Scientific paper

Synthesis and *in vitro* Study of Some New Bis(thiadiazolyl-2*H*-pyrazolo[3,4-*d*][1,3]thiazole)-methanes as Potential Nematicides

Cherkupally Sanjeeva Reddy,[1,*] Dasari Chandrashekar Rao,[1] Vookanti Yakub[1] and Adki Nagaraj[2]

[1] *Department of Chemistry, University College, Kakatiya University, Warangal 506 009, Andhra Pradesh, India*

[2] *Department of Pharmaceutical Chemistry, Telangana University, Nizamabad 503 175, Andhra Pradesh, India*

* *Corresponding author: E-mail: chsrkuc@yahoo.co.in*
Tel: +91-870-2573788, Fax:(off) +91-870-2439600

Received: 04-02-2010

Abstract

A new series of bis(pyrazolo[3,4-*d*][1,3]thiazoles) **7a**–**j** has been synthesized and characterized *via* IR, ^{1}H NMR, ^{13}C NMR, MS and elemental analyses. All the newly synthesized compounds **7a**–**j** have been assayed for their nematicidal activity against *Ditylenchus myceliophagus* and *Caenorhabditis elegans* by aqueous *in vitro* screening technique. The screened data reveal that the compound **7e** is most effective against *D. myceliophagus* and *C. elegans* with LD_{50} of 160 and 180 ppm respectively and is almost equal to the activity of the standard levamisole. The compounds **7h** and **7j** are also most active against *C. elegans* with LD_{50} of 190 ppm and *D. myceliophagus* with LD_{50} of 180 ppm, respectively. Further, **7a**–**j** were screened for their antibacterial activity. Most of these new compounds showed potent activity against the test bacteria and emerged as potential molecules for further development.

Keywords: Bis(thiadiazolyl-2*H*-pyrazolo[3,4-*d*][1,3]thiazole)methane, synthesis, nematicidal activity, antibacterial activity

1. Introduction

In recent years, attention has been increasingly paid to the synthesis of heterocyclic compounds which exhibit various biological activities,[1–6] including antibacterial, fungicidal, tuberculostatic and plant growth regulative properties. Further, it was indicated that bis-heterocyclic compounds displayed much better antibacterial activity than the simple heterocyclic compounds.[7]

Thiadiazoles exhibit a broad spectrum of biological effectiveness, such as anti-parkinsonism,[8] hypoglycaemic,[9] anti-histaminic,[10] anticancer,[11] anti-inflammatory,[12] anti-asthmatic[13] and anti-hypertensive.[14] Further, there has been a considerable interest in the chemistry of thiazolidin-4-one ring system, which is a core structure in various synthetic pharmaceuticals and displays a broad spectrum of biological activities.[15–17] Thiazolidin-4-one ring also occurs in nature; thus actithiazic acid isolated from *Streptomyces* strains exhibits highly specific *in vitro* activity against *Mycobacterium tuberculosis*.[18] Thiazolidin-4-one derivatives are also known to exhibit diverse bioactivities, such as anti-convulsant,[19] antidiarrheal,[20] anti-platelet activating factor,[21,22] anti-histaminic,[23,24] anti-diabetic,[25] cyclooxygenase (COX) inhibitory,[26] Ca^{2+}-channel blocker,[27] platelet activating factor (PAF) antagonist,[28] cardioprotective,[29] anti-ischemic,[30] anti-cancer,[31] tumor necrosis factor-á antagonist[32] and nematicidal activities.[33] Similarly, pyrazole and its derivatives could be considered as potential antimicrobial agents.[34,35] The other activities include antidepressant,[36] inhibitors of protein kinases,[37] antiagregating,[38] antiarthritic[39] and cerebroprotecting.[40] Some aryl pyrazoles were reported to have non-nucleoside HIV-1 reverse transcriptase inhibitory,[41] COX-2 inhibitory,[42,43] activator of the nitric oxide receptor and soluble guanylate cyclase activity.[44]

In view of all these reports and in continuation of our ongoing research on the synthesis of new heterocyclic derivatives,[45–51] it was thought of interest to accommodate

thiadiazole, thiazolidin-4-one and pyrazole moieties in a single molecular framework and to obtain new bis-heterocyclic compounds with potential biological activity. In the present study we performed the synthesis and biological evaluation of some new bis[thiadiazolyl-2*H*-pyrazolo[3,4-*d*][1,3]thiazole]methanes.

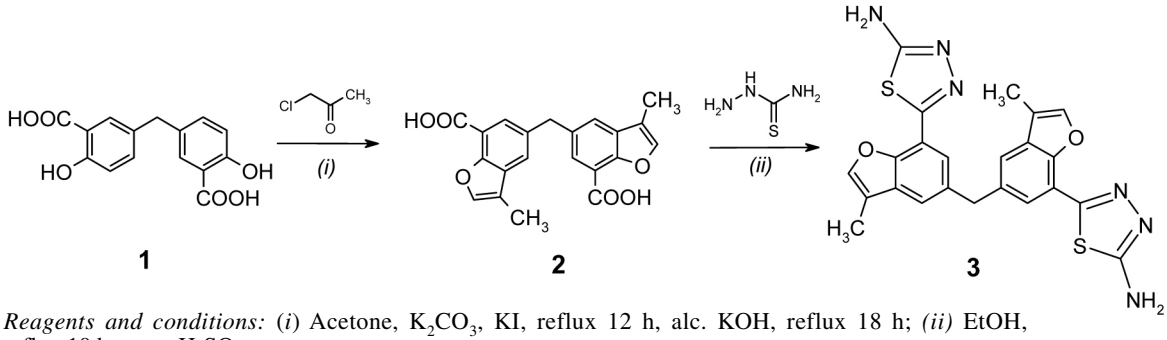
2. Results and Discussion

The synthesis of the title compounds is summarized in the Schemes 1–3. Salicylic acid derivative **1** has been prepared according to the literature procedure.[52] The compound **1** on condensation with chloroacetone, in the presence of K_2CO_3 and catalytic amount of KI, at reflux for 12 h followed by cyclization in alc. KOH at reflux for 18 h gave 5,5'-methylenebis(3-methylbenzo[*b*]furan-7-carboxylic acid) (**2**) in 72% yield. Further, condensation of the compound **2** with thiosemicarbazide in ethanol at reflux for 10 h, followed by cyclization in conc. H_2SO_4 at room temperature afforded the bis(1,3,4-thiadiazol) derivative **3** in 78% yield (Scheme 1).

The compound **3** on reaction with 4-methylbenzaldehyde, in the presence of acetic acid at reflux for 3 h, furnished the corresponding bis(methylideneamine) derivative **4** in 74% yield. The compounds **4** when reacted with thioglycolic acid, in the presence of $ZnCl_2$ in DMF at reflux temperature for 6 h, afforded the bis(1,3-thiazolan-4-one) derivative **5** in 71% yield (Scheme 2).

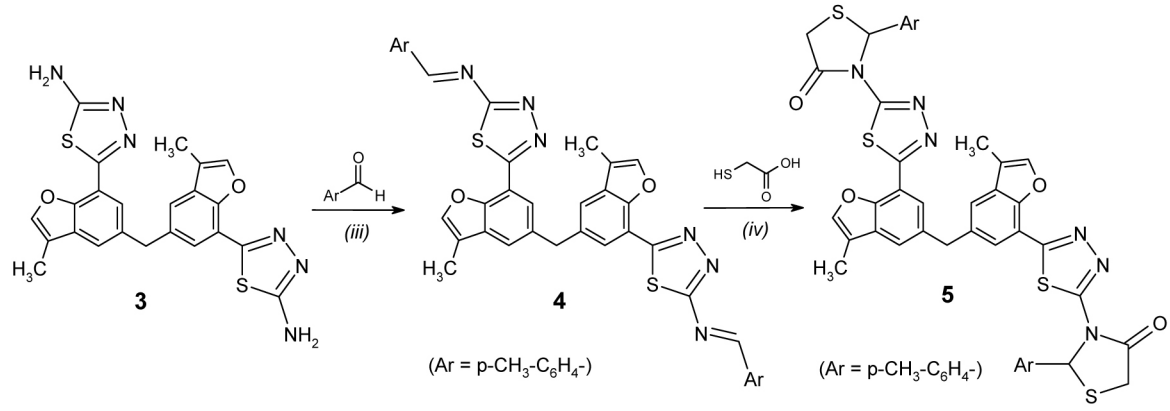
The compound **5** when reacted with the corresponding arylaldehyde, in the presence of anhydrous NaOAc in glacial AcOH at reflux tempereature for 6 h, formed the bis(arylmethylidene-1,3-thiazolan-4-ones) **6a**–**j** in 82–88% yield. Further, **6a**–**j** on cyclocondensation with hydrazine in the presence of anhydrous NaOAc in glacial AcOH at reflux temperature for 8 h, gave bis(pyrazolo[3,4-*d*][1,3]thiazoles) **7a**–**j** in 67–76% yield (Scheme 3). Chemical structures of the newly prepared compounds were confirmed by their elemental analysis, IR, ^{1}H NMR, ^{13}C NMR and MS spectral data.

