 °C, yield 93%. Anal. Calcd. For $C_{16}H_{16}N_2O_3S_2$: C, 55.15; H, 4.62; N, 8.03%. Found: C, 55.00; H, 4.69; N, 7.92%. IR υ_{max} (KBr, cm^{-1}): 3191 (NH stretching), 1720 (C=O ring), 1645 (C=O). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 1.55 (d, *J* = 6.9 Hz, 3H, C**H**$_3$CHS), 3.31 (s, 3H, ArOCH_3), 4.13 (q, *J* = 7.3 Hz, 1H, CH_3C**H**S), 5.93 (s, 1H, NCHS), 7.07-7.38 (m, 1H, C_4-H thiophene), 7.52-7.98 (m, 5H, ar and, C_3-H thiophene), 8.26 (broad s, 1H, C_5-H thiophene), 10.85 (s, 1H, NH).

2-(4-Fluorophenyl)-5-methyl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6e)

White solid, mp, 183-184 °C, yield 91%. Anal. Calcd. For $C_{15}H_{13}FN_2O_2S_2$: C, 53.55; H, 3.89; N, 8.32%. Found: C, 53.00; H, 3.85; N, 8.08%. IR υ_{max} (KBr, cm^{-1}): 3228 (NH), 1697 (C=O ring), 1664 (C=O). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 1.55 (d, *J* = 4.5 Hz, 3H, C**H**$_3$CHS), 4.08 (q, *J* = 6.8 Hz, 1H, CH_3C**H**S), 5.90 (s, 1H, NCHS), 7.10-7.25 (m, 3H, Ar and C_4-H thiophene), 7.50-7.57 (m, 2H, Ar), 7.69 (d, *J* = 3.7Hz, 1H, C_3-H thiophene), 7.83 (d, *J* = 4.9 Hz, 1H, C_5-H thiophene), 10.68 (s, 1H, NH). EIMS m/z (%): 336 (M^+,5), 111 (100).

2-(3-Chlorophenyl)-5-methyl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6f)

White solid, mp, 154-155 °C, yield 80%. Anal. Calcd. For $C_{15}H_{13}ClN_2O_2S_2.1.5H_2O$: C, 47.43; H, 4.25; N, 7.37%. Found: C, 47.42; H, 3.25; N, 6.91%. IR υ_{max} (KBr, cm^{-1}): 3185 (NH/OH), 1697 (C=O ring), 1658 (C=O). ^{1}HNMR (200 MHz) (DMSO-d_6/TMS) δ (ppm): 1.55 (d, *J* = 4.5 Hz, 3H, C**H**$_3$CHS), 4.11 (q, *J* = 6.9Hz, 1H, CH_3CHS), 5.92 (s, 1H, NCHS), 7.14 (t, *J* = 4.3 Hz, 1H, C_4-H thiophene), 7.42 (s, 3H, Ar), 7.56 (s, 1H, Ar), 7.71 (d, *J* = 3.7 Hz, 1H, C3-H thiophene), 7.85 (d, *J* = 4.9 Hz, 1H, C_5-H thiophene), 10.75 (s, 1H, NH).

2-(4-Chlorophenyl)-5-methyl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6g)

White solid, mp, 190-191 °C, yield 98%. Anal. Calcd. For $C_{15}H_{13}ClN_2O_2S_2.0.5H_2O$: C, 49.79; H, 3.89; N, 7.74%. Found: C, 49.59; H, 3.53; N, 7.46%. IR υ_{max} (KBr, cm^{-1}): 3235 (NH/OH), 1701 (C=O ring), 1660 (C=O). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 1.54 (d, *J* = 6.8 Hz, 3H, CH_3CHS), 2.29 (s, 3H, Ar-CH3), 4.06 (q, *J* = 7.0Hz, 1H, CH_3C**H**S), 5.85 (s, 1H, NCHS), 7.09-7.20 (m, 3H, Ar and C4-H thiophene), 7.35 (d, *J* = 8.0 Hz, 2H, Ar), 7.71 (d, *J* = 3.8 Hz, 1H, C_3-H thiophene), 7.82 (d, *J* = 4.8 Hz, 1H, C_5-H thiophene), 10.66 (s, 1H, NH).

