Design and Synthesis of Novel Methylene-bis-fused Pyrazoles as Biologically Active Molecules

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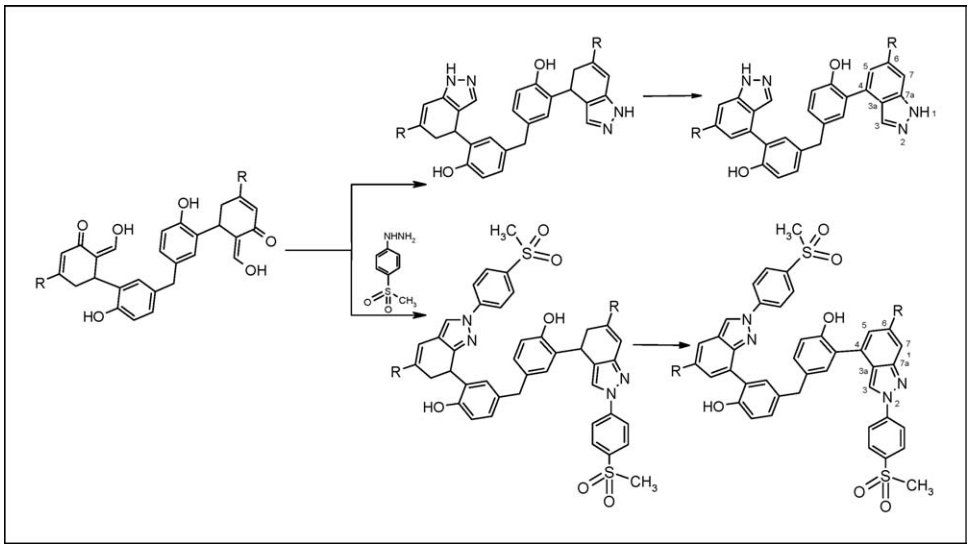

A series of novel methylene-bis-fused pyrazoles **12a–e** and methylene-bis-2-(4-methylsulfonyl)-phenyl substituted fused pyrazoles **15a–d** have been synthesized by the reaction of methylene-bis-aryl-6-hydroxymethylene-2-cyclohexenones **10** with hydrazine hydrate or (4-methylsulfonyl)-phenyl hydrazine **13**. Chemical structures of all the newly synthesized compounds were elucidated by their IR, ^{1}H NMR, ^{13}C NMR, and MS spectral data. The compounds **15a–d** were evaluated for their cyclooxygenase-2 (COX-2) inhibitory activity, and the compound **15b** showed appreciable COX-2 inhibition and selectivity. Further, all the new compounds were screened for their antimicrobial activity against Gram-positive, Gram-negative bacteria, and fungi. Amongst the screened compounds, **12c**, **15a**, and **15b** were found to be the most active against almost all the test bacteria. The compound **15b** displayed notable antibacterial activity against *Bacillus subtilis* (ATCC 6633), *Staphylococcus aureus* (ATCC 6538p), *Micrococcus luteus* (IFC 12708), *Proteus vulgaris* (ATCC 3851), and *Salmonella typhimurium* (ATCC 14028), equal to that of ampicillin. Similarly, these compounds also showed potent antifungal activity against *Candida albicans* (ATCC 10231), *Aspergillus fumigatus* (HIC 6094), *Trichophyton rubrum* (IFO 9185), and *Trichophyton mentagrophytes* (IFO 40996).

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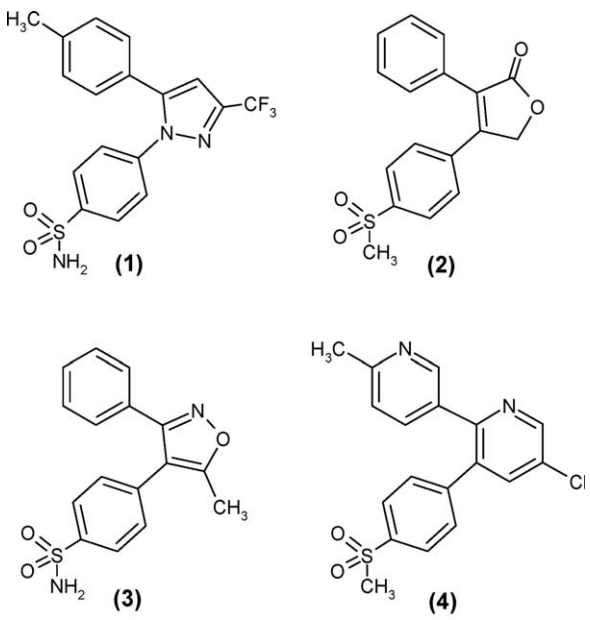
INTRODUCTION

Pyrazole and its derivatives could be considered as possible antimicrobial agents [1,2]. The other activities include antidepressant [3], inhibitors of protein kinases [4], antiagreegating [5], antiarthritic [6], and cerebroprotectors [7]. Recently some aryl pyrazoles were reported to have non-nucleoside HIV-1 reverse transcriptase inhibitors [8], cyclooxygenase-2 (COX-2) inhibitors [9], potent activator of the nitric oxide receptor, and soluble guanylate cyclase [10] activity. Besides, great interest in the pyrazole molecule has been stimulated by some promising pharmacological, agrochemical, and analytical applications of its derivatives [11].

The role of COX-2 isoform in inflammation and the attractiveness of COX-2 as a therapeutic target for the development of anti-inflammatory drugs are very well recognized [12]. The traditional nonsteroidal anti-inflammatory drugs (NSAIDs) are nonselectively inhibit both COX-1 and COX-2, and hence, downregulate prostaglandin formation in almost all cells and tissues, which may induce gastrointestinal side effect, adversely affect the mucus-bicarbonate secretion, acid secretion, and mucosal blood flow. COX-1 inhibition may also elicit an

Scheme 1. COX-2 selective inhibitors.