In the IR spectra of compounds **7a**–**j**, disappearance of amide carbonyl (C=O) absorption at about 1700 cm^{-1}, olefinc (C=C) absorption at 1610 cm^{-1}, which was present in compounds **6a**–**j**, confirms the cyclization with the involvement of α,β-unsaturated carbonyl system. In addition, the absorption bands corresponding to C=N of the pyrazole moiety were observed at about 1600 cm^{-1}. Additional support was obtained from the ^{1}H NMR spectra. The N–CH–S protons of thiazole ring appeared at 7.58 ppm, 5-CH fused protons at 4.31 ppm as a doublet and



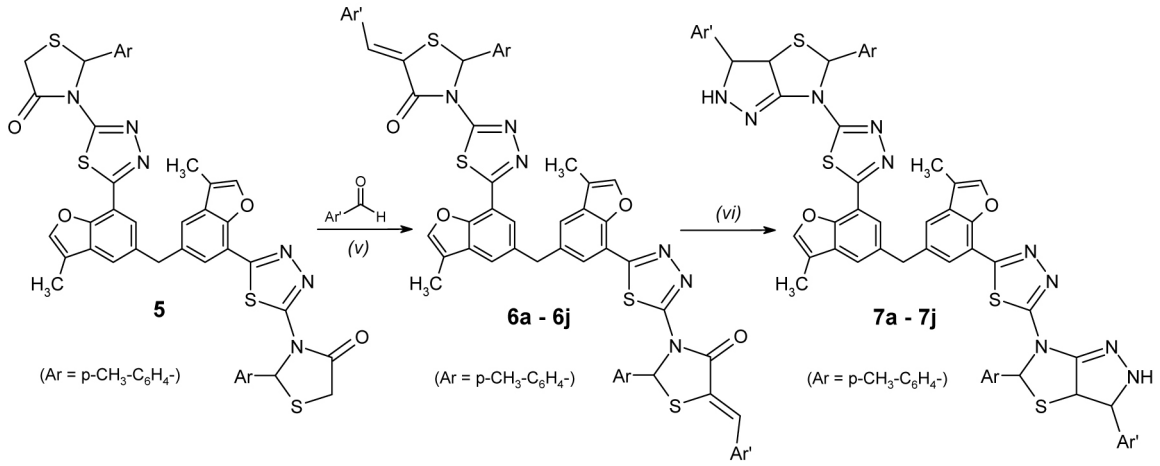
Reagents and conditions: (*i*) Acetone, K_2CO_3, KI, reflux 12 h, alc. KOH, reflux 18 h; *(ii)* EtOH, reflux 10 h, conc. H_2SO_4, rt.

Scheme 1. Synthetic route to bis(1,3,4-thiadiazol) derivative **3**



Reagents and conditions: (*iii*) AcOH, reflux 3 h; (*iv*) DMF, $ZnCl_2$, reflux 6 h.

Scheme 2. Synthetic route to bis(1,3-thiazolan-4-one) derivative **5**



6/7: Ar' = (**a**) 4-CH_3-C_6H_4; (**b**) 4-Cl-C_6H_4; (**c**) 4-NO_2-C_6H_4; (**d**) 3-NO_2-C_6H_4; (**e**) 4-OH-C_6H_4; (**f**) 2-OH-C_6H_4; (**g**) 4-N(CH_3)$_2$-C_6H_4; (**h**) 4-OH-3-OCH_3-C_6H_3; (**i**) 2-furyl; (**j**) 5-(1,3-benzodioxole)

Reagents and conditions: (*v*) AcOH/NaOAc, reflux 6 h; (*vi*) NH_2–NH_2 × HCl, AcOH/NaOAc, reflux 8 h.

Scheme 3. Synthetic route to bis(pyrazolo[3,4-*d*][1,3]thiazoles) **7a–j**

Ar'–CH–N proton of pyrazole ring appeared at 5.18 ppm as a doublet. These signals demonstrate that the cyclization has occurred. In the ^{13}C NMR spectra, the prominent signals corresponding to the carbons of pyrazolo-thiazole ring, for all the compounds, observed around 152.4, 67.1, 56.3 and 52.0 ppm, are a further evidence of their structures. In summary, all the newly synthesized compounds exhibited satisfactory spectral data and elemental analyses consistent with the proposed structures.

3. Nematicidal Evaluation

All novel compounds **7a–j** were assayed for their nematicidal activity against *Ditylenchus myceliophagus* and *Caenorhabditis elegans* by aqueous *in vitro* screening technique[53] at various concentrations. The *D. myceliophagus* was extracted form the cultivated mushrooms (*Agaricus bisporus*) infected with the nematode. *C. elegans* was grown on 10 cm 8P plates on a Na22 bacteria diet; they grow in a very thick layer and constitute an abundant food source for large quantities of nematode. The nematode water suspension was collected in petri dishes. Suspension of adult worms from five day old culture was diluted with approximately 100 to 250 nematodes/mL of water, 100 μL of the nematode suspension was introduced into a solution of each test compound at various concentrations in a well of 24-well plates and incubated at 25 °C. The percentage of immobile nematodes was recorded after two days. The nematicidal activity of each compound tested was compared with the standard drug levamisole. The results are expressed in terms of LD_{50} *i.e.* median lethal dose at which 50% of nematodes became immobile (dead).

The nematicidal screening data (Table 1) reveal that the compound **7e** is the most effective against *D. myceliophagus* and *C. elegans* with LD_{50} of 160 and 180 ppm, respectively, and is almost equally active as the standard levamisole. Compounds **7h** and **7j** are also most active against *C. elegans* with LD_{50} of 190 ppm and *D. myceliophagus* with LD_{50} of 180 ppm, respectively. The other compounds tested showed moderate activity. The comparison of LD_{50} values (in ppm) of the selected compounds **7e**, **7f**, **7h** and **7j** and the standard drug levamisole against nematodes is presented in Figure 1.

4. Antibacterial Evaluation

All novel compounds **7a–j** were also assayed for their antibacterial activity against Gram-positive bacteria *viz. Bacillus subtilis* (ATCC 6633), *Staphylococcus au-*

Table 1. Median Lethal Dose (LD_{50}, ppm) of compounds **7a–j**

Compound	7a	7b	7c	7d	7e	7f	7g	7h	7i	7j	Levamisole
D. myceliophagus	780	840	550	590	160	260	940	420	560	180	160
C. elegans	750	760	350	540	180	210	860	190	600	760	170

LD_{50}, (median lethal dose is the concentration at which 50% of nematodes became immobile).

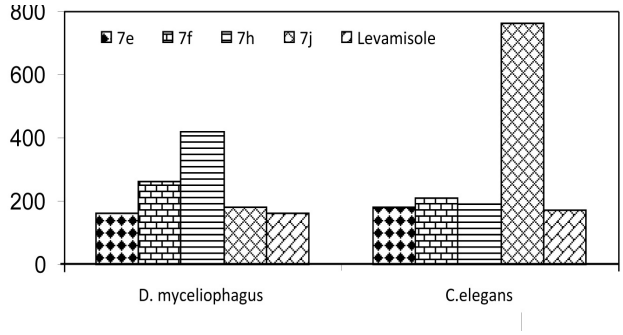


Fig. 1. Comparison of LD_{50} values (in ppm) of selected compounds and standard drug

reus (ATCC 6538p) and *Micrococcus luteus* (IFC 12708), and Gram-negative bacteria *viz. Proteus vulgaris* (ATCC 3851), *Salmonella typhimurium* (ATCC 14028) and *Escherichia coli* (ATCC 25922) by the broth dilution method, recommended by National Committee for Clinical Laboratory Stadards (NCCLS).[54] The minimum inhibitory concentration (MIC, μg/mL), was determined for all the compounds and compared with the control. The MIC values of the assayed compounds are presented in Table 2. All assays include the solvent and reference controls. ampicillin was used as the standard drug.

