

Original article:

**N1-BENZENESULFONYL-2-PYRAZOLINE HYBRIDS IN
NEUROLOGICAL DISORDERS: SYNTHESSES, BIOLOGICAL
SCREENING AND COMPUTATIONAL STUDIES**

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ABSTRACT

A novel series of 1,3,5-trisubstituted-2-pyrazolines (**5a-5t**) was prepared via Claisen Schmidt condensation, followed by heterocyclization with hydrazine hydrate, substitution of N1 hydrogen of 2-pyrazoline nucleus with 4-chlorobenzenesulfonylchloride, applying conventional and green chemistry approaches. Among the two, microwave assisted organic synthesis (MAOS) emerged as a better synthetic tool in terms of faster reaction rate and high yield. Various physicochemical and spectral studies were conducted to characterize the synthesized derivatives including- IR, Mass, ¹H-NMR, ¹³C-NMR and elemental analysis. During pharmacological evaluation, compound **5b** showed excellent anti-anxiety activity and compound **5k** exhibited the best antidepressant effect at the tested doses, 50 and 100 mg/kg b.w., being comparable to diazepam and imipramine, respectively. The docking experiments confirmed the probable mechanism of neuropharmacological action, showing excellent affinity towards MAO-A target protein, which was also evidenced from some of the key interactions with binding site residues Ala68, Tyr69 and Phe352. Furthermore, complimentary *in silico* pharmacokinetic recital without any potential risk of neurotoxicity (as evaluated by rotarod and actophotometer tests), or carcinogenicity, mutagenicity, reproductive toxicity, acute toxicity and irritancy (as predicted by LAZAR and OSIRIS programs) signified their probable use in depression and anxiety disorders.

Keywords: 2-Pyrazolines, antidepressant and anti-anxiety, neurotoxicity, microwave synthesis, molecular docking, *in silico* ADME prediction

INTRODUCTION

Monoamine oxidase (MAO) regulates monoaminergic homeostasis and neurotransmission in the nervous system. Low level of certain neurotransmitters (NTs) in

the brain, like dopamine (DA), norepinephrine (NE), serotonin (5-HT), and gamma amino butyric acid (GABA), is the main cause of depressive mental disorders. These NTs are released during neurotransmission and are degraded by the MAOs enzymes.

Monoamine oxidase inhibitors (MAOIs) block the action of MAOs, and thus increase the concentration of NTs in the brain (Mito-ma and Ito, 1992; Meyer et al., 2006). The antidepressant effect of a hydrazine based anti-tubercular drug, iproniazid (a structural analog of isoniazid) started the journey of development of MAOIs. This discovery showed the way to design and produce some more effective analogs such as phenelzine (Zeller and Barsky, 1952). MAOIs belong to the first generation antidepressants used for decades to treat the patients suffering from high level of anxiety, atypical depression (Pletscher, 1991), anergic bipolar depression and treatment resistant depression (Thase, 2012), specific phobias, post-traumatic stress disorder and migraine headaches resistant to other therapies (Gareri et al., 2000). Two major therapeutic categories of MAOI: the MAO-A inhibitors in certain mental disorders such as depression and anxiety (Amrein et al., 1999) and MAO-B inhibitors proved their remedial value in neurodegenerative diseases (Youdim et al., 2006; Foley et al., 2000) including Parkinson's (Cesura and Pletscher, 1992) and Alzheimer's (Volz and Gleiter, 1998). The initial hydrazine class of MAO inhibitors was associated with some severe adverse effects such as liver toxicity and cheese reaction (Brown et al., 1989). These side-effects were correlated to nonselective and irreversible MAO inhibition. These findings prompted research to develop selective and reversible type of MAO (A and B) inhibitors. Pyrazoline derivatives have attracted substantial attention for years, chiefly 1,3,5-trisubstituted-2-pyrazoline pharmacophore is allied with encouraging neurological activities such as tranquilizer, anticonvulsant and antidepressant (Kaplancikli et al., 2010; Ozdemir et al., 2008; Palaska et al., 2001; Rajendra Prasad et al., 2005; Ruhoglu et al., 2005), psychoanaleptic, MAO inhibitory and other biological activities (Chimenti et al., 2004; 2010; Gokhan et al., 2003; Gokhan-Kelekci et al., 2009; Jagrat et al., 2011; Ja-

yaprakash et al., 2008; Karuppasamy et al., 2010; Maccioni et al., 2010; Manna et al., 2002; Mishra and Sasmal, 2011; Sahoo et al., 2010; Agrawal et al., 2012; Gautam et al., 2010). Considering these facts, some 1,3,5-trisubstituted-2-pyrazolines were synthesized by integrating some novel aromatic and hetero-aromatic substitutions at 3rd and 5th positions, respectively. Microwave facilitated synthetic technique was also utilized to prepare the proposed compounds and the results were compared to that of the conventional heating methods (Lidstrom et al., 2001). This work is in continuation to the previous communications by our group (Tripathi et al., 2016, Upadhyay et al., 2017; Bhandari et al., 2013) with some novel derivatives having promising potential in the field.

MATERIALS AND METHODS

S. D. Fine Chemicals and Sigma Aldrich, Mumbai, India supplied the chemicals and reagents for synthesis and the pre-coated TLC sheets were obtained from Merck Chemicals, India and were used as such. Microwave assisted organic synthesis (MAOS) was performed on Raga's Scientific Microwave System (Ragatech, Pune, Maharashtra, India).

Chemistry

The reaction of suitably substituted aromatic/heteroaromatic ketones with different aldehydes, in alkaline medium, yielded substituted chalcones (**1a-1t**) via Claisen-Schmidt condensation. Then, the formed chalcones were reacted with hydrazine hydrate (in excess) to give 2-pyrazoline derivatives (**2a-2t**). Finally, the N1 hydrogen of the 2-pyrazoline nucleus was replaced with 4-chlorobenzenesulfonylchloride (**5a-5t**), as given in Figure 1. The reaction progress was examined by thin layer chromatography (TLC), using pre-coated silica gel G plates as stationary phase and iodine vapors/UV light as the visualizing agents.

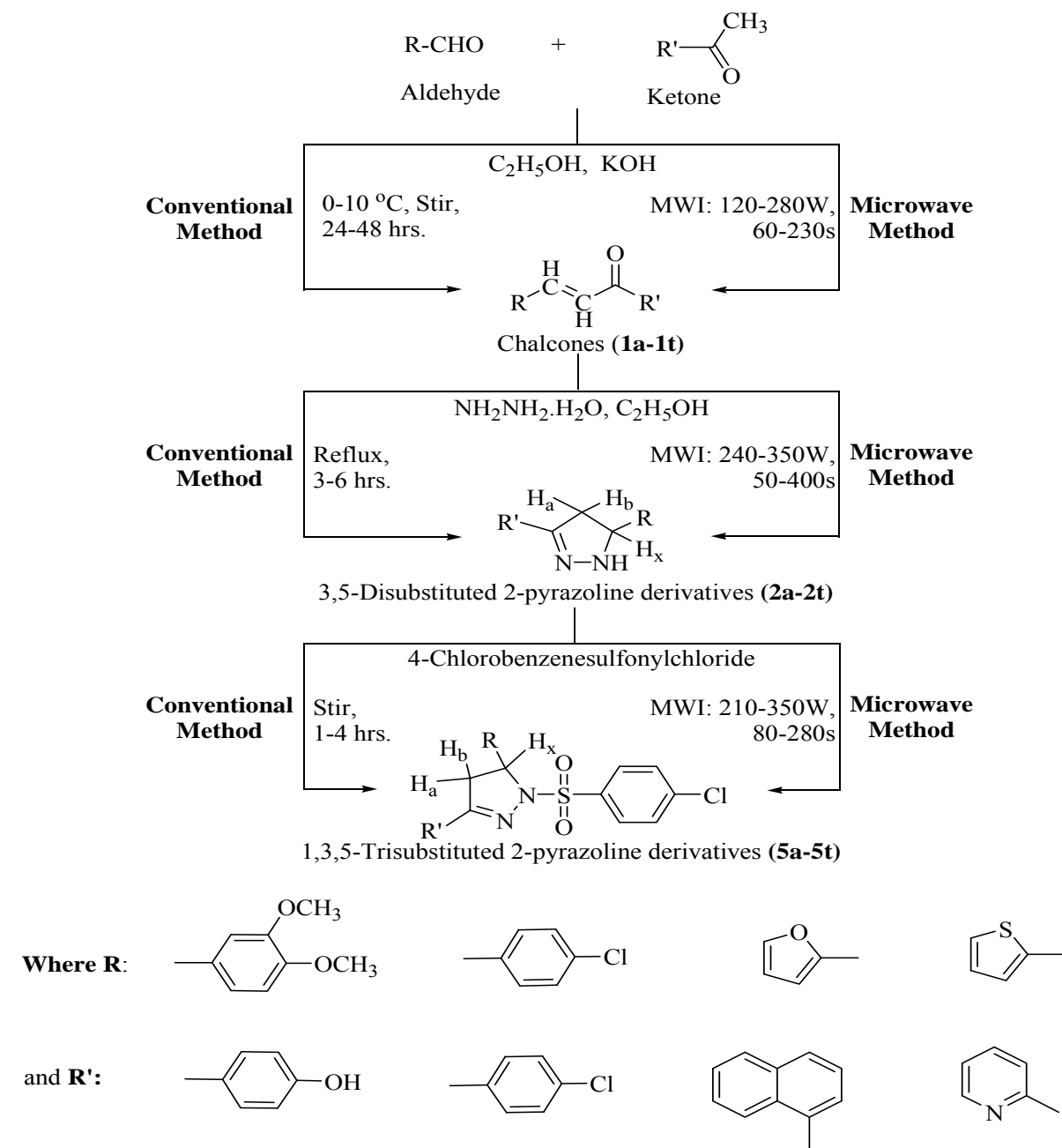


Figure 1: Synthesis of 1,3,5-Trisubstituted-2-pyrazoline derivatives

Synthesis of chalcone derivatives (1a-1t)

The proposed chalcone derivatives of the first step were synthesized by reacting suitably substituted aldehydes and ketones, in equimolar ratio, via conventional and microwave assisted organic synthesis (MAOS) procedures (Tripathi et al., 2016; Upadhyay et al., 2017; Agrawal et al., 2012; Gautam et al., 2010).

Synthesis of 3,5-disubstituted-2-pyrazoline derivatives (2a-2t)

The 3,5-disubstituted-2-pyrazoline derivatives of the second step were synthesized by reacting different chalcone derivatives of the first step with hydrazine hydrate (excess), using conventional and MAOS procedures (Tripathi et al., 2016; Upadhyay et al., 2017).

Synthesis of 1,3,5-trisubstituted-2-pyrazoline derivatives (5a-5t)

Conventional synthesis: Appropriately substituted 2-pyrazoline derivatives (0.001M) of the second step reacted with 4-chlorobenzenesulfonylchloride (0.002M) by stirring, using THF (10 mL) as the solvent. Continued the stirring for 1-4 hrs, poured the reaction mixture in a petri plate and evaporated the solvent up to dryness. Re-precipitated the crude product using acetonitrile/methanol and re-crystallized from acetonitrile/methanol to obtain the pure product (Upadhyay et al., 2017).

MAOS: Appropriately substituted 3,5-disubstituted-2-pyrazolines (0.001M) were reacted with 4-chlorobenzenesulfonylchloride (0.002M) under microwave irradiation (MWI: 210-350W; 80-280s), taking THF (10 mL) as the solvent. The reaction mixture was poured in a petri plate and evaporated the solvent up to dryness. The crude product was re-precipitated using acetonitrile/methanol and recrystallized from acetonitrile/methanol to get the purified derivative (Upadhyay et al., 2017).

Characterization of the synthesized 1,3,5-trisubstituted-2-pyrazoline derivatives

First and second step intermediate compounds were characterized by TLC, melting point (determined by open capillary method and are uncorrected) and mass spectrometric techniques. However, comprehensive physicochemical (Table 1) and spectral characterization was undertaken for the final derivatives and the values were found to be in agreement.

4-[1-(4-Chloro-benzenesulfonyl)-5-(3,4-dimethoxy-phenyl)-4,5-dihydro-1H-pyrazol-3-yl]-phenol (5a)

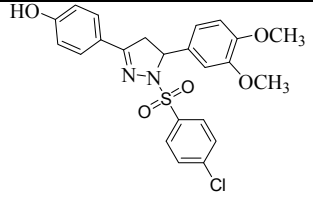
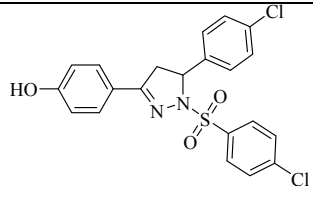
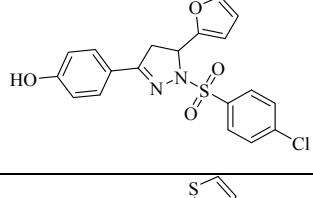
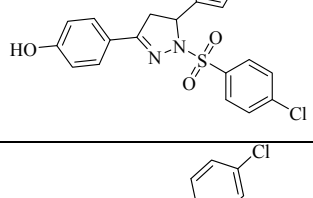
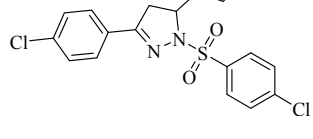
IR spectra were recorded on Bruker FT-IR, ALPHA-T (Eco-ATR) spectrophotometers, (Bruker Corporation., USA) and the values are expressed in cm^{-1} . 3220 (N-H stretch), 2865 (C-H Aromatic), 1650 (C=N stretch), 1515 (C-H deform), 1159, 1350 (sym., asym $\text{S}(=\text{O})_2$ stretch). $^1\text{H-NMR}$ and $^{13}\text{C-NMR}$ spectra were recorded on Bruker

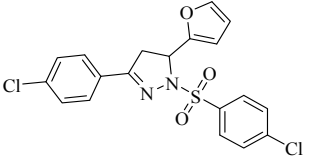
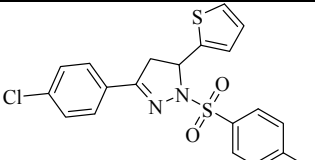
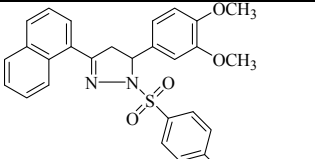
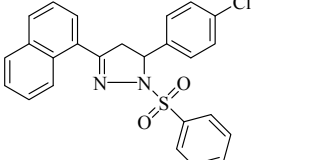
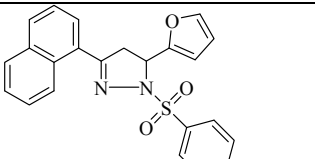
Avance-400, FTNMR spectrometer (Bruker, Tech. Pvt. Ltd., USA) at 400MHz and the chemical shifts are reported in parts per million (δ value), taking TMS (δ 0 ppm for ^1H NMR) as the internal standard: 2.03-2.09 (dd, J_{ab} : 16.77 Hz, J_{ax} : 3.58 Hz, 1H, H_a), 2.73-2.78 (dd, J_{ab} : 3.85 Hz, J_{bx} : 17.11 Hz, 1H, H_b), 3.81-3.93 (dd, J_{ax} : 3.50 Hz, J_{bx} : 17.05 Hz, 1H, H_x), 3.97-4.10 (m, 6H, methyl), 5.23-5.25 (s, 1H, Ar-OH), 6.56-7.90 (m, 11H, Ar). ^{13}C NMR (DMSO, ppm): 37.9 (CH_2 pyrazoline), 43.0 (CH pyrazoline), 113.8-146.3 (12CH benzene), 147.5-159.2 (7C benzene), 160.1 (C pyrazoline). Mass spectra were recorded on Waters UPLC-TQD Mass Spectrometer instrument (Waters Corporation, USA) using LC-ESI or APCI-MS Technique; MS (m/z): 473 (M^+ , 100 %). Elemental analysis was performed on Perkin Elmer-2400, Series-II Analyzer (Waltham, Massachusetts, USA). Anal. Calcd. for $\text{C}_{23}\text{H}_{21}\text{ClN}_2\text{O}_5\text{S}$: C, 58.41; H, 4.48; N, 5.92. Found: C, 58.39; H, 4.50; N, 5.89.

4-[1-(4-Chloro-benzenesulfonyl)-5-(4-chloro-phenyl)-4,5-dihydro-1H-pyrazol-3-yl]-phenol (5b)

IR (cm^{-1}): 3359 (N-H stretch), 3142 (C-H Aromatic), 1606 (C=N stretch), 1516 (C-H deform), 1165, 1351 (sym., asym $\text{S}(=\text{O})_2$ stretch). $^1\text{H-NMR}$ (δ ppm, DMSO): 1.98-1.99 (dd, J_{ab} : 17.12 Hz, J_{ax} : 3.41 Hz, 1H, H_a), 3.40-3.48 (dd, J_{ab} : 4.01 Hz, J_{bx} : 16.23 Hz, 1H, H_b), 3.70-3.80 (dd, J_{ax} : 3.28 Hz, J_{bx} : 17.46 Hz, 1H, H_x), 5.39-5.45 (s, 1H, Ar-OH), 6.88-7.89 (m, 12H, Ar). ^{13}C NMR (DMSO, ppm): 38.6 (CH_2 pyrazoline), 43.2 (CH pyrazoline), 115.6-140.3 (12CH benzene), 158.3-160.1 (3C benzene), 161.2 (C pyrazoline). MS (m/z): 447 (M^+ , 100 %). Anal. Calcd. for $\text{C}_{21}\text{H}_{16}\text{Cl}_2\text{N}_2\text{O}_3\text{S}$: C, 56.38; H, 3.61; N, 6.26. Found: C, 57.42; H, 3.59; N, 6.24.