2-(4-Bromophenyl)-5-methyl-3-(2-thienylcarbonyl)amino-4-thiazolidinone (6h)

White solid, mp, 198-199, yield 94%. Anal. Calcd. For $C_{15}H_{13}BrN_2O_2S_2.H_2O$: C, 43.37; H, 3.64; N, 6.74%. Found: C, 43.65; H, 3.43; N, 6.76%. IR υ_{max} (KBr, cm^{-1}): 3232 (NH/OH), 1698 (C=O ring), 1651 (C=O). ^{1}HNMR (60 MHz) (DMSO-d_6/TMS) δ (ppm): 1.54 (d, *J* = 6.8 Hz, 3H, C**H**$_3$CHS), 2.29 (s, 3H, Ar CH_3), 4.06 (q, *J* = 7.0 Hz, 1H, CH_3C**H**S), 5.85 (s, 1H, NCHS), 7.09-7.20 (m, 3H, Ar and C4-H thiophene), 7.35 (d, *J* = 8.0 Hz, 2H, Ar), 7.71 (d, 3.8 Hz, 1H, C3-H thiophene), 7.82 (d, *J* = 4.8 Hz, 1H, C5-H thiophene), 10.66 (s, 1H, NH).

5.2. X-Ray analysis of 2e

The data were collected on a Nonius CAD4 diffractometer using a graphite-monochromated Mo-Kα radiation (λ = 0.71073Å). An empirical ψ-scan absorption correction was applied from the MolEN which has been used to carry out all calculations. The structure was solved by direct methods. Refinements were carried

out by full-matrix least square techniques. Non-hydrogen atoms were anisotropically refined. The hydrogen atoms except H3 were generated in idealized positions 0.95 Å from the bonded carbon atom and refined isotropically using a riding model for a few cycles and then fixed. H3 atom was taken from the difference fourier map. The view of the molecule was performed using ORTEP [23].

5.3. Microbiology

5.3.1. Antimycobacterial activity

Primary screen was conducted at 12.5 μg/ml against Mycobacterium tuberculosis H37Rv in BACTEC 12B medium using the BACTEC 460 radiometric system. Compounds effecting < 90 % inhibition in the primary screen (MIC > 12.5 μg/ml) were not evaluated further. Compounds demonstrating at least 90 % inhibition in the primary screen were selected for further evaluation at lower concentration to determine the actual MIC in the CABTEC 460. The MIC is defined as the lowest concentration inhibiting 99 % of the inoculum.

5.3.1.1. BACTEC radiometric method of susceptibility testing

Inocula for susceptibility testing were either from a positive BACTEC isolation vial with a growth index (GI) of 500 or more, or a suspension of organisms isolated earlier on a conventional medium. The culture was well mixed with a syringe and 0.1 ml of a positive BACTEC culture was added to each of the vials containing the test compounds (12.5 μg/ml). The standard vials contain rifampin (RMP) (0.25 μg/ml). A control vial was inoculated with a 1:100 dilution of the culture. Each vial was tested immediately on a BACTEC instrument to provide CO2 in the headspace. The vials were incubated at 37°C and tested daily with a BACTEC instrument. When the GI in the control read at least 30, the increase in GI (Δ GI) from the previous day in the control was compared with that in the drug vial. The following formula was used to interpret the results:

$$\Delta \text{ GI control} > \Delta \text{ GI drug} = \text{susceptible}$$

$$\Delta \text{ GI control} < \Delta \text{ GI drug} = \text{resistant}$$

If a clear susceptibility pattern (the difference of Δ GI of control and the drug bottle) was not seen at the time the control GI was 30 the vials were read for 1 or 2 additional days to establish a definite pattern of Δ GI differences.

5.4. Antifungal activity

All the compounds to be tested were dissolved in DMSO at a concentration of 4000 μg/ml and the final concentration was reduced to 200 μg/ml with sterile distilled water. No effect of DMSO (5%) was observed upon growth of dermatophytes. The dermatophyte strains which were grown on slant medium of Sabouraud (Difco) were transferred to 3.5 ml nutrient broth (NB, Diagnostic Pasteur) and incubated for three to five days at 25°C. At the end of the incubation period these strains were transferred into screwcapped bottles containing sterilized beads and shaken for 4-5 min in a vortex (IKA-VF, Germany). The suspensions of the cultures were adjusted to have an absorbance degree of 0.6 at 450 nm. Eight different dilutions of the test compounds between 50-0.75 μg/ml were prepared in microplates by serial dilutions from top to bottom. Then all the wells (12th wells positive control) were filled with 10 μl of the standardized strains. These plates were incubated at 25°C for five or six days. The minimum concentration at which no growth was observed was taken as the MIC value.