The investigation of antibacterial screening data (Table 2) revealed that the compound **7c** containing 3-nitrophenyl moiety at the pyrazole ring, is highly active against all the microorganisms employed (except *E. coli)* at 1.56 μg/mL concentration, which is equal to the standard. The compound **7j** containing 1,3-benzodioxole moiety at the pyrazole ring is also highly active against *M. luteus* and *P. vulgaris* at the same concentration as **7c**. The compound **7e** containing 4-hydroxyphenyl moiety at the pyrazole ring also showed good antibacterial activity against all the organisms tested. The compound **7a**, containing 4-methylphenyl moiety at the pyrazole ring, is almost inactive towards *M. luteus* and *E. coli*. The remaining compounds showed moderate to good activity.

5. Experimental

Commercial grade reagents were used as supplied. Solvents, except analytical reagent grade, were dried and purified according to the literature when necessary. Reaction progress and purity of the compounds were checked by thin-layer chromatography (TLC) on pre-coated silica gel F_{254} plates from Merck and compounds were visualized by exposure to UV light. Silica gel chromatographic columns (70–230 mesh) were used for separations. Melting points were determined on a Fisher–Johns apparatus and are uncorrected. IR spectra were recorded as KBr disks on a Perkin–Elmer FTIR spectrometer. The ^{1}H NMR and ^{13}C NMR spectra were recorded on a Varian Gemini spectrometer (300 MHz for ^{1}H and 75 MHz for ^{13}C). Chemical shifts are reported as δ ppm against TMS as internal standard and coupling constants (*J*) are reported in Hz units. Mass spectra were recorded on a VG micro mass 7070H spectrometer. Elemental analyses (C, H, N), determined by a Perkin–Elmer 240 CHN elemental analyzer, were within ±0.4% of theoretical values.

Preparation of 5,5'-methylenebis(3-methylbenzo[*b*]furan-7-carboxylic acid) (2): To a stirred solution of the compound **1** (5 mmol), anhydrous potassium carbonate (3 mmol) and a catalytic amount of potassium iodide in dry acetone (30 mL), was added drop-wise a solution of chloroacetone (10 mmol) in dry acetone (20 mL) at reflux temperature. Reflux was continued for 12 h. The reaction mixture was evaporated to dryness, and then transferred into ice-cold water, and the solid separated was collected by filtration. The crude product was dissolved in the ethanolic potassium hydroxide (10%, 100 mL) and further refluxed for 18 h. The excess ethanol was then removed by

Table 2. Antibacterial Activity of Compounds **7a**–**j**

Compound	Minimum Inhibitory Concentration (MIC) in μg/mL					
	B. subtilis	***S. aureus***	***M. luteus***	***P. vulgaris***	***S. typhimurium***	***E. coli***
7a	12.5	12.5	–	12.5	12.5	25.0
7b	6.25	12.5	6.25	6.25	6.25	12.5
7c	1.56	1.56	1.56	1.56	1.56	12.5
7d	6.25	6.25	6.25	–	12.5	6.25
7e	3.12	6.25	1.56	1.56	3.12	1.56
7f	12.5	6.25	3.12	12.5	6.25	12.5
7g	6.25	25.0	25.0	6.25	50.0	–
7h	3.12	6.25	12.5	12.5	6.25	25.0
7i	3.12	3.12	6.25	6.25	12.5	12.5
7j	6.25	12.5	1.56	1.56	12.5	12.5
Ampicillin	1.56	1.56	1.56	3.12	3.12	12.5

MIC, minimum inhibitory concentration (the lowest concentration that inhibited the bacterial growth).
– Indicates bacteria are resistant to the compound >50 μg/mL concentration. Standard deviation 0.05

distillation *in vacuo*, the reaction mixture was poured into the ice-cold aq. HCl and the solid separated was collected by filtration, purified by column chromatography using petroleum ether (b.p. 60–80 °C) as eluent to get the pure compound **2** as a yellow solid; yield 72%, mp 182–184 °C; IR (KBr) ν 3300–3200, 3037, 1695, 1030 cm^{-1}; ^1H NMR (DMSO-d_6): δ 9.90 (s, 2H, 2 × OH), 7.79 (s, 2H, ArH), 7.65–7.60 (m, 4H, ArH), 4.11 (s, 2H, CH_2), 2.39 (s, 6H, 2 × CH_3); ^{13}C NMR (DMSO-d_6): δ 172.6, 152.7, 143.9, 135.2, 132.9, 132.0, 124.6, 121.2, 119.1, 42.7, 9.2; Anal. Calcd for $C_{21}H_{16}O_6$: C, 69.23; H, 4.43. Found: C, 69.18; H, 4.40. MS: *m/z* 365 (M^++1, 10%), 106 (100%).

Preparation of 5,5'-[5,5'-methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazol-2-amine) (3): A mixture of compound **2** (5 mmol) and thiosemicarbazide (10 mmol) in acetone (20 mL) was refluxed for 10 h. The reaction mixture was allowed to cool and the solid separated was collected by filtration. The crude product was dissolved in conc. H_2SO_4 (5 mL) and stirred at room temperature for few minutes and left overnight. It was then poured on crushed ice; the resulting suspension was kept in ammonical water (25 mL) for 4 h, the solid was filtered and recrystallized from ethanol to get the pure compound **3** as a yellow solid; yield 78%, mp 192–194 °C; IR (KBr) ν 3350, 3050, 2985, 1605, 1030, 712 cm^{-1}; ^1H NMR (DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH), 7.47 (s, 2H, ArH), 4.92 (s, 4H, 2 × NH_2), 4.10 (s, 2H, CH_2), 2.36 (s, 6H, 2 × CH_3); ^{13}C NMR (DMSO-d_6): δ 168.2, 163.4, 153.4, 141.7, 136.1, 130.6, 128.2, 127.1, 123.4, 119.1, 42.6, 9.1; Anal. Calcd for $C_{23}H_{18}N_6O_2S_2$: C, 58.21; H, 3.82; N, 17.71. Found: C, 58.16; H, 3.80; N, 17.69. MS: *m/z* 475 (M^++1, 18%), 106 (100%).

Preparation of 5,5'-[5,5'-methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis[*N*-(4-methylbenzylidene)-1,3,4-thiadiazol-2-amine] (4): A mixture of compound **3** (5 mmol), 4-methylbenzaldehyde (10 mmol) and acetic acid (0.5 mL) was refluxed in toluene for 3 h using a Dean–Stark apparatus and the water formed was removed azeiotropically. The progress of the reaction was checked by TLC using toluene : ethyl acetate (4:1) as an eluent. After completion of the reaction, solvent was removed by distillation to give the solid, which was filtered, and recrystallized from ethyl alcohol to get the pure compound **4** as a yellow solid; yield 74%, mp 186–188 °C; IR (KBr) ν 3052, 2988, 1625, 1610, 1070, 714 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.76 (s, 2H, 2 × N-CH), 7.70–7.60 (m, 6H, ArH), 7.55–7.50 (m, 4H, ArH), 7.00–6.95 (m, 4H, ArH), 4.12 (s, 2H, CH_2), 2.44 (s, 6H, 2 × CH_3), 2.21 (s, 6H, 2 × CH_3); ^{13}C NMR (DMSO-d_6): δ 165.3, 162.7, 161.4, 150.6, 143.4, 135.6, 135.0, 133.9, 131.7, 129.6, 128.7, 126.4, 122.8, 118.5, 42.0, 20.7, 9.7; Anal. Calcd for $C_{39}H_{30}N_6O_2S_2$: C, 69.01; H, 4.45; N, 12.38. Found: C, 68.95; H, 4.40; N, 12.33. MS: *m/z* 678 (M^+).