Table 1: Comparative study of physicochemical properties of synthesized 1,3,5-trisubstituted-2-pyrazoline derivatives (5a-5t)

Com- pounds	Structure	Molecular Formula	Color and State	Solubility	R _f Value	Melting Range (°C)	Conventional Synthesis (Reflux- ing)		Microwave Assisted Organic Synthesis (MAOS)		
							Reaction Time (h)	% Yield	Microwave Power (W)	Reaction Time (s)	% Yield
5a		C ₂₃ H ₂₁ ClN ₂ O ₅ S	Cream yellow colored amorphous solid	Acetonitrile, acetone, DMSO	0.67	206- 208	2.5	47	210-240	130	66
5b		C ₂₁ H ₁₆ Cl ₂ N ₂ O ₃ S	Colorless crystalline solid	Methanol, DMSO, chloroform, acetone	0.92	198- 200	2.0	68	210	85	70
5c		C ₁₉ H ₁₅ ClN ₂ O ₄ S	Light brown colored amorphous solid	Methanol, acetonitrile, chloroform, acetone, DMSO	0.88	162- 164	4.5	58	240-280	210	81
5d		C ₁₉ H ₁₅ ClN ₂ O ₃ S ₂	Brown col- ored amor- phous solid	Methanol, acetonitrile, acetone, DMSO	0.75	148- 150	3.5	57	240-350	200	82
5f		C ₂₁ H ₁₅ Cl ₃ N ₂ O ₂ S	Grey col- ored amor- phous solid	Methanol, acetonitrile, chloroform, acetone, DMSO	0.52	185- 187	2.0	45	210-240	150	78

Com- pounds	Structure	Molecular Formula	Color and State	Solubility	R _f Value	Melting Range (°C)	Conventional Synthesis (Reflux- ing)		Microwave Assisted Organic Synthesis (MAOS)		
							Reaction Time (h)	% Yield	Microwave Power (W)	Reaction Time (s)	% Yield
5g		C ₁₉ H ₁₄ Cl ₂ N ₂ O ₃ S	Black col- ored amor- phous solid	Acetonitrile, acetone, DMSO	0.80	120- 122	4.0	45	240-350	130	86
5h		C ₁₉ H ₁₄ Cl ₂ N ₂ O ₂ S ₂	Yellowish brown col- ored amor- phous solid	Acetonitrile, acetone, DMSO	0.66	193- 195	2.5	57	280-350	100	70
5i		C ₂₇ H ₂₃ ClN ₂ O ₄ S	Orange brown col- ored amor- phous solid	Methanol, acetone, DMSO	0.92	168- 170	1.0	48	210-280	100	63
5j		C ₂₅ H ₁₈ Cl ₂ N ₂ O ₂ S	Cream col- ored amor- phous solid	Acetonitrile, acetone, DMSO	0.58	178- 180	2.5	51	280	80	85
5k		C ₂₃ H ₁₇ ClN ₂ O ₃ S	Black col- ored amor- phous solid	Acetone, DMSO, ace- tonitrile	0.77	218- 220	3.0	32	210-280	220	86

Com- pounds	Structure	Molecular Formula	Color and State	Solubility	R _f Value	Melting Range (°C)	Conventional Synthesis (Reflux- ing)		Microwave Assisted Organic Synthesis (MAOS)		
							Reaction Time (h)	% Yield	Microwave Power (W)	Reaction Time (s)	% Yield
5l		C ₂₃ H ₁₇ ClN ₂ O ₂ S ₂	Black col- ored amor- phous solid	Acetonitrile, acetone, DMSO	0.64	112- 114	3.0	97	280-350	180	94
5q		C ₂₂ H ₂₀ ClN ₃ O ₄ S	Brown col- ored amor- phous solid	Methanol, acetonitrile, chloroform, acetone, DMSO	0.90	180- 182	2.5	30	280	160	71
5r		C ₂₀ H ₁₅ Cl ₂ N ₃ O ₂ S	Yellowish brown col- ored amor- phous solid	Methanol, acetonitrile, chloroform, acetone, DMSO	0.78	116- 118	2.0	93	210-350	175	68
5t		C ₁₈ H ₁₄ ClN ₃ O ₂ S ₂	Brown col- ored amor- phous solid	Methanol, acetonitrile, acetone, DMSO	0.60	138- 140	3.0	48	280-350	260	72

4-[1-(4-Chloro-benzenesulfonyl)-5-furan-2-yl-4,5-dihydro-1H-pyrazol-3-yl]-phenol (5c)

IR (cm⁻¹): 3472 (N-H stretch), 3239 (C-H Aromatic), 1602 (C=N stretch), 1434 (C-H deform), 1159, 1348 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 2.14-2.16 (dd, *J*_{ab}: 17.00 Hz, *J*_{ax}: 3.18 Hz, 1H, H_a), 3.27-3.36 (dd, *J*_{ab}: 3.88 Hz, *J*_{bx}: 16.69 Hz, 1H, H_b), 3.42-3.84 (dd, *J*_{ax}: 3.05 Hz, *J*_{bx}: 16.96 Hz, 1H, H_x), 5.20 (s, 1H, Ar-OH), 6.04-7.66 (m, 11H, Ar). ¹³C NMR (DMSO, ppm): 40.1 (CH₂ pyrazoline), 44.5 (CH pyrazoline), 104.2-142.9 (11CH Ar), 135.3-158.6 (4C benzene), 159.0 (C furan), 160.7 (C pyrazoline). MS (*m/z*): 229 (M⁺, 100 %). Anal. Calcd. for C₁₉H₁₅ClN₂O₄S: C, 56.65; H, 3.75; N, 6.95. Found: C, 56.66; H, 3.77; N, 6.93.

4-[1-(4-Chloro-benzenesulfonyl)-5-thiophen-2-yl-4,5-dihydro-1H-pyrazol-3-yl]-phenol (5d)

IR (cm⁻¹): 3270 (N-H stretch), 3010 (C-H Aromatic), 1615 (C=N stretch), 1463 (C-H deform), 1189, 1382 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 1.99-2.00 (dd, *J*_{ab}: 15.72 Hz, *J*_{ax}: 3.86 Hz, 1H, H_a), 3.40-3.48 (dd, *J*_{ab}: 3.94 Hz, *J*_{bx}: 16.70 Hz, 1H, H_b), 3.71-3.85 (dd, *J*_{ax}: 3.28 Hz, *J*_{bx}: 17.46 Hz, 1H, H_x), 5.45 (s, 1H, Ar-OH), 6.75-7.80 (m, 11H, Ar). ¹³C NMR (DMSO, ppm): 39.4 (CH₂ pyrazoline), 44.8 (CH pyrazoline), 113.8-138.6 (11CH Ar), 120.1-151.7 (4C benzene), 161.3 (C thiophene), 162.4 (C pyrazoline). MS (*m/z*): 419 (M⁺, 95 %). Anal. Calcd. for C₁₉H₁₅ClN₂O₃S₂: C, 54.47; H, 3.61; N, 6.69. Found: C, 54.43; H, 3.62; N, 6.67.

1-(4-Chloro-benzenesulfonyl)-3,5-bis-(4-chloro-phenyl)-4,5-dihydro-1H-pyrazole (5f)

IR (cm⁻¹): 3315 (N-H stretch), 3100 (C-H Aromatic), 1616 (C=N stretch), 1465 (C-H deform), sym., asym S(=O)₂ stretch (1164, 1389). ¹H NMR (δ ppm, DMSO): 1.91-1.96 (dd, *J*_{ab}: 17.56 Hz, *J*_{ax}: 3.50 Hz, 1H, H_a), 3.29-3.48 (dd, *J*_{ab}: 4.01 Hz, *J*_{bx}: 17.23 Hz, 1H, H_b), 3.66-3.84 (dd, *J*_{ax}: 3.75 Hz, *J*_{bx}: 16.54 Hz, 1H, H_x), 7.00-7.77 (m, 12H, Ar). ¹³C NMR (DMSO, ppm): 39.0 (CH₂ pyrazoline), 42.5 (CH pyrazoline), 125.8-131.2

(12CH benzene), 136.1-141.9 (4C benzene), 161.2 (C pyrazoline). MS (*m/z*): 463 (M⁺, 55 %). Anal. Calcd. for C₂₁H₁₅Cl₃N₂O₂S: C, 54.15; H, 3.25; N, 6.01. Found: C, 54.11; H, 3.22; N, 6.00.

1-(4-Chloro-benzenesulfonyl)-3-(4-chloro-phenyl)-5-furan-2-yl-4,5-dihydro-1H-pyrazole (5g)

IR (cm⁻¹): 3330 (N-H stretch), 3092 (C-H Aromatic), 1685 (C=N stretch), 1539 (C-H deform), 1168, 1355 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 2.08-2.13 (dd, *J*_{ab}: 15.31 Hz, *J*_{ax}: 2.98 Hz, 1H, H_a), 3.39-3.44 (dd, *J*_{ab}: 4.16 Hz, *J*_{bx}: 15.57 Hz, 1H, H_b), 3.61-3.79 (dd, *J*_{ax}: 3.58 Hz, *J*_{bx}: 16.95 Hz, 1H, H_x), 6.53-7.60 (m, 11H, Ar). ¹³C NMR (DMSO, ppm): 39.9 (CH₂ pyrazoline), 42.5 (CH pyrazoline), 107.7-128.5 (11CH Ar), 130.2-136.8 (3C benzene), 159.0 (C furan), 161.7 (C pyrazoline). MS (*m/z*): 421 (M⁺, 65 %). Anal. Calcd. for C₁₉H₁₄Cl₂N₂O₃S: C, 54.17; H, 3.35; N, 6.65. Found: C, 54.20; H, 3.39; N, 6.61.

1-(4-Chloro-benzenesulfonyl)-3-(4-chloro-phenyl)-5-thiophen-2-yl-4,5-dihydro-1H-pyrazole (5h)

IR (cm⁻¹): 3345 (N-H stretch), 3056 (C-H Aromatic), 1621 (C=N stretch), 1471 (C-H deform), 1165, 1393 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 2.10-2.19 (dd, *J*_{ab}: 16.88 Hz, *J*_{ax}: 3.29 Hz, 1H, H_a), 4.15-4.22 (dd, *J*_{ab}: 4.01 Hz, *J*_{bx}: 16.78 Hz, 1H, H_b), 4.20-4.23 (dd, *J*_{ax}: 3.63 Hz, *J*_{bx}: 16.91 Hz, 1H, H_x), 6.65-7.79 (m, 11H, Ar). ¹³C NMR (DMSO, ppm): 39.7 (CH₂ pyrazoline), 41.3 (CH pyrazoline), 121.4-136.5 (11CH Ar), 128.6-134.1 (3C benzene), 139.4 (C thiophene), 155.8 (C pyrazoline). MS (*m/z*): 437 (M⁺, 40 %). Anal. Calcd. for C₁₉H₁₄Cl₂N₂O₂S₂: C, 52.18; H, 3.23; N, 6.41. Found: C, 52.17; H, 3.20; N, 6.47.

1-(4-Chloro-benzenesulfonyl)-5-(3,4-dimethoxy-phenyl)-3-naphthalen-1-yl-4,5-dihydro-1H-pyrazole (5i)

IR (cm⁻¹): 3345 (N-H stretch), 3056 (C-H Aromatic), 1577 (C=N stretch), 1462 (C-H deform), 1165, 1362 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 1.97-1.98 (dd, *J*_{ab}: 17.22 Hz, *J*_{ax}: 3.52 Hz, 1H, H_a),

3.39-3.47 (dd, J_{ab} : 3.87 Hz, J_{bx} : 16.73 Hz, 1H, H_b), 3.54-3.69 (dd, J_{ax} : 3.18 Hz, J_{bx} : 17.26 Hz, 1H, H_x), 3.80-3.94 (m, 6H, methyl), 6.78-7.64 (m, 14H, Ar). ¹³C NMR (DMSO, ppm): 39.7 (CH₂ pyrazoline), 42.5 (CH pyrazoline), 61.7 (2CH₃), 113.7-132.5 (14CH Ar), 130.8-137.2 (7C Ar), 159.3 (C pyrazoline). MS (m/z): 507 (M⁺, 50 %). Anal. Calcd. for C₂₇H₂₃ClN₂O₄S: C, 63.96; H, 4.57; N, 5.53. Found: C, 64.07; H, 4.61; N, 5.46.

1-(4-Chloro-benzenesulfonyl)-5-(4-chlorophenyl)-3-naphthalen-1-yl-4,5-dihydro-1H-pyrazole (5j)

IR (cm⁻¹): 3266 (N-H stretch), 3011 (C-H Aromatic), 1570 (C=N stretch), 1495 (C-H deform), 1166, 1362 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 2.15-2.19 (dd, J_{ab} : 16.68 Hz, J_{ax} : 3.27 Hz, 1H, H_a), 3.83-3.88 (dd, J_{ab} : 3.95 Hz, J_{bx} : 17.10 Hz, 1H, H_b), 3.50-3.67 (dd, J_{ax} : 4.12 Hz, J_{bx} : 16.97 Hz, 1H, H_x), 6.90-7.73 (m, 14H, Ar). ¹³C NMR (DMSO, ppm): 39.2 (CH₂ pyrazoline), 46.4 (CH pyrazoline), 124.2-129.0 (14CH Ar), 128.5-136.1 (7C benzene), 158.7 (C pyrazoline). MS (m/z): 481 (M⁺, 50 %). Anal. Calcd. for C₂₅H₁₈Cl₂N₂O₂S: C, 62.37; H, 3.77; N, 5.82. Found: C, 62.49; H, 3.80; N, 5.78.

1-(4-Chloro-benzenesulfonyl)-5-furan-2-yl-3-naphthalen-1-yl-4,5-dihydro-1H-pyrazole (5k)

IR (cm⁻¹): 3290 (N-H stretch), 3050 (C-H Aromatic), 1635 (C=N stretch), 1469 (C-H deform), 1166, 1355 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 2.48-2.51 (dd, J_{ab} : 17.12 Hz, J_{ax} : 3.41 Hz, 1H, H_a), 3.04-3.89 (dd, J_{ab} : 4.01 Hz, J_{bx} : 16.23 Hz, 1H, H_b), 4.88-5.21 (dd, J_{ax} : 3.28 Hz, J_{bx} : 17.46 Hz, 1H, H_x), 6.80-6.93 (d, 2H, Ar), 7.43-7.54 (m, 4H, Ar), 7.66-7.85 (m, 7H, Ar). ¹³C NMR (DMSO, ppm): 50.7 (CH₂ pyrazoline), 77.2 (CH pyrazoline), 124.4-126.9 (2CH furan), 129.0-131.3 (7CH naphthalene), 133.7-138.2 (4CH benzene), 159.4 (C pyrazoline). MS (m/z): 437 (M⁺, 50 %), 438 (30 %). Anal. Calcd. for C₂₃H₁₇ClN₂O₃S: C, 63.23; H, 3.92; N, 6.41. Found: C, 63.26; H, 3.97; N, 6.40.

1-(4-Chloro-benzenesulfonyl)-3-naphthalen-1-yl-5-thiophen-2-yl-4,5-dihydro-1H-pyrazole (5l)

IR (cm⁻¹): 3292 (N-H stretch), 3047 (C-H Aromatic), 1581 (C=N stretch), 1509 (C-H deform), 1177, 1322 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 2.08-2.09 (dd, J_{ab} : 17.48 Hz, J_{ax} : 2.99 Hz, 1H, H_a), 3.10-3.18 (dd, J_{ab} : 3.74 Hz, J_{bx} : 17.04 Hz, 1H, H_b), 3.83-3.86 (dd, J_{ax} : 3.26 Hz, J_{bx} : 16.36 Hz, 1H, H_x), 6.60-7.92 (m, 14H, Ar). ¹³C NMR (DMSO, ppm): 40.6 (CH₂ pyrazoline), 46.1 (CH pyrazoline), 118.5-129.2 (14CH Ar), 137.8-148.1 (6C Ar), 161.5 (C pyrazoline). MS (m/z): 463 (M⁺, 50 %). Anal. Calcd. for C₂₃H₁₇ClN₂O₂S₂: C, 60.98; H, 3.78; N, 6.18. Found: C, 61.06; H, 3.76; N, 6.21.

2-[1-(4-Chloro-benzenesulfonyl)-5-(3,4-dimethoxy-phenyl)-4,5-dihydro-1H-pyrazol-3-yl]-pyridine (5q)

IR (cm⁻¹): 3321 (N-H stretch), 3084 (C-H Aromatic), 1624 (C=N stretch), 1466 (C-H deform), 1170, 1337 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 1.89-1.91 (dd, J_{ab} : 15.62 Hz, J_{ax} : 3.17 Hz, 1H, H_a), 3.33-3.39 (dd, J_{ab} : 3.74 Hz, J_{bx} : 17.11 Hz, 1H, H_b), 3.65-3.80 (dd, J_{ax} : 3.54 Hz, J_{bx} : 16.91 Hz, 1H, H_x), 3.73-3.94 (m, 6H, methyl), 6.97-8.21 (m, 11H, Ar). ¹³C NMR (DMSO, ppm): 40.2 (CH₂ pyrazoline), 45.7 (CH pyrazoline), 49.5 (6C, CH₃), 114.9-135.8 (11CH Ar), 143.4-152.8 (6C benzene), 161.9 (C pyrazoline). MS (m/z): 458 (M⁺, 30 %). Anal. Calcd. for C₂₂H₂₀ClN₃O₄S: C, 57.70; H, 4.40; N, 9.18. Found: C, 57.64; H, 4.45; N, 9.14.

2-[1-(4-Chloro-benzenesulfonyl)-5-(4-chloro-phenyl)-4,5-dihydro-1H-pyrazol-3-yl]-pyridine (5r)

IR (cm⁻¹): 3421 (N-H stretch), 3091 (C-H Aromatic), 1616 (C=N stretch), 1471 (C-H deform), 1170, 1383 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 1.91-1.94 (dd, J_{ab} : 17.36 Hz, J_{ax} : 3.21 Hz, 1H, H_a), 3.64-3.48 (dd, J_{ab} : 4.12 Hz, J_{bx} : 17.44 Hz, 1H, H_b), 3.69-3.85 (dd, J_{ax} : 3.28 Hz, J_{bx} : 16.55 Hz, 1H, H_x), 7.29-8.69 (m, 12H, Ar). ¹³C NMR (DMSO, ppm): 40.2 (CH₂ pyrazo-

line), 44.5 (CH pyrazoline), 124.6-149.2 (12CH Ar), 137.1-139.4 (2C benzene), 154.2 (C pyridine), 160.7 (C pyrazoline). MS (m/z): 432 (M^+ , 55%). Anal. Calcd. for $C_{20}H_{15}Cl_2N_3O_2S$: C, 55.56; H, 3.50; N, 9.72. Found: C, 55.60; H, 3.51; N, 9.68.

2-[1-(4-Chloro-benzenesulfonyl)-5-thiophen-2-yl-4,5-dihydro-1H-pyrazol-3-yl]-pyridine (5t)

IR (cm^{-1}): 3387 (N-H stretch), 3083 (C-H Aromatic), 1612 (C=N stretch), 1468 (C-H deform), 1167, 1389 (sym., asym S(=O)₂ stretch). ¹H NMR (δ ppm, DMSO): 1.90-1.93 (dd, J_{ab} : 17.23 Hz, J_{ax} : 3.32 Hz, 1H, H_a), 3.46-3.50 (dd, J_{ab} : 3.87 Hz, J_{bx} : 16.65 Hz, 1H, H_b), 3.82-3.89 (dd, J_{ax} : 3.14 Hz, J_{bx} : 16.91 Hz, 1H, H_x), 6.53-8.97 (m, 11H, Ar). ¹³C NMR (DMSO, ppm): 38.6 (CH₂ pyrazoline), 43.2 (C pyrazoline), 115.6-140.3 (12CH benzene), 144.9-157.1 (4C Ar), 161.3 (C pyrazoline). MS (m/z): 403 (M^+ , 25%). Anal. Calcd. for $C_{18}H_{14}ClN_3O_2S_2$: C, 53.53; H, 3.49; N, 10.40. Found: C, 53.49; H, 3.53; N, 10.37.

Biological evaluation

Study animals

The study protocols were authenticated by the Institutional Animal Ethics Committee (IAEC) with the protocol number BBDNIIT/IAEC/009/2014.

