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ARTICLE TYPE

Benzothiazolyl Substituted Iminothiazolidinones and Benzamido-oxothiazolidines as Potent and Partly Selective Aldose Reductase Inhibitors

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Two new series of oxothiazolidine benzoate and acetate derivatives were synthesized and evaluated as aldehyde reductase (ALR1) and aldose reductase (ALR2) inhibitors. Methyl 2-[2-benzamido-3-(benzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene]acetates (**2a-k**) and ethyl 4-[2-benzamido-5-(2-methoxy-2-oxyethylidene)-4-oxothiazolidin-3-yl]benzoates (**4a-j**) were obtained in good to excellent yields by heterocyclization of 1-aroyl-3-(2-benzothiazolyl) thioureas (**1a-j**) and ethyl 4-(3-aroylthioureido)benzoates (**3a-j**) respectively with dimethyl acetylenedicarboxylate (DMAD) in dry methanol. Among the tested compounds, **2d, 2g, 2h, 2i, 2j, 4b, 4f** and **4h** showed potent inhibitory activity on ALR1, whereas **2a, 2g, 2h, 4d, 4f, 4h** and **4j** exhibited potent inhibition on ALR2. Docking analysis suggested likely binding modes of the inhibitors within the active site of ALR2. The new aldose reductase inhibitors are thought to represent useful lead structures for the generation of candidate compounds to target a number of pathological conditions, most notably the long-term diabetic complications. FTIR, 1H NMR, 13C NMR, GC-MS and elemental analyses data confirmed the assigned structures to the synthesized compounds. Further the structure and geometry of compound ethyl 4-((2Z,5Z)-22,4-di-(2,4-dichlorobenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4c**) was unequivocally confirmed by single crystal X-Ray diffraction data.

Introduction

Diabetes mellitus is a major human disease. Given its present rate of increase, it is anticipated that it might become one of the world's most common diseases over the next decades and largest public health problems, affecting a minimum of half-a-billion cases[1]. Therapeutic intervention of hyperglycemia-induced diabetic complications is actively pursued because it is very difficult to maintain normoglycemia by any means in patients with diabetes mellitus[2, 3, 4]. Hyperglycemia, as a result of compromised glucose regulation in the diabetic individual, is considered as the cause of the onset and progression of diabetes' chronic complications. Under hyperglycemic conditions, and especially in cells where the intake of glucose is non-insulin dependent, there is observed activation of the polyol pathway to metabolize excess glucose. This increased flux through the polyol pathway reaches up to 30% of the total glucose turnover[1]. As a consequence, osmotic, oxidative, reductive, glycative and protein kinase C (PKC) stress are induced[5] with devastating consequences for affected cells. The polyol pathway was first linked to secondary complications in diabetes during the mid '60s[6]. Aldose reductase (ALR2, EC 1.1.1.21), an enzyme of the polyol metabolic pathway, was first implicated in the etiology of secondary complications of diabetes[7, 8]. This enzyme is a member of the aldo-keto reductase (AKR) superfamily that catalyzes the NADPH-dependent reduction of a broad range of aldehydes and ketones[9, 10]. It is widely believed that the unfavorable profile of many aldose reductase inhibitors (ARIs) in clinical trials is due to their concurrent inhibition of the closely related aldehyde reductase (ALR1, EC 1.1.1.2). ALR1 is responsible for the reduction of many aldehydes and metabolizes methyl glyoxal and 3-deoxyglucosone, representing intermediates for the formation of toxic glycation endproducts[11]. ALR1 also is a member of the AKR superfamily and responsible for a wide variety of biological functions like regulation of proinflammatory response via the reduction of aldehyde phospholipids[12]. Although several ARIs having progressed to clinical trials, only one is currently on the market (Epalrestat, ONO Pharmaceutical, Osaka, Japan)[13, 14]. However, the inhibition of the polyol pathway is considered to be a promising approach to control diabetes complications as well as a number of other pathological conditions such as ischemia, abnormal vascular smooth muscle cell proliferation, cancers, and mood disorders[5, 15, 16]. Prolonged exposure to chronic

hyperglycemia in diabetes can lead to various complications, affecting the cardiovascular, renal, neurological and visual systems[17]. Therefore, the inhibition of ALR2 has been an attractive approach to the prevention and treatment of diabetic complications. Thus, attention is currently paid to discover ARIs of distinct chemical structures, which are neither derivatives of hydantoin nor carboxylic acid because these derivatives are known to cause toxicity and they possess a narrow spectrum of tissue activity[18].

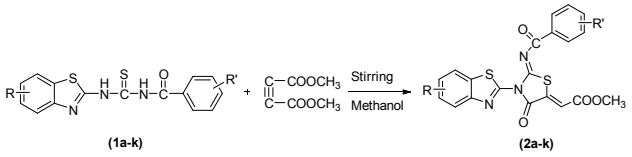
Iminothiazolidin-4-one acetate derivatives can be used for treatment of diabetic complications[19] and substituted benzenesulfonamides as (ALR2) inhibitors with antioxidant activity[15]. N,N'-bis(5-arylidene-4-oxo-4,5-dihydrothiazoline-2-yl)diamine derivatives exhibited *in vitro* antiproliferative as well as kinase inhibitor activities[20]. In the present work, we have focused on the synthesis of two series of new oxothiazolidine derivatives as potent *in vitro* inhibitors of ALR2 as well as ALR1.

Results and Discussion

Chemistry

1-aroyl-3-(2-benzothiazolyl) thioureas (**1a-j**) and ethyl 4-(3-aroylthioureido)benzoates (**3a-j**) were converted into five membered sulphur and nitrogen containing heterocycles with exocylic imino moieties *viz*. methyl 2-(2-benzamido-3-(2-benzothiazolyl)-4-oxothiazolidin-5-ylidene)acetates (**2a–k**) (Scheme 1) and **(4a-j)** (Scheme 2) by heterocyclization of these thioureas with dimethyl acetylenedicarboxylate (DMAD) in dry methanol in the absence of any catalyst.

In the FTIR data of methyl 2-[2-benzamido-3-(benzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene] acetates (**2a-k**) the absence of N-H peaks around 3200-3400 cm^{-1} and appearance of characteristic strong absorption peaks for the ester moiety (-$COOCH_3$) in the range of 1721-1728 cm^{-1} was noticed. Other characteristic absorptions include 1650-1670 (amidic C=O) and 1670-1685 cm^{-1}(ring C=O), besides the aromatic C=C at 1540-1575 cm^{-1}.

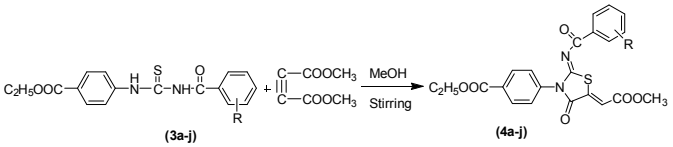


2a: **R** = **R'** = H
2b: **R** = H **R'** = 2-OMe
2c: **R** = H **R'** = 4-Me
2d: **R** = 6-Br **R'** = 3-Cl
2e: **R** = 6-Br **R'** = 2-F
2f: **R** = 6-Me **R'** = 3-Cl
2g: **R** = 6-Me **R'** = 2-Br
2h: **R** = 6-OMe **R'** = H
2i: **R** = 6-OMe **R'** = 2,4-di-Cl
2j: **R** = 5,6-di-Cl **R'** = 2,4-di-Cl
2k: **R** = 5,6-di-Cl **R'** = H

Scheme 1 Synthesis of methyl 2-[2-benzamido-3-(benzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene]acetates (**2a-k**)

In the IR spectra of ethyl 4-[2-benzamido-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl]benzoates (**4a-j**) absorptions for N-H protons of thioureas were not observed but the characteristic carbonyl carbon (C=O) absorption peaks for the ester moiety in the range of 1718-1726 cm^{-1} and for the other ester group from 1720-1727 cm^{-1} were detected. The characteristic carbonyl carbon (C=O) absorption peaks for the amidic moiety were observed at 1653-1668 cm^{-1} and another carbonyl absorption of thiazolidinone rings appeared in the range of 1672-1779 cm^{-1}.

In the ^{1}H NMR spectra of **(4a-j)** revealed with characteristic singlets for the ethylidene moieties appeared in the range of δ 7.11-7.21 ppm. In ^{13}C NMR the characteristic carbonyl carbon (C=O) absorption peaks for the ester moiety appeared in the range of δ 164.4-166.8 ppm and for the other ester group from δ 164.3-165.8 ppm. The carbonyl carbon (C=O) absorption peaks for the amidic moiety were observed from δ 175.2-176.4 ppm and another carbonyl absorption of thiazolidinone rings appeared in the range of δ 165.6-167.7 ppm.



3a: **R** = H
3b: **R** = 3-Cl
3c: **R** = 2,4-di-Cl
3d: **R** = 4-Me
3e: **R** = 3-Me
3f: **R** = 4-OMe
3g: **R** = 3,4-di-OMe
3h: **R** = 2-Br
3i: **R** = 2-OMe
3j: **R** = 2-F

Scheme 2 Synthesis of ethyl 4-[2-benzamido-5-(2-methoxy-2-oxyethylidene)-4-oxothiazolidin-3-yl]benzoates (**4a-j**)

Molecular structure of compound ethyl 4-((2Z,5Z)-22,4-di-(2,4-dichlorobenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4c**) was determined by single crystal X-ray analysis. It belong to Triclinic Space group P-1, with a = = 8.0592(12) Å, b = 11.0011(16)Å, c = 13.471(2) Å, α = 109.374(3)°, β = 92.452(3)°, γ = 105.902(3)° Z = 2 and V = 1604.78(18) Å^3.· (Figure 2). CCDC deposition number: CCDC 1007068.