Preparation of 3,3'-{5,5'-[5,5'-methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[2-(4-methylphenyl)thiazolidin-4-one] (5): A mixture of compound **4** (5 mmol), thioglycolic acid (12 mmol) in *N*,*N*-dimethylformamide (40 mL) with a pinch of anhydrous $ZnCl_2$, was refluxed for 6 h. The progress of the reaction was checked by TLC using toluene : ether (3:1) as an eluent. The reaction mixture was cooled to room temperature and then poured into crushed ice. It was set aside overnight at room temperature. The solid thus separated was filtered, washed several times with water, and purified by column chromatography on silica-gel with hexane-ethyl acetate as eluent to get the pure compound **5** as a brown solid; yield 71%, mp 210–212 °C; IR (KBr) ν 3062, 1698, 1612, 1604, 1475, 1066, 712 cm^{-1}; ^1H NMR (DMSO-d_6): δ 7.64 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.25–7.19 (m, 6H, ArH), 7.10–7.05 (m, 4H, ArH), 5.94 (s, 2H, 2 × N-CH), 4.20 (s, 2H, CH_2), 3.67 (s, 4H, 2 × CH_2), 2.36 (s, 6H, 2 × CH_3), 2.24 (s, 6H, 2 × CH_3); ^{13}C NMR (DMSO-d_6): δ 173.2, 170.6, 154.9, 150.8, 142.9, 137.9, 135.2, 135.0, 132.0, 127.4, 126.9, 125.8, 124.6, 123.7, 118.9, 72.0, 42.0, 33.9, 22.1, 9.20; Anal. Calcd for $C_{43}H_{34}N_6O_4S_4$: C, 62.45; H, 4.14; N, 10.16. Found: C, 62.90; H, 4.10; N, 10.11. MS: *m/z* 828 (M^+).

General procedure for the synthesis of bis(arylmethylidene-1,3-thiazolan-4-ones) 6a–j: A mixture of compound **5** (5 mmol), arylaldehyde (10 mmol) and sodium acetate (5 mmol) in anhydrous glacial acetic acid (10 mL), was refluxed for 6 h. The reaction mixture was concentrated and poured into ice cold water, the solid thus separated was filtered, washed with water. The crude product thus obtained was purified by column chromatography on silica gel with hexane-ethyl acetate as an eluent to afford pure compounds.

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-methylbenzylidene)-2-(4-methylphenyl)thiazolidine-4-one] (6a). Yield 83%, mp 184–186 °C; IR (KBr) ν 3056, 2942, 1720, 1610, 1604, 1270, 1066, 715 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.80 (s, 2H, 2 × CH=C), 7.64 (s, 2H, ArH), 7.57–7.54 (m, 6H, ArH), 7.35–7.25 (m, 14H, ArH), 6.65 (s, 2H, 2 × CH-S), 4.17 (s, 2H, CH_2), 2.51 (s, 6H, 2 × CH_3), 2.40 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 172.6, 164.2, 154.5, 149.5, 143.4, 139.7, 136.7, 136.1, 134.9, 134.0, 132.9, 132.4, 131.2, 129.4, 128.1, 126.7, 126.1, 125.2, 124.8, 116.4, 72.1, 43.4, 22.9, 22.0, 9.2; Anal. Calcd for $C_{59}H_{46}N_6O_4S_4$: C, 68.71; H, 4.50; N, 8.15. Found: C, 68.66; H, 4.45; N, 8.12. MS: *m/z* 1032 (M^+).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-chlorobenzylidene)-2-(4-methylphenyl)thiazolidine-4-one] (6b). Yield 77%, mp 178–180 °C; IR (KBr) ν 3047, 1716, 1617, 1595, 1272, 715, 686 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.80 (s, 2H, 2 × CH=C), 7.65–7.60 (m, 12H, ArH), 7.30–7.20 (m, 10H, ArH), 6.65 (s, 2H, 2 × CH-S), 4.17 (s, 2H, CH_2), 2.50 (s, 6H, 2 × CH_3), 2.20 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 171.2, 163.2, 152.3, 148.4, 143.2, 139.6, 136.1, 135.1, 134.9, 133.8,

131.9, 131.0, 130.6, 129.4, 129.9, 127.1, 126.1, 125.3, 125.1, 124.5, 116.5, 72.0, 43.3, 22.0, 9.2; Anal. Calcd for $C_{57}H_{40}Cl_2N_6O_4S_4$: C, 63.86; H, 3.76; N, 7.84. Found: C, 63.81; H, 3.80; N, 7.79. MS: *m/z* 1072 (M^+).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-nitrobenzylidene)-2-(4-methylphenyl)thiazolidine-4-one] (6c). Yield 72%, mp 189–191 °C; IR (KBr) *ν* 3048, 1717, 1612, 1590, 1562, 1370, 715 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 8.15–8.10 (m, 4H, ArH), 7.80 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 14H, ArH), 6.64 (s, 2H, 2 × CH-S), 4.20 (s, 2H, CH_2), 2.50 (s, 6H, 2 × CH_3), 2.21 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 172.0, 163.7, 152.0, 149.7, 149.0, 143.1, 142.0, 139.7, 136.1, 134.9, 132.7, 131.9, 131.1, 129.2, 128.3, 126.1, 125.4, 125.0, 124.9, 124.4, 116.4, 72.2, 43.3, 22.0, 9.2; Anal. Calcd for $C_{57}H_{40}N_8O_8S_4$: C, 62.62; H, 3.69; N, 10.25. Found: C, 62.57; H, 3.64; N, 10.20. MS: *m/z* 1094 (M^+).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(3-nitrobenzylidene)-2-(4-methylphenyl)thiazolidine-4-one] (6d). Yield 70%, mp 194–196 °C; IR (KBr) *ν* 3052, 1720, 1610, 1592, 1560, 1367, 716 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 8.27 (s, 2H, ArH), 7.80–7.70 (m 6H, ArH), 7.82 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 10H, ArH), 6.65 (s, 2H, 2 × CH-S), 4.18 (s, 2H, CH_2), 2.51 (s, 6H, 2 × CH_3), 2.20 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 170.1, 163.4, 152.7, 151.7, 149.3, 143.0, 139.7, 138.2, 137.1, 136.1, 134.9, 134.0, 132.0, 131.3, 129.1, 128.4, 126.1, 126.9, 125.8, 125.0, 124.3, 123.4, 116.2, 72.0, 43.0, 22.0, 9.3; Anal. Calcd for $C_{57}H_{40}N_8O_8S_4$: C, 62.62; H, 3.69; N, 10.25. Found: C, 62.59; H, 3.66; N, 10.18. MS: *m/z* 1094 (M^+).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-hydroxybenzylidene)-2-(4-methylphenyl)thiazolidine-4-one] (6e). Yield 76%, mp 192–194 °C; IR (KBr) *ν* 3270, 3030, 2928, 1719, 1612, 1270, 1066, 720 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.80 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 14H, ArH), 7.00–6.95 (m, 4H, ArH), 6.65 (s, 2H, 2 × CH-S), 5.17 (s, 2H, 2 × OH), 4.19 (s, 2H, CH_2), 2.50 (s, 6H, 2 × CH_3), 2.20 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 172.0, 163.4, 158.1, 152.2, 149.4, 143.1, 139.6, 136.1, 134.6, 132.7, 131.9, 131.0, 129.2, 128.7, 126.8, 126.0, 125.8, 125.0, 124.2, 117.4, 116.3, 72.1, 42.9, 22.0, 9.3; Anal. Calcd for $C_{57}H_{42}N_6O_6S_4$: C, 66.13; H, 4.09; N, 8.12. Found: C, 66.08; H, 4.04; N, 8.07. MS: *m/z* 1037 (M^+ +1).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(2-hydroxybenzylidene)-2-(4-methylphenyl)thiazolidine-4-one] (6f). Yield 72%, mp 179–181 °C; IR (KBr) *ν* 3230, 2972, 1718, 1615, 1272, 1070, 725 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.84 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 16H, ArH), 6.90 (m, 2H, ArH), 6.64 (s, 2H, 2 × CH-S), 4.76 (s, 2H, 2 × OH), 4.19 (s, 2H, CH_2), 2.51 (s, 6H, 2 × CH_3), 2.23 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 171.4, 163.2, 159.7, 152.4, 149.3, 143.2, 139.5, 136.1, 134.6, 132.9, 131.2, 131.9, 129.1, 128.8, 128.0, 126.2, 125.9, 125.1, 124.5, 123.0, 118.4, 116.2, 115.7, 72.0, 43.0, 22.0, 9.2; Anal. Calcd for $C_{57}H_{42}N_6O_6S_4$: C, 66.13; H, 4.09; N, 8.12. Found: C, 66.10; H, 4.06; N, 8.05. MS: *m/z* 1036 (M^+).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis{5-[4-(dimethylamino)benzylidene]-2-(4-methylphenyl)thiazolidine-4-one} (6g). Yield 68%, mp 191–193 °C; IR (KBr) *ν* 3062, 2965, 1715, 1620, 1270, 1065, 728 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.80 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 10H, ArH), 6.95–6.80 (m, 8H, ArH), 6.65 (s, 2H, 2 × CH-S), 4.18 (s, 2H, CH_2), 3.10 (s, 12H, 4 × N-CH_3), 2.50 (s, 6H, 2 × CH_3), 2.25 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 171.0, 163.1, 152.4, 151.6, 149.3, 143.1, 139.6, 134.6, 136.1, 132.8, 131.2, 130.6, 129.1, 128.0, 126.9, 126.0, 125.4, 124.6, 120.4, 116.4, 113.4, 72.1, 43.0, 38.7, 22.0, 9.4; Anal. Calcd for $C_{61}H_{52}N_8O_4S_4$: C, 67.26; H, 4.81; N, 10.29. Found: C, 67.22; H, 4.76; N, 10.24. MS: *m/z* 1091 (M^+ +1).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-hydroxy-3-methoxybenzylidene)-2-(4-methylphenyl) thiazolidine-4-one] (6h). Yield 69%, mp 184–186 °C; IR (KBr) *ν* 3275, 3042, 2932, 1718, 1612, 1270, 1070, 710 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.69 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 10H, ArH), 7.00–6.90 (m, 6H, ArH), 6.63 (s, 2H, 2 × CH-S), 5.21 (s, 2H, 2 × OH), 4.18 (s, 2H, CH_2), 3.78 (s, 6H, 2 × OCH_3), 2.52 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 172.3, 163.2, 152.7, 149.8, 149.1, 148.1, 143.2, 139.6, 136.1, 134.6, 132.0, 131.0, 129.0, 128.1, 127.6, 126.7, 126.0, 125.9, 125.0, 124.3, 116.1, 114.7, 113.4, 72.1, 55.9, 43.1, 22.1, 9.1; Anal. Calcd for $C_{59}H_{46}N_6O_8S_4$: C, 64.70; H, 4.23; N, 7.67. Found: C, 64.66; H, 4.25; N, 7.61. MS: *m/z* 1096 (M^+).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(2-furylmethylene)-2-(4-methylphenyl)thiazolidine-4-one] (6i). Yield 75%, mp 210–212 °C; IR (KBr) *ν* 3030, 1720, 1610, 1270, 1075, 715 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.90 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 10H, ArH), 7.10–7.00 (m, 2H, ArH), 6.60 (s, 2H, 2 × CH-S), 6.20–6.10 (m, 4H, ArH), 4.18 (s, 2H, CH_2), 2.49 (s, 6H, 2 × CH_3), 2.24 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 171.2, 162.0, 154.1, 152.3, 149.0, 143.9, 143.0, 139.6, 136.1, 135.3, 134.6, 131.0, 129.2, 128.2, 126.2, 126.0, 125.7, 125.2, 124.5, 116.2, 110.1, 68.7, 43.2, 22.1, 9.0; Anal. Calcd for $C_{53}H_{38}N_6O_6S_4$: C, 64.75; H, 3.90; N, 8.55. Found: C, 64.70; H, 3.85; N, 8.51. MS: *m/z* 984 (M^+).

3,3'-{5,5'-[5,5'-Methylenebis(3-methylbenzo[*b*]furan-

7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(benzo[*d*][1,3]dioxol-5-ylmethylene)-2-(4-methylphenyl)thiazolidine-4-one] (6j). Yield 71%, mp 202–204 °C; IR (KBr) *ν* 3030, 1715, 1615, 1260, 1072, 712 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.65 (s, 2H, 2 × CH=C), 7.60–7.50 (m, 4H, ArH), 7.30–7.20 (m, 12H, ArH), 7.00–6.90 (m, 4H, ArH), 6.62 (s, 2H, 2 × CH-S), 5.76 (s, 4H, 2 × O-CH_2-O), 4.19 (s, 2H, CH_2), 2.49 (s, 6H, 2 × CH_3), 2.20 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 171.0, 163.1, 154.0, 149.9, 149.1, 147.3, 143.2, 139.4, 136.1, 134.7, 132.6, 131.1, 129.0, 128.3, 127.4, 126.2, 125.9, 125.1, 124.9, 124.0, 116.4, 110.7, 108.9, 102.3, 72.1, 43.0, 22.0, 9.2; Anal. Calcd for $C_{59}H_{42}N_6O_8S_4$: C, 64.94; H, 3.88; N, 7.70. Found: C, 64.90; H, 3.82; N, 7.66. MS: *m/z* 1092 (M^+).

General procedure for the synthesis of bis(pyrazolo[3,4-*d*][1,3]thiazoles) 7a–j: A mixture of compound **6** (5 mmol), hydrazine hydrochloride (10 mmol) and anhydrous sodium acetate (5 mmol) in glacial acetic acid (20 mL), was refluxed for 8 h. The reaction mixture was concentrated and cooled to room temperature, the solid thus separated, was filtered, washed thoroughly with water. The crude product thus obtained was purified by column chromatography on silica gel with hexane-ethyl acetate as an eluent to get the pure compounds.

Bis(7-{5-[3,5-di(4-methylphenyl)-3,3a-dihydro-2*H*-pyrazolo[3,4-*d*]thiazol-6(5*H*)-yl]-1,3,4-thiadiazol-2-yl}-3-methylbenzofuran-5-yl)methane (7a). Yield 69%; mp 196–198 °C; IR (KBr) *ν* 3400–3300, 3078, 2947, 1604, 1598, 1065, 710 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.35–7.20 (m, 8H, ArH), 7.10–7.05 (m, 8H, ArH), 5.61 (bs, 2H, 2 × NH), 5.18 (d, *J* = 1.9 Hz, 2H, 2 × CH-N), 4.31 (d, *J* = 1.9 Hz, 2H, 2 × CH-S), 4.18 (s, 2H, CH_2), 2.47 (s, 6H, 2 × CH_3), 2.31 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-*d*6): δ 166.9, 157.6, 152.4, 150.6, 141.7, 139.3, 138.0, 136.9, 133.7, 132.0, 130.0, 129.1, 128.9, 128.2, 128.0, 127.5, 125.1, 123.0, 117.9, 67.1, 56.3, 52.0, 42.0, 22.1, 21.0, 9.7; Anal. Calcd for $C_{59}H_{50}N_{10}O_2S_4$: C, 66.89; H, 4.76; N, 13.22. Found: C, 66.82; H, 4.72; N, 13.19. MS: *m/z* 1061 (M^+ +1).

Bis(7-{5-[3-(4-chlorophenyl)-5-(4-methylphenyl)-3,3a-dihydro-2*H*-pyrazolo[3,4-*d*]thiazol-6(5*H*)-yl]-1,3,4-thiadiazol-2-yl}-3-methylbenzofuran-5-yl)methane (7b). Yield 71%; mp 208–210 °C; IR (KBr) *ν* 3400–3300, 3067, 2949, 1610, 1592, 1063, 715, 685 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.66 (m, 4H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.40–7.35 (m, 4H, ArH), 7.22 (m, 8H, ArH), 7.10–7.05 (m, 4H, ArH), 5.61 (bs, 2H, 2 × NH), 5.18 (d, *J* = 1.9 Hz, 2H, 2 × CH-N), 4.31 (d, *J* = 1.9 Hz, 2H, 2 × CH-S), 4.18 (s, 2H, CH_2), 2.47 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.9, 157.6, 152.3, 150.6, 141.5, 138.0, 136.9, 134.5, 133.7, 132.6, 130.0, 129.9, 129.1, 128.2, 127.5, 127.0, 125.1, 123.0, 117.9, 67.1, 56.3, 52.0, 42.0, 22.1, 9.7; Anal. Calcd for $C_{57}H_{44}Cl_2N_{10}O_2S_4$: C, 62.23; H, 4.03; N, 12.73. Found: C, 62.18; H, 4.00; N, 12.70. MS: *m/z* 1100 (M^+).