Acknowledgements: We thank Dr. Joseph A. Maddry from the Tuberculosis Antimicrobial Acquisition and Coordinating Facility (TAACF), National Institute of Allergy and Infectious Diseases Southern Research Institute, GWL Hansen's Disease Center, Colorado State University, Birmingham, Alabama, USA for the *in vitro* evaluation of antimycobacterial activity. We thank Prof. Dr. Gültaze Çapan for her valuable contributions to this study and Dr. Gökçe Cihan-Üstündağ for her support related to the spectral analysis of title compounds. This work was supported by Istanbul University Scientific Research Project (Project No: 673/301194 and 36795).

Author contributions: Concept – E.D.D., N.U.G., D.Ş., S.Ö.; Design – E.D.D., N.U.G., D.Ş., S.Ö.; Supervision – N.U.G; Resources – E.D.D., N.U.G., D.Ş., S.Ö.; Materials – E.D.D., N.U.G., D.Ş., S.Ö.; Data Collection and/or Processing –

E.D.D., N.U.G., D.Ş., S.Ö.; Analysis and/or Interpretation – E.D.D., N.U.G., D.Ş., S.Ö.; Literature Search – E.D.D., N.U.G., D.Ş., S.Ö.; Writing – E.D.D., N.U.G.; Critical Reviews – E.D.D., N.U.G., D.Ş., S.Ö..

Conflict of interest statement: "The authors declared no conflict of interest" in the manuscript.

REFERENCES

[1] Omar K, Geronikaki A, Zoumpoulakis P, Camoutsis C, Sokovic M, Ciric A, Glamoclija J. Novel 4-thiazolidinone derivatives as potential antifungal and antibacterial drugs. Bioorg Med Chem. 2010; 18(1): 426-432. [CrossRef]

[2] Ulusoy N, Çapan G, Ergenç N, Ötük Sanış G, Kiraz M, Kaya D. Synthesis and antimicrobial activity of novel imidazo[2,1-b]thiazolylacetylamino/hydrazono 4-thiazolidinones. Acta Pharm. Turcica. 1997; 39(4), 181-186.

[3] Ouf SA, Gomha SM, Eweis M, Ouf AS, Sharawy IA. Efficiency of newly prepared thiazole derivatives against some cutaneous fungi. Bioorg Med Chem. 2018; 26(12): 3287-3295. [CrossRef]

[4] Samadhiya P, Sharma R, Srivastava SK, Srivastava SD. Synthesis and biological evaluation of 4-thiazolidinone derivatives as antitubercular and antimicrobial agents. Arab J Chem. 2014; 7(5): 657-665. [CrossRef]

[5] Kaminskyy D, Bednarczyk-Cwynar B, Vasylenko O, Kazakova O, Zimenkovsky B, Zaprutko L, Lesyk R. Synthesis of new potential anticancer agents based on 4-thiazolidinone and oleanane scaffolds. Med Chem Res. 2012; 21: 3568-3580. [CrossRef]

[6] Deep A, Jain S, Sharma PC, Phogat P, Malhotra M. Synthesis of 2-(aryl)-5-(arylidene)-4-thiazolidinone derivatives with potential analgesic and anti-inflammatory activity. Med Chem Res. 2012; 21: 1652-1659. [CrossRef]

[7] Vigorita MG, Ottana R, Monforte F, Maccari R, Trovato A, Monforte MT, Taviano M. Synthesis and anti-inflammatory, analgesic activity of 3,3'-(1,2,-Ethanediyl)-bis[2-aryl-4-thiazolidinone] chiral compounds. Part 10. Bioorganic Med Chem Lett. 2001; 11(21): 2791-2794. [CrossRef]

[8] Gürsoy A, Terzioğlu N. Synthesis and isolation of new regioisomeric 4-thiazolidinones and their anticonvulsant activity. Turk J Chem. 2005; 29(3): 247-254.