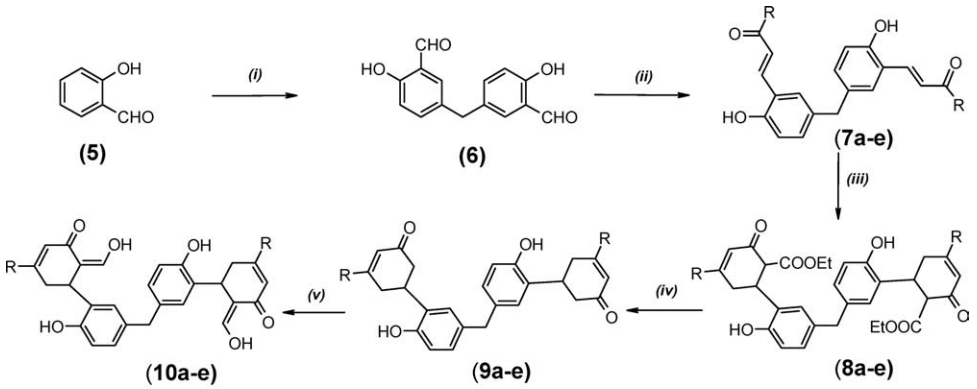
increase in 5-lipoxygenase (5-LO) activity that would potentiate production of leukotriene-B_4 (LTB_4) and vasoconstrictor peptide-leukotrienes (p-LTs) by the lipoxygenase pathway, and this may also contribute to the vascular and other mucosal damage by NSAIDs [13]. Hence, it was proposed that a selective inhibitor of COX-2 would be an attractive approach to the treatment of inflammatory conditions, without concomitant gastric and renal side effects. There are four COX-2 selective inhibitors, such as celecoxib (1), rofecoxib (2), valdecoxib (3), and etoricoxib (4) (Scheme 1), which are currently prescribed for the treatment of arthritis and inflammatory diseases. They show anti-inflammatory activity with reduced gastro intestinal side effects to traditional NSAIDs. However, the long term use of both traditional NSAIDs and coxibs has been reported to cause significant cardiovascular side effects [14].

Following the successful introduction of antimicrobial agents and COX-2 inhibitors, in continuation of our research on biologically active heterocycles [15–18] it was considered worthwhile to design and synthesize more selective COX-2 analogs, incorporating two active pharmacophores would enhance further the activity and selectivity towards the COX-2 enzyme. In this article, we explored fused pyrazoles **12a–e** and 2-substituted fused pyrazoles **15a–d** in which central five-membered scaffold is similar to that of celecosib 1, and 4-(methylsulfonyl) benzene, used as a COX-2 pharmacophore, and evaluated their *in vitro* antimicrobial and COX-2 inhibitory activity.

RESULTS AND DISCUSSION

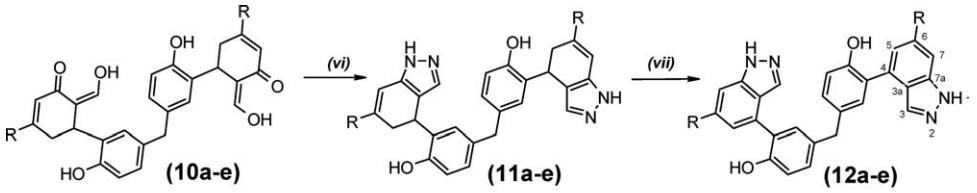
Synthesis. The key intermediate, **10** required for the synthesis of title compounds was prepared according to the procedure outlined in the Scheme 2. Condensation of commercially available salicylaldehyde **5** and trioxane in the presence of a mixture of conc. sulfuric acid and acetic acid gave methylene-bis-salicylaldehyde **6** in good yield [19]. Compound **6** was then reacted with the corresponding acetophenone in presence of alc. KOH at room temperature to give methylene-bis-chalcones **7a–e** (yield over 90%) [20]. Knoevenagel condensation of compounds **7a–e** with ethyl acetoacetate gave methylene-bis-aryl-6-carbethoxycyclohexenones **8a–e** (yield over 80%). Decarboxylation of **8a–e** in the presence of HCl/AcOH at reflux temperature resulted methylene-bis-aryl-cyclohexenones **9a–e** (yield over 80%), which on Claisen-like condensation with ethylformate in the

Scheme 2. Reagents and conditions: (i) trioxane, H_2SO_4/AcOH, reflux, 81%; (ii) $RCOCH_3$, KOH/EtOH, rt, 82–95%; (iii) EAA, NaOEt/EtOH, reflux, 78–86%; (iv) HCl/AcOH, reflux, 74–82%; (v) HCOOEt, NaOMe/C_6H_6, rt, 79–88%.



R = **a)** C_6H_5; **b)** 4-Br-C_6H_4; **c)** 4-NH_2-C_6H_4; **d)** 4-Cl-C_6H_4; **e)** 4-MeO-C_6H_4

Scheme 3. Reagents and conditions: (vi) hydrazinehydrate/AcOH, reflux, 80%; (vii) DDQ, N_2-atm, reflux, 76–83%.



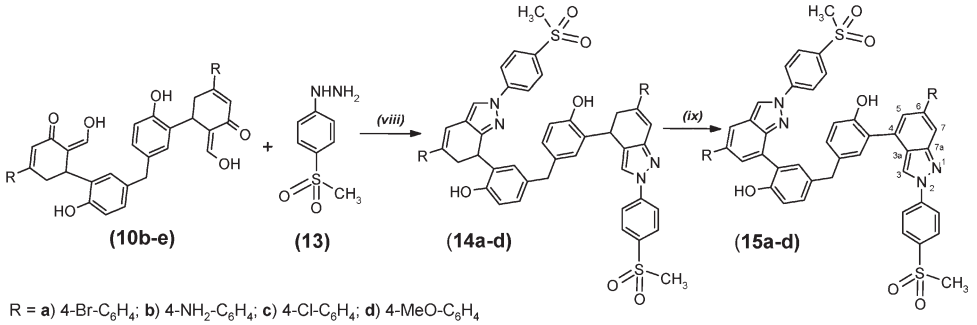
R = **a**) C_6H_5; **b**) 4-Br-C_6H_4; **c**) 4-NH_2-C_6H_4; **d**) 4-Cl-C_6H_4; **e**) 4-MeO-C_6H_4

presence of sodium methoxide at room temperature afforded methylene-bis-aryl-6-hydroxy-methylene-2-cyclohexenones **10a–e** in good yields.

The compounds **10a–e** on cyclocondensation with the hydrazine hydrate in refluxing acetic acid resulted dihydropyrazole derivatives **11a–e** in good to excellent yields (yield over 80%). Subsequent aromatization of **11a–e**, with dichlorodicyanoparabenzoquinone (DDQ) under N_2-atmosphere at reflux temperature gave fused pyrazoles **12a–e** in good yields (Scheme 3). The chemical structure of the compounds was elucidated by their IR, ^{1}H NMR, ^{13}C NMR, and MS spectral data. In the IR spectra of compounds **12a–e**, C═N and N─H bands were observed in the regions 1560–1585, 3390–3410 cm^{-1} respectively. According to the IR spectral data, the compounds **12a–e** have pyrazole structure. In the ^{1}H NMR spectra of **12a–e**, the absence of signals corresponding to methine and methylene protons of cyclohexadiene ring indicates that aromatization has indeed taken place. The ─NH proton of the pyrazole ring was observed as a broad singlet at about 8.87–8.80 ppm. The signal because of the methylene bridge proton, present in all compounds, appeared at 4.06–4.00 ppm as singlet. The N═CH proton of compounds **12a–e** appeared at 7.95–7.90 ppm as singlet. All the other aromatic and aliphatic protons of compounds **12a–e** were observed at the expected regions. In the ^{13}C NMR spectra of compounds **12a–e**, the prominent signals corresponding to C-3, C-3a, C-4, and C-7a, for all the compounds, observed nearly at 137.2, 137.4, 155.6, 145.3 ppm, respectively.

Further, the intermediate **10** when treated with the commercially available 4-(methylsulfonyl)-phenylhydrazine **13** in refluxing ethanol gave methylsulfonyl derivative of pyrazole **14a–d** in good yield. This reaction is a regioselective transformation and the 2-substituted pyrazole could be generated almost exclusively by carrying out the condensation in the presence of hydrochloric acid and one equivalent of 4-(methyl- sulfonyl)phenyl hydrazine **13**. The compounds **14a–d** when aromatized with DDQ under N_2-atmosphere at reflux temperature gave 4-(methylsulfonyl)-phenyl substituted fused pyrazoles **15a–d** in good yields (Scheme 4). The chemical structures of all the synthesized compounds were confirmed by their IR, ^{1}H NMR, ^{13}C NMR, and MS spectral data. In the IR spectra of compounds **15a–d**, C═N band was observed in the region 1560–1585 cm^{-1} and SO_2 group symmetric, asymmetric stretching bands at about 1300–1328, 1155–1165 cm^{-1}. In the ^{1}H NMR spectrum of compounds **15a–d**, absence of the signal corresponding to NH group proved that these compounds have pyrazole nucleus with (4-methyl- sulfonyl)-phenyl group at nitrogen. Further, the ─CH_3 proton signal was seen as singlet at about 2.96–2.90 ppm, and the signal of N═CH proton of compounds **15a–d** appeared at 6.68–6.65 ppm proved that these compounds have pyrazole nucleus with (4-methylsulfonyl)phenyl group at nitrogen. All the other aromatic and aliphatic protons of

Scheme 4. Reagents and conditions: (viii) HCl/EtOH, reflux, 78–86%; (ix) DDQ/dry C_6H_6, N_2-atm, reflux, 79–86%.



R = **a**) 4-Br-C_6H_4; **b**) 4-NH_2-C_6H_4; **c**) 4-Cl-C_6H_4; **d**) 4-MeO-C_6H_4

Table 1

Antibacterial activity of compounds **12a–e** and **15a–d**.

Compound	Minimum inhibitory concentration (MIC, μg/mL)					
	B. subtilis	*S. aureus*	*M. luteus*	*P. vulgaris*	*S. typhimurium*	*E. coli*
12a	—	—	—	—	—	—
12b	3.12	6.25	3.12	6.25	3.12	—
12c	3.12	3.12	1.56	1.56	3.12	25.0
12d	25.0	—	25.0	—	—	—
12e	12.5	12.5	12.5	12.5	12.5	—
15a	3.12	3.12	3.12	3.12	6.25	—
15b	1.56	1.56	1.56	1.56	1.56	25.0
15c	12.5	12.5	12.5	12.5	12.5	—
15d	12.5	6.25	6.25	6.25	6.25	—
Ampicillin	1.56	1.56	1.56	3.12	3.12	12.5

—, Indicates bacteria are resistant to the compound >50 μg/mL concentration.

compounds **15a–d** were observed at the expected regions. In the ^{13}C NMR spectra of compounds **15a–d**, the prominent signals corresponding to C-3, C-3a, C-4, and C-7a, for all the compounds, observed nearly at 125.4, 124.0, 136.0, 153.1 ppm, respectively, provide further evidence for their structures. Mass spectra of all the synthesized compounds showed $M^+/M^+ + 1$ peaks, in agreement with their molecular formulae.

Antibacterial activity. The *in vitro* antibacterial activity of the newly prepared compounds, **12a–e** and **15a–d**, was assayed against gram-positive bacteria *viz. Bacillus subtilis* (ATCC 6633), *Staphylococcus aureus* (ATCC 6538p), and *Micrococcus luteus* (IFC 12708), 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 standards (NCCLS) [21]. Amphicillin was used as a standard drug, the lowest concentration (highest dilution) required to arrest the growth of bacteria was regarded as minimum inhibitory concentration (MIC, μg/mL), was determined and compared with the controls, the MIC values of the compounds assayed are presented in Table 1.

Investigation of the antibacterial screening data revealed that all the tested compounds exhibit interesting biological activity, however, with a degree of variation. Compound **15b** is highly active against all the microorganisms used (accept *E. coli*) at 1.56 μg/mL concentration, and is almost equal to the standard. Compound **12c** is also highly active against *M. luteus* and *P. vulgaris* only at the same concentration as **15b**. The compound **15a** also showed good antibacterial activity against *B. subtilis*, *S. aureus*, *M. luteus,* and *P. vulgaris*. Compound **12a** is almost inactive towards all the microorganisms used. Other compounds were also inactive towards *E. coli* bacteria. The remaining compounds showed moderate to good activity.

Antifungal activity. The newly prepared compounds were screened for their antifungal activity against *Candida albicans* (ATCC 10231), *Aspergillus fumigatus* (HIC 6094), *Trichophyton rubrum* (IFO 9185), and *Trichophyton mentagrophytes* (IFO 40996). The antifungal activity of each compound was compared with standard drug Amphotericin B. MIC (μg/mL) was determined and compared with controls; the MIC values of the compounds screened are given in Table 2. The antifungal screening data showed only moderate activity of the test compounds. Among the screened compounds, only **15b** showed the highest activity against all the microorganisms used. Similarly compound **12c** is also highly active but only against *T. rubrum* and *T. mentagrophytes*. The activities of these two compounds are almost equal to

Table 2

Antifungal activity of compounds **12a–e** and **15a–d**.

Compound	Minimum inhibitory concentration (MIC, μg/mL)			
	C. albicans	*A. fumigatus*	*T. rubrum*	*T. mentagrophytes*
12a	—	—	—	25.0
12b	12.5	25.0	12.5	25.0
12c	12.5	12.5	6.25	6.25
12d	—	—	50.0	25.0
12e	50.0	25.0	50.0	25.0
15a	12.5	12.5	12.5	12.5
15b	6.25	6.25	6.25	6.25
15c	50.0	50.0	25.0	25.0
15d	25.0	25.0	12.5	25.0
Amphotericin B	6.25	3.12	3.12	3.12

—, Indicates fungi are resistant to the compound >50 μg/mL concentration.

the standard, the remaining compounds showed moderate to good activity.

COX-2 inhibitory activity. The compounds synthesized **15a–d** was evaluated as COX inhibitors in human whole blood (HWB). Among the four compounds, **15b** showed good inhibition and selectivity. Some interesting features can be deduced from the comparison of the structure of compound **15b** with that of celecoxib **1**. (i) The sulfonamide group present in celecoxib is replaced by methyl sulfonyl group, believed to be crucial for increasing the COX-2 selectivity. (ii) The 4-methylphenyl and the trifluoromethyl groups on pyrazole ring are replaced and is fused with 1,3-diarylbenzene. (iii) Two similar pharmacophores were introduced in a single molecular frame work, linked by a methylene bridge. We evaluated the compound **15b** for COX inhibition in the HWB assay, performed essentially as described [22]. The 2-(4-methylsulfonyl)phenyl-substituted compound **15b** showed 69.51% of COX-2 inhibition at 3 μM in HWB assay, compared with 90.90% inhibition observed for celecoxib **1** at the same concentration. The compound **15b** showed only moderate COX-1 inhibition at a very high concentration (50 μM) for about 21.02%.

In conclusion, a series of novel methylene-bis-fused pyrazoles **12a–e** and methylene-bis-2(4-methylsulfonyl)-phenyl substituted fused pyrazoles **15a–d** have been designed and synthesized. The antimicrobial activity of these compounds was evaluated against various Gram-positive, Gram-negative bacteria, and fungi. Among the synthesized compounds, **12c**, **15a**, and **15b** showed good activity against bacteria and fungi and emerged as potential molecules for further development. The compounds were also evaluated for their COX-2 selective inhibition, **15b** showed an appreciable COX-2 inhibition and selectivity. With this set of analogs, we are now in a position to investigate the multiple biological activities for these compounds.

EXPERIMENTAL

Research chemicals were either purchased from Aldrich Company or Fluka or used without further purification in the reactions, or were prepared according to procedures described in the literature. Reactions were monitored by thin layer chromatography on silica gel plated (60 F_{254}; Merck) visualizing with ultraviolet light or iodine. Column chromatography was performed on silica gel 60 (0.043–0.060 mm), Merck. The reported yields of the products are unoptimized. Melting points were determined with a Fisher-Johns apparatus and are uncorrected. IR spectra were recorded on a Perkin-Elmer FTIR 5000 spectrometer, using KBr pellet. ^{1}H NMR, ^{13}C NMR spectra were recorded on a Varian Gemini spectrometer, operating at 300, 75 MHz, respectively. Chemical shifts (δ) are reported as parts per million downfield from tetramethyl silane. Mass spectra were obtained on a VG micromass 7070H spectrometer.

Ethyl-6-(5-{3-[6-(ethoxycarbonyl)-5-oxo-3-phenyl-3-cyclohexenyl]-4-hydroxybenzyl}-2-hydroxyphenyl)-2-oxo-4-phenyl- 3-cyclohexene-1-carboxylate (8a). In a solution sodium metal (2 g) in ethanol (30 mL), a mixture of freshly distilled ethylacetoacetate (3.9 mL, 0.03 mol) and compound **7a** (4.6 g, 0.01 mol) dissolved in ethanol (20 mL) was added. The resulting solution was refluxed on a water bath for 4 h. Allowing the reaction mixture to cool and crystallization of the ppt. from ethanol to give **8a** as brown solid; Yield 82%; m.p. 150–52°C; IR (KBr): ν 3452, 3065, 1702, 1695, 1597, 1245 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ 7.10–7.14 (m, 10 H, ArH), 6.80 (s, 2H, ArH), 6.79 (d, J = 9.2 Hz, 2H, ArH), 6.62 (d, J = 9.2 Hz, 2H, ArH), 6.10 (s, 2H, CH), 5.20 (s, 2H, OH), 4.06 (q, 4H, CH_2), 3.87 (q, 2H, CH), 3.81 (d, 2H, CH), 3.72 (s, 2H, CH_2), 2.87 (d, 4H, CH_2), 1.10 (t, 6H, CH_3); ^{13}C NMR (DMSO-d_6): δ 190.1, 176.7, 154.6, 149.5, 142.3, 133.4, 130.0, 128.7, 128.2, 127.9, 125.5, 123.8, 121.9, 117.4, 61.2, 60.6, 42.1, 37.0, 30.7, 17.0. MS: m/z 685 (M^+ + 1). The other compounds **8b–e** were prepared by the similar procedure.

5-{2-Hydroxy-5-[4-hydroxy-3-(5-oxo-3-phenyl-3-cyclohexenyl)benzyl]phenyl-3-phenyl}-2-cyclohexen-1-one (9a). To a mixture of glacial acetic acid (100 mL) and conc. HCl (50 mL) was added compound **8a** (6.5 g, 0.01 mol) in portions. The mixture was heated to reflux for 10 h. After cooling to room temperature, the reaction mixture was concentrated *in vacuo*. The residue was taken up with ethyl acetate and washed with water and brine, dried over $MgSO_4$, filtered, and evaporated in *vacuo* to give oil, which soon solidified; it was purified by recrystallization from ethanol to give the compound **9a** as brown solid; Yield 79%; m.p. 132–34°C; IR (KBr): ν 3357, 3025, 2932, 1687, 1596 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ 7.10–7.14 (m, 10H, ArH), 7.00 (d, J = 9.2 Hz, 2H, ArH), 6.82 (s, 2H, ArH), 6.62 (d, J = 9.2 Hz, 2H, ArH), 6.10 (s, 2H, CH), 5.20 (s, 2H, OH), 3.83–3.87 (m, 2H, CH), 3.72 (s, 2H, CH_2), 2.70 (d, 4H, CH_2), 2.67 (d, 4H, CH_2); ^{13}C NMR (DMSO-d_6): δ 190.0, 155.2, 150.1, 140.5, 132.3, 131.2, 128.5, 127.8, 127.4, 126.2, 124.5, 122.6, 116.1, 47.5, 42.0, 38.9, 31.0; MS: m/z 541 (M^+ + 1). The other compounds **9b–e** were prepared by the similar procedure.

5-[2-Hydroxy-5-(4-hydroxy-3-{6-[(*Z*)-1-hydroxymethylidene]-5-oxo-3-phenyl-3-cyclohexenyl}benzyl)phenyl]-6-[(*Z*)-1-hydroxymethylidene]-3-phenyl-2-cyclohexen-1-one (10a). In a solution of 10% sodium methoxide (10 mL) in benzene (25 mL), ethylformate (2.24 mL, 0.03 mol) was added and afterward over 30 min, compound **9a** (5.4 g, 0.01 mol) dissolved in benzene (10 mL) was added. The resulting solution was stirred for 10 h at room temperature and allowed to stand over night, then evaporated to dryness. The suspension obtained was mixed with cold water and acidified with dil HCl (20 mL) and extracted three times with ether (40 mL). The organic layer was dried over $MgSO_4$ and evaporated *in vacuo* to give solid, was purified by crystallization in ethanol to afford pure **10a** as yellow solid; Yield 81%; m.p. 143–45°C; IR (KBr): ν 3320, 3028, 2952, 1662, 1620, 1597 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ 8.97 (s, 2H, OH), 7.92 (s, 2H, CH), 7.14–7.10 (m, 10H, ArH), 6.80 (s, 2H, ArH), 6.49–6.40 (m, 4H, ArH), 5.67 (s, 2H, CH), 4.12 (t, 2H, CH), 3.72 (s, 2H, CH_2), 3.22 (d, 4H, CH_2); ^{13}C NMR (DMSO-d_6): δ 191.3, 167.6, 156.1, 149.7, 142.0, 131.1, 130.6, 130.2, 128.9, 128.0, 127.9, 127.6, 126.2, 116.7, 115.5, 44.1, 42.0, 37.8; MS: m/z 596 (M^+). The other compounds **10b–e** were prepared by the similar procedure.

4-[4-Hydroxy-3-(6-phenyl-4,5-dihydro-1*H*-4-indazolyl)benzyl]-2-(6-phenyl-4,5-dihydro-1*H*-4-indazolyl)phenol (11a). To a solution of **10a** (5.9 g, 0.01 mol) in glacial acetic acid (50 mL), hydrazine hydrate (1.5 g, 0.03 mol) was added. After stirring at 80°C for 10 h, the mixture was concentrated *in vacuo*. To the residue was added water and twice extracted with ether, washed the organic layer with saturated $NaHCO_3$ solution, subsequently with water and brine, dried over $MgSO_4$, and evaporated to dryness. The residue could be recrystallized from ethanol to afford **11a** as brown solid; Yield 79%; m.p. 123–25°C; IR (KBr): ν 3390, 3037, 2972, 1595, 1585 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.12 (bs, 2H, NH), 7.65 (s, 2H, ArH), 7.63 (s, 2H, ArH), 7.32 (d, J = 9.2 Hz, 4H, ArH), 7.21 (s, 2H, ArH), 6.99–7.05 (m, 6H, ArH), 6.84 (d, J = 9.1 Hz, 2H, ArH), 6.73 (d, J = 9.0 Hz, ArH), 6.62 (s, 2H, OH), 4.92 (t, 2H, CH), 3.72 (s, 2H, CH_2), 2.92 (d, 4H, CH_2); ^{13}C NMR (DMSO-d_6): δ 156.2, 148.7, 143.4, 140.3, 132.8, 132.0, 130.9, 130.0, 128.9, 127.8, 126.9, 126.5, 125.4, 118.7, 117.2, 42.1, 39.3, 38.1; MS: m/z 589 (M^+ + 1). The other compounds **11b–e** were prepared by the similar procedure.

4-[4-Hydroxy-3-(6-aryl-1*H*-4-indazolyl)benzyl]-2(6-aryl-1*H*-4-indazolyl)phenol (12a–e). To a solution of corresponding compound **11** (0.01 mol) in dry benzene (20 mL), DDQ (0.03 mol) dissolved in dry benzene (20 mL) was added in portions. The mixture was heated to reflux and stirred for 5 h under a nitrogen atmosphere. The precipitated DDQ-H_2 was filtered off and the filtrate was subjected to column chromatography on silica gel (60–120 mesh) to afford pure compounds.

4-[4-Hydroxy-3-(6-phenyl-1*H*-4-indazolyl)benzyl]-2(6-phenyl-1*H*-4-indazolyl)phenol (12a). This was obtained as yellow solid; Yield 83%; m.p. 137–39°C; IR (KBr): ν 3392, 3062, 2985, 1584 cm^{-1}. ^1H NMR (DMSO-d_6): δ 8.87 (bs, 2H, NH), 7.47 (s, 2H, ArH), 7.32–7.26 (m, 8H, ArH) 6.94 (d, J = 9.0 Hz, 2H, ArH) 4.05 (s, 2H, CH_2); ^{13}C NMR (DMSO-d_6): δ 159.0, 155.6, 147.9, 146.1, 145.3, 141.7, 139.0, 137.2, 136.5, 134.2, 128.8, 126.9, 126.5, 125.4, 123.8, 118.2, 112.7, 42.1; MS: m/z 585 (M^+ + 1).

2-[6-(4-Bromophenyl)-1*H*-4-indazolyl]-4-3-[6-(4-bromophenyl)-1*H*-4-indazolyl]-4-hydroxybenzylphenol (12b). This was obtained as brown solid; Yield 82%; m.p. 142–44°C; IR (KBr): ν 3400, 3037, 2962, 1585, 712 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.86 (bs, 2H, NH), 8.15 (s, 2H, ArH), 8.10 (s, 2H, ArH), 7.94 (s, 2H, ArH), 7.47 (s, 2H, ArH), 7.41 (d, J = 8.3 Hz, 4H, ArH), 7.32–7.26 (m, 6H, ArH), 6.94 (d, J = 9.0 Hz, 2H, ArH), 4.02 (s, 2H, CH_2); ^{13}C NMR (DMSO-d_6): δ 159.0, 155.6, 149.3, 146.1, 141.7, 139.1, 137.4, 137.1, 136.5, 134.3, 131.2, 129.4, 125.4, 123.9, 118.3, 112.7, 42.1; MS: m/z 743 (M^+ + 1).

2-[6-(4-Aminophenyl)-1*H*-4-indazolyl]-4-3-[6-(4-aminophenyl)-1*H*-4-indazolyl]-4-hydroxybenylphenol (12c). This was obtained as orange solid; Yield 76%; m.p. 151–53°C; IR (KBr): ν 3395, 3061, 2937, 1585 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.84 (bs, 2H, NH), 8.15 (s, 2H, ArH), 8.10 (s, 2H, ArH), 7.94 (s, 2H, ArH), 7.62 (d, J = 8.4 Hz, 4H, ArH), 7.47 (s, 2H, ArH), 7.28 (d, J = 9.0 Hz, 2H, ArH), 6.94 (d, J = 9.0 Hz, 2H, ArH), 6.62 (d, J = 8.4 Hz, 4H, ArH), 6.32 (bs, 4H, NH_2), 4.02 (s, 2H, CH_2); ^{13}C NMR (DMSO-d_6): δ 159.1, 155.6, 149.7, 146.1, 142.4, 141.6, 139.0, 137.2, 136.5, 134.9, 134.2, 130.2, 124.2, 123.8, 118.2, 113.1, 112.7, 42.2; MS: m/z 615 (M^+ + 1).

2-[6-(4-Clorophenyl)-1*H*-4-indazolyl]-4-{3-[6-(4-chlorophenyl)-1*H*-4-indazolyl]-4-hydroxy}benzylphenol (12d). This was obtained as yellow solid; Yield 81%; m.p. 115–17°C; IR (KBr): ν 3387, 3042, 2932, 1585, 686 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.86 (bs, 2H, NH), 8.15 (s, 2H, ArH), 8.10 (s, 2H, ArH), 7.94 (s, 2H, ArH), 7.82 (d, J = 8.2 Hz, 4H, ArH), 7.41 (s, 2H, ArH), 7.32–7.26 (m, 6H, ArH), 7.00 (s, 2H, ArH) 6.94 (d, J = 9.0 Hz, 2H, ArH) 4.02 (s, 2H, CH_2); ^{13}C NMR (DMSO-d_6): δ 158.7, 155.6, 147.4, 146.1, 141.7, 139.0, 137.2, 136.5, 134.9, 134.2, 134.0, 128.7, 126.4, 125.4, 123.8, 118.3, 112.7, 42.2; MS: m/z 654 (M^+).

4-{4-Hydroxy-3-[6-(4-methoxyphenyl)-1*H*-4-indazolyl]benzyl}-2-[6-(4-methoxyphenyl)-1*H*-4-indazolyl]phenol (12e). This was obtained as brown solid; Yield 80%; m.p. 122–24°C; IR (KBr): ν 3400, 3042, 2962, 1584, 1030 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.87 (bs, 2H, NH), 8.14 (s, 2H, ArH), 8.10 (s, 2H, ArH), 7.94 (s, 2H, ArH), 7.47 (s, 2H, ArH), 7.39 (d, J = 8.4 Hz, 4H, ArH), 7.32 (d, J = 9.0 Hz, 2H, ArH), 6.94 (d, J = 9.0 Hz, 2H, ArH), 6.84 (d, J = 8.4 Hz, 4H, ArH), 4.02 (s, 2H, CH_2), 3.84 (s, 6H, OCH_3); ^{13}C NMR (DMSO-d_6): δ 161.2, 159.0, 155.6, 149.5, 146.1, 141.7, 139.2, 137.4, 136.5, 134.2, 133.2, 129.4, 129.0, 125.7, 124.3, 123.8, 118.2, 112.7, 112.0, 54.7, 42.2; MS: m/z 645 (M^+ + 1).

4-{6-(4-Bromophenyl)-4-[5-(3-{6-(4-bromophenyl)-2-[4-(methylsulfonyl)phenyl]-4,5-dihydro-2*H*-4-indazolyl}-4-hydroxy benzyl)-2-hydroxyphenyl]-4,5-dihydro-2*H*-2-indazolyl}-1-benzene sulfonic acid (14a). To a stirred solution of **10b** (7.5 g, 0.01 mol) in ethanol (100 mL) and 6 *N* HCl (14.8 mL, 0.892 mol) was added 4-(methylsulfonyl)phenyl hydrazine **13** (4.1 g, 0.022 mol). The mixture was heated to reflux and stirred for 10 h. After cooling to room temperature, the reaction mixture was concentrated *in vacuo*. The residue was taken up with ethyl acetate and washed with water and brine, dried over $MgSO_4$, filtered, and evaporated *in vacuo* to give a solid that was crystallized from diisopropyl ether (100 mL) to give pyrazole **14a** as brown solid; Yield 82%; m.p. 222–24°C; IR (KBr): ν 3400, 3062, 2937, 1585, 1328, 1165, 732 cm^{-1}; ^1H NMR (DMSO-d_6): δ 7.91 (s, 2H, ArH), 7.67–7.62 (m, 6H, ArH), 7.50–7.42 (m, 12H, ArH), 7.23 (s, 2H, ArH), 6.94 (s, 2H, OH), 6.84 (d, J = 8.9 Hz, 2H, ArH), 6.77 (d, J = 8.9 Hz, 2H, ArH), 4.87 (t, 2H, CH), 3.72 (s, 2H, CH_2), 3.12 (d, 4H, CH_2), 2.94 (s, 6H, CH_3); ^{13}C NMR (DMSO-d_6): δ 156.1, 154.2, 153.5, 142.7, 138.8, 133.5, 132.3, 130.4, 129.9, 129.1, 128.0, 126.8, 126.4, 124.6, 124.2, 122.4, 120.7, 119.4, 118.2, 43.5, 42.3, 41.6, 38.7; MS: m/z 1056 (M^+). The other compounds **14c–d** were also prepared by the similar procedure.

4-{6-(aryl)-4-[5-(3-{6-(aryl)-2-[4-(methylsulfonyl)phenyl]-2*H*-4-indazolyl}-4-hydroxybenzyl)-2-hydroxyphenyl]-2*H*-2-indazolyl}-1-benzenesulfonic acid (15a–d). To a solution of **14** (0.005 mol) in dry benzene (20 mL), DDQ (0.015 mol) in dry benzene (20 mL) was added in portions. The mixture was heated to reflux and stirred for 5 h under a nitrogen atmosphere. The precipitated DDQ-H_2 was filtered off and the filtrate was subjected to column chromatography on silica gel (60–120 mesh), to afford pure compounds.

4-{6-(4-Bromophenyl)-4-[5-(3-{6-(4-bromophenyl)-2-[4-(methylsulfonyl)phenyl]-2*H*-4-indazolyl}-4-hydroxybenzyl)-2-hydroxyphenyl]-2*H*-2-indazolyl}-1-benzenesulfonic acid (15a). This was obtained as brown solid; Yield 80%; m.p. 210–12°C; IR (KBr): ν 3400, 3062, 2937, 1585, 1328, 1165, 736 cm^{-1}. ^1H NMR (DMSO-d_6): δ 8.14 (s, 2H, ArH), 7.80 (s, 2H, ArH), 7.72 (d, J = 8.3 Hz, 4H, ArH), 7.53–7.48 (m, 8H, ArH), 7.32–7.28 (m, 6H, ArH), 6.84–6.78 (m, 4H, ArH, OH), 6.72 (s, 2H, ArH), 4.02 (s, 2H, CH_2), 2.94 (s, 6H, CH_3); ^{13}C NMR

(DMSO-d_6): δ 153.1, 152.0, 143.3, 142.9, 136.7, 136.2, 133.3, 130.7, 129.8, 129.0, 128.7, 127.9, 125.4, 124.0, 123.9, 122.3, 119.7, 117.6, 111.3, 110.4, 44.3, 42.4; MS: *m*/*z* 1050 (M^+).

4-{6-(4-aminophenyl)-4-[5-(3-{6-(4-aminophenyl)-2-[4-(methylsulfonyl)phenyl]-2*H*-4-indazolyl}-4-hydroxybenzyl)-2-hydroxyphenyl]-2*H*-2-indazolyl}-1-benzenesulfonic acid (15b). This was obtained as orange solid; Yield 84%; m.p. 240–42°C; IR (KBr): ν 3400, 3065, 2932, 1589, 1328, 1162 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.14 (s, 2H, ArH), 7.80 (s, 2H, ArH), 7.72 (d, J = 8.2 Hz, 4H, ArH), 7.67 (d, J = 8.6 Hz, 4H, ArH), 7.53 (d, J = 8.2 Hz, 4H, ArH), 7.32 (d, J = 8.6 Hz, 2H, ArH), 6.94 (d, J = 9.1 Hz, 2H, ArH), 6.72 (d, J = 8.6 Hz, 4H, ArH), 5.96 (s, 2H, OH), 4.96 (bs, 4H, NH_2), 4.02 (s, 2H, CH_2), 2.94 (s, 6H, CH_3); ^{13}C NMR (DMSO-d_6): δ 153.1, 152.0, 147.3, 145.2, 143.3, 138.2, 136.7, 130.7, 129.7, 129.0, 126.2, 125.6, 124.0, 123.9, 122.3, 121.0, 119.7, 117.6, 111.9, 110.4, 44.3, 42.4; MS: *m*/*z* 924 ($M^+ + 1$).

4-{6-(4-Chlorophenyl)-4-[5-(3-{6-(4-chlorophenyl)-2-[4-(methylsulfonyl)phenyl]-2*H*-4-indazolyl}-4-hydroxybenzyl)-2-hydroxyphenyl]-2*H*-2-indazolyl}-1-benzenesulfonic acid (15c). This was obtained as yellow solid; Yield 79%; m.p. 196–98°C; IR (KBr): ν 3410, 3065, 2932, 1586, 1328, 1162, 686 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.14 (s, 2H, ArH), 7.95 (d, J = 8.6 Hz, 4H, ArH), 7.80 (s, 2H, ArH), 7.68 (d, J = 8.9 Hz, 4H, ArH), 7.59 (d, J = 8.9 Hz, 4H, ArH), 7.47 (d, J = 8.6 Hz, 4H, ArH), 7.32 (d, J = 9.0 Hz, 2H, ArH), 6.90 (d, J = 9.0 Hz, 2H, ArH), 6.68 (s, 2H, ArH), 5.92 (s, 2H, OH), 4.02 (s, 2H, CH_2), 2.96 (s, 6H, CH_3); ^{13}C NMR (DMSO-d_6): δ 153.1, 152.0, 146.6, 143.3, 136.9, 136.0, 134.9, 133.3, 131.2, 130.0, 129.8, 129.0, 128.6, 125.6, 124.0, 123.9, 122.7, 122.0, 119.7, 117.6, 110.4, 109.7, 44.3, 42.2; MS: *m*/*z* 963 ($M^+ + 1$).

4-[4-[2-Hydroxy-5-(4-hydroxy-3-{6-(4-methoxyphenyl)-2-[4-(methylsulfonyl)phenyl]-2H-4-indazolyl}benzyl)phenyl]-6-(4-methoxyphenyl)-2H-2-indazolyl]-1-benzene sulfonic acid (15d). This was obtained as brown solid; Yield 86%; m.p. 209–11°C; IR (KBr): ν 3400, 3065, 2937, 1589, 1328, 1162 cm^{-1}; ^1H NMR (DMSO-d_6): δ 8.14 (s, 2H, ArH), 7.68 (d, J = 8.1 Hz, 4H, ArH), 7.68 (d, J = 8.9 Hz, 4H, ArH), 7.59 (d, J = 8.9 Hz, 4H, ArH), 7.47 (d, J = 8.1 Hz, 4H, ArH), 7.32 (d, J = 9.0 Hz, 2H, ArH), 6.90 (d, J = 9.0 Hz, 2H, ArH), 6.89 (d, J = 8.1 Hz, 4H, ArH), 6.68 (s, 2H, ArH), 5.96 (s, 2H, OH), 4.02 (s, 2H, CH_2), 3.89 (s, 6H, OCH_3), 2.96 (s, 6H, CH_3); ^{13}C NMR (DMSO-d_6): δ 159.6, 153.9, 153.4, 152.0, 143.3, 136.7, 133.8, 133.1, 130.7, 129.8, 129.0, 125.6, 124.7, 124.0, 123.8, 122.3, 119.7, 117.6, 113.0, 110.9, 110.4, 55.6, 44.3, 42.4; MS: *m*/*z* 955 ($M^+ + 1$).

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