Animal house, Faculty of Pharmacy, BBDNIIT, Lucknow, U.P., India provided animals (male albino mice) for the *in-vivo* study which were treated humanely and maintained under standard conditions of temperature (26 ± 2 °C), percent humidity (55 ± 5), light and dark cycles and free access to food and water.

Study design

The studies were performed as per reported procedures (Tripathi et al., 2016; Upadhyay et al., 2017). The detailed description of the groups and respective doses are given in Table 2.

Antidepressant activity

The antidepressant potential of the synthesized derivatives (**5a-5t**) was evaluated

using behavioral *in vivo* tests in mice model, such as forced swim test (FST) and tail suspension test, (TST) as given by Porsolt (1981) and Willner and Mitchell (2002).

- Porsolt's behavioral despair or FST

In FST, the state of despair can be minimized by different therapeutically effective antidepressants (Porsolt et al., 1977; Vogel, 2002). The duration of immobility in animals, produced by the test compounds, was measured by known procedures (Tripathi et al., 2016; Upadhyay et al., 2017).

- Tail suspension test (TST)

TST is another well known method (Steru et al., 1985; Vogel, 2002), employed to evaluate the antidepressant potential of the test compounds. In this test, the duration of immobility was measured by known procedures (Tripathi et al., 2016; Upadhyay et al., 2017).

Anti-anxiety activity

The maze model was used to evaluate anti-anxiety potential of the test compounds, which selectively identified the anxiolytic (open arm exploration time increased and time spent in closed arms decreased) and anxiogenic drugs (Vogel, 2002).

Elevated plus-maze test

The elevated plus-maze was used to determine anxiety-related behavior, measured by the extent to which the mouse avoids visiting the open arm of the maze (Lister, 1987; Pellow et al., 1985). After one hour of oral administration of the test compound, observations (i.e. time spent in closed arm and total number of the arm entries) were recorded during the 6 minute test duration by known procedures (Tripathi et al., 2016; Upadhyay et al., 2017).

Table 2: Data showing antidepressant, anti-anxiety and neurotoxicity studies of the synthesized 2-pyrazoline derivatives (5a-5t)

Compounds	Group Number	Doses (mg/kg b.w.)	Antidepressant Activity ^a		Anti-anxiety Activity ^b		Neurotoxicity Studies			
			Duration of Im-mobility in FST (sec) (Mean±SD)	Duration of Im-mobility in TST (sec) (Mean±SD)	Number of En-tries in Closed Arms (Mean±SD)	Time Spent in Closed Arms (sec) (Mean±SD)	Rotarod Test ^c		Actophotometer Test ^d (Mean Counts)	
							Before Dose	After Dose (After 1h)	Before Dose	After Dose (Af-ter 1h)
5a	1	50	125.17±8.93	177.83±17.27	10.83±3.66	162.00±21.54	0/6	0/6	291.83	298.00
	2	100	89.50±11.11	174.00±19.97	14.00±2.53	126.33±11.50	1/6	0/6	277.33	284.50
5b	3	50	79.33±12.39	107.67±27.44	12.33±2.58	143.83±11.67	0/6	2/6	284.17	284.17
	4	100	85.50±10.50	101.67±15.71	15.67±2.94	102.83±9.58	0/6	0/6	317.17	317.17
5c	5	50	82.67±6.41	136.50±18.29	6.83±1.47	194.33±13.59	0/6	0/6	440.17	430.67
	6	100	68.50±15.73	122.83±10.03	7.67±2.94	188.33±15.68	0/6	0/6	275.50	277.67
5d	7	50	124.33±9.71	201.17±12.54	8.83±3.06	146.00±14.21	1/6	1/6	446.67	473.33
	8	100	97.67±14.19	155.33±9.73	9.33±4.18	138.67±11.94	0/6	0/6	277.33	272.33
5f	9	50	105.17±13.85	97.50±30.25	5.17±2.32	196.33±13.43	0/6	0/6	470.33	461.17
	10	100	87.83±18.79	76.50±20.69	6.50±1.87	211.83±15.25	0/6	1/6	286.17	283.50
5g	11	50	104.17±15.56	149.67±8.45	6.50±1.05	273.17±12.58	0/6	0/6	275.50	270.83
	12	100	99.50±14.64	150.50±8.31	9.00±2.37	275.17±26.50	0/6	0/6	284.17	258.33
5h	13	50	155.00±11.64	93.00±11.17	3.67±1.63	198.50±19.38	0/6	0/6	477.83	517.50
	14	100	101.67±12.26	149.17±42.10	5.17±2.32	255.67±28.39	0/6	0/6	286.17	301.00
5i	15	50	94.33±14.18	133.33±23.85	6.50±3.27	253.17±15.18	0/6	0/6	446.67	445.67
	16	100	73.33±25.40	107.67±17.96	8.67±3.27	208.17±24.26	0/6	0/6	298.00	297.00
5j	17	50	112.50±19.15	129.50±37.25	8.00±2.28	222.17±16.80	0/6	0/6	486.67	488.67
	18	100	84.67±13.34	132.17±11.32	9.50±1.87	193.67±20.22	0/6	0/6	286.17	281.83
5k	19	50	58.00±7.97	65.83±13.79	7.00±1.79	205.17±14.02	0/6	0/6	485.50	482.67
	20	100	23.00±6.20	37.83±10.25	8.83±2.04	175.17±24.00	0/6	0/6	286.17	282.83

Compounds	Group Number	Doses (mg/kg b.w.)	Antidepressant Activity ^a		Anti-anxiety Activity ^b		Neurotoxicity Studies			
			Duration of Im-mobility in FST (sec) (Mean±SD)	Duration of Im-mobility in TST (sec) (Mean±SD)	Number of En-tries in Closed Arms (Mean±SD)	Time Spent in Closed Arms (sec) (Mean±SD)	Rotarod Test ^c		Actophotometer Test ^d (Mean Counts)	
							Before Dose	After Dose (After 1h)	Before Dose	After Dose (Af-ter 1h)
5l	21	50	59.67±9.93	129.17±17.93	7.67±3.20	164.83±14.74	1/6	1/6	275.50	269.33
	22	100	43.50±8.04	54.83±15.88	9.17±2.32	137.83±10.46	0/6	0/6	284.17	282.50
5q	23	50	73.00±17.13	107.50±17.90	8.83±2.32	214.17±15.22	0/6	0/6	286.17	288.33
	24	100	70.50±16.96	76.67±6.59	12.50±2.07	170.33±18.00	0/6	1/6	275.50	274.67
5r	25	50	158.33±9.07	114.50±13.13	2.50±2.43	225.17±22.44	0/6	0/6	284.17	279.17
	26	100	103.83±10.11	100.83±11.75	4.00±3.03	233.33±22.65	0/6	0/6	279.17	276.00
5t	27	50	107.50±8.87	101.17±14.97	7.33±2.25	274.17±19.03	0/6	0/6	281.67	272.83
	28	100	79.67±11.48	76.67±6.59	8.50±2.81	257.67±34.37	0/6	0/6	277.33	287.00
Control (CMC)	29	0.5%	179.67± 18.67	219.00±17.56	0.83±0.75	281.67±41.09	0/6	0/6	410.33	394.17
Standard drug (Imipramine)	30	10	29.33±8.43	60.33±9.33	---	---	0/6	1/6	399.50	404.83
Standard drug (Diazepam)	31	2	---	---	9.33±3.08	177.33±45.18	0/6	0/6	228.00	239.67

Bold data represents the most active compounds in the series.

n=6 (Number of animals tested at each dose level); p < 0.05.

Control: Carboxy methyl cellulose (CMC, 0.5 % suspension).

Standard: Imipramine (10 mg/kg, b.w.) for antidepressant activity and Diazepam (2 mg/kg, b.w.) for anti-anxiety activity.

^a The reduction in time of immobility in FST and TST are an established way to evaluate effectiveness of antidepressants.

^b The number of entries is increased in anxiolytic agents and decrease in anxiogenic agents and the amount of time spent in closed arms is decreased in anxiolytic agents and increase in anxiogenic agents.

^c Neurotoxicity was evaluated in Rotarod test before dosing and after 1 hour of dosing (number of animals exhibiting toxicity/number of animals tested).

^d Neurotoxicity recorded in actophotometer test (Counts recorded in 10 minutes duration)

Neurotoxicity study

This study was used to evaluate the effect of the compounds on the CNS, employing various behavioral tests, such as rotarod, open-field/actophotometer, turning on flat surface and inclined plane tests in mice model (Vogel, 2002; Parasuraman, 2011).

Neuromuscular coordination study

The effect of the compounds on motor coordination, where relaxation of skeletal muscle is produced, was examined, as per method given by Dunham and Miya in 1957, with slight modification (Vogel et al., 2002).

Locomotor activity (actophotometer test)

Locomotor activities of the animals were used as an index of their mental alertness, which is affected (increased or decreased) by majority of the CNS acting drugs (Dhingra and Goyal, 2008; Tripathi et al., 2016; Upadhyay et al., 2017).

Acute toxicity study

The consequences (changes in skin and fur, behavior patterns, convulsions, tremors and death) of a single dose on a particular animal species was determined by acute toxicity testing. In this study, acute oral toxicity (LD₅₀) of two most active final derivatives (**5b** and **5k**) was performed as per guidelines laid by Organization for Economic Cooperation and Development (OECD) guideline No 423 “Acute Oral Toxicity - Acute Toxic Class Method” using known procedures (OECD, 2001).

Computational studies

Molecular docking simulations were used to predict binding affinity and binding orientations of the synthesized compounds to the MAO-A protein (PDB ID: 2Z5X), with the help of GLIDE program (Schrödinger, LLC, 2014), using known procedures (Tripathi et al., 2016; Upadhyay et al., 2017). QikProp module of Schrödinger software program was successfully employed for *in silico* prediction of ADME properties of the synthesized derivatives. Furthermore, for *in silico* toxicity prediction of the synthesized

compounds, two freely accessible computer programs (LAZAR and OSIRIS Property explorer) were employed. Among them, LAZAR provided a generic tool for predicting complex toxicological end points (like long-term toxicity, reproductive toxicity and carcinogenicity). OSIRIS program calculated various drug-related properties of chemical structures and risks of untoward effects, which are considered as an indication of drug-conform behavior (Klebe, 2000).

Statistical analysis

Statistical analysis was carried out using Graph Pad Prism 5.0 (Graph Pad Software, San Diego, CA). Experimental results are expressed as mean±SD, analyzed by one-way ANOVA followed by Dunnett’s test for the possible significance (P<0.05) between various groups.

RESULTS AND DISCUSSION

A total of fourteen 1,3,5-trisubstituted 2-pyrazoline derivatives (**5a-5t**) were prepared through Claisen-Schmidt condensation, followed by heterocyclization and substitution with 4-chlorobenzenesulfonylchloride using both, conventional and MAOS methods. The microwave assisted procedures were developed for the first time to synthesize the final 1,3,5-trisubstituted-2-pyrazoline derivatives. The research outcomes suggested that almost all the derivatives synthesized by this technique were obtained at a faster rate and in a better synthetic yield in comparison to conventional procedures. The prepared derivatives were analyzed by various physico-chemical (Table 1) and spectral techniques and results comply with the proposal. The IR spectra reflect some characteristic absorption bands in the corresponding regions to C=N str (1509-1612 cm⁻¹), N-H str (3449-3107 cm⁻¹) and C-H deform (1431-1354 cm⁻¹). In the third step derivatives, a characteristic peak of sulfonyl group was also observed in the IR spectra. ¹H-NMR spectra showed that two dissimilar methylene protons (H_a/H_b) were visible at δ 2.92–3.38 ppm, 3.70–3.93 which coupled themselves and with the me-

thine proton (H_x) at δ 6.67–7.03 in vicinity. The other values were in congruence with the corresponding aliphatic and aromatic protons.

The pharmacological results (Table 2 and Figure 2) demonstrated that number of entries in closed and open arms, in the elevated plus maze test, increased significantly with a lesser closed arm exploration time (Figure 2a and 2b) for Compound **5b** and Compound **5k** showed the best antidepressant action (Figure 2c and 2d) in FST and TST models at the

evaluated doses. The pharmacological actions of the prepared analogs were observed in a dose dependent manner, as the magnitude of the effects increased at higher tested doses (100 mg/kg b.w.). The structure activity relationship studies established that C-3 naphthalen-1-yl substitution and C-5 furan-2-yl substitution at 2-pyrazoline nucleus were crucial for the antidepressant activity (**5k**). However, a polar substitution of 4-hydroxyphenyl group at 3rd position (**5b**) was essential in eliciting anxiolytic activity.

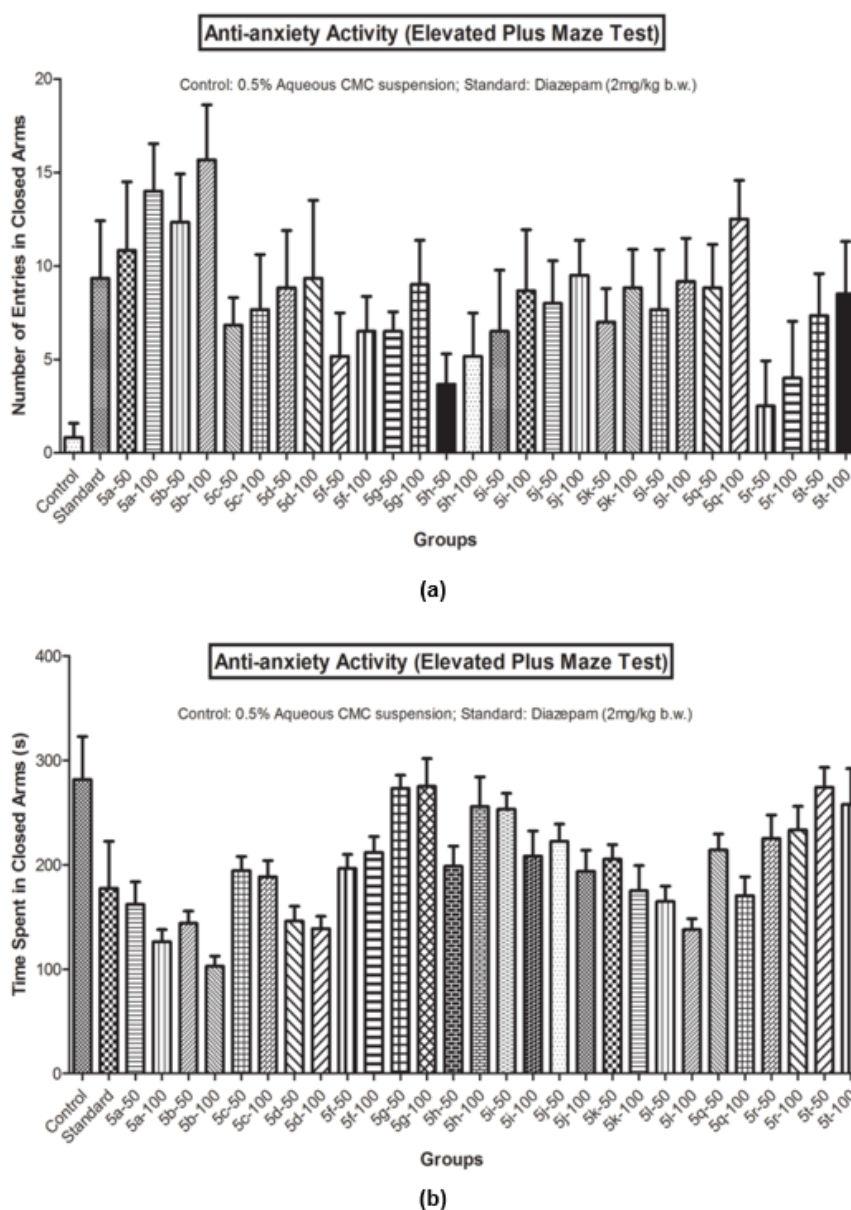


Figure 2: (a) Anti-anxiety activity (Number of entries in closed arms in Elevated Plus Maze Test); (b) Anti-anxiety activity (Time spent in closed arms in Elevated Plus Maze Test).

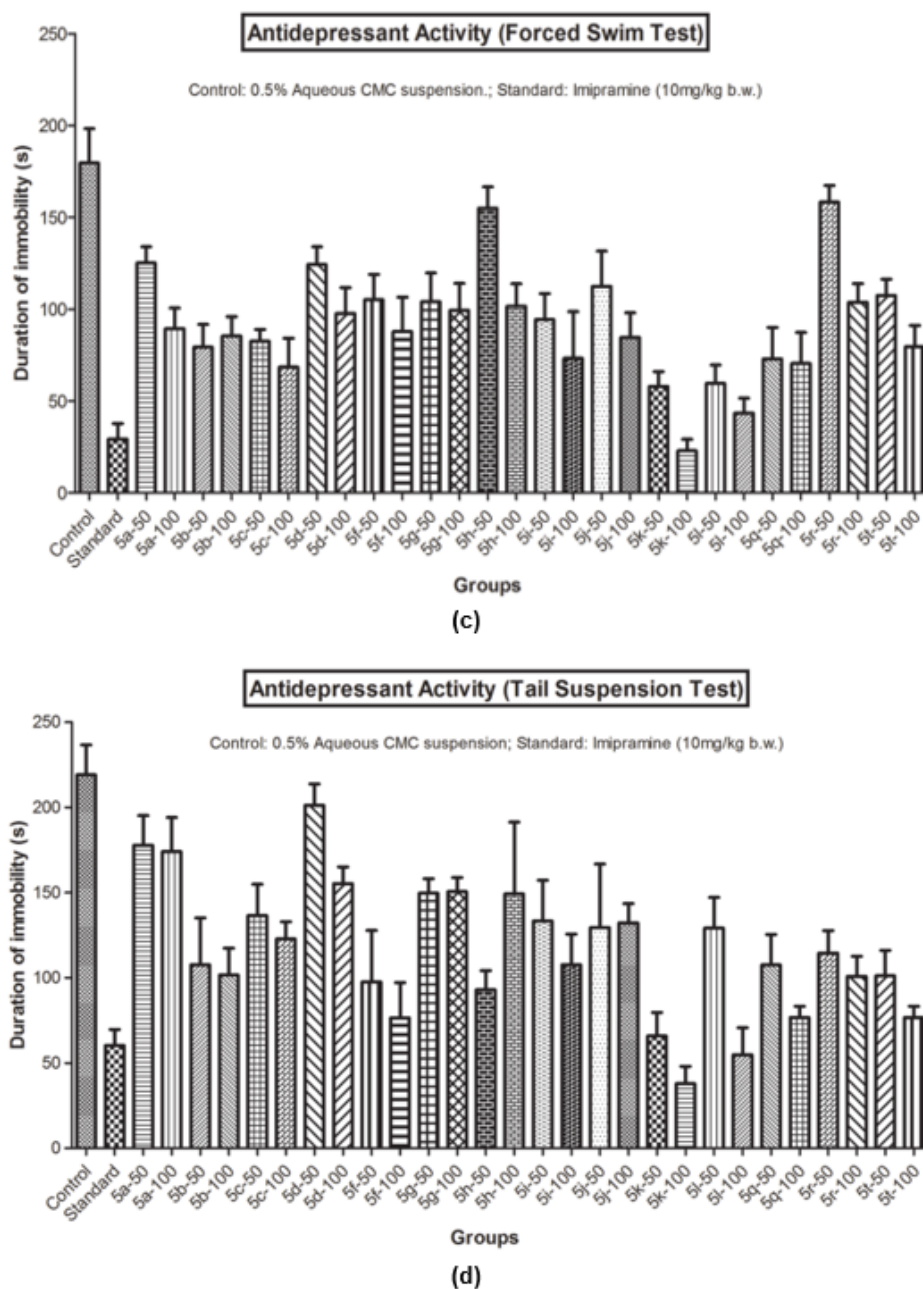


Figure 2 (cont.): (c) Antidepressant activity (Forced Swim Test); (d) Antidepressant activity (Tail Suspension Test)

MAOs are considered as an important biological target to develop drugs against certain mental disorders, such as depression and anxiety. It has been also established that the 2-pyrazoline derivatives possess excellent affinity towards MAO isoforms, as reported in previous literatures. Docking studies allowed to gain some structural inputs towards binding characteristics of the prepared derivatives with the MAO-A target

protein. The most stable conformation of ligand-protein complex and the interaction studies of the most active compounds, **5b** and **5k** at the MAO-A protein binding pocket have been depicted in Figure 3 and 4. Docking studies inferred that Ala 68, Tyr69 and Phe352 are some important residues interacting at the MAO-A binding pocket. It was evident that at 2-pyrazoline nucleus, N1 *p*-chlorobenzenesulfonyl substitution proved to

be very useful and all the compounds having this substitution showed better activity than the N1 unsubstituted pyrazolines (Tripathi et al., 2016). The presence of sulfonyl group at N1 position of 2-pyrazoline nucleus evidenced H-bonding interaction between sulfonyl oxygen and Ala68 and Tyr69 residues at the MAO-A binding pocket. It was demonstrated in previous works that ligands having such type of hydrogen bond interactions are well placed in the aromatic cage of the MAO protein possessing C5 furyl/ substituted phenyl ring and N1-benzenesulfonyl ring of 2-pyrazoline nucleus. The naphthyl/4-chlorophenyl rings showed, imperative pi-pi stacking interactions with Phe352 backbone residue. It was also evident that most of the interactions of PDB co-crystal ligand 2Z5X were conserved in compounds **5b** and **5k**. Furthermore, the predicted binding affinity (Glide gscores) of the prepared analogs complemented well with the *in vivo* anti-anxiety and antidepressant activities data.

The synthesized compounds were also tested for their probable neurotoxicological effects, such as motor coordination (rotarod test) and locomotor activity (actophotometer test). The obtained results suggested that these derivatives were completely free from severe neurotoxicity (motor co-ordinations and locomotor disturbances) threats at the evaluated doses (Table 2). In Rotarod test, the mean fall-off time of most of the mice was greater than 180 seconds, and hence these compounds may not affect the motor coordination of the animals. Also, the mean counts in actophotometer test in pre-dose studies, and after 1 hour of dosing, were not significantly different and therefore, any probability of CNS stimulating or depressing effects of the tested compounds was ruled

out. Additionally, most potent derivatives were evaluated for their acute toxicity at the corresponding doses. None of them showed any changes in behavioral pattern, convulsions, tremors, skin changes and death throughout the 14 days acute toxicity studies. Also, encouraging *in silico* pharmacokinetic properties (Table 3) were observed. Moreover, the screened compounds were not liable to cause carcinogenicity, mutagenicity, reproductive toxicity, acute toxicity and irritancy. Therefore, they were regarded to be safe as was corroborated by the computational programs such as LAZAR and OSIRIS (Table 4).

CONCLUSION

This study concludes that the synthesized 2-pyrazoline analogs possess excellent to good antidepressant and anti-anxiety potential, as evaluated using various *in vivo* methods. The presence of C-3 naphthalen-1-yl substitution and C-5 furan-2-yl substitution at 2-pyrazoline nucleus was decisive for the antidepressant activity (**5k**). However, a polar substitution of 4-hydroxyphenyl group at 3rd position (**5b**) was essential in eliciting anxiolytic activity. Nearly all the compounds were completely free from any neurotoxic indications, with a complementary pharmacokinetic behavior as predicted by *in silico* methods. Molecular docking experiments ascertained some imperative interactions of these compounds with the target protein, playing a pivotal role in neuropharmacology. Thus, in a nutshell, the synthesized 2-pyrazoline derivatives may prove their worth in the treatment and management of certain mental disorders, possibly through MAO-A inhibition.

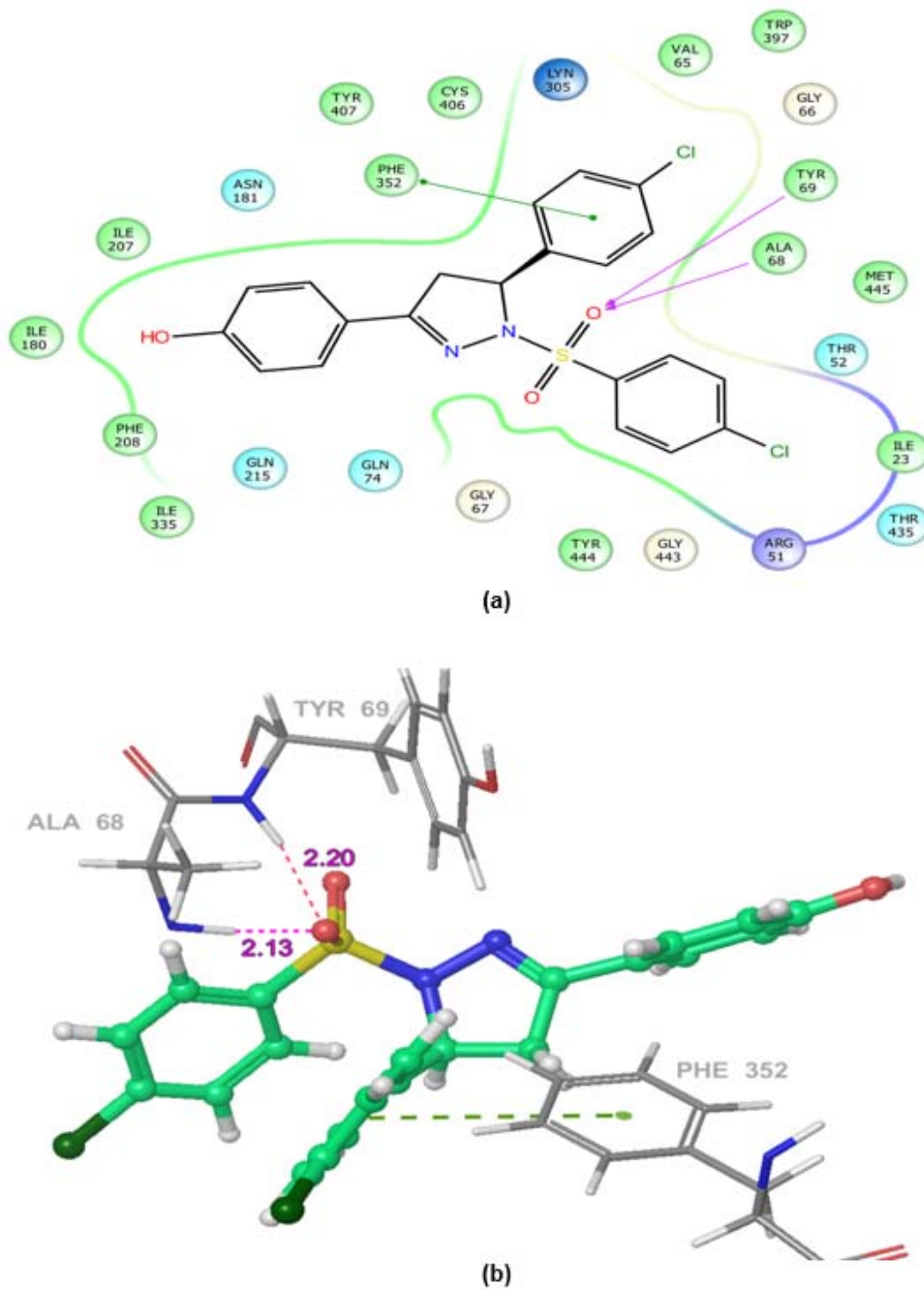


Figure 3: Ligand receptor interaction diagram of compound **5b** at the binding site of MAO-A protein (PDB ID: 2Z5X) showing best anti-anxiety activity. **(a)** 2D Ligand receptor interaction diagram. **(b)** 3D Ligand receptor interaction diagram.

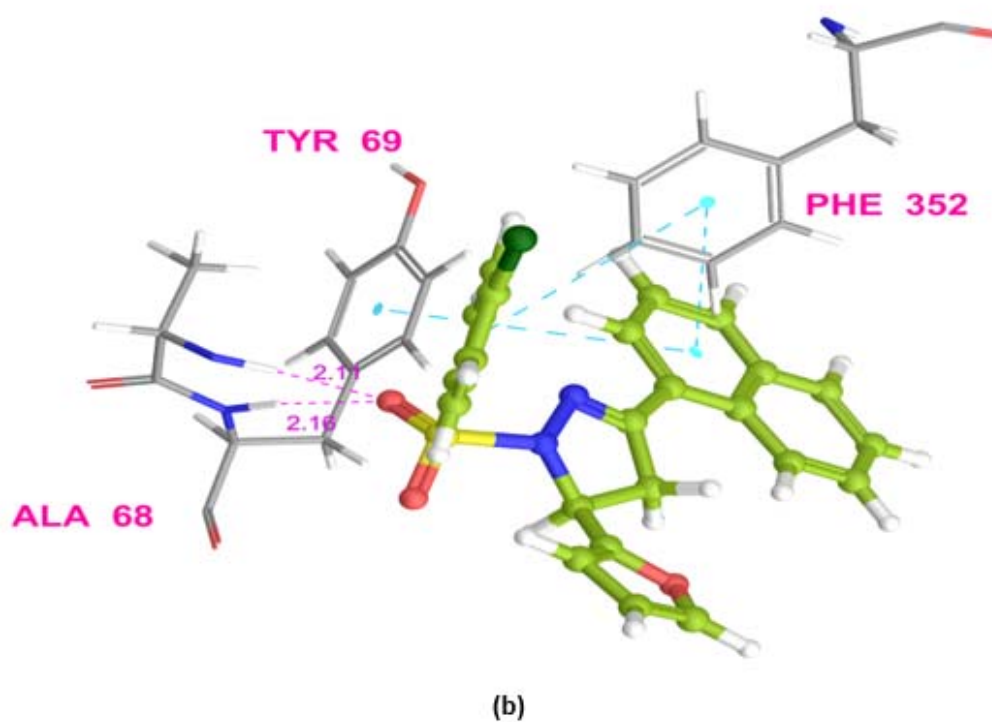
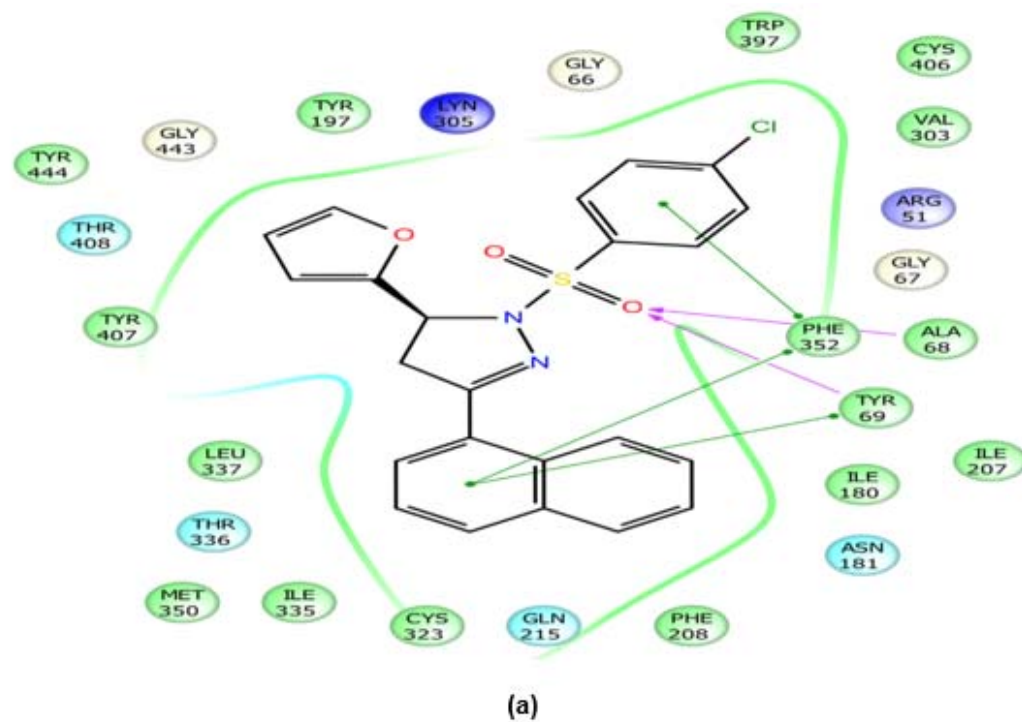


Figure 4: Ligand receptor interaction diagram of compound **5k** at the binding site of MAO-A protein (PDB ID: 2Z5X) showing best antidepressant activity. **(a)** 2D Ligand receptor interaction diagram. **(b)** 3D Ligand receptor interaction diagram.

Table 3: *In silico* prediction of binding affinity (Glide gscore) and ADME parameters of the synthesized 1,3,5-trisubstituted-2-pyrazoline derivatives (**5a-5t**)

Com-pounds	Glide gscore	#Stars	MW	Volume	PSA	SASA	Donar-HB	Ac-cptH B	CNS	QPlog BB	QlogP o/w	QPlog Khsa	QPP Caco	Site of metab	% Human Oral absorption	Violations of Rule of Five
5a	-7.36	0	472.94	1272.98	86.63	666.85	1	7.75	-1	-0.80	3.70	0.35	582.26	5	100	0
5b	-8.91	0	447.34	1184.44	73.71	639.41	1	6.25	0	-0.53	4.08	0.50	557.41	3	100	0
5c	-8.75	0	402.85	1094.53	82.24	602.32	1	6.75	0	-0.68	3.09	0.16	520.10	4	93.64	0
5d	-8.10	0	418.91	1130.25	74.61	622.66	1	6.25	0	-0.60	3.65	0.35	538.82	4	100	0
5f	-7.73	1	465.78	1205.86	51.17	651.08	0	5.50	1	0.20	5.09	0.56	1836.33	2	100	1
5g	-8.82	0	421.30	1115.91	59.71	613.96	0	6.00	1	0.05	4.04	0.17	1713.80	3	100	0
5h	-7.80	1	437.36	1151.77	52.08	634.43	0	5.50	1	0.14	4.66	0.41	1775.53	3	100	0
5i	-8.78	0	507.00	1389.36	64.30	720.45	0	7.00	0	-0.21	5.16	0.61	2166.91	4	90.92	2
5j	-9.06	1	481.40	1294.00	51.34	690.76	0	5.50	1	0.078	5.579	0.82	2146.71	2	100	1
5k	-9.24	1	436.91	1204.01	59.87	652.93	0	6.00	0	-0.09	4.52	0.42	1972.42	3	100	0
5l	-8.36	0	452.97	1240.41	52.23	673.58	0	5.50	1	0.01	5.14	0.66	2055.46	3	100	1
5q	-7.20	0	457.93	1240.78	76.47	652.37	0	8.00	-1	-0.36	3.46	-0.09	1384.40	5	100	0
5r	-8.59	0	432.32	1152.67	63.55	624.66	0	6.50	0	-0.10	3.92	0.14	1315.15	3	100	0
5t	-8.12	0	403.90	1099.01	64.46	608.64	0	6.50	0	-0.17	3.48	-0.02	1263.71	4	100	0
2Z5X	-6.89	0	212.25	733.39	33.50	381.35	1	1.75	0	0.18	3.11	0.222	840.62	3	100	0
Std. values	---	0-5	130 to 725	500 to 2000	7 to 200	300 to 1000	0 to 6	2 to 20	-2 to +2	-3.0 to 1.2	-2.0 to 6.5	-1.5 to 1.5	<25poor >500 great	1 to 8	<20% low, >80% high	Maximum is 4

#stars: Number of property or descriptor values that fall outside the 95 % range of similar values for known drugs; **Volume:** Total solvent accessible volume in cubic angstrom using a probe with 1.4 Å radius; **PSA:** Van der Waals polar surface area of nitrogen and oxygen atoms; **SASA:** Total solvent accessible surface area in square angstrom using a probe with 1.4 Å radius; **donarHB:** Hydrogen bond donar; **acctHB:** Hydrogen bond acceptor; **CNS:** Predicted central nervous system activity at -2 (inactive) to +2 (active) scale; **QPlogBB:** Predicted brain/blood partition coefficient; **QlogPo/w:** Predicted octanol-water partition coefficient. **QPlogKhsa:** Prediction of binding to human serum albumin; **QPPCaco:** Predicted apparent Caco-2 cell permeability in nm/sec; **metab:** Number of likely metabolic reactions; Number of violations of Lipinski's rule of five.

Table 4: *In silico* toxicity prediction data of the synthesized 1,3,5-trisubstituted -2-pyrazoline derivatives (**5a-5t**)

Compounds	Toxicity prediction							
	OSIRIS PROPERTY EXPLORER				LAZAR			
	Mutagenic	Tumorigenic	Irritant	Reproductive effect	Maximum recommended daily dose (mmol)	Mutagenicity	Acute toxicity LC ₅₀ (mmol)	Carcinogenic potency
5a	-	-	-	-	4.23e+00	-	5.51e+00	-
5b	-	-	-	-	4.62e+00	-	1.38e+01	-
5c	-	-	-	-	1.44e+01	-	2.31e+01	-
5d	-	-	-	-	1.50e+01	-	2.61e+01	-
5f	-	-	-	-	6.86e+00	-	8.78e+00	-
5g	-	-	-	-	1.25e+01	-	1.01e+01	-
5h	-	-	-	-	1.30e+01	-	9.11e+00	-
5i	+	-	-	-	4.52e+00	-	4.10e+00	+
5j	+	-	-	-	8.67e+00	-	7.05e+00	-
5k	+	-	-	-	1.47e+01	-	8.28e+00	-
5l	+	-	-	-	1.37e+01	-	1.03e+01	+
5q	-	-	-	-	1.02e+01	-	3.03e+00	-
5r	-	-	-	-	4.24e+01	-	5.34e+00	-
5t	-	-	-	-	2.16e+02	-	7.31e+00	+

No risk: (-); Medium risk: (+); High risk: (++)

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Conflict of interest

No conflict of interest is reported by the authors.

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