

Predictions and correlations of structure activity relationship of some aminoantipyrine derivatives on the basis of theoretical and experimental ground

Parvez Ali · Jyotsna Meshram · Javed Sheikh ·
Vandana Tiwari · Rajendra Dongre · Taibi Ben Hadda

Received: 1 February 2010 / Accepted: 3 November 2010 / Published online: 7 December 2010
© Springer Science+Business Media, LLC 2010

Abstract A series of 4-aminoantipyrine derivatives were produced by condensing aromatic primary amine, namely, 4-aminoantipyrine with different aryl carbonyls which occurred cleanly and efficiently without using any catalyst at room temperature. Their structures were confirmed on the basis of ^1H NMR, IR, Mass spectra, and elemental analysis. The synthesized library was screened virtually using versatile bioinformatics parameters owing to the quick access, biological response, and utility of the produced moieties. The virtually screened molecules were subjected to screen practically against certain some strain of microorganisms: *Staphylococcus aureus* (Gram positive), *Pseudomonas vulgaris* (Gram positive), *Pseudomonas Aeruginosa* (Gram negative), and *Escherichia coli* (Gram negative). A correlation of structure and activities relationship of these compounds with respect to molecular modeling, Lipinski rule of five, drug-likeness, toxicity profiles, and other physico-chemical properties of drugs is described and verified experimentally.

Keywords Azomethines · Synthesis · Molecular properties · Screening

Introduction

Pyrazolone moiety is a five-membered lactam ring containing two nitrogens and ketone in the same molecule or alternatively a derivative of pyrazole possessing an additional carbonyl/hydroxy group. It has been the focus of medicinal chemists over the past ten decades because of the outstanding pharmacological properties shown by several of its derivatives (Kees *et al.*, 1996), e.g., ampyrone, metamizole, etc. Ludwig Knorr discovered antipyrine. Owing to its promising antipyretic and analgesic activities antipyrine was launched by Hoechst Pharmaceuticals. For the next 20 years, antipyrine became the most widely used drug in the world, proving highly successful for treating fever and flu-like infections, until acetylsalicylic acid (aspirin) began to outsell it (Goder, 1985). Recently, a new pyrazolone compound, edaravone (3-methyl-1-phenyl-2-pyrazolin-5-one, also known as MCI-186), has been developed as a promising drug for brain ischemia and has also been reported to be effective for myocardial ischemia (Watanabe *et al.*, 1994; Kawai *et al.*, 1997). More recently, a series of pyrazolone derivatives have been synthesized as potent inhibitors of protease-resistant prion protein accumulation for the treatment of fatal neurodegenerative diseases (Tarafder *et al.*, 2002). Antipyrine shows minimal protein binding and is rapidly and completely absorbed from the gastrointestinal tract and extensively metabolized by the cytochrome P450 liver enzymes. Estimates of half-life and systemic clearance of antipyrine have been used for the in vivo assessment of hepatic drug oxidation in different

P. Ali (✉) · J. Meshram · V. Tiwari · R. Dongre
Department of Chemistry, Rashtrasant Tukadoji Maharaj,
Nagpur University, Nagpur 440033, India
e-mail: parvezali_81@yahoo.com

J. Meshram
e-mail: drjsmeshram@rediffmail.com

J. Sheikh
National Environmental Engineering and Research Institute,
CSIR, Nagpur 440010, India

T. B. Hadda
Laboratoire de Chimie des Matériaux, Université Mohammed
Premier, 60000 Oujda, Morocco

species (Kimata *et al.*, 2007). Owing to its low pK_a value and small degree of plasma protein binding, antipyrine is distributed in total body water. In view of their high medicinal value and due to our interest in the synthesis of compounds of potential pharmacological interest (Pericherla *et al.*, 2007; Pal *et al.*, 2007; Jayaselli *et al.*, 2008), we became interested in constructing a library based on pyrazolone scaffold. Azomethine belong to a widely used group of organic intermediates important for production of specialty chemicals, e.g., pharmaceuticals, or rubber additives (Macho *et al.*, 2004) and as amino protective groups in organic synthesis (Bey and Vevert, 1977; Lucas *et al.*, 1960; Bezas and Zervas, 1961). They also used as liquid crystals, (Adams, 2000) and in analytical (Layer, 1963) medicinal (Jarrahpour *et al.*, 2004) and polymer chemistry (Higuchi and Yamamoto, 1999). Based on the significant biological and pharmacological properties the pyrazolone moiety possesses a new class of such compounds is reported by combining the chemistry of antipyrine with azomethine with varying functionalities and to explore their biological activities with the aim of obtaining more potent antibacterial compounds. These reported molecules in this article formulate a new class of antibacterial agents that may become excellent candidates for globally alarming drug resistance issues in clinically used therapeutics. There is considerable interest in these compounds as combined antibacterial agents, exhibiting potency similar pharmacophore pockets to that observed for Ampicillin. Several groups have reported on the failed attempts to get a clear idea concerning the origin of each activity in this standard reference, casting doubt over the structural needs assignment. We present here the results of our virtual screening investigation into possible alternative structures for these compounds.

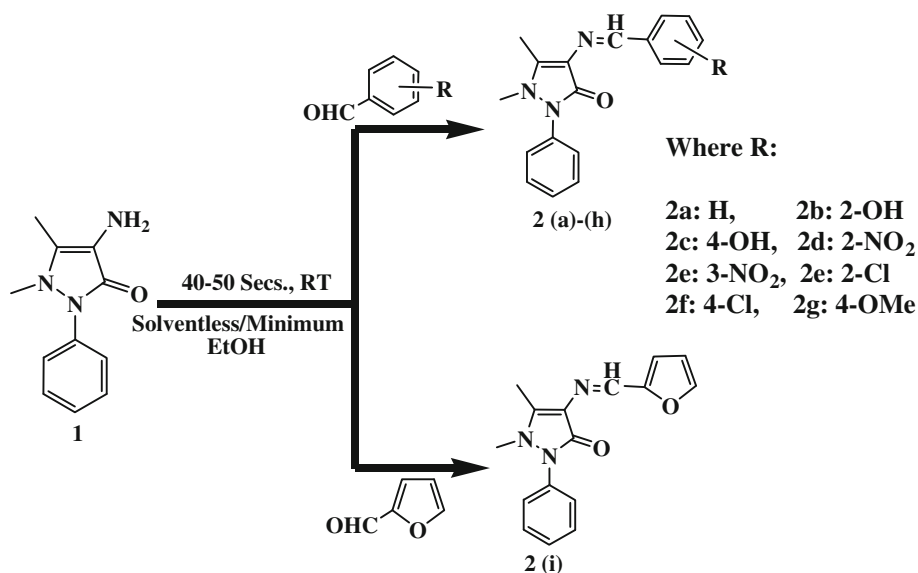
A comparison between experiment and theoretical predictions of the antibacterial activity has enabled us to identify alternative combined pharmacophore sites structures. The structural assignment of the synthesized compounds was based on its ^1H NMR, IR, Mass, and Elemental analysis.

Result and discussions

Chemistry

The azomethine derivatives **2a–2i** were synthesized in good yields (85–95%) by condensation of 4-aminoantipyrine with various substituted aromatic aldehