Synthesis, Characterization, and Antimicrobial Studies of New Rigid Linker-Based Bispyrazolines

Mohamad Yusuf,* Varsha Samdhian, and Payal Jain

Department of Chemistry, Punjabi University, Patiala-147002, Punjab, India
*E-mail: yusuf_sah04@yahoo.co.in

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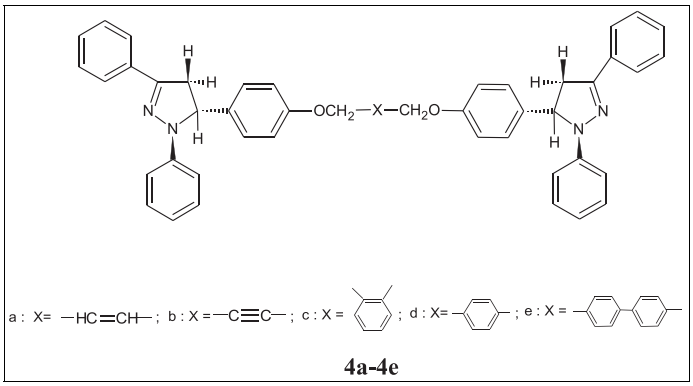

4a-4e

The bispyrazolines **4a–4e** were synthesized from the cyclization reaction of bischalcones **3a–3e** with phenyl hydrazine by refluxing under alcoholic medium in the presence of glacial acetic acid. The bischalcones were obtained from the Claisen–Schmidt reaction of acetophenone with dibenzaldehydes **1a–1e** and later were obtained in good yield from the O-alkylation of 4-hydroxybenzaldehyde with suitable alkylating agents. The structures of the prepared compounds were determined from the rigorous analysis of their spectral data (UV–vis, IR, ^{1}H-NMR, ^{13}C-NMR, and ESI-MS). The elemental analysis also confirmed the purity of these compounds. All the bischalcones **3a–3e** and bispyrazolines **4a–4e** were screened for their antimicrobial activity using the serial dilution method. Seven bacterial and five fungal species were used as the antimicrobial test strains, namely *Klebsiella pneumoniae* (MTCC 3384), *Pseudomonas aeruginosa* (MTCC 424), *Escherichia coli* (MTCC 443), *Staphylococcus aureus* (MTCC 443), *Bacillus subtilis* (MTCC 441), *Pseudomonas fluorescens* (MTCC 103), and *Staphylococcus pyrogens* (MTCC 442), and *Aspergillus janus* (MTCC 2751), *Penicillium glabrum* (MTCC 4951), *Fusarium oxysporum* (MTCC 2480), *Aspergillus sclerotiorum* (MTCC 1008), and *Aspergillus niger* (MTCC 281), respectively. The minimum inhibitory concentrations (MIC in μg/mL) were determined by using different dilutions.

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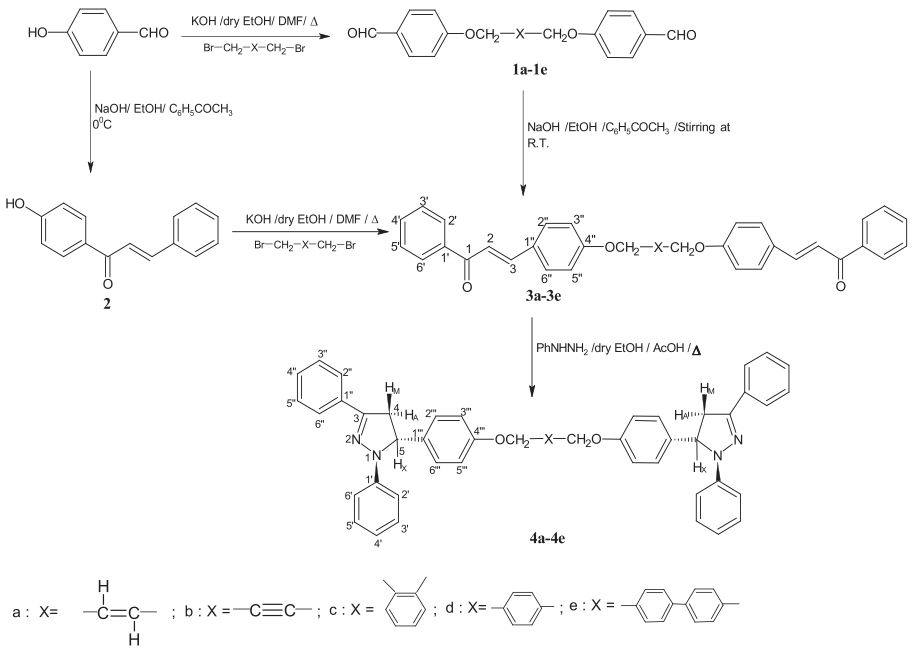
INTRODUCTION

The chalcones are the significant class of the flavanoid derivatives that are associated with wide spectrum of biological applications. The availability of prop-2-ene-1-one moiety in the chalcones is responsible for their biological activities [1–3]. The chalcones are the very important synthones in the organic synthesis, and with proper planning and designing, these molecules can be used for the preparation of various heterocyclic compounds [4–7]. The cyclocondesation reaction of chalcones with hydrazine and their derivatives lead to the formation of pyrazolines. Pyrazolines are the important nitrogen-containing five-membered heterocyclic compounds, and their derivatives have been found to possess a broad spectrum of biological activities such as tranquilizing, muscle relaxant, psychoanaleptic, anticonvulsant and antihypertensive, and antidepressant activities [8–13]. Antibacterial, antifungal, and anti-inflammatory activities [14,15] of pyrazolines and their derivatives have given much needed impetus to their synthesis. Pyrazole moiety is also found in drugs such as celecobix [16], sildenefil [17], and rimonabant [18]. Bispyrazolines are the molecules that are formed by joining two pyrazoline moieties through the carbon chains of varying lengths and structures. By keeping this aspect in view and in continuation to our study on the bishetrocyclics [19], present researches have been focused on the synthesis of bispyrazolines built around five rigid linkers (*trans*-2-butene, 2-butyne, *o*-xylene, *p*-xylene, and 4,4′-bis-methyl-diphenyl).

RESULTS AND DISCUSSION

The bispyrazolines **4a–4e** in the present study were synthesized from the cyclization reaction of bischalcones **3a–3e** with phenyl hydrazine by refluxing under alcoholic medium in the presence of glacial acetic acid (Scheme 1). The bischalcones were obtained from the Claisen–Schmidt reaction of acetophenone with dibenzaldehydes **1a–1e** and later were obtained in good yield from the O-alkylation of 4-hydroxybenzaldehyde with suitable alkylating agents (*trans*-1,4-dibromo-2-butene, 1,4-dichloro-2-butyne,

Scheme 1



α,α′-dibromo-*o*-xylene, α,α′-dibromo-*p*-xylene and 4, 4′-bis-chloromethyldiphenyl). The structures of the prepared compounds were determined from the rigorous analysis of their spectral data (UV–vis, IR, ^{1}H-NMR, ^{13}C-NMR, and ESI-MS). The elemental analysis also confirmed the purity of these compounds.

The compounds **1a–1e** were reacted with acetophenone in the presence of NaOH/EtOH at room temperature, and the reaction mixture was acidified using HCl furnishing the respective solid product. Individual crude product was subjected to crystallization in a mixture of $CHCl_3$/MeOH (1:1) to produce the corresponding products **3a–3e**.

The structures of **3a–3e** were based on their various spectral analyses. Its IR spectrum exhibited major absorptions at 3057–3054 (aromatic C–H), 2926–2916, 2870–2828 (methylene C–H), 1662–1657 (CO), 1601–1597 (CC), and 1251–1215, 1035–1016 cm^{-1} (C–O). The UV–vis spectrum of **3a–3e** exhibited two maxima at 343–328 and 292–247 nm, which may be assigned to $n \rightarrow \pi^*$ and $\pi \rightarrow \pi^*$ transitions, respectively.

^{1}H-NMR spectra (400 MHz, $CDCl_3$) of **3a–3e** were quite informative, and the aromatic protons (H-2′, 6′, and 3′, 5′) produced well-defined signals at δ 8.00–7.99 (4H, dd, $J_{p,o} = 1.2$, 7.5 Hz) and 7.54–7.52 (4H, d, $J_o = 7.6$ Hz). The downfield resonance of former as compared with latter protons could be ascribed to its proximity to the CO group. The proton H-4′ resonates at δ 7.60–7.52 (2H, d, $J_o = 7.3$ Hz), which is ortho to the H-3′ and 5′. In this region, two broad doublets centered at δ 7.82–7.78 (2H) and 7.51–7.42 (2H) were denoted by H-3 and H-2, respectively. The coupling constant of 15.8–15.5 Hz between these protons describes the *trans* geometry around C-2 and C-3 double bond. Regarding the remaining signals in the aromatic region, two doublets integrating for four protons each at δ 7.78–7.60 ($J_o = 8.8$ Hz) and 7.06–6.96 ($J_o = 8.8$ Hz) can be easily ascribed to H-2″, 6″, and H-3″, 5″, respectively. A triplet at δ 5.19–4.65 (4H, $J_{vic} = 2.5$ Hz) may be given to O***C****H*$_2$ protons.

^{13}C-NMR (100 MHz, $CDCl_3$) spectra of **3a–3e** were very instrumental to corroborate its proposed structure. Here, the presence of carbonyl group was confirmed by the appearance of a downfield signal at δ 190.61–189.31 (C-1). The carbon atoms belonging to the double bond appeared at δ 144.65–143.89 (C-3) and 120.09–119.30 (C-2), the downfield occurrence of the former can be ascribed to its β-position in the enone moiety. Other downfield resonance present at δ 160.85–160.07 could be generated by C-4″ due to its direct linkage to the electronegative oxygen atom. The remaining signals in the aromatic region were found at δ 138.51–137.81 (C-1′), 132.30–130.62 (C-2′, 6′), 136.46–131.16 (C-1‴), 130.28–129.21 (C-2″, 6″), 128.80–127.89 (C-4′), 128.12–127.43 (C-3′, 5′), and 115.34–114.75 (C-3″, 5″). Toward the right-hand side of the spectra, a signal was also placed at δ 69.92–55.92, which could be attributed to O***C***H$_2$ group.

Alternatively, bischalcones **3a–3e** were also prepared from the O-alkylation of chalcone **2** with suitable alkylating agents (*trans*-1,4-dibromo-2-butene, 1,4-dichloro-2-butyne, α,α′-dibromo-*o*-xylene, α,α′-dibromo-*p*-xylene, and 4, 4′-bis-chloromethyldiphenyl) under KOH/dry EtOH/DMF medium. The resulting reaction mixture was decomposed into iced HCl to give a solid compound that was

recrystallized from $CHCl_3$/MeOH (1:1) to yield pure compounds **3a–3e**. The chalcone **2** (68%, mp 168–170°C) in turn was obtained from the Claisen–Schmidt reaction of acetophenone with 4-hydroxybenzaldehyde under the alkaline medium.

The IR spectrum of **4a–4e** was very helpful to interpret its structure, which did not exhibit a strong absorption at 1662–1657 cm^{-1} indicating the involvement of carbonyl group during cyclization reaction of **3a–3e**. Here, a significant band was observed at 1600–1596 cm^{-1} may be due to the CN moiety of the pyrazoline ring.

A comparison of the ^{1}H-NMR spectra (400 MHz, DMSO-d_6) of **3a–3e** and **4a–4e** shows that resonances present at δ 7.82–7.78 (H-3) and 7.51–7.42 (H-2) in former were found to be missing together in the latter thereby pronouncing the involvement of these protons in the chemical transformation. Here, aromatic protons H-2″, 6″, H-3″, 5″, and H-4″ were also found resonating at δ 7.80–7.66 (4H, dd, $J_{p,o}$ = 1.2, 8.5 Hz), 7.43–7.10 (4H, m) and 7.51–7.29 (2H, m), respectively. The phenyl ring (C-5) protons furnished doublet of doublet at δ 7.63–7.28 (4H, $J_{p,o}$ = 1.3, 8.1 Hz, H-2‴, 6‴) and a doublet at δ 7.01–6.86 (4H, J_o = 7.7 Hz, H-3‴, 5‴). The protons belonging to the *N*-phenyl ring were also resonating in the aromatic region at δ 7.30–6.97 (4H, td, $J_{p,o}$ = 1.0, 7.4 Hz, H-3′, 5′), 7.11–6.87 (4H, m, H-2′, 6′), and 6.71-6.66 (2H, t, J = 7.3 Hz, H-4′). The major feature of these spectra was the signals of the pyrazoline ring protons (H-X, M, and A), which were centered at δ 5.33–5.26 (dd, J_{XA} = 7.1–6.6 Hz, J_{XM} = 12.3–12.1 Hz), 3.87–3.82 (2H, dd, J_{MX} = 12.3–12.1 Hz, J_{MA} = 17.5–17.3 Hz) and 3.09–3.04 (2H, dd, J_{XA} = 7.1–6.6 Hz, J_{AM} = 17.5–17.3 Hz), respectively. A broad singlet was also present at δ 5.08–4.53 (4H), which may be assigned to O***C*H**$_2$ group protons. Its UV–vis spectrum showed two maxima at 357–355 and 229–228 nm, which could be ascribed to n → π* and π → π* transitions, respectively.