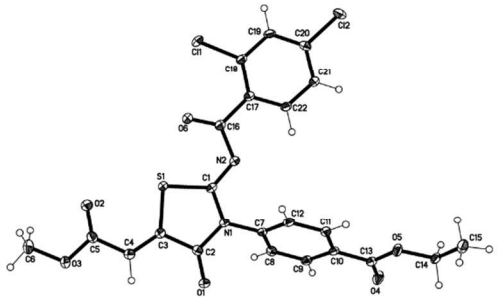


Figure 1. Molecular structure of Ethyl 4-((2Z,5Z)-22,4-di-(2,4-dichlorobenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate **(4c).** Displacement ellipsoids are drawn at the 50% probability level.

Enzyme inhibition studies

The major objective of this study was to identify highly potent, selective, and efficacious ALR2 inhibitors for treatment of diabetic complications. All synthesized compounds were initially tested *in vitro* for inhibitory potency against ALR1 and ALR2. The results from these experiments are listed in Table 1. Newly

synthesized benzothiazolyl substituted iminothiazolidinones (**2a-k**) and benzamido-oxothiazolidines derivatives (**4a-j**) were evaluated for their inhibition of the enzymes ALR1 and ALR2 isolated from the calf eye and bovine kidney, respectively. Compounds were first tested at 1 mM, and those compounds that exhibited potent inhibition > 50% were further evaluated at eight different concentrations to generate dose-response curves, from which IC_{50} values were calculated. Valproic acid and sulindac were used as standard inhibitors for ALR1 and ALR2, respectively. The performed assay was based on the spectrophotometric monitoring of NADPH oxidation, which has proven to be a reliable method[21]. In general, most of the tested compounds were found to be more potent inhibitors of ALR1 than ALR2. Among substituted oxothiazolidine acetates, compounds **2b, 2d, 2g, 2h, 2i** and **2j** were more potent inhibitors of ALR1 than the standard inhibitor (valproic acid). Compound **2a** having unsubstituted rings showed lower inhibitory activity on ALR1 than the reference inhibitor; however, it was a more potent inhibitor of ALR2 with an IC_{50} value of 0.04 ± 0.02 µM. The presence of halogen substituents further increased the inhibitory activity. Methyl 2-((Z)-3-(4,6-dichlorobenzo[d]thiazol-2-yl)-2-((2,4-dichlorobenzoyl)imino)-4-oxothiazolidin-5-ylidene)acetate (**2j**) having di-halogen substitution at both rings was the most potent compound in the methyl substituted oxothiazolidine acetate series. It showed an IC_{50} value of 0.01 ± 0.01 and 0.39 ± 0.05 µM for ALR1 and ALR2, respectively. Among the evaluated compounds, methyl 2-((Z)-3-(6-bromobenzo[d]thiazol-2-yl)-2-((4chlorobenzoyl)imino-4-oxothiazo-lidin-5-ylidene)acetate (**2d**), methyl 2-((Z)-2-((2-bromobenzoyl)imino)-3-(6-methoxybenzo[d]thiazol-2-yl)-4-oxothia-zolidin-5-ylidene)acetate (**2g**) and methyl 2-((Z)-2-((2,4-dichlorobenzoyl)imino)-3-(6-methoxybenzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene)acetate (**2i**) yielded IC_{50} values of 0.02 ± 0.01, 0.01 ± 0.01 and 0.02 ± 0.03 µM, respectively, for ALR1. For ALR2, methyl 2-((Z)-3-(benzo[d]thiazol-2-yl)-2-(benzoylimino)-oxothiazolidin-5-ylidene)acetate (**2a**) and methyl 2-((Z)-2-((2-bromobenzoyl)imino)-3-(6-methoxybenzo[d]thiazol-2-yl)-4-oxothia-zolidin-5-ylidene)acetate (**2g**) exhibited 0.04 ± 0.02 and 0.45 ± 0.09 µM, respectively. These compounds were even more potent inhibitors of ALR2 than sorbinil[22]. Among the ethyl substituted oxothiazolidine benzoates, ethyl 4-[2-(3-chlorobenzamido)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl]benzoate (**4b**), ethyl 4-[5-(2-methoxy-2-oxoethylidene)-2-(4-methoxybenzamido)-4-oxothiazolidin-3-yl]benzoate (**4f**) and ethyl 4-[-2-(2-bromobenzamido)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl]benzoate (**4h**) exhibited IC_{50} values of 0.07 ± 0.01, 0.02 ± 0.01 and 0.02 ± 0.01 µM, respectively, for ALR1. For ALR2, ethyl 4-[5-(2-methoxy-2-oxoethylidene)-2-(4-methylbenzamido)-4-oxothiazolidin-3-yl]benzoate (**4d**) and ethyl 4-[-2-(2-fluorobenzamido)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl] benzoate (**3j**) had IC_{50} values of 0.28 ± 0.03 and 0.20 ± 0.08 µM, respectively, both of them are more potent than sorbinil. Compounds (**4c)** and (**4e)** showed 53.1% and 55.3% inhibition for ALR2, while (**4a)** and (**4i)** exhibited less than 50% inhibition. Some of the compounds showed excellent inhibition on ALR1, it may due the fact that the side chain orientations for the anion-binding-site residues Tyr50, His113 and Trp114 and Leu300 make an electrostatic interactions with the benzothiazolyl substitution in ALR1 resulting in a strong inhibitory activities.

Table 1 Aldehyde reductase and aldose reductase inhibitory activities of methyl substituted oxothiazolidine acetates (**2a-k**) and ethyl substituted oxothiazolidine benzoates (**4a-j**).

Compounds	ALR1	ALR2
	IC_{50} ± SEM (µM) or (% inhibition)[a]	
2a	(12.2)[a]	0.04 ± 0.02
2b	2.17 ± 0.03	(46.6)[a]
2c	(1.11)[a]	38.5 ± 0.02
2d	0.02 ± 0.01	(26.6)[a]
2e	0.01 ± 0.01	(20.4)[a]
2f	(38.9)[a]	(67.6)[a]
2g	0.01 ± 0.01	0.45 ± 0.09
2h	0.02 ± 0.01	6.08 ± 0.10
2i	0.02 ± 0.03	8.21 ± 0.41
2j	0.01 ± 0.01	0.39 ± 0.05
2k	0.01 ± 0.01	4.21 ± 0.01
4a	(22.2)[a]	(23.3)[a]
4b	0.07 ± 0.01	1.28 ± 0.02
4c	(37.1)[a]	(53.1)[a]
4d	0.61 ± 0.08	0.28 ± 0.03
4e	0.62 ± 0.06	(55.3)[a]
4f	0.02 ± 0.01	0.92 ± 0.40
4g	(38.5)[a]	(35.7)[a]
4h	0.02 ± 0.01	0.7 ± 0.14
4i	(17.8)[a]	(16.7)[a]
4j	0.16 ± 0.02	0.20 ± 0.08
Valproic acid	57.4 ± 0.89	–
Sulindac	–	0.29 ± 0.08

[a]% inhibition; "–" means not tested.

Computational studies of aldose reductase inhibitors

It was also attempted to model putative binding modes of selected ALR2 inhibitors. Due to the availability of several X-ray structures of human ALR2, which include different side chain conformations within the active site of the enzyme, docking experiments were performed on selected template structures, representing the currently known active site conformational space. The structure with PDB code 1US0 was found to be best suited in preliminary docking studies to model the class of inhibitors presented herein.

Figure 2 shows the putative binding mode of compound (**2a)**, the most potent representative of the benzothiazolyl substituted iminothiazolidinone compound series. The methyl ester side chain is directed into the anionic binding site with the carbonyl group in possible hydrogen bonding distance to residue His110. In addition, the carbonyl group of the thiazolidinone ring might form another hydrogen bond to the side chain NH of residue Trp111. The benzothiazol substituent occupies the specificity pocket formed by residues Phe122, Trp111, Leu300 and Ala299

with a high degree of shape complementarily and is in a perfect position for π-stacking interactions with residue Trp111. Furthermore the benzoyl substituent is directed toward the entrance of the active site binding pocket and within π-interaction distance to residue Phe122.

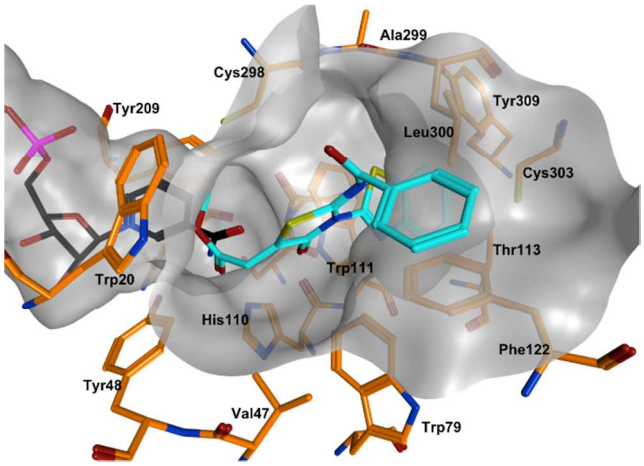


Fig.2 Putative binding mode of compound **2a** (carbon atoms colour cyan) within the active site of ALR2 (orange) including the cofactor NADP+ (carbon atoms colour black).