Bis(3-methyl-7-{5-[3-(4-nitrophenyl)-5-(4-methylphenyl)-3,3a-dihydro-2*H*-pyrazolo[3,4-*d*]thiazol-6(5*H*)-yl]-1,3,4-thiadiazol-2-yl}benzofuran-5-yl)methane (7c). Yield 70%; mp 204–206 °C; IR (KBr) *ν* 3400–3300, 3067, 2965, 1610, 1590, 1570, 1320, 1063, 710 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 8.10–8.00 (m, 4H, ArH), 7.65–7.60 (m, 8H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.20–7.15 (m, 8H, ArH), 5.61 (bs, 2H, 2 × NH), 5.19 (d, *J* = 1.9 Hz, 2H, 2 × CH-N), 4.30 (d, *J* = 1.9 Hz, 2H, 2 × CH-S), 4.18 (s, 2H, CH_2), 2.47 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.7, 157.6, 152.2, 150.6, 144.7, 141.6, 138.0, 136.9, 136.7, 136.0, 133.7, 130.0, 129.1, 128.2, 127.5, 125.1, 123.0, 122.6, 117.9, 67.0, 56.3, 52.1, 42.0, 22.1, 9.7; Anal. Calcd for $C_{57}H_{44}N_{12}O_6S_4$: C, 61.06; H, 3.96; N, 14.99. Found: C, 61.00; H, 3.90; N, 14.93. MS: *m/z* 1122 (M^+).

Bis(3-methyl-7-{5-[3-(3-nitrophenyl)-5-(4-methylphenyl)-3,3a-dihydro-2*H*-pyrazolo[3,4-*d*]thiazol-6(5*H*)-yl]-1,3,4-thiadiazol-2-yl}benzofuran-5-yl)methane (7d). Yield 76%; mp 222–224 °C; IR (KBr) *ν* 3400–3300, 3032, 2981, 1603, 1585, 1061, 705 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 8.10–8.00 (m, 4H, ArH), 7.65–7.60 (m, 6H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (m, 4H, ArH), 7.20–7.15 (m, 8H, ArH), 5.64 (bs, 2H, 2 × NH), 5.19 (d, *J* = 1.9 Hz, 2H, 2 × CH-N), 4.30 (d, *J* = 1.9 Hz, 2H, 2 × CH-S), 4.20 (s, 2H, CH_2), 2.47 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.8, 157.6, 152.4, 150.5, 147.3, 141.5, 138.0, 136.9, 134.4, 133.7, 130.0, 129.1, 128.4, 128.7, 128.0, 127.5, 126.4, 125.1, 123.3, 123.0, 122.7, 117.9, 67.1, 56.3, 52.0, 42.0, 22.1, 9.7; Anal. Calcd for $C_{57}H_{44}N_{12}O_6S_4$: C, 61.06; H, 3.96; N, 14.99. Found: C, 61.02; H, 3.90; N, 14.93. MS: *m/z* 1122 (M^+).

4,4'-(6,6'-{5,5'-[5,5'-Methylenebis(3-methylbenzofuran-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-methylphenyl)-3,3a,5,6-tetrahydro-2*H*-pyrazolo[3,4-*d*]thiazole-6,3-diyl])diphenol (7e). Yield 74%; mp 218–220 °C; IR (KBr) *ν* 3400–3300, 3061, 2991, 1602, 1067, 715 cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.20–7.10 (m, 12H, ArH), 6.85–6.80 (m, 4H, ArH), 5.75 (s, 2H, 2 × OH), 5.63 (bs, 2H, 2 × NH), 5.19 (d, *J* = 1.9 Hz, 2H, 2 × CH-N), 4.30 (d, *J* = 1.9 Hz, 2H, 2 × CH-S), 4.20 (s, 2H, CH_2), 2.47 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.9, 158.0, 153.4, 152.4, 150.4, 141.6, 138.0, 136.9, 133.7, 130.0, 129.1, 128.2, 128.0, 127.5, 127.1, 125.1, 123.0, 116.9, 66.8, 56.3, 52.0, 42.0, 22.1, 9.7; Anal. Calcd for $C_{57}H_{46}N_{10}O_4S_4$: C, 64.39; H, 4.36; N, 13.17. Found: C, 64.40; H, 4.32; N, 13.13. MS: *m/z* 1065 (M^++ 1).

2,2'-(6,6'-{5,5'-[5,5'-Methylenebis(3-methylbenzofuran-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-methylphenyl)-3,3a,5,6-tetrahydro-2*H*-pyrazolo[3,4-

***d*]thiazole-6,3-diyl])diphenol (7f).** Yield 73%; mp 210–212 °C; IR (KBr) ν 3400–3300, 3065, 2997, 1604, 1071, 712 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.30–7.15 (m, 10H, ArH), 6.82–6.78 (m, 6H, ArH), 5.62 (bs, 2H, 2 × NH), 5.19 (d, J = 1.9 Hz, 2H, 2 × CH-N), 4.91 (s, 2H, 2 × OH), 4.30 (d, J = 1.9 Hz, 2H, 2 × CH-S), 4.19 (s, 2H, CH_2), 2.48 (s, 6H, 2 × CH_3), 2.23 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.9, 158.0, 156.4, 152.3, 150.3, 147.6, 144.5, 141.4, 136.9, 136.0, 133.7, 130.0, 129.1, 128.2, 127.5, 127.3, 125.1, 123.0, 120.4, 117.9, 117.0, 115.8, 66.9, 56.3, 52.0, 42.0, 22.0, 9.7; Anal. Calcd for $C_{57}H_{46}N_{10}O_4S_4$: C, 64.39; H, 4.36; N, 13.17. Found: C, 64.37; H, 4.38; N, 13.12. MS: *m/z* 1064 (M^+).

4,4'-(6,6'-{5,5'-[5,5'-Methylenebis(3-methylbenzofuran-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-methylphenyl)-3,3a,5,6-tetrahydro-2*H*-pyrazolo[3,4-*d*]thiazole-6,3-diyl])bis(*N*,*N*-dimethylaniline) (7g). Yield 70%; mp 232–234 °C; IR (KBr) ν 3400–3300, 3061, 2991, 1601, 1410, 1052, 712 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH, 2 × N-H-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.20–7.15 (m, 10H, ArH), 6.55–6.50 (m, 6H, ArH), 5.64 (bs, 2H, 2 × NH), 5.19 (d, J = 1.9 Hz, 2H, 2 × CH-N), 4.30 (d, J = 1.9 Hz, 2H, 2 × CH-S), 4.18 (s, 2H, CH_2), 2.89 (s, 12H, 4 × CH_3), 2.47 (s, 6H, 2 × CH_3), 2.24 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.8, 157.7, 152.3, 150.3, 148.3, 142.7, 141.3, 138.0, 136.9, 133.7, 130.0, 129.1, 128.2, 127.5, 125.1, 123.0, 122.9, 117.9, 111.2, 67.0, 56.3, 52.0, 43.7, 42.0, 22.1, 9.7; Anal. Calcd for $C_{61}H_{56}N_{12}O_2S_4$: C, 65.57; H, 5.05; N, 15.04. Found: C, 65.55; H, 5.02; N, 15.00. MS: *m/z* 1118 (M^+).