^{13}C-NMR (100 MHz, DMSO-d_6) data of **4a–4e** provided enough evidence in favor of the proposed expression. C-4‴ was resonating at δ 158.17–157.21 due to its direct bonding to the oxygen atom and carbon atoms C-3 and C-1′ directly connected to nitrogen atom that produced suitable resonances at δ 146.68–146.43 and 144.28–144.19, respectively. The remaining aromatic carbon atoms were found to be placed at the appropriate position in the aromatic region (see Experimental section). The upfield placement of C-3‴, 5‴ at δ 112.90–112.81 could be ascribed to the electron-donating effect of alkoxy (OCH_2) group at C-4‴. The signals present at δ 62.98–62.91 and 43.02–42.98 could be easily generated by pyrazoline rings carbons C-5 and C-4, respectively, and downfield resonance of former may be attributed to its benzylic nature and its placement adjacent to nitrogen. A signal in the upfield region was also found at δ 69.00–55.30, which could be generated by O***C*H**$_2$ group.

Table 1

In vitro minimum inhibitory concentration (μg/mL) for bischalcones **3a–3e**

	Gram-negative bacteria				Gram-positive bacteria			Fungi				
Compound	*Escherichia coli*	*Klebsiella pneumoniae*	*Pseudomonas aeruginosa*	*Pseudomonas fluorescens*	*Staphylococcus aureus*	*Bacillus subtilis*	*Streptococcus pyrogens*	*Aspergillus janus*	*Penicillium glabrum*	*Aspergillus niger*	*Fusarium oxysporum*	*Aspergillus sclerotiorum*
3a	16	**8**	**8**	**8**	**4**	16	16	32	16	32	32	16
3b	32	16	32	16	32	**8**	16	**8**	**8**	16	16	**8**
3c	16	**8**	**8**	16	16	16	16	16	16	32	32	16
3d	32	16	16	16	**8**	16	16	16	32	64	32	16
3e	32	16	32	16	16	16	16	**8**	16	16	16	**8**
Amoxicillin	**4**	**4**	**4**	**4**	2	2	**4**	–	–	–	–	–
Fluconazole	–	–	–	–	–	–	–	2	2	2	2	2

The bold emphasis indicates the significant activity given by the corresponding compounds.

ANTIBACTERIAL AND ANTIFUNGAL ACTIVITIES

All the prepared bischalcones **3a–3e** and bispyrazolines **4a–4e** were screened for their antibacterial and antifungal activities using the serial dilution method [20]. Seven bacterial and five fungal species were used as the antimicrobial test strains, namely, *Klebsiella pneumoniae* (MTCC 3384), *Pseudomonas aeruginosa* (MTCC 424), *Escherichia coli* (MTCC 443), *Staphylococcus aureus* (MTCC 443), *Bacillus subtilis* (MTCC 441), *Pseudomonas fluorescens* (MTCC 103), and *Staphylococcus pyrogens* (MTCC 442), and *Aspergillus janus* (MTCC 2751), *Penicillium glabrum* (MTCC 4951), *Fusarium oxysporum* (MTCC 2480), *Aspergillus sclerotiorum* (MTCC 1008), and *A. niger* (MTCC 281), respectively. Amoxicillin and fluconazole were used as standard drugs against bacteria and fungi, respectively. The minimum inhibitory concentration (MIC in μg/mL) was determined by using different dilutions of the concerned compound. The results were compared with positive controls, the standard drugs amoxicillin and fluconazole. Serial dilution of the test compounds previously dissolved in DMSO were prepared to final concentrations of 128, 64, 32, 16, 8, 4, 2 and 1 μg/mL. All the bacterial strains were grown at 37°C for 24 h in nutrient broth, and fungi were grown in malt extract at 28°C for 72 h. Each test compound was dissolved in DMSO, and MIC thus obtained were compared with control. The observed MIC of the aforementioned prepared compound has been given in Tables 1 and 2, respectively.

It is evident from Table 1 that compound **3a** showed very significant activity (MIC 4 μg/mL) against bacterial strain *S. aureus*, whereas compounds **3a–3e** also showed noticeable activity (MIC 8 μg/mL) against bacterial and fungal strains, namely, *K. pneumoniae*, *P. fluorescens*, *P. aeruginosa*, *S. aureus*, *B. subtilis*, *A. janus*, *P. glabrum*, and *A. sclerotiorum* respectively.

Similarly, Table 2 describes that compounds **4a** and **4c** displayed significant activity (MIC 4 μg/mL) against *A. sclerotiorum* and *A. janus*, respectively, whereas compounds **4a**, **4c**, **4d**, and **4e** showed better activity against fungal strains *A. janus*, *F. oxysporum*, *A. sclerotiorum*, and *P. glabrum*, and bacterial strain *S. pyrogens* (MIC 8 μg/mL).