The hypothetical binding conformation of compound (**4j**) of the ethyl 4-[2-benzamido-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl] benzoates series is depicted in Figure 3. In this class of compounds, the methoxy-oxoethylidene substituent is most likely to occupy the anionic binding site. Again, the carbonyl group of this side chain might form a hydrogen bond with residue His110. In contrast to the binding mode of compound (**2a**) of the benzothiazolyl substituted iminothiazolidinone series, the 2-fluoro substituted benzoyl moiety of (**4j**) is directed into the specificity pocket. Nonetheless, π-stacking interactions to residue Trp111 are also possible for this class of compounds. Similar to compound **2a**, a branching residue of inhibitor (**4j**) is directed toward the entrance of the binding site. The ethyl benzoate group of **4j** is in π-interaction distance to residue Phe122.

In summary, the docking studies helped to identify three plausible interaction hotspots for the compound series reported herein within the active site of ALR2 and two of these sites were shared with the crystallographic inhibitor IDD 594. Figure 4 shows an overlay of the putative binding modes of compounds **2a** and **4j** and the crystallographic pose of the known inhibitor IDD 594. All three binding conformations share a hydrogen bonding interaction of a carbonyl function with His110 within the anionic binding site, which is a known common feature of ALR2 inhibitors[23]. In addition, the specificity pocket is targeted by aromatic residues of all three inhibitors. A possible aromatic interaction with Phe122 at the entrance of the binding site represents an additional feature shared by compounds **2a** and **4j** that is not present in IDD 594 and might contribute to the activity of the new inhibitors.

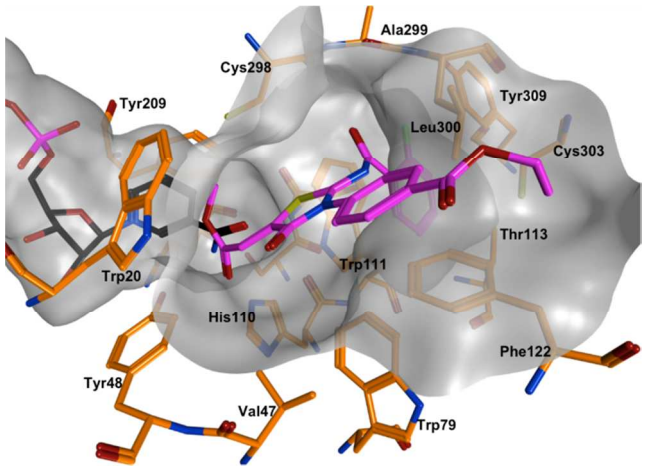


Fig.3 Putative binding mode of compound **4j** (carbon atoms colour magenta) within the active site of ALR2 (orange) including the cofactor $NADP^+$ (black).

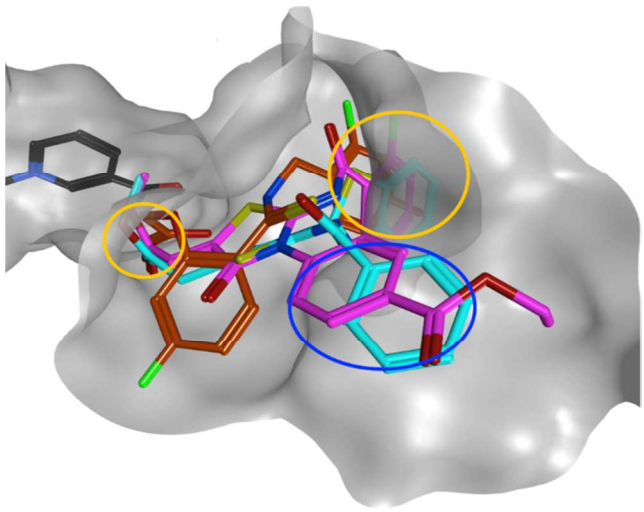


Fig.4 Comparison of predicted and crystallographic binding modes. An overlay of the docking poses of compounds **2a** (cyan) and **4j** (magenta) and the crystallographic binding mode of IDD 594 (carbon atoms colour brown) is shown. Circled in yellow are two interaction hotspots shared by all three inhibitors. In addition, a third interaction hotspot shared only by compounds **2a** and **4j** is circled in blue.

Experimental Section

Chemistry

Melting points were recorded using a digital Gallenkamp (SANYO, Loughborough,UK) model MPD BM 3.5 apparatus and are uncorrected. ^1H and ^{13}C NMR spectra were determined in $CDCl_3$ at 300 and 75 MHz, respectively, using Bruker AM-300 spectrophotometer (Billerica, Middlesex, MA, USA). FT-IR spectra were recorded using Rad Excalibur FTS 3000 MX spectrophotometer (Madison, WI, USA). Mass spectra (EI, 70 eV) were recorded on a GC–MS instrument (Agilent Technologies 1200 series, Santa Clara, CA, USA), and elemental analyses were carried out with a LECO-183 CHNS analyzer (LECO Corporation, St Joseph, MI, USA). Thin layer chromatography (TLC) was carried out on 0.25 mm silica gel plates (60 F254, Merck). Visualization was made with ultraviolet light.

a) General procedures for the synthesis of methyl 2-[2-benzamido-3-(benzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene] acetates (2a-k)

To a stirred solution of suitable 1-aroyl-3-(2-benzothiazolyl)thioureas (**1a-k**) (0.5 g, 0.0015 mol) in 20 mL dry methanol was added dropwise dimethyl acetylene dicarboxylate (DMAD) (0.4 mL, 0.003 mol) and the reaction mixture was stirred at room temperature for 2-3 h until the product was separated as white precipitate. The reaction mixture was filtered to obtain the crude products methyl 2-[2-benzamido-3-(benzo[d]thiazol-2-yl)-4-oxo-thiazolidin-5-ylidene]acetates **(2a-k)**, which were recrystallized from suitable solvents.

*(Z)-methyl-2-((Z)-3-(benzo[d]thiazol-2-yl)-2-(benzoylimino)-oxothiazolidin-5-ylidene)acetate (**2a**):*

(Yield: 72%): Rf[a]: 0.43;mp: 195-196 ^{o}C; IR (KBr) ν_{max}: 2921 (C=C−H), 1721 ($COOCH_3$), 1692 (ring C=O), 1653 (Ar C=O), 1568 (C=N), 1534 (C=C), 1457 (C−N), 1161 (C−S) cm^{-1}; **^{1}H NMR** (300 MHz, $CDCl_3$):8.17 (1H, d, *J* = 7.6 Hz), 8.12 (1H, d, *J* = 7.4 Hz), 8.03-7.66 (5H, m, Ar), 7.57 (1H, dd, *J* = 7.2,7.4 Hz), 7.36 (1H, dd, *J* = 7.2,7.3 Hz), 7.11 (1H, s, C=C-*H*), 3.74 (-OCH_3); **^{13}CNMR** ($CDCl_3$, δ ppm): 174.3 (Ar-C=O), 166.2 (N-C=O), 165.5 ($COOCH_3$), 162.4 (C=N), 156.5 (S-C=N), 146.3 (C-9), 140.6 (=CH), 136.7 (C-1'), 134.5 (C-2',C-6'), 133.4 (C-3',C-5'), 131.5 (S-C=), 128.2 (C-4'), 127.4 (C-6), 125.6 (C-5), 124.3 (C-8), 123.7 (C-4), 122.5 (C-7), 54.2 (-OCH_3); MS (70eV): *m/z* (%):[M^+] 423 (64), 318 (37), 184 (100%), 134 (27), 125 (35), 105 (48), 77 (18): Anal. Calcd. for $C_{20}H_{13}N_3O_4S_2$; C: 56.73, H = 3.07, N = 9.93, S = 15.13, found: C = 56.62, H = 2.96, N = 9.87, S =15.09.

*(Z)-methyl2-((Z)-3-(benzo[d]thiazol-2-yl)-2-((2-methoxybenzoyl)imino-oxothiazolidin-5-ylidene)acetate (**2b**):*

(Yield: 83%): Rf[a]: 0.35; mp: 233-234 ^{o}C; IR (KBr) ν_{max}: 2925 (C=C−H), 1719 ($COOCH_3$), 1691 (ring C=O), 1651 (Ar C=O), 1573 (C=N), 1542 (C=C), 1461 (C−N), 1158 (C−S) cm^{-1}; **^{1}H NMR,** ($CDCl_3$, δ ppm): 8.28 (1H, d, *J* = 7.4 Hz), 8.16 (1H, d, *J* = 7.4 Hz), 7.81 (1H, d, *J* = 7.2 Hz), 7.76 (1H, dd, *J* = 7.8,7.6 Hz), 7.63 (1H, dd, *J* = 7.6,7.8 Hz), 7.52 (1H, dd, *J* = 7.1,7.2 Hz), 7.45 (1H, dd, *J* = 7.3,7.2 Hz), 7.32 (1H, d, *J* = 7.3 Hz), 7.13 (1H, s, C=C-*H*), 3.84 (3H, s, Ar-OCH_3),3.76 (3H, s, -OCH_3),**^{13}CNMR** ($CDCl_3$, δ ppm): 174.4 (Ar-C=O), 166.4 (N-C=O), 165.2 ($COOCH_3$),162.2 (C=N), 156.4 (S-C=N), 146.4 (C-9), 140.5 (=CH),137.8 (C-1'), 136.6 (C-2'), 135.5 (C-6'), 134.4 (C-3'), 133.5 (C-5'), 131.4 (S-C=),127.8 (C-4'), 126.6 (C-6), 125.3 (C-5), 124.7 (C-8), 123.5 (C-4), 122.6 (C-7), 54.4 (-OCH_3),MS (70eV): *m/z* (%): [M^+] 453 (73), 318 (24), 184 (100%), 135 (37), 134 (17), 125 (25), 107 (53), 77 (21), Anal. Calcd. for $C_{21}H_{15}N_3O_5S_2$: C = 55.63, H = 3.31, N = 9.27, S = 14.13, found: C = 55.54, H = 3.22, N = 9.16, S= 14.09.