4,4'-(6,6'-{5,5'-[5,5'-Methylenebis(3-methylbenzofuran-7,5-diyl)]bis(1,3,4-thiadiazole-5,2-diyl)}bis[5-(4-methylphenyl)-3,3a,5,6-tetrahydro-2*H*-pyrazolo[3,4-*d*]thiazole-6,3-diyl])bis(2-methoxyphenol) (7h). Yield 75%; mp 221–223 °C; IR (KBr) ν 3400–3300, 3064, 2992, 1600, 1077, 1030, 706 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.20–7.10 (m, 8H, ArH), 7.00–6.90 (m, 6H, ArH), 5.62 (bs, 2H, 2 × NH), 5.49 (s, 2H, 2 × OH), 5.20 (d, J = 1.9 Hz, 2H, 2 × CH-N), 4.30 (d, J = 1.9 Hz, 2H, 2 × CH-S), 4.18 (s, 2H, CH_2), 3.82 (s, 6H, 2 × OCH_3), 2.45 (s, 6H, 2 × CH_3), 2.23 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.7, 157.8, 152.4, 150.4, 147.5, 144.8, 141.3, 138.1, 136.9, 133.7, 130.0, 129.8, 129.1, 128.2, 127.6, 127.0, 125.1, 123.1, 117.9, 114.7, 110.1, 66.8, 59.3, 56.3, 52.0, 42.0, 22.1, 9.7; Anal. Calcd for $C_{59}H_{50}N_{10}O_6S_4$: C, 63.08; H, 4.49; N, 12.47. Found: C, 63.02; H, 4.45; N, 12.44. MS: *m/z* 1124 (M^+).

Bis(7-{5-[3-(2-furyl)-5-(4-methylphenyl)-3,3a-dihydro-2*H*-pyrazolo[3,4-*d*]thiazol-6(5*H*)-yl]-1,3,4-thiadiazol-2-yl}-3-methylbenzofuran-5-yl)methane (7i). Yield 74%, mp 211–213 °C; IR (KBr) ν 3400–3300, 3062, 2987, 1610, 1044, 715 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (s, 2H, ArH), 7.30–7.20 (m, 10H, ArH), 6.20–6.15 (m, 2H, ArH), 5.63 (bs, 2H, 2 × NH), 5.20 (m, 4H, ArH, 2 × CH-N), 4.30 (d, J = 1.9 Hz, 2H, 2 × CH-S), 4.19 (s, 2H, CH_2), 2.45 (s, 6H, 2 × CH_3), 2.22 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.5, 157.9, 152.3, 150.3, 145.7, 143.5, 141.2, 138.0, 136.0, 133.7, 130.0, 129.1, 128.2, 127.5, 125.1, 123.1, 117.9, 108.2, 104.9, 66.9, 56.9, 56.2, 42.0, 22.1, 9.7; Anal. Calcd for $C_{53}H_{42}N_{10}O_4S_4$: C, 62.95; H, 4.19; N, 13.85. Found: C, 62.91; H, 4.18; N, 13.80. MS: *m/z* 1012 (M^+).

Bis(7-{5-[3-(benzo[*d*][1,3]dioxol-5-yl)-5-(4-methylphenyl)-3,3a-dihydro-2*H*-pyrazolo[3,4-*d*]thiazol-6(5*H*)-yl]-1,3,4-thiadiazol-2-yl}-3-methylbenzofuran-5-yl) methane (7j). Yield 75%; mp 227–229 °C; IR (KBr) ν 3400–3300, 3067, 2987, 1604, 1067, 7106 cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ 7.65–7.60 (m, 4H, ArH, 2 × N-CH-S), 7.58 (s, 2H, ArH), 7.49 (m, 6H, ArH), 7.22 (m, 6H, ArH), 7.10–7.05 (m, 4H, ArH), 5.63 (bs, 2H, 2 × NH), 5.20 (d, J = 1.9 Hz, 2H, 2 × CH-N), 4.62 (s, 4H, 2 × CH_2), 4.30 (d, J = 1.9 Hz, 2H, 2 × CH-S), 4.19 (s, 2H, CH_2), 2.45 (s, 6H, 2 × CH_3), 2.24 (s, 6H, 2 × CH_3); ^{13}C NMR (75 MHz, DMSO-d_6): δ 166.8, 157.8, 152.4, 150.9, 150.1, 146.7, 141.2, 138.1, 136.0, 133.6, 130.0, 129.1, 128.2, 127.5, 126.7, 125.1, 117.9, 109.9, 108.1, 101.4, 67.0, 56.3, 52.0, 42.0, 22.1, 9.7; Anal. Calcd for $C_{59}H_{46}N_{10}O_6S_4$: C, 63.31; H, 4.14; N, 12.51. Found: C, 63.30; H, 4.10; N, 12.55. MS: *m/z* 1120 (M^+).

6. Conclusions

A new series of bis(pyrazolo[3,4-*d*][1,3]thiazoles) **7a–j** has been synthesized and evaluated for their nematicidal and antibacterial activity. Most of the newly synthesized compounds showed appreciable activity and emerged as potential molecules for further development.

7. Acknowledgements

The authors are grateful to the Director, Indian Institute of Chemical Technology, Hyderabad, India, for providing NMR and mass spectral data. Financial assistance from the UGC SAP (Phase-I)-DRS Programme, New Delhi, India, is greatly acknowledged.

8. References

1. D. P. Singh, R. Kumar, M. Kamboj, K. Jain, *Acta Chim. Slov*. **2009**, *56*, 780–785.
2. L. Pezdirc, D. Bevk, S. Pirc, J. Svete, *Acta Chim. Slov*. **2009**, *56*, 545–558.
3. S. C. Madhukar, A. P. Kiran, A. B. Pravin, R. K. Rahul, S. V. Deepak, *J. Braz. Chem. Soc*. **2008**, *19*, 42–52.

4. P.-F. Xu, X.-W. Sun, L.-M. Zhang, Z.-Y. Zhang, *J. Chem. Res. (S)* **1999**, 170–171.
5. Y. N. Mabkhoot, *Molecules* **2009**, *14*, 1904–1914.
6. F. Q. He, X. H. Liu, B. L. Wang, Z. M. Li, *Heteroatom Chem*. **2008**, *19*, 21–27.
7. V. Padmavathi, T. Radhalakshmi, K. Mahesh, A. V. N. Mohan, *Indian J. Chem*. **2008**, *47B*, 1707–1712.
8. F. Azam, A. I. Ibn-Rajab, A. A. Alruiad, *Pharmazie* **2009**, *64*, 771–795.
9. A. K. Avetisyan, T. R. Ovsepyan, N. O. Stepanyan, L. G. Sapondzhyan, *Pharm. Chem. J*. **1981**, *15*, 416–418.
10. I. Lalezari, A. Shafiee, A. Badaly, M. M. Salimi, M. A. Khoyi, F. Abtahi, M. R. Zarrindast, *J. Pharm. Sci*. **1975**, *64*, 1250–1252.
11. C. Parkanyi, H. L. Yuan, B. H. E. Stromberg, A. Evenzahav, *J. Heterocycl. Chem*. **1992**, *29*, 749–753.
12. D. H. Boschelli, D. T. Connor, D. A. Bornemeier, R. D. Dyer, J. A. Kennedy, P. J. Kuipers, G. C. Okonkwo, D. J. Schrier, C. D. Wright, *J. Med. Chem*. **1993**, *36*, 1802–1810.
13. P. Bhattacharya, J. T. Leonard, K. Roy, *Bioorg. Med. Chem*. **2005**, *13*, 1159–1165.
14. L. Vio, M. G. Mamolo, A. Laneve, *Il Farmaco* **1989**, *44*, 165–172.
15. M. G. Vigorita, R. Ottanà, F. Monforte, R. Maccari, A. Trovato, M. T. Monforte, M. F. Taviano, *Bioorg. Med. Chem. Lett*. **2001**, *11*, 2791–2794.
16. M. S. Chande, V. Suryanarayan, *J. Chem. Res*. **2005**, 345–347.
17. C. V. Kavitha, Basappa, S. N. Swamy, K. Mantelingu, S. Doreswamy, M. A. Sridhar, J. S. Prasad, K. S. Rangappa, *Bioorg. Med. Chem*. **2006**, *14*, 2290–2299.
18. B. A. Sobin, *J. Am. Chem. Soc*. **1952**, *74*, 2947–2948.
19. M. R. Shiradkar, M. Ghodake, K. G. Bothara, S. V. Bhandari, A. Nikalje, K. C. Akula, N. C. Desai, P. J. Burange, *Arkivoc* **2007**, *xiv*, 58–74.
20. A. A. Ahmadu, A. U. Zezi, A. H. Yaro, *Afr. J. Tradit. Complement. Altern. Med*. **2007**, *4*, 524–528.
21. Y. Tanabe, G. Suzukamo, Y. Komuro, N. Imanishi, S. Morooka, M. Enomoto, A. Kojima, Y. Sanemitsu, M. Mizutani, *Tetrahedron Lett*. **1991**, *32*, 379–382
22. Y. Tanabe, H. Yamamoto, M. Murakami, K. Yanagi, Y. Kubota, H. Okumura, Y. Sanemitsu, G. Suzukamo, *J. Chem. Soc. Perkin Trans 1* **1995**, 935–947.
23. M. V. Diurno, O. Mazzoni, G. Correale, I. G. Monterry, A. Calignano, G. La Rana, A. Bolognese, *Il Farmaco* **1999**, *54*, 579–583.
24. T. Previtera, M. G. Vigorita, M. Basile, F. Orsini, F. Benetollo, G. Bombieri, *Eur. J. Med. Chem*. **1994**, *29*, 317–324.
25. S. D. Firke, B. M. Firake, R. Y. Chaudhari, V. R. Patil, *Asian J. Research Chem*. **2009**, *2*, 157–161.
26. R. Ottaná, E. Mazzon, L. Dugo, F. Monforte, R. Maccari, L. Sautebin, G. De Luca, M. G. Vigorita, S. Alcaro, F. Ortuso, A. P. Caputi, S. Cuzzocrea, *Eur. J. Pharmacol*. **2002**, *448*, 71–80.
27. T. Kato, T. Ozaki, K. Tamura, Y. Suzuki, M. Akima, N. Ohi, *J. Med. Chem*. **1999**, *42*, 3134–3146.
28. N. Imanishi, Y. Komuro, S. Morooka, *Lipids* **1991**, *26*, 1391–1395.
29. T. Kato, T. Ozaki, N. Ohi, *Tetrahedron Asymm.* **1999**, *10*, 3963–3968.
30. Y. Adachi, Y. Suzuki, N. Homma, M. Fukazawa, K. Tamura, I. Nishie, O. Kuromaru, *Eur. J. Pharmacol*. **1999**, *367*, 267–273.
31. H. Dmytro, Z. Borys, L. Roman, *Phosphorus, Sulfur and Silicon* **2009**, *184*, 638–650.
32. M. E. Voss, P. H. Carter, A. J. Tebben, P. A. Scherle, G. D. Brown, L. A. Thompson, M. Xu, Y. C. Lo, G. Yang, R.-Q. Liu, P. Strzemienski, J. G. Everlof, J. M. Trzaskos, C. P. Decicco, *Bioorg. Med. Chem. Lett*. **2003**, *13*, 533–538.
33. A. Srinivas, A. Nagaraj, C. Sanjeeva Reddy, *J. Heterocycl. Chem*. **2008**, *45*, 999–1003.
34. P. G. Baraldi, M. G. Pavani, M. Nunez, P. Brigidi, B. Vitali, R. Gambari, R. Romagnoli, *Pharm. Chem. J*. **2001**, *35*, 602–604
35. K. Y. Lee, J. M. Kim, J. N. Kim, *Tetrahedron Lett*. **2003**, *44*, 6737–6740.
36. E. Palaska, M. Aytemir, Ý. T. Uzbay, D. Erol, *Eur. J. Med. Chem*. **2001**, *36*, 539–543.
37. T. Ma, J. R. Thiagarajah, H. Yang, N. D. Sonawane, C. Folli, L. J. V. Galietta, A. S. Verkman, *J. Clin. Invest*. **2002**, *110*, 1651–1658.
38. O. Bruno, F. Bondavalli, A. Ranise, P. Schenone, C. Losasso, L. Cilenti, C. Matera, E. Marmo, *Il Farmaco* **1990**, *45*, 147–166.
39. R. A. Nugent, M. Murphy, S. T. Schlachter, C. J. Dunn, R. J. Smith, N. D. Staite, L. A. Galinet, S. K. Shields, D. G. Aspar, K. A. Richard, N. A. Rohloff, *J. Med. Chem*. **1993**, *36*, 134–139.
40. K. Hiroshi, T. Yasuhiro, S. Yoshio, S. Fumiki, O. Norio, T. Akira, *Jpn. J. Pharmacol*. **1997**, *73*, 317–324.
41. M. J. Genin, C. Biles, B. J. Keiser, S. M. Poppe, S. M. Swaney, W. G. Tarpley, Y. Yagi, D. L. Romero, *J. Med. Chem*. **2000**, *43*, 1034–1040.
42. A. G. Habeeb, P. N. P. Rao, E. E. Knaus, *J. Med. Chem*. **2001**, *44*, 3039–3042.
43. H. Hashimoto, K. Imamura, J.-i. Haruta, K. Wakitani, *J. Med. Chem*. **2002**, *45*, 1511–1517.
44. D. L. Selwood, D. G. Brummell, J. Budworth, G. E. Burtin, R. O. Campbell, S. S. Chana, I. G. Charles, P. A. Fernandez, R. C. Glen, M. C. Goggin, A. J. Hobbs, M. R. Kling, Q. Liu, D. J. Madge, S. Meillerais, K. L. Powell, K. Reynolds, G. D. Spacey, J. N. Stables, M. A. Tatlock, K. A. Wheeler, G. Wishart, C.-K. Woo, *J. Med. Chem*. **2001**, *44*, 78–93.
45. C. Sanjeeva Reddy, L. Sanjeeva Rao, A. Nagaraj, *Acta Chim. Slov*. **2010**, *57*, 726–732
46. A. Srinivas, A. Nagaraj, C. Sanjeeva Reddy, *Eur. J. Med. Chem*. **2010**, *45*, 2353–2358.
47. C. Sanjeeva Reddy, A. Srinivas, A. Nagaraj, *Chem. Pharm. Bull*. **2009**, 57, 685–693.
48. A. Srinivas, A. Nagaraj, C. Sanjeeva Reddy, *J. Heterocycl. Chem.* **2009**, *46*, 497–502.

49. A. Nagaraj, C. Sanjeeva Reddy, *J. Iran. Chem. Soc.* **2008**, *5*, 262–267.
50. M. Raghu, A. Nagaraj, C. Sanjeeva Reddy, *J. Heterocycl. Chem.* **2009**, *46*, 261–267.
51. C. Sanjeeva Reddy, G. P. Reddy, A. Nagaraj, A. Srinivas, *Org. Commun*. **2008**, *1*, 84–94.
52. E. Clemmensen, A. H. C. Heitman, *J. Am. Chem. Soc.* **1911**, *33*, 733–745.
53. C. W. McBeth, G. B. Bergeson, *Phytopathology* **1953**, *43*, 264–270.
54. National Committee for Clinical Laboratory Standards (NCCLS). Standard methods for dilution antimicrobial susceptibility tests for bacteria, which grows aerobically. *Nat. Comm. Lab. Stands*. Villanova, **1982**, pp. 242.

Povzetek

Pripravili smo novo serijo bis(pirazolo[3,4-*d*][1,3]tiazolov) **7a**–**j** ter spojine karakterizirali z IR, ^{1}H NMR, ^{13}C NMR, MS in elementnimi analizami. Vsem novim spojinam **7a**–**j** smo določili nematicidno aktivnost proti *Ditylenchus myceliophagus* in *Caenorhabditis elegans* z *in vitro* testiranjem v vodi. Rezultati kažejo, da je najbolj učinkovita spojina **7e** z LD_{50} vrednostmi proti *D. myceliophagus* in *C. elegans* 160 oz. 180 ppm in je torej skoraj enako aktivna kot standardni levamizol. Tudi spojini **7h** in **7j** sta zelo aktivni: LD_{50} vrednost proti *C. elegans* je 190 ppm in proti *D. myceliophagus* 180 ppm. Spojine **7a**–**j** smo testirali tudi za morebitno antibakterijsko aktivnost. Večina spojin je pokazala močno aktivnost proti testiranim bakterijam in s tem se tudi odpirajo možnosti za nadaljnje modifikacije pripravljenih molekul.