Table 2
In vitro MIC (μg/mL) for bispyrazolines **4a–4e**

Compound	Gram-negative bacteria				Gram-positive bacteria			Fungi				
	Escherichia coli	*Klebsiella pneumoniae*	*Pseudomonas aeruginosa*	*Pseudomonas fluoresecns*	*Staphylococcus aureus*	*Bacillus subtilis*	*Streptococcus pyrogens*	*Aspergillus janus*	*Penicillium glabrum*	*Aspergillus niger*	*Fusarium oxysporum*	*Aspergillus sclerotiorum*
4a	16	16	16	16	32	32	32	**8**	16	32	16	**4**
4b	16	16	32	16	16	16	16	16	16	32	32	16
4c	16	16	32	32	16	32	16	**4**	32	16	**8**	**8**
4d	16	16	16	16	32	32	**8**	**8**	**8**	16	**8**	32
4e	32	16	32	16	32	16	**8**	16	**8**	32	32	16
Amoxicillin	**4**	**4**	**4**	**4**	2	2	**4**	–	–	–	–	–
Fluconazole	–	–	–	–	–	–	–	2	2	2	2	2

The bold emphasis indicates the significant activity given by the corresponding compounds.

CONCLUSION

The present study represents a general and efficient method for the synthesis of bispyrazolines linked through the rigid chains. The bispyrazolines are found to be better antimicrobial agents than the bischalcones.

EXPERIMENTAL

Melting points reported are uncorrected. IR spectra were scanned in KBr pellets on a Perkin Elmer RXIFT (Buckinghamshire, England) Infrared spectrophotometer. ^{1}H-NMR spectra (Fallanden,

Switzerland) were recorded on a 400 MHz Bruker spectrometer using TMS as the internal standard. The mass spectra have been scanned on the Waters Micromass Q-T of Micro (ESI) spectrometer (Vernon Hills, IL). TLC plates were coated with silica gel suspended in MeOH–$CHCl_3$, and iodine vapors were used as visualizing agent. Dibenzaldehydes **1a–1e** were synthesized according to the reported methods [21–23].

Synthesis of 3-(4-hydroxyphenyl)-1-phenylprop-2-en-1-one 2. A solution of 4-hydroxybenzaldehyde (5 g, 0.04 mol), acetophenone (4.8 g, 0.04 mol), and NaOH (2.5 g) in ethanol (20.0 mL) was stirred at 0°C for 8 h, and the resulting reddish mass was kept in refrigerator overnight. The reaction mixture was poured into iced HCl to yield a yellow solid that was filtered, thoroughly washed with water, and dried. The crude product thus obtained was crystallized from $CHCl_3$/MeOH (1:1, v/v) to yield pure compound **2**.

2. Yield (1.5 g, 68%); yellow solid; mp 168–170°C; IR (KBr): υ_{max} (cm^{-1}) 3217 (OH), 1651 (CO), 1599 (CC); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 9.82 (1H, brs, OH), 8.00 (2H, dd, J_o = 8.4 Hz, H-2″,6″), 7.66 (1H, d, J_{trans} = 15.5 Hz, H-3), 7.54 (3H, t, J = 5.0 Hz, H-2′,4′, 6′), 7.46 (2H, d, J_o = 7.4 Hz, H-3′, 5′), 7.44 (1H, d, J_{trans} = 15.5 Hz, H-2), 6.83 (2H, dd, J_o = 8.4 Hz, H-3″, 5″); ^{13}C-NMR (100 Hz, $CDCl_3$): δ 189.21 (C-1), 160.06 (C-4″), 144.54 (C-3), 138.00 (C-1′), 132.24 (C-1″), 130.34 (C-2″, 6″), 128.26 (C-2′, 6′), 127.96 (C-4′), 125.57 (C-3′, 5′), 118.14 (C-2), 115.75 (C-3″, 5″); ESI-MS: *m/z* 247 (M + Na, 100%), 225 (M + 1, 89%), 198 (67%), 122 (54%), 106 (18%); *Anal*. Calcd for $C_{15}H_{12}O_2$: Calcd C, 80.35%; H, 5.35%; Found: C, 80.42%; H, 5.40%.

Synthesis of (2E,2′E)-3,3′-(4,4′-(Z)-but-2-ene-1,4-diylbis(oxy) bis(4,1-phenylene)) bis(1-phenylprop-2-en-1-one 3a. A solution of **1a** (1.0 g, 0.003 mol), acetophenone (0.72 g, 0.006 mol), and NaOH (2.5 g) in ethanol (20.0 mL) was stirred for 12 h at room temperature. The progress of reaction was monitored by TLC. After the completion of reaction, the resulting reaction mixture was poured into iced HCl to give a yellow solid that was filtered, thoroughly washed with water, and dried. The crude product was crystallized from $CHCl_3$/MeOH (1:1, v/v) to yield pure compound **3a**.

3a. Yield (1.4 g, 70%); yellow solid; mp 138–140°C; IR (KBr): υ_{max} (cm^{-1}) 3054 (aromatic C–H), 2926, 2870 (methylene C–H), 1660 (CO), 1601 (CC), 1251, 1035 (C–O); UV-vis (MeOH): λ_{max} (nm) 340, 264; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.00 (4H, dd, $J_{p,o}$ = 1.2, 7.5 Hz, H-2′, 6′), 7.79 (2H, d, J_{trans} = 15.6 Hz, H-3), 7.61 (4H, d, J_o = 8.8 Hz, H-2″, 6″), 7.58 (2H, d, J_o = 7.3 Hz, H-4′), 7.51 (4H, d, J_o = 7.6 Hz, H-3′, 5′), 7.42 (2H, d, J_{trans} = 15.6 Hz, H-2), 6.96 (4H, d, J_o = 8.8 Hz, H-3″, 5″), 6.12 (2H, t, J_{vic} = 2.5 Hz, C***H***=), 4.65 (4H, d, J_{vic} = 2.5 Hz, OC***H***$_2$); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 189.31 (C-1), 160.07 (C-4″), 143.89 (C-3), 137.81 (C-1′), 132.30 (C-2′, 6′), 131.16 (C-1″), 130.03 (C-2″, 6″), 128.23 (***C***H), 127.94 (C-4′), 127.68 (C-3′, 5′), 119.30 (C-2), 114.75 (C-3″, 5″), 67.18 (O***C***H$_2$); ESI-MS: *m/z* 524 (M + Na + 1, 37%), 523 (M + Na, 100%), 502 (M + 2, 16%), 501 (M + 1, 43%), 475 (37%), 453 (20%), 371 (17%), 355 (23%), 275 (13%), 257 (22%), 223 (17%), 195 (14%), 159 (20%), 140 (32%), 117 (15%), 116 (37%); *Anal*. Calcd for $C_{34}H_{28}O_4$: Calcd C, 81.60%; H, 5.60%; Found: C, 81.52%; H, 5.54%.