*(Z)-methyl 2-((Z)-3-(benzo[d]thiazol-2-yl)-2-((4methylbenzoyl)imino-4-oxothiazolidin-5-ylidene)acetate (**2c**):*

(Yield: 81%): Rf[a]: 0.31; mp: 287-288 ^{o}C; IR (KBr) ν_{max}: 2928 (C=C−H), 1722 ($COOCH_3$), 1693 (ring C=O), 1653 (Ar C=O),1576 (C=N), 1538 (C=C), 1457 (C−N), 1162 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.25 (1H, d, *J* = 7.4 Hz), 8.14 (1H, d, *J* = 7.4 Hz), 7.91 (1H, d, *J* = 7.2 Hz). 7.82 (1H, d, *J* = 7.2Hz), 7.61 (1H, dd, *J* = 7.2,7.6 Hz), 7.34 (1H, dd, *J* = 7.2,7.6 Hz), 7.12 (1H, s, C=C-*H*), 3.75 (3H, s, -OCH_3), 2.55 (3H, s, Ar-CH_3),**^{13}CNMR** ($CDCl_3$, δ ppm): 174.5 (Ar-C=O), 166.3 (N-C=O), 165.4 ($COOCH_3$), 162.3 (C=N), 156.4 (S-C=N),146.6 (C-9), 140.4 (=CH),135.4 (C-1'), 134.6 (C-2',C-6'), 132.7 (C-3',C-5'), 131.6 (S-C=),128.4 (C-4'), 127.3 (C-6), 126.5 (C-5), 125.7 (C-8), 124.6 (C-4), 123.4 (C-7), 54.3 (-OCH_3), 22.4 (Ar-CH_3):MS (70eV): *m/z* (%): [M^+] 437 (67), 318 (43), 184 (100%), 134 (28), 125 (30), 119 (56), 91 (41), 77 (29), Anal. Calcd. for $C_{21}H_{15}N_3O_4S_2$; C = 57.66, H = 3.43, N = 9.6, S = 14.65, found: C = 57.41, H = 3.34, N = 9.48, S = 14.52.

*(Z)-methyl 2-((Z)-3-(6-bromobenzo[d]thiazol-2-yl)-2-((4chlorobenzoyl)imino-4-oxothiazo-lidin-5-ylidene)acetate (**2d**):*

(Yield: 76%):Rf[a]: 0.45; mp: 266-267 ^{o}C; IR (KBr) ν_{max}: 2934 (C=C−H), 1724 ($COOCH_3$), 1689 (ring C=O), 1652 (Ar C=O), 1571 (C=N), 1533 (C=C), 1452 (C−N), 1165 (C−S) cm^{-1}; **^{1}H NMR,** ($CDCl_3$, δ ppm): 8.16 (1H, d, *J* = 7.4 Hz), 8.10 (1H, d, *J* = 2.4 Hz), 7.95 (1H, d, *J* = 7.4 Hz), 7.83 (1H, d, *J* = 7.2 Hz), 7.76 (1H, d, *J* = 2.3 Hz),7.69 (1H, dd, *J* = 7.2,2.3 Hz), 7.32 (1H, dd, *J* = 7.1,7.2 Hz), 7.14 (1H, s, C=C-*H*), 3.77 (3H, s, -OCH_3); **^{13}CNMR** ($CDCl_3$, δ ppm): 174.7 (Ar-C=O),166.5 (N-C=O), 165.6 ($COOCH_3$),162.6 (C=N), 156.7 (S-C=N),146.6 (C-9), 140.6 (=CH), 138.6 (C-1'), 137.5 (C-3'), 136.3 (C-6'), 135.7 (C-4'), 134.5 (C-6), 133.4 (C-5'), 132.6 (C-5), 131.7 (S-C=), 128.6 (C-2'), 127.5 (C-8), 125.6 (C-4), 124.3 (C-7), 123.4 (C-6), 54.5 (-OCH_3), MS (70eV): *m/z* (%), [(^{79}Br)M^+] 535.5 (57), [(^{81}Br)M^+] 537.5 (43), 396 (26), 212 (18), 184 (100%), 139.5 (33), 134 (28), 125 (23), 111.5 (43), 77 (16), Anal. Calcd. for $C_{20}H_{11}N_3O_4S_2ClBr$; C = 44.73, H = 2.05, N = 7.83, S = 11.93, found: C = 44.56, H = 1.97, N = 7.74, S = 11.86.

*(Z)-methyl 2-[3-(6-bromobenzo[d]thiazol-2-yl)-2-(2-fluorobenzamido)-4-oxothiazolidin-5-ylidene]acetate (**2e**):*

(Yield: 72%): Rf[a]: 0.5; mp: 151-153 ^{o}C; IR (KBr) ν_{max}: 2936 (C=C−H), 1725 ($COOCH_3$), 1690 (ring C=O), 1661 (Ar C=O), 1573 (C=N), 1536 (C=C), 1457 (C−N), 1163 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.19 (1H, d, *J* = 7.4 Hz), 8.13 (1H, d, *J* = 2.3 Hz), 7.87 (1H, d, *J* = 7.4 Hz), 7.76 (1H, d, *J* = 7.2 Hz),7.68 (1H, dd, *J* = 7.2,7.3 Hz), 7.54 (1H, dd, *J* = 7.4,7.2 Hz),7.34 (1H, d, *J* = 7.4 Hz), 7.15 (1H, s, C=C-H), 3.76 (3H, s, -OCH_3); **^{13}CNMR** ($CDCl_3$, δ ppm): 174.5 (Ar-C=O), 166.8 (N-C=O), 165.7 ($COOCH_3$),162.7 (C=N), 156.8 (S-C=N), 146.5 (C-9), 140.8 (=CH), 140.5 (C-2'), 137.6 (C-1'), 136.5 (C-6'), 134.6 (C-4'), 131.4 (C-5), 131.9 (S-C=),128.6 (C-5'), 127.8 (C-3'), 126.5 (C-8), 125.7 (C-4), 124.8 (C-7), 122.4 (C-6), 54.7 (-OCH3); MS (70eV): *m/z* (%); [(^{79}Br)M^+] 519 (60), [(^{81}Br)M^+] 521 (46), 396 (32), 212 (28), 184 (100%), 134 (18), 123 (37), 95 (40), 77 (21); Anal. Calcd. for $C_{20}H_{11}N_3O_4S_2ClF$; C = 46.15, H = 2.12, N = 8.0, S = 12.31, found: C = 46.08, H = 2.04, N = 7.95, S = 12.21.

*(Z)-methyl 2-((Z)-2-((3-chlorobromobenzoyl)imino)-3-(6-methoxybenzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene)acetate (**2f**):*

(Yield: 74%): Rf[a]: 0.4; mp: 223-224 ^{o}C; IR (KBr) ν_{max}: 2947 (C=C−H), 1723 ($COOCH_3$), 1688 (ring C=O), 1653 (Ar C=O),1574 (C=N), 1536 (C=C), 1456 (C−N), 1162 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.13 (1H, d, *J* = 7.4 Hz), 8.08 (1H, d, *J* = 2.4 Hz), 7.91 (1H, d, *J* = 7.6 Hz), 7.83 (1H, d, *J* = 7.2 Hz),7.74 (1H, d, *J* = 2.3 Hz), 7.62 (1H, dd, *J* = 7.1,2.3 Hz), 7.36 (1H, d, *J* = 7.2,7.1 Hz), 7.14 (1H, s, C=C-H), 3.75 (3H, s, -OCH_3), 2.57 (3H, s, Ar-CH_3); **^{13}CNMR** ($CDCl_3$, δ ppm): 174.6 (Ar-C=O), 166.4 (N-C=O), 165.5 ($COOCH_3$),162.7 (C=N), 156.6 (S-C=N), 146.5 (C-9), 140.7 (=CH), 138.6 (C-1'), 137.5 (C-3'), 136.4 (C-6'), 135.7 (C-4'), 134.5 (C-6), 133.4 (C-5'), 131.7 (S-C=),128.5 (C-2'), 126.6 (C-5), 124.7 (C-8), 123.3 (C-4), 122.5 (C-7), 54.6 (-OCH_3), 23.4 (Ar-CH_3): MS (70eV): *m/z* (%), [M^+]

471.5 (68), 332 (43), 184 (100%), 148 (38), 139 (32), 134 (21), 111.5 (29), 77 (15); Anal. Calcd. for $C_{21}H_{14}N_3O_4S_2Cl$; C = 53.45, H = 2.97, N = 8.91, S = 13.57, found: C = 53.33, H = 2.88, N = 8.75, S = 13.37.

*(Z)-methyl 2-((Z)-2-((2-bromobenzoyl)imino)-3-(6-methoxybenzo[d]thiazol-2-yl)-4-oxothia-zolidin-5-ylidene)acetate **(2g)**:*

(Yield: 76 %): Rf[a]: 0.45; mp: 134-136 ^oC; IR (KBr) ν_{max}: 2945 (C=C−H), 1722 ($COOCH_3$), 1687 (ring C=O), 1653 (Ar C=O), 1572 (C=N), 1537 (C=C), 1453 (C−N), 1164 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.13 (1H, d, *J* = 7.6 Hz), 8.06 (1H, d, *J* = 2.4 Hz), 7.82 (1H, d, *J* = 7.4 Hz), 7.73 (1H, d, *J* = 7.2 Hz),7.42 (1H, dd, *J* = 7.3,7.2 Hz), 7.31 (1H, dd, *J* = 7.2,7.1 Hz),7.18 (1H, d, *J* = 7.1 Hz), 7.15 (1H, s, C=C-H), 3.76 (3H, s, -OCH_3), 2.56 (3H, s, Ar-CH_3); **^{13}CNMR** ($CDCl_3$, δ ppm):174.2 (Ar-C=O),166.1 (N-C=O), 165.3 ($COOCH_3$),162.5 (C=N), 156.3 (S-C=N),146.4 (C-9), 140.4 (=CH), 138.4 (C-1'), 137.5 (C-6'), 136.6 (C-4'), 135.7 (C-3'), 134.5 (C-5'), 133.6 (C-6), 131.5 (S-C=),127.8 (C-2'), 126.7 (C-5), 125.6 (C-8), 124.5 (C-4), 122.6 (C-7), 54.3 (-OCH_3), 23.2 (Ar-CH_3); MS (70eV): *m/z* (%); [(^{79}Br)$M^{+\cdot}$] 515 (71), [(^{81}Br)$M^{+\cdot}$] 517 (55), 332 (38), 184 (100%), 155 (45), 148 (30), 134 (22), 125 (15), 77 (28); Anal. Calcd. for $C^{21}H^{14}N^3O^4S^2Br$; C = 48.84, H = 2.71, N = 8.14, S = 12.40, found: C = 48.73, H = 2.54, N = 8.07, S = 12.28.

*(Z)-methyl 2-((Z)-2-(benzoylimino)-3-(6-methoxybenzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene)acetate **(2h)**:*

(Yield: 78%): Rf[a]: 0.36; mp: 183-184 ^oC; IR (KBr) ν_{max}: 2943 (C=C−H), 1721 ($COOCH_3$), 1688 (ring C=O), 1654 (Ar C=O), 1576 (C=N), 1542 (C=C), 1457 (C−N), 1166 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.11 (1H, d, *J* = 7.6 Hz), 8.04 (1H, d, *J* = 2.3 Hz), 7.81 (1H, d, *J* = 7.4 Hz), 7.74-7.37 (5H, m, Ar),7.13 (1H, s, C=C-H), 3.86 (3H, s,Ar-OCH3), 3.74 (3H, s, -OCH3); **^{13}CNMR** ($CDCl_3$, δ ppm):174.4 (Ar-C=O),166.4 (N-C=O), 165.4 ($COOCH_3$),162.3 (C=N), 156.4 (S-C=N),146.2 (C-9), 140.5 (=CH), 137.6 (C-6), 135.7 (C-1'), 134.5 (C-2',C-6'), 132.6 (C-3',C-5'), 131.6 (S-C=),128.7 (C-4'), 127.5 (C-8), 126.4 (C-4), 124.5 (C-5), 122.3 (C-7), 56.2 (Ar-OCH_3), 54.4 (-OCH_3); MS (70eV): *m/z* (%); [$M^{+\cdot}$] 453 (74), 348 (40), 184 (100%), 164 (48), 134 (20), 125 (17), 105 (35), 77 (24); Anal. Calcd. for $C_{21}H_{15}N_3O_5S_2$; C = 55.63, H = 3.31, N = 9.27, S = 14.13. found: C = 55.51, H = 3.26, N = 9.16, S = 14.02.

*(Z)-methyl 2-((Z)-2-((2,4-dichlorobenzoyl)imino)-3-(6-methoxybenzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene)acetate **(2i)**:*

(Yield: 77%): Rf[a]: 0.41; mp: 146-147 ^oC; IR (KBr) ν_{max}: 2953 (C=C−H), 1726 ($COOCH_3$), 1694 (ring C=O), 1657 (Ar C=O),1578 (C=N), 1547 (C=C), 1459 (C−N), 1168 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.19 (1H, d, *J* = 7.6 Hz), 8.08 (1H, d, *J* = 2.4 Hz), 7.81 (1H, d, *J* = 7.6 Hz), 7.73 (1H, d, *J* = 7.4 Hz), 7.66(1H, d, *J* = 2.4 Hz), 7.37 (1H, d, *J* = 7.6 Hz), 7.16 (1H, s, C=C-H), 3.87 (3H, s, Ar-OCH3), 3.75 (3H, s, -OCH_3);**^{13}CNMR** ($CDCl_3$, δ ppm): 174.8 (Ar-C=O),166.5 (N-C=O), 165.7 ($COOCH_3$),162.6 (C=N), 156.8 (S-C=N),146.3 (C-9), 141.5 (=C-S), 138.5 (C-1'), 137.3 (C-2'), 136.7 (C-4'), 135.6 (C-'6), 134.5 (C-3'), 133.4 (C-5'), 132.1 (S-C=),127.5 (C-8), 126.6 (C-4), 124.5 (C-5), 122.7 (C-7), 56.5 (Ar-OCH_3), 54.6 (-OCH_3); MS (70eV): *m/z* (%); [$M^{+\cdot}$] 521 (56), 348 (30), 184 (100%), 173 (38), 164 (41), 145 (32), 134 (24), 125 (14), 111.5 (44), 77 (20); Anal. Calcd. for $C_{21}H_{13}N_3O_5S_2Cl_2$; C = 48.37, H = 2.49, N = 8.06, S = 12.28, found: C = 48.21, H = 2.37, N = 7.94, S = 12.16.

*(Z)-methyl 2-((Z)-3-(4,6-dichlorobenzo[d]thiazol-2-yl)-2-((2,4-dichlorobenzoyl)imino)-4-oxothiazolidin-5-ylidene)acetate **(2j)**:*

(Yield: 75%): Rf[a]: 0.5; mp: 116-117 ^oC; IR (KBr) ν_{max}: 2963 (C=C−H), 1727 ($COOCH_3$), 1696 (ring C=O), 1658 (Ar C=O), 1578 (C=N), 1547 (C=C), 1460 (C−N), 1167 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.26 (1H, d, *J* = 2.3 Hz), 8.18 (1H, d, *J* = 2.3 Hz), 7.88 (1H, d, *J* = 7.2 Hz), 7.75(1H, d, *J* = 2.3 Hz), 7.67 (1H, d, *J* = 7.2 Hz), 7.19 (1H, s, C=C-H), 3.78 (3H, s, -OCH_3); **^{13}CNMR** ($CDCl_3$, δ ppm): 175.7 (Ar-C=O), 166.8 (N-C=O), 165.8 ($COOCH_3$),162.8 (C=N), 156.9 (S-C=N),146.5 (C-9), 141.8 (=CH), 138.6 (C-1'), 137.5 (C-2'), 136.6 (C-4'), 135.8 (C-6'), 134.4 (C-6), 133.5 (C-4), 132.6 (C-3'), 132.4 (=C-S), 128.5 (C-5'), 126.4 (C-8), 124.6 (C-5), 123.7 (C-7), 54.8 (-OCH_3); MS (70eV): *m/z* (%); [$M^{+\cdot}$] 559 (52), 386 (47), 202 (34), 184 (100%), 173 (41), 168.5 (54), 145 (32), 134 (19), 125 (12), 111.5 (40), 77 (25); Anal. Calcd. for $C_{20}H_9N_3O_4S_2Cl_4$; C = 42.93, H = 1.61, N = 7.51, S = 11.45, found: C = 42.84, H = 1.54, N = 7.36, S = 11.38.