[9] Barbosa VA, Barea P, Mazia RS, Ueda-Nakamura T, da Costa WF, Foglio MA, Ruiz ALTG, de Carvalho JE, Vendramini-Costa DB, Nakamura CV, Sarragiotto MH. Synthesis and evaluation of novel hybrids β-carboline-4-thiazolidinones as potential antitumor and antiviral agents. Eur J Med Chem. 2016; 124: 1093-1104. [CrossRef]

[10] Gomha SM, Khalil KD. A convenient ultrasound-promoted synthesis of some new thiazole derivatives bearing a coumarin nucleus and their cytotoxic activity. Molecules. 2012; 17(8): 9335-9347. [CrossRef]

[11] Bhutani R, Pathak DP, Kapoor G, Husain A, Iqbal MA. Novel hybrids of benzothiazole-1,3,4-oxadiazole-4-thiazolidinone: Synthesis, *in silico* ADME study, molecular docking and *in vivo* anti-diabetic assessment. Bioorg Chem. 2019; 83: 6-19. [CrossRef]

[12] Türe A, Ergül M, Ergül M, Altun A, Küçükgüzel İ. Design, synthesis, and anticancer activity of novel 4-thiazolidinone-phenylaminopyrimidine hybrids. Mol. Divers. 2021; 25: 1025-1050. [CrossRef]

[13] Çakır G, Küçükgüzel İ, Guhamazumder R, Tatar E, Manvar D, Basu A, Patel BA, Zia J, Talele TT, Kaushik-Basu N. Novel 4-Thiazolidinones as Non-Nucleoside Inhibitors of Hepatitis C Virus NS5B RNA-Dependent RNA Polymerase. Arch. Pharm. 2015; 348: 10-22. [CrossRef]

[14] Küçükgüzel İ, Satılmış G, Gurukumar KR, Basu A, Tatar E, Nichols DB, Talele TT, Kaushik-Basu N. 2-Heteroarylimino-5-arylidene-4-thiazolidinones as a new class of non-nucleoside inhibitors of HCV NS5B polymerase. Eur. J. Med. Chem. 2013; 69: 931-941. [CrossRef]

[15] Tatar E, Küçükgüzel İ, De Clercq E, Şahin F, Güllüce M. Synthesis, characterization and screening of antimicrobial, antituberculosis, antiviral and anticancer activity of novel 1,3-thiazolidine-4-ones derived from 1-[2-(benzoylamino)-4-(methylthio)butyryl]-4-alkyl/arylalkyl thiosemicarbazides. Arkivoc. 2008; 14: 191-210. [CrossRef]

[16] Tatar E, Küçükgüzel İ, Küçükgüzel ŞG, Yılmaz-Demircan F, De Clercq E, Andrei G, Snoeck R, Pannecouque C, Şahin F, Bayrak ÖF. Synthesis, Anti-Tuberculosis and Antiviral Activity of Novel 2-isonicotinoylhydrazono-5-arylidene-4-thiazolidinones. Int. J. Drug Discov. 2010; 1: 19-32.

[17] Kline T, Felise HB, Barry KC, Jackson ST, Nguyen HV, Miller SI. Substituted 2-Imino-5-arylidenethiazolidin-4-one inhibitors of bacterial type III secretion. J Med Chem. 2008; 51(22): 7065-7074. [CrossRef]

[18] Cesur N, Cesur Z, Gürsoy A. New acylthiosemicarbazides, thiazolidinones, and 1,3,4-oxadiazoles as possible anticonvulsants. Arch. Pharm. 1992; 325: 623-624.

[19] Ashour FA, Almazroa SA. Synthesis of certaine thiosemicarbazide and triazole derivatives as potential antimicrobial agents. Farmaco. 1990; 45: 1207-1218.

[20] Dincel ED, Ulusoy-Güzeldemirci N, Şatana D, Küçükbasmacı Ö. Design, synthesis, characterization and antimicrobial evaluation of some novel hydrazinecarbothioamide, 4-thiazolidinone and 1,2,4-triazole-3-thione derivatives. J Heterocycl Chem. 2021; 58(1): 195-205. [CrossRef]

[21] Ulusoy N, Çapan G, Ergenç N, Ekinci AC, Vidin A. Synthesis, characterization and anticonvulsant activity of new 4-thiazolidinone and 1,2,4-triazole-3-thione derivatives. Acta Pharm. Turcica. 1998; 40(1): 5-8.

[22] Ulusoy-Güzeldemirci N, Pehlivan E, Naesens L. Synthesis and antiviral activity evaluation of novel 4-thiazolidinones bearing an imidazo[2,1-*b*]thiazole moiety. Marmara Pharm J. 2018; 22(2): 237-248. [CrossRef]

[23] Johnson CK, ORTEP II. Report ORNL-5138, Oak Ridge National Laboratory, Tennessee, USA, 1976.