Synthesis of (2E, 2′E)-3,3′-(4,4′-(but-2-yne-1,4-diylbis(oxy)) bis(4,1-phenylene)) bis(1-phenylprop-2-en-1-one) 3b. The compound **3b** was prepared by reacting **1b** (1.0 g, 0.003 mol) with acetophenone (0.72 g, 0.006 mol) under the similar conditions as described earlier for **3a**.

3b. Yield (0.6 g, 65%); brown solid; mp 98–100°C; IR (KBr): υ_{max} (cm^{-1}) 3057 (aromatic C–H), 2919 (methylene C–H), 1660 (CO), 1597 (CC), 1217, 1017 (C–O); UV-vis (MeOH): λ_{max} (nm) 328, 247; ^{1}H-NMR (400 MHz, DMSO-d_6): δ 8.02 (4H, dd, $J_{p,o}$ = 1.3, 7.6 Hz, H-2′, 6′), 7.80 (2H, d, J_{trans} = 15.5 Hz, H-3), 7.78 (4H, dt, J = 2.6, 4.7 Hz, H-2″, 6″), 7.60 (2H, d, J_o = 7.3 Hz, H-4′), 7.51 (2H, d, J_{trans} = 15.5 Hz, H-2), 7.49 (4H, d, J_o = 7.6 Hz, H-3′, 5′), 7.1 (4H, dt, J = 2.6, 4.5 Hz, H-3″, 5″), 4.89 (4H, s, OC***H***$_2$); ^{13}C-NMR (100 MHz, DMSO-d_6): δ 190.56 (C-1), 160.55 (C-4″), 143.99 (C-3), 137.96 (C-1′), 132.46 (C-2′, 6′), 131.25 (C-1″), 130.14 (C-2″, 6″), 128.01 (C-3′, 5′), 127.89 (C-4′), 120.0 (C-2), 114.83 (C-3″, 5″), 82.00 (C-2‴), 55.92 (O***C***H$_2$); ESI-MS: *m/z* 521 (M + Na, 100%), 500 (M + 2, 35%), 499 (M + 1, 57%), 473 (39%), 421 (47%), 394 (18%), 368 (22%), 355 (27%), 198 (16%), 122 (28%); *Anal*. Calcd for $C_{34}H_{26}O_4$: Calcd C, 81.90%; H, 5.20%; Found: C, 81.97%; H, 5.25%.

The compound **3c** was prepared by reacting **1c** (1.0 g, 0.002 mol) with acetophenone (0.48 g, 0.004 mol) under the similar conditions as described earlier for **3a**.

3c. Yield (1.0 g, 63%); yellow solid; mp 128–130°C; IR (KBr): υ_{max} (cm^{-1}) 3055 (aromatic C–H), 2929, 2855 (methylene C–H), 1662 (CO), 1597 (CC), 1215, 1019 (C–O); UV-vis (MeOH): λ_{max} (nm) 340, 292; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.99 (4H, dt, $J_{p,o}$ = 1.3, 7.0 Hz, H-2′, 6′), 7.78 (2H, d, J_{trans} = 15.7 Hz, H-3), 7.60 (4H, d, J_o = 7.8 Hz, H-2″, 6″), 7.52 (8H, m, H-3′, 4′, 5′, 5‴), 7.42 (2H, d, J_{trans} = 15.7 Hz, H-2), 7.41 (2H, t, J = 3.8 Hz, H-6‴), 7.0 (4H, d, J_o = 8.8 Hz, H-3″, 5″), 5.22 (4H, s, OC***H***$_2$); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 190.56 (C-1), 160.43 (C-4″), 144.50 (C-3), 138.46 (C-1′), 134.64 (C-1″), 132.62 (C-2′, 6′), 130.28 (C-1‴,2‴), 129.21 (C-2″,6″'), 128.80 (C-4′′), 128.59 (C-3‴, 6‴), 128.43 (C-4‴,5‴), 128.12 (C-3′, 5′), 120.09 (C-2), 115.24 (C-3″, 5″), 68.19 (O***C***H$_2$); ESI-MS: *m/z* 575 (9%), 574 (M + Na + 1, 40%), 573 (M + Na, 100%), 552 (M + 2, 20%), 551 (M + 1, 40%), 476 (14%), 475 (50%), 453 (27%), 355 (19%), 327 (40%), 275 (36%), 257 (36%), 223 (19%), 195 (20%), 177 (15%), 159 (28%), 140 (40%), 117 (23%), 116 (47%); *Anal*. Calcd for $C_{38}H_{30}O_4$: Calcd C, 82.90%; H, 5.45%; Found: C, 82.83%; H, 5.40%.

Synthesis of (2E, 2E′)-3,3′-(4,4′-(1,4-phenylenebis (methylene))bis(oxy)bis(4,1-phenylene)bis(1-phenylprop-2-en-1-one) 3d. The compound **3d** was obtained by treating **1d** (1.0 g, 0.002 mol) with acetophenone (0.48 g, 0.004 mol) under the same conditions as used earlier for **3a**.

3d. Yield (1.2 g, 75%); light yellow solid; mp 135–137°C; IR (KBr): υ_{max} (cm^{-1}) 3057 (aromatic C–H), 2930, 2862 (methylene C–H), 1660 (CO), 1600 (CC), 1249, 1017 (C–O); UV-vis (MeOH): λ_{max} (nm) 343, 265; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.02 (4H, dd, $J_{p,o}$ = 1.3, 7.2 Hz, H-2′, 6′), 7.81 (2H, d, J_{trans} = 15.6 Hz, H-3), 7.63 (4H, d, J_o = 8.7 Hz, H-2″, 6″), 7.59 (2H, d, J_o = 7.3 Hz, H-4′), 7.53 (4H, d, J_o = 7.7 Hz, H-3′, 5′), 7.50 (4H, s, H-2‴, 3‴, 5‴, 6‴), 7.43 (2H, d, J_{trans} = 15.6 Hz, H-2), 7.03 (4H, d, J_o = 8.7 Hz, H-3″, 5″), 5.15 (4H, s, OC***H***$_2$); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 190.59 (C-1), 160.73 (C-4″), 144.58 (C-3), 138.49 (C-1′), 136.46 (C-1″), 132.60 (C-2′, 6′), 130.26 (C-2″, 6″), 128.59 (C-4′), 128.43 (C-1‴), 127.98 (C-3′, 5′), 127.80 (C-2‴,6‴), 120.0 (C-2), 115.3 (C-3″, 5″), 69.79 (O***C***H$_2$); ESI-MS: *m/z* 574 (M + Na + 1, 9%), 573 (M + Na, 30%), 476 (27%), 475 (100%), 393 (6%), 249 (5%), 187 (5%), 154 (4%), 135 (4%), 105 (11%); *Anal*. Calcd for $C_{38}H_{30}O_4$: Calcd C, 82.90%; H, 5.45%; Found: C, 82.96%; H, 5.49%.

Synthesis of (E)-3-(4-((4′-(4-((E)-3-oxo-3-phenylprop-1-enyl) benzyloxy)biphenyl-4yl)methoxy)phenyl)-1-phenylprop-2-en-1-one 3e. The compound **3e** was prepared by reacting **1e** (1.0 g,

0.002 mol) with acetophenone (0.48 g, 0.004 mol) under the similar conditions as described earlier for **3a**.

3e. Yield (1.2 g, 85%); Yellow solid; mp 126–128°C; IR (KBr): υ_{max} (cm^{-1}) 3037 (aromatic C–H), 2916, 2828 (methylene C–H), 1657 (CO), 1599 (CC), 1249, 1016 (C–O); UV-vis (MeOH): λ_{max} (nm) 340, 255; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.02 (4H, dd, $J_{p,o}$ = 1.4, 7.0 Hz, H-2′ 6′), 7.82 (2H, d, J_{trans} = 15.8 Hz, H-3), 7.66 (8H, m, H-2″, 6″, 3‴, 5‴), 7.59 (2H, d, J = 7.3 Hz, H-4′), 7.54 (8H, m, H-3′, 5′, 2‴, 6‴), 7.45 (2H, J_{trans} = 15.8 Hz, H-2), 7.06 (4H, d, J_o = 8.8 Hz, H-3″, 5″), 5.19 (4H, s, OC***H***$_2$); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 190.61 (C-1), 160.85 (C-4″), 144.65 (C-3), 139.9 (C-1‴), 138.51 (C-1′), 137.90 (C-4‴), 135.36 (C-1″), 132.6 (C-2′, 6′), 130.28 (C-2″, 6″), 128.59 (C-4′), 128.21 (C-3‴, 5‴), 128.01 (C-2‴, 6‴), 127.43 (C-3′, 5′), 119.96 (C-2), 115.34 (C-3″, 5″), 69.9 (O***C***H$_2$); ESI-MS: *m/z* 648 (M + Na, 73%), 626 (M + 1, 100%), 627 (M + 2, 32%), 599 (58%), 493 (21%), 467 (25%), 437 (19%), 361 (38%), 285 (7%); *Anal*. Calcd for $C_{44}H_{34}O_4$: Calcd C, 84.34%; H, 5.43%; Found: C, 84.40%; H, 5.47%.

Synthesis of (Z)-1-(4-((S)-1,3-diphenyl-4,5-dihydro-1H-pyrazol-5-yl)phenoxy)-4-(4-(1,3-diphenyl-4,5-dihydro-1H-pyrazol-5-yl)phenoxy)but-2-ene 4a. A mixture of **3a** (0.5 g, 0.001 mol), phenyl hydrazine (0.22 g, 0.002 mol), and glacial acetic acid (5.0 mL) in dry EtOH (25.0 mL) was refluxed for 8 h. The progress of reaction was monitored by TLC. After the completion of reaction, the resulting reaction mixture was concentrated under vacuum, which upon cooling in an ice bath, yielded a yellow solid. The crude product thus obtained was crystallized from MeOH to provide pure compound **4a**.

4a. Yield (0.5 g, 71%); light yellow solid; mp 98–100°C; IR (KBr): υ_{max} (cm^{-1}) 3025 (aromatic C-H), 2910, 2858 (methylene C–H), 1599 (CN), 1172, 1010 (C–O); UV-vis (MeOH): λ_{max} (nm) 355, 228; ^{1}H-NMR (400 MHz, DMSO-d_6): δ 7.72 (4H, dd, $J_{p,o}$ = 1.2, 8.5 Hz, H-2″, 6″), 7.42 (2H, m, H-4″), 7.34 (4H, dd, $J_{p,o}$ = 1.3, 8.1 Hz, H-2‴, 6‴), 7.19 (4H, m, H-3″, 5″), 7.11 (4H, td, $J_{p,o}$ = 1.0, 7.4 Hz, H-3′, 5′), 7.01 (4H, d, J_o = 7.7 Hz, H-3‴, 5‴), 6.87 (4H, m, H-2′, 6′), 6.70 (2H, t, J = 7.3 Hz, H-4′), 6.00 (2H, brs, C***H***), 5.33 (2H, dd, J_{XA} = 6.7 Hz, J_{XM} = 12.3 Hz, H-X), 4.53 (4H, brs, OC***H***$_2$), 3.87 (2H, dd, J_{MX} = 12.3 Hz, J_{MA} = 17.3 Hz, H-M), 3.09 (2H, dd, J_{AX} = 6.7 Hz, J_{AM} = 17.3 Hz, H-A); ^{13}C-NMR (100 MHz, DMSO-d_6): δ 157.21 (C-4‴), 146.55 (C-3), 144.2 (C-1′), 134.36 (C-1‴), 128.78 (C-2‴, 6‴), 128.76 (C-2″, 6″), 128.58 (***C***H), 128.50 (C-4″), 128.27 (C-1″), 127.96 (C-3″, 5″), 125.37 (C-3′, 5′), 118.42 (C-4′), 114.85 (C-2′, 6′), 112.90 (C-3‴, 5‴), 67.09 (O***C***H$_2$), 62.91 (C-5), 43.00 (C-4); ESI-MS: *m/z* 682 (M + 2, 47%), 681 (M + 1, 100%), 679 (47%), 588 (23%), 476 (85%), 475 (85%), 453 (50%), 435 (18%), 380 (18%), 313 (17%), 282 (18%), 275 (41%), 257 (73%), 233 (62%), 223 (30%), 195 (42%), 163 (28%), 159 (57%), 140 (89%), 116 (90%); *Anal*. Calcd for $C_{46}H_{40}N_4O_2$: Calcd C, 81.17%; H, 5.88%, N, 8.23%; Found: C, 81.24%; H, 5.93%; N, 8.27%.

Synthesis of 1-(4-((S)-1,3-diphenyl-4,5-dihydro-1H-pyrazol-5-yl)phenoxy)-4-(4-(1,3-diphenyl-4,5-dihydro-1H-pyrazol-5-yl) phenoxy)but-2-yne 4b. The compound **4b** was prepared by reacting **3b** (0.5 g, 0.001 mol) with phenyl hydrazine (0.22 g, 0.002 mol) under the similar conditions as described earlier for **4a**.

4b. Yield (0.4 g, 62%); brown solid; mp 148–150°C; IR (KBr): υ_{max} (cm^{-1}) 3023 (aromatic C–H), 2912, 2858 (methylene C–H), 1600 (CN), 1177, 1015 (C–O); UV-vis (MeOH): λ_{max} (nm) 357, 228; ^{1}H-NMR (400 MHz, DMSO-d_6): δ 7.80 (4H, dd, $J_{p,o}$ = 1.2, 8.5 Hz, H-2″, 6″), 7.51 (2H, m, H-4″), 7.40 (4H, dd, $J_{p,o}$ = 1.2, 8.1, H-2‴, 6‴), 7.22 (4H, m, H-3″, 5″), 7.20 (4H, td, $J_{p,o}$ = 1.0, 7.4 Hz, H-3′, 5′), 6.98 (4H, d, J_o = 7.7 Hz, H-3‴, 5‴), 6.90 (4H, m, H-2′, 6′), 6.71 (2H, t, J_o = 7.3 Hz, H-4′), 5.30 (2H, dd, J_{XA} = 6.7 Hz, J_{XM} = 12.3 Hz, H-X), 4.79 (4H, s, H-1‴′), 3.85 (2H, dd, J_{MX} = 12.3 Hz, J_{MA} = 17.3 Hz, H-M), 3.06 (2H, dd, J_{XA} = 6.7 Hz, J_{AM} = 17.3 Hz, H-A); ^{13}C-NMR (100 MHz, DMSO-d_6): δ 157.35 (C-4″), 146.68 (C-3), 144.22 (C-1′), 135.03 (C-1‴), 128.89 (C-2″, 6″), 128.53 (C-4″), 128.43 (C-1″), 127.63 (C-3″, 5″), 126.98 (C-2‴, 6‴), 125.36 (C-3′, 5′), 118.40 (C-4′), 114.99 (C-2′, 6′), 112.81 (C-3‴, 5‴), 83.00 (C-2‴′), 55.30 (C-1‴′), 62.98 (C-5), 43.02 (C-4); ESI-MS: *m/z* 701 (M + Na, 40%), 679 (M + 1, 100%), 648 (23%), 626 (18%), 618 (11%), 550 (19%), 474 (59%), 405 (38%), 205 (43%), 184 (28%), 94 (12%); *Anal*. Calcd for $C_{46}H_{38}N_4O_2$: Calcd C, 81.41%; H, 5.60%; N, 8.25%; Found: C, 81.45%; H, 567%; N, 8.28%.

Synthesis of 1,2-bis [(4-(1,3-diphenyl-4,5-dihydro-1H-pyrazol-5-yl)phenoxy methyl)benzene] 4c. The compound **4c** was obtained by reacting **3c** (0.5 g, 0.0009 mol) with phenyl hydrazine (0.19 g, 0.0018 mol) under the similar conditions as used earlier for **4a**.

4c. Yield (0.4 g, 66%); yellow solid; mp 78–80°C; IR (KBr): υ_{max} (cm^{-1}) 3026 (aromatic C–H), 2917 (methylene C–H), 1596 (CN), 1172, 1012 (C–O); UV-vis (MeOH): λ_{max} (nm) 356, 228; ^{1}H-NMR (400 MHz, DMSO-d_6): δ 7.66 (4H, d, J_o = 7.4 Hz, H-2″, 6″), 7.43 (4H, dd, J = 3.6, 5.5 Hz, H-3″, 5″), 7.42 (2H, dd, J = 3.6, 5.9 Hz, H-4″), 7.33 (2H, d, J_o = 7.2 Hz, H-4‴′, 5‴′), 7.28 (4H, m, H-2‴, 6‴), 7.13 (2H, dd, $J_{m,o}$ = 2.2, 8.6 Hz, H-3‴′, 6‴′), 7.08 (8H, m, H-2′, 3′, 5′, 6′), 6.86 (4H, d, J_o = 7.6 Hz, H-3‴, 5‴), 6.67 (2H, t, J_o = 6.8 Hz, H-4′), 5.26 (2H, dd, J_{XA} = 6.7 Hz, J_{XM} = 12.2 Hz, H-X), 5.08 (4H, s, OC***H***$_2$), 3.82 (2H, dd, J_{MX} = 12.2 Hz, J_{MA} = 17.5 Hz, H-M), 3.04 (2H, dd, J_{AX} = 6.3 Hz, J_{AM} = 16.9 Hz, H-A); ^{13}C-NMR (100 MHz, DMSO-d_6): δ 157.44 (C-4‴), 146.43 (C-3), 144.19 (C-1′), 134.75 (C-1‴), 134.49 (C-1″), 132.22 (C-1‴′), 128.50 (C-2″, 6″), 128.45 (C-4″), 128.20 (C-5‴′), 127.8 (C-6‴′), 127.38 (C-3″, 5″), 126.73 (C-2‴, 6‴), 125.30 (C-3′, 5′), 118.42 (C-4′), 114.91 (C-2′, 6′), 112.8 (C-3‴, 5‴), 67.27 (O***C***H$_2$), 62.97 (C-5), 42.98 (C-4); ESI-MS: *m/z* 732 (M + 2, 16%), 731 (M + 1, 30%), 729 (37%), 701 (30%), 679 (24%), 491 (13%), 476 (23%), 475 (70%), 453 (40%), 380 (23%), 355 (49%), 299 (23%), 275 (50%), 257 (90%), 223 (47%), 195 (47%), 177 (35%), 159 (66%), 140 (94%), 116 (100%); *Anal*. Calcd for $C_{50}H_{42}N_4O_2$: Calcd C, 82.19%; H, 5.75%, N, 7.67%; Found: C, 82.13%; H, 5.71%; N, 7.71%.