*(Z)-methyl2-((Z)-2-(benzoylimino)-3-(4,6-dichlorobenzo[d]thiazol-2-yl)-4-oxothiazolidin-5-ylidene)acetate **(2k)**:*

(Yield: 78%): Rf[a]: 0.4; mp: 169-170 ^oC; IR (KBr) ν_{max}: 2961 (C=C−H), 1725 ($COOCH_3$), 1693 (ring C=O), 1656 (Ar C=O), 1576 (C=N), 1545 (C=C), 1458 (C−N), 1163 (C−S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 8.22 (1H, d, *J* = 2.3 Hz), 8.14 (1H, d, *J* = 2.3 Hz), 7.93-7.66 (5H, m, Ar), 7.17 (1H, s, C=C-H), 3.77 (3H, s, -OCH_3); **^{13}CNMR** ($CDCl_3$, δ ppm): 175.3 (Ar-C=O),166.7 (N-C=O), 165.6 ($COOCH_3$),162.7 (C=N), 156.7 (S-C=N), 146.3 (C-9), 141.6 (=CH), 136.7 (C-1'), 135.6 (C-6), 134.5 (C-4), 133.6 (C-2',C-6'), 132.4 (C-3',C-5'), 132.2 (S-C=),127.4 (C-4'), 125.5 (C-8), 124.6 (C-5), 122.8 (C-7), 54.6 (-OCH_3); MS (70eV): *m/z* (%); [$M^{+\cdot}$] 491 (66), 386 (51), 202 (41), 184 (100%), 168.5 (47), 134 (29), 125 (23), 105 (46), 77 (26); Anal. Calcd. for $C_{20}H_{11}N_3O_4S_2Cl_2$: C = 48.88, H = 2.24, N = 8.55, S = 13.03, found: C = 48.78, H = 2.17, N 8.41, S = 12.94.

b) General procedure for synthesis of ethyl 4-[2-benzamido-5-(2-methoxy-2-oxoethylidene)-4-oxothi-azolidin-3-yl] benzoates (4a-j)

A solution of appropriate ethyl 4-(3-aroylthioureido)benzoate (**3a-j**) (0.5 g, 1.5 mmol) in 20 mL dry methanol was stirred in 100 mL two neck round bottom flask. Dimethyl acetylenedicarboxylate (DMAD) (0.4 mL, 3.0 mmol) was added drop wise and the reaction mixture stirred at room temperature for 2-3 h until the product precipitated out. The reaction mixture was filtered afford crude methyl 2-[2-benzamido-3-(benzo[d]thiazol-2-yl)-4-oxo-thiazolidin-5-ylidene]acetates **(4a-j)** which were recrystallized from suitable solvents.

*Ethyl 4-((2Z,5Z)-2-(benzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate **(4a)**:*

(Yield: 65%): Rf[a]: 0.65; m.p.: 121-122 oC; IR (KBr) ν_{max}: 2946 (C=C-H), 1724 (-C_2H_5 ester C=O), 1721 (-CH_3 ester C=O), 1676 (ring C=O), 1653 (amide C=O), 1563 (C=N), 1546 (C=C), 1437 (C-N), 1253 (C-S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.91 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.64-7.83 (5H, m, Ar), 7.57 (1H, d, *J* = 7.6Hz, H-3,H-5), 7.13 (1H, s, C=C-H), 4.26 (2H, q, *J* = 7.1 Hz, -CH2), 3.76 (3H, s, -OCH_3), 2.23 (3H, t, *J* = 7.1 Hz, -CH_3);**^{13}CNMR** ($CDCl_3$, δ ppm): 168.4 (amide C=O), 167.1 (ring C=O), 166.2 (-CH_3 ester C=O), 165.4 (-C_2H_5 ester C=O), 162.6 (C=N), 140.2 (=CH), 137.4 (C-4), 136.3 (C-1'), 134.7 (C-2',C-

6'), 132.4 (C-3',C-5'), 131.3 (S-C=), 130.4 (C-2,C-6), 128.2 (C-4'), 126.7 (C-1), 122.4 (C-3,C-5), 59.4 (-OCH_2), 53.4 (-OCH_3), 15.2 (-CH_3);MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 438 (49), 333 (19), 183 (34), 149 (100 %), 124 (26), 105 (43), 77 (26); Anal. Calcd. for $C_{22}H_{18}N_2O_6S$: C= 60.27, H= 4.11, N= 6.39, S= 7.31. Found: C= 60.19, H= 4.03, N= 6.28, S= 7.24.

*Ethyl 4-((2Z,5Z)-2-(3-chlorobenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4b**):*

(Yield: 69%): Rf[a]: 0.7; m.p: 137-138°C; IR (KBr) ν_{max}: 2951 (C=C-H), 1725 (-C2H5 ester C=O), 1722 (-CH3 ester C=O), 1677 (ring C=O), 1656 (amide C=O), 1561 (C=N), 1544 (C=C), 1435 (C-N), 1250 (C-S) cm^{-1}; **^{1}H NMR**, ($CDCl_3$, δ ppm): 7.94 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.85 (1H, d, *J* = 2.3 Hz, H-2'), 7.78 (1H, d, *J* = 7.2 Hz, H-6'), 7.67 (1H, d, *J* = 7.6 Hz, H-3,H-5), 7.58 (1H, dd, *J* = 7.2,7.3 Hz, H-5'), 7.56 (1H, dd, *J* = 7.3,2.3 Hz, H-4'), 7.17 (1H, s, C=C-H), 4.28 (2H, q, *J* = 7.1 Hz, -CH_2), 3.82 (3H, s, -OCH_3), 2.26 (3H, t, *J* =7.2 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.6 (amide C=O), 167.3 (ring C=O), 166.4 (-CH_3 ester C=O), 165.5 (-C_2H_5 ester C=O), 162.7 (C=N), 140.4 (=CH), 137.5 (C-4), 136.5 (C-1'), 135.2 (C-3'), 134.8 (C-4'), 133.2 (C-5'), 132.5 (C-2'), 131.6 (S-C=), 130.5 (C-2,C-6), 128.6 (C-6'), 126.6 (C-1), 122.7 (C-3,C-5), 59.8 (-OCH_2), 53.5 (-OCH_3), 15.6 (-CH_3); MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 472.5 (54), [$(M^{+2})^{+\cdot}$] 474.5 (41), 333 (35), 183 (29), 149 (100 %), 139.5 (38), 124 (17), 111.5 (23); Anal. Calcd. for $C_{22}H_{17}N_2O_6ClS$: C= 55.93, H= 3.60, N= 5.93, S= 6.78. Found: C= 55.81, H= 3.46, N= 5.78, S= 6.56.

*Ethyl 4-((2Z,5Z)-22,4-di -(2,4-dichlorobenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4c**):*

(Yield: 67%): Rfa: 0.65; m.p: 130-131 °C; IR (KBr) ν_{max}: 2958 (C=C-H), 1731 (-C_2H_5 ester C=O), 1726 (-CH_3 ester C=O), 1678 (ring C=O), 1664 (Ar C=O), 1567 (C=N), 1554 (C=C), 1451 (C−N), 1260 (C−S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.96 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.87 (1H, d, *J* = 2.3 Hz, H-3', 7.74 (1H, d, *J* = 7.2 Hz, H-6'), 7.65 (1H, dd, *J* = 7.2,2.3 Hz, H-5'), 7.56 (1H, d, *J* = 7.6 Hz, H-3,H-5), 7.21 (1H, s, C=C-H), 4.31 (2H, q, *J* = 7.1 Hz,-CH_2), 3.86 (3H, s, -OCH_3), 2.29 (3H, t, *J* = 7.1 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.8 (amide C=O), 167.6 (ring C=O), 166.7 (-CH_3 ester C=O), 165.8 (-C_2H_5 ester C=O), 162.9 (C=N), 140.5 (=CH), 138.8 (C-4'), 137.6 (C-4), 136.4 (C-2'), 135.5 (C-1'), 133.6 (C-6'), 131.8 (S-C=), 131.2 (C-3'), 130.5 (C-2,C-6), 127.6 (C-5'), 126.6 (C-1), 122.7 (C-3,C-5), 60.1 (-OCH_2), 53.7 (-OCH_3), 15.8 (-CH_3); MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 506 (36), 333 (47), 183 (25), 173 (19), 149 (100 %), 145 (56), 138.5 (33), 124 (18), 111.5 (20); Anal. Calcd. for $C_{22}H_{16}N_2O_6SCl_2$: C= 52.17, H= 3.16, N= 5.53, S= 6.32. Found: C= 52.13, H= 3.09, N= 5.43, S= 6.26.

*Ethyl 4-((2Z,5Z)-2-(4-methylbenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4d**):*

(Yield: 72%): Rf[a]: 0.6; m.p: 119-120 °C; IR (KBr) ν_{max}: 2952 (C=C−H), 1725 (-C_2H_5 ester C=O), 1721 (-CH_3 ester C=O), 1672 (ring C=O), 1663 (Ar C=O), 1556 (C=N), 1564 (C=C), 1456 (C−N), 1247 (C−S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.92 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.84 (1H, d, *J* = 7.4Hz, H-2',H-6'), 7.75 (1H, d, *J* = 7.4 Hz, H-3',H-5'), 7.63 (1H, d, *J* = 7.6Hz, H-3,H-5), 7.16 (1H, s, C=C-H), 4.26 (2H, q, *J* = 7.0 Hz,-CH_2), 3.86 (3H, s, -OCH_3), 2.68 (3H, s, Ar-CH_3), 2.26 (3H, t, *J* = 7.0 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.3 (amide C=O), 167.2 (ring C=O), 166.6 (-CH_3 ester C=O), 165.5 (-C_2H_5 ester C=O), 162.5 (C=N), 140.4 (=CH), 137.5 (C-4), 134.3 (C-1'), 132.6 (C-4'), 131.5 (S-C=), 130.5 (C-2,C-6), 129.7 (C-2',C-6'), 129.3 (C-3',C-5'), 126.7 (C-1), 122.6 (C-3,C-5), 59.6 (2H, s, -OCH_2), 53.5 (3H, s, -OCH_3), 23.6 (3H, s, Ar-CH_3),15.3 (3H, s, -CH_3); MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 452 (51), 361 (16), 333 (22), 183 (39), 149 (100 %), 119 (37), 124 (41), 91 (27); Anal. Calcd. for $C_{23}H_{20}N_2O_6S$: C= 61.06, H= 4.42, N= 6.19, S= 7.08. Found: C= 60.94, H= 4.31, N= 6.13, S= 7.01.

*Ethyl 4-((2Z,5Z)-2-(3-methylbenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4e**):*

(Yield: 68%): Rf[a]: 0.6; m.p: 122-123 °C; IR (KBr) ν_{max}: 2954 (C=C−H), 1727 (-C_2H_5 ester C=O), 1723 (-CH_3 ester C=O), 1674 (ring C=O), 1665 (Ar C=O), 1561 (C=N), 1553 (C=C), 1452 (C−N), 1263 (C−S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.87 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.79 (1H, d, *J* = 7.1 Hz, H-6'), 7.72 (1H, d, *J* = 2.4 Hz, H-2'), 7.68 (1H, d, *J* = 7.6 Hz, H-3,H-5), 7.61 (1H, dd, *J* = 7.1,7.2 Hz, H-5'), 7.55 (1H, dd, *J* = 7.2,2.4 Hz, H-4'), 7.15 (1H, s, C=C-H), 4.24 (2H, q, *J* = 7.2 Hz, -CH_2), 3.83 (3H, s, -OCH_3), 2.66 (3H, s, Ar-CH_3), 2.25 (3H, t, J = 7.2 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.2 (amide C=O), 167.1 (ring C=O), 166.3 (-CH_3 ester C=O), 165.4 (-C_2H_5 ester C=O), 162.5 (C=N), 140.3 (=CH), 137.5 (C-4), 135.7 (C-1'), 134.6 (C-3'), 133.5 (C-6'), 132.7 (C-4'), 131.6 (S-C=), 130.3 (C-2,C-6), 129.5 (C-5'), 126.7 (C-1), 122.5 (C-3,C-5), 59.6 (2H, s, -OCH_2), 53.5 (3H, s, -OCH_3), 23.8 (3H, s, Ar-CH_3),15.4 (3H, s, -CH_3); MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 452 (57), 361 (19), 333 (24), 183 (44), 149 (100 %), 119 (33), 124 (44), 91 (25); Anal. Calcd. for $C_{23}H_{20}N_2O_6S$: C= 61.07, H= 4.43, N= 6.18, S= 7.07. Found: C= 60.96, H= 4.34, N= 6.11, S= 6.98.

*Ethyl 4-((2Z,5Z)-2-(4-methoxybenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4f**):*

(Yield: 71%): Rf[a]: 0.55; m.p: 126-127 °C; IR (KBr) ν_{max}: 2945 (C=C−H), 1724 (-C_2H_5 ester C=O), 1720 (-CH_3 ester C=O), 1681 (ring C=O), 1661 (Ar C=O), 1558 (C=N), 1557 (C=C), 1447 (C−N), 1253 (C−S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.88 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.78 (1H, d, *J* = 7.4Hz, H-2',H-6'), 7.67 (1H, d, *J* = 7.4 Hz, H-3',H-5'), 7.56 (1H, d, *J* = 7.6Hz, H-3,H-5), 7.14 (1H, s, C=C-H), 4.24 (2H, q, *J* = 7.1 Hz,-CH_2),3.86 (3H, s, Ar-OCH_3), 3.78 (3H, s, -OCH_3), 2.23 (3H, t, *J* = 7.1 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.5 (amide C=O), 167.7 (ring C=O), 166.3 (-CH_3 ester C=O), 165.5 (-C_2H_5 ester C=O), 162.4 (C=N), 140.3 (=CH), 138.6 (C-4'), 137.4 (C-4), 131.5 (S-C=), 130.8 (C-2',C-6'), 130.3 (C-2,C-6), 128.4 (C-1'), 126.5 (C-1), 122.4 (C-3,C-5), 117.6 (C-3',C-5'), 59.3 (2H, s, -OCH_2), 56.4 (3H, s, Ar-OCH_3),53.2 (3H, s, -OCH_3), 15.3 (3H, s, -CH_3); MS (70eV): *m/z* (%); [$M^{+\cdot}$] 468 (44), 333 (27), 319 (25), 183 (47), 149 (37), 135 (100 %), 124 (32); Anal. Calcd. for $C_{23}H_{20}N_2O_7S$: C= 58.97, H= 4.27, N= 5.98, S= 6.84. Found: C= 58.83, H= 4.16, N= 5.91, S= 6.73.

*Ethyl 4-((2Z,5Z)-2-(2-methoxybenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4g**):*

(Yield: 74%): Rf[a]: 0.55; m.p: 147-148 °C; IR (KBr) ν_{max}: 2937 (C=C−H), 1722 (-C_2H_5 ester C=O), 1718 (-CH_3 ester C=O), 1674 (ring C=O), 1664 (Ar C=O), 1546 (C=N), 1564 (C=C), 1436 (C−N), 1264 (C−S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.92 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.85 (1H, d, *J* = 7.2Hz, H-6'), 7.74 (1H, s, Ar-2'), 7.65 (1H, d, *J* = 7.2 Hz, H-5'), 7.57 (1H, d, *J* = 7.6Hz, H-3,H-5), 7.12 (1H, s, C=C-H), 4.25 (2H, q, *J* = 7.1 Hz, -CH_2), 3.85 (6H, s, Ar-OCH_3), 3.77 (3H, s, -OCH_3), 2.26 (3H, t, *J* = 7.2 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.7 (amide C=O), 167.6

(ring C=O), 166.2 (-CH_3 ester C=O), 165.3 (-C_2H_5 ester C=O), 162.5 (C=N), 140.2 (=CH), 138.5 (C-4'), 137.4 (C-4), 136.7 (C-3'), 131.3 (S-C=), 130.5 (C-2,C-6), 129.4 (C-1'), 126.6 (C-1), 124.3 (C-6'), 122.2 (C-3,C-5), 118.3 (C-5'), 117.6 (C-2'), 59.2 (2H, s, -OCH_2), 56.5 (6H, s, Ar-OCH_3),53.1 (3H, s, -OCH_3),15.2 (3H, s, -CH_3); MS (70eV): *m/z* (%); [$M^{+\cdot}$] 498 (62), 333 (25), 183 (29), 165 (100 %), 149 (72), 135 (42), 105 (36); Anal. Calcd. for $C_{24}H_{22}N_2O_8S$: C= 57.83, H= 4.42, N= 5.62, S= 6.42. Found: C= 57.72, H= 4.33, N= 5.48, S= 6.34.

*Ethyl 4-((2Z,5Z)-2-(2-bromobenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4h**):*

(Yield: 72%): Rf[a]: 0.7; m.p: 127-128 °C; IR (KBr) ν_{max}: 2945 (C=C−H), 1726 (-C_2H_5 ester C=O), 1723 (-CH_3 ester C=O), 1676 (ring C=O), 1662 (Ar C=O), 1556 (C=N), 1554 (C=C), 1452 (C−N), 1261 (C−S) cm^{-1}; **^{1}HNMR** ($CDCl_3$, δ ppm): 7.89 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.58-7.84 (4H, m, Ar), 7.53 (1H, d, *J* = 7.6Hz, H-3,H-5), 7.11 (1H, s, C=C-H), 4.24 (2H, q, *J* = 7.1 Hz,-CH_2), 3.76 (3H, s, -OCH_3), 2.25 (3H, t, *J* = 7.6 Hz,-CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.3 (amide C=O), 165.6 (ring C=O), 165.4 (-CH_3 ester C=O), 164.3 (-C_2H_5 ester C=O), 162.6 (C=N), 140.5 (=CH), 138.2 (C-1'), 137.6 (C-4), 136.7 (C-4'), 134.6 (C-6'), 133.5 (C-3'), 131.6 (S-C=), 130.4 (C-2,C-6), 128.5 (C-5'), 126.6 (C-1), 123.5 (C-2'), 122.4 (C-3,C-5), 59.5 (2H, s, -OCH_2), 53.3 (3H, s, -OCH_3), 15.6 (3H, s, -CH_3); MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 516 (47), [M^{+2}]$^{+\cdot}$ 518 (22), 333 (16), 183 (36), 154 (26), 149 (100 %), 124 (17); Anal. Calcd. for $C_{22}H_{17}N_2O_6SBr$: C= 51.16, H= 3.29, N= 5.43, S= 6.20. Found: C= 51.11, H= 3.16, N= 5.36, S= 6.14.

*Ethyl 4-((2Z,5Z)-2-(3-methoxybenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4i**):*

(Yield: 66%): Rf[a]: 0.6; m.p: 134-135 °C; IR (KBr) ν_{max}: 2957 (C=C−H), 1724 (-C_2H_5 ester C=O), 1722 (-CH_3 ester C=O), 1679 (ring C=O), 1668 (Ar C=O), 1565 (C=N), 1543 (C=C), 1452 (C−N), 1263 (C−S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.88 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.56-7.82 (4H, m, Ar), 7.52 (1H, d, *J* = 7.6Hz, H-3,H-5), 7.13 (1H, s, C=C-H), 4.26 (2H, q, *J* = 7.1 Hz,-CH_2), 3.84 (3H, s, Ar-OCH_3), 3.75 (3H, s, -OCH_3), 2.24 (3H, t, *J* = 7.1 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 168.5 (amide C=O), 167.7 (ring C=O), 166.3 (-CH_3 ester C=O), 165.6 (-C_2H_5 ester C=O), 162.5 (C=N), 140.4 (=CH), 138.7 (C-3'), 137.4 (C-4), 136.3 (C-4'), 133.6 (C-6'), 131.4 (S-C=), 130.5 (C-2,C-6), 126.3 (C-1), 124.8 (C-1'), 123.5 (C-5'), 122.6 (C-3,C-5), 118.3 (C-2'), 59.2 (2H, s, -OCH_2), 56.1 (3H, s, Ar-OCH_3), 53.5 (3H, s, -OCH_3), 15.4 (3H, s, -CH_3); MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 468 (52), 333 (34), 319 (21), 183 (54), 149 (37), 135 (100 %), 124 (44); Anal. Calcd. for $C_{23}H_{20}N_2O_7S$: C= 58.97, H= 4.26, N= 5.97, S= 6.83. Found: C= 58.84, H= 4.16, N= 5.86, S= 6.71.

*Ethyl 4-((2Z,5Z)-2-(2-fluorobenzoylimino)-5-(2-methoxy-2-oxoethylidene)-4-oxothiazolidin-3-yl)benzoate (**4j**):*

(Yield: 65%): Rf[a]: 0.71; m.p: 142-143 °C; IR (KBr) ν_{max}: 2963 (C=C−H), 1731 (-C_2H_5 ester C=O), 1728 (-CH_3 ester C=O), 1682 (ring C=O), 1667 (Ar C=O), 1571 (C=N), 1564 (C=C), 1454 (C−N), 1276 (C−S) cm^{-1}; **^{1}H NMR** ($CDCl_3$, δ ppm): 7.97 (1H, d, *J* = 7.6 Hz, H-2,H-6), 7.92-7.65 (4H, m, Ar), 7.61 (1H, d, *J* = 7.6Hz, H-3,H-5), 7.18 (1H, s, C=C-H), 4.28 (2H, q, *J* = 7.1 Hz,-CH_2), 3.79 (3H, s, -OCH_3), 2.31 (3H, t, *J* = 7.5 Hz, -CH_3); **^{13}C NMR** ($CDCl_3$, δ ppm): 169.4 (amide C=O), 167.5 (ring C=O), 166.8 (-CH_3 ester C=O), 165.7 (-C_2H_5 ester C=O), 163.2 (C=N), 141.4 (=CH), 139.3 (C-2'), 137.8 (C-4), 136.5 (C-4'), 134.3 (C-6'), 131.7 (S-C=), 130.4 (C-2,C-6), 128.7 (C-5'), 126.8 (C-1), 125.5 (C-1'), 122.7 (C-3,C-5), 118.5 (C-3'), 60.4 (2H, s, -OCH_2), 53.6 (3H, s, -OCH_3), 15.7 (3H, s, -CH_3); MS (70 eV): *m/z* (%); [$M^{+\cdot}$] 456 (26), 397 (31), 333 (20), 183 (44), 152 (35), 149 (100 %), 124 (15), 105 (23), 95 (12); Anal. Calcd. for $C_{22}H_{17}N_2O_6SF$: C= 57.89, H= 3.73, N= 6.14, S= 7.02. Found: C= 57.49, H= 3.61, N= 6.05, S= 6.95.

Computational studies

The X-ray structure of human ALR2 (PDB ID 1US0)[23] in complex with $NADP^+$ and the inhibitor IDD 594 was obtained from the RCSB Protein Bank[24] and used as template for flexible ligand docking with AutoDock 4.2[25]. Prior to docking, crystallographic ligand and water molecules except $NAPD^+$ were removed from the X-ray structure and explicit hydrogen atoms were added using the Molecular Operating Environment (MOE 2012.10)[26]. Atomic partial charges were calculated using AutoDock Tools[25]. For docking, $NADP^+$ was kept rigid and was retained in its crystallographic position. Selected inhibitors were docked into the active site of ALR2 using AutoDock 4.2 with standard parameter settings. On the basis of visual inspection, high-scoring docking poses were selected and further explored as putative binding modes.

Extraction and activity of ALR1 and ALR2

Isolation and purification of aldehyde reductase (ALR1)

Bovine kidneys were obtained from a local slaughterhouse and processed within 2 h of release. The tissue was sliced into small pieces and then homogenized (Glas-Potter) in three volumes of 0.25 M sucrose, 2.0 mM EDTA, 2.5 mM β-mercaptoethanol and 10 mM sodium phosphate, pH 7.2. The homogenate was centrifuged at 10000 *g* at 0–4 °C for 30 min and the supernatant was subjected to a 40-60% ammonium sulfate fractionation. The concentration of ammonium sulfate was increased up to 75% to precipitate pure ALR1. All operations were carried out at 4 °C. The fractions possessing ALR1 activity, were re-dissolved in 10 mM sodium phosphate buffer, pH 7.2, containing 2.0 mM EDTA dipotassium salt and 2.0 mM β-mercaptoethanol and was dialyzed overnight using the same buffer. The dialyzed material was subsequently used in the enzymatic assays[27].

Isolation and purification of aldose reductase (ALR2)

Calf lenses were used for the isolation and purification of ALR2 using the previously described method with some modifications[28]. Calf lenses were homogenized with 10 volume of 10 mM phosphate buffer (pH 7.0) containing 1 mM β-mercaptoethanol and 1mM EDTA, and centrifuged at 18,000 *g* for 20 min. Ammonium sulfate was added slowly to the stirred supernatant fraction to yield a 35% saturated solution. After stirring for 3 h, the solution was centrifuged at 10,000 *g* for 20 min, and the supernatant was added gradually to 80% saturated ammonium sulfate. The solution was stirred for 3 h and centrifuged at 18,000 *g* for 20 min to obtain the precipitate. The precipitates obtained were dissolved in phosphate buffer and were dialyzed for 48 h in the same buffer, the dialyzed material was stored at -80°C until use in the enzyme assays.

Determination of aldehyde and aldose reductase activities

Enzyme activity was assayed spectrophotometrically using a microplate reader (Bio-Tek ELx 800™, Instruments Inc.,

Winooski, VT, USA) by measuring the decrease in absorption of NADPH at 340 nm, which accompanies the oxidation of NADPH catalyzed by ALR1 and ALR2. ALR1 activity was measured using the method of Costantino *et al*.[29]. The reaction mixture contained 20 µL of the inhibitor, 0.1 mM NADPH, 0.1 M sodium phosphate buffer, pH 6.2, 10 mM of sodium D-glucoronate (substrate) and enzyme preparation in a total volume of 200 µL. The reaction was started by the addition of substrate after incubation at 37 ºC for 10 min. The change in absorbance at 340 nm was measured on a micro plate reader after 5 mins incubation at 37 °C. Appropriate blank controls were employed for correction of oxidation associated with NADPH. One unit of enzyme was defined as an amount of enzyme required to catalyze the oxidation of 1 µM of NADPH per minute in the above mentioned assay conditions. ALR2 activity was assayed according to the method described by Da Settimo *et al*.[30]. The reaction mixture contained 40 µL of 10 mM DL-glyceraldehyde as a substrate and the other procedures were similar to the ALR1 assay. A 20 µL of valproic acid (10 mM) was used as positive control for ALR1 and sulindac for ALR2. All reactions were performed in triplicate. The absorbance was recorded and the data analyzed using PRISM 5.0 (GraphPad, San Diego, California, USA) to calculate the IC_{50} of the test samples. The percentage inhibition was calculated from the formula $100 - (OD_{testwell}/OD_{control}) \times 100$. [31].

Conclusion

Two novel series of methyl substituted oxothiazolidine acetate derivatives and ethyl substituted oxothiazolidine benzoate derivatives were synthesized and investigated for *in vitro* inhibitory activities on ALR1 and ALR2 using valproic acid and sulindac as reference inhibitors. Compounds **2d** and **2j** exhibited significant activity against ALR1in the nanomolar range with IC_{50} values of 0.01 ± 0.01 µM. Compounds **2a** and **4j** also displayed significant activity against ALR2 with IC_{50} values of 0.04 ± 0.02 µM and 0.20 ± 0.08 µM, respectively. Many new inhibitors were at least as active as or more active against ALR1 and ALR2 than the well-known standard inhibitors valproic acid and sulindac, respectively. Furthermore, a number of our new inhibitors displayed differential activity against ALR1 and ALR2. For example, compounds **2h**, **2i** and **2k** were much more active against ALR1 than ALR2. Moreover, opposite activity trends were also observed. Most notably, compound **2a** was highly active against ALR2, but only weakly active against ALR1, thus providing a prototypic inhibitor with selectivity for ALR2 over ALR1. In addition, slight selectivity for ALR2 over ALR1 was also observed for compound **4d**. Putative binding modes of selected (most active) inhibitors were explored by docking into the active site of theALR2. Preferred putative binding modes were in accord with the *in vitro* inhibition studies. On the basis of the present study, ethyl/methyl substituted oxothiazolidine derivatives may be considered as a new class of potent and selective inhibitors of ALR2. These compounds can be further modified to optimize ALR2 inhibitors and their selectivity over ALR1 for the prevention and treatment of diabetic complications. Understanding the pharmacophore requirements in more detail, for which computational studies reported herein lay a foundation, may lead to the rational design of further advanced ARIs.

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