Synthesis of 1,4bis[(4-(1,3-diphenyl-4,5-dihydro-1H-pyrazol-5yl)phenoxy) methyl)benzene] 4d. The compound **4d** was prepared by treating **3d** (0.5 g, 0.0009 mol) with phenyl hydrazine (0.19 g, 0.0018 mol) under the same conditions as used earlier for **4a**.

4d. Yield (0.4 g, 68%); yellow solid; mp 156–158°C; IR (KBr): υ_{max} (cm^{-1}) 3027 (aromatic C–H), 2910, 2865 (methylene C–H), 1599 (CN), 1168, 1011 (C–O); UV-vis (MeOH): λ_{max} (nm) 355, 228; ^{1}H-NMR (400 MHz, DMSO-d_6): δ 7.67 (4H, d, J_o = 7.4 Hz, H-2″, 6″), 7.51 (4H, d, J_o = 8.6 Hz, H-2‴, 6‴), 7.43 (4H, s, H-2‴′, 3‴′, 5‴′, 6‴′), 7.29 (2H, d, J_o = 8.6 Hz, H-4″), 7.10 (4H, m, H-3″, 5″), 7.00 (4H, d, J_o = 8.3 Hz, H-3‴, 5‴), 6.97 (8H, m, H-2′, 3′, 5′, 6′), 6.66 (2H, t, J_o = 7.2 Hz, H-4′), 5.27 (2H, dd, J_{XA} = 6.7 Hz, J_{XM} = 12.2 Hz, H-X), 5.08 (4H, s, OC***H***$_2$), 3.85 (2H, dd, J_{MX} = 12.2 Hz, J_{MA} = 17.5 Hz, H-M), 3.06 (2H, dd, J_{XA} = 6.6 Hz, J_{AM} = 17.5 Hz, H-A); ^{13}C-NMR (100 MHz, DMSO-d_6): δ 158.1 (C-4‴), 146.4 (C-3), 144.21 (C-1′), 136.32 (C-1‴), 136.15 (C-1‴′), 128.68 (C-2″, 6″), 128.47 (C-4″), 128.23 (C-1″), 127.40 (C-3″, 5″), 126.75 (C-2‴, 6‴), 125.33 (C-3′, 5′), 118.43 (C-4′), 118.17 (C-2‴′, 6‴′), 114.93 (C-2′, 6′), 112.89 (C-3‴, 5‴), 69.0

(OCH_2), 62.96 (C-5), 43.0 (C-4); ESI-MS: *m/z* 731 (M + 1, 30%), 729 (45%), 701 (53%), 679 (37%), 476 (27%), 475 (100%), 453 (63%), 380 (24%), 282 (21%), 275 (42%), 257 (69%), 223 (30%), 195 (37%), 163 (26%), 159 (54%), 140 (77%), 116 (78%); *Anal.* Calcd for $C_{50}H_{42}N_4O_2$: Calcd C, 82.19%; H, 5.75%; N, 7.67%; Found: C, 82.24%; H, 5.80%; N, 7.72%.

Synthesis of bis[4-(4-(S)-1,3-diphenyl-4,5-dihydro-1H-pyrazol-5-yl)phenoxy) methyl] 4e. The compound **4e** was obtained by treating **3e** (0.5 g, 0.0007 mol) with phenyl hydrazine (0.15 g, 0.0014 mol) under the similar conditions as used earlier for **4a**.

4e. Yield (0.4 g, 65%); yellow solid; mp 208–210°C; IR (KBr): υ_{max} (cm^{-1}) 3036 (aromatic C–H), 2916, 2859 (methylene C–H), 1598 (CN), 1174, 1015 (C–O); UV-vis (MeOH): λ_{max} (nm) 356, 229; ^{1}H-NMR (400 MHz, DMSO-d_6): δ 7.72 (4H, dd, J_o = 8.1 Hz, H-2″, 6″), 7.63 (4H, d, J_o = H-2‴, 6‴), 7.50 (6H, m, H-4″, 3⁗, 5⁗), 7.30 (8H, m, H-3′, 5′, 3″, 5″), 7.11 (4H, m, H-2′, 6′), 6.98 (4H, d, J_o = 7.4 Hz, H-2⁗, 6⁗), 6.92 (4H, d, J_o = 8.0 Hz, H-3‴, 5‴), 6.67 (2H, t, J_o = 7.2 Hz, H-4′), 5.32 (2H, dd, J_{XA} = 7.1 Hz, J_{XM} = 12.4 Hz, H-X), 5.05 (4H, s, OCH_2), 3.84 (2H, dd, J_{MX} = 12.1 Hz, J_{MA} = 17.3 Hz, H-M), 3.05 (2H, dd, J_{AX} = 7.1 Hz, J_{AM} = 17.0 Hz, H-A); ^{13}C-NMR (100 MHz, DMSO-d_6): δ 157.81 (C-4‴), 146.45 (C-3), 144.28 (C-1′), 137.95 (C-4⁗), 135.42 (C-1‴), 130.01 (C-1⁗), 128.65 (C-2″, 6″), 128.58 (C-4″), 128.35(C-1″), 127.94 (C-2‴, 6‴), 127.55 (C-3″, 5″), 126.98 (C-2⁗, 6⁗), 126.87 (C-3⁗, 5⁗), 125.35 (C-3′, 5′), 118.43 (C-4′), 114.89 (C-2′, 6′), 112.87 (C-3‴, 5‴), 63.00 (OCH_2), 62.95 (C-5), 43.00 (C-4); ESI-MS: *m/z* 829 (M + Na, 100%), 807 (M + 1, 57%), 652 (47%), 585 (33), 555 (39%), 479 (11%), 478 (18%), 403 (26%), 401 (20%), 371 (13%), 297 (22%), 230 (56%), 153 (43%); *Anal.* Cal. for $C_{56}H_{46}N_4O_2$: Calcd C, 83.37%; H, 5.70%; N, 6.94%; Found: C, 83.44%; H, 5.74%; N, 6.99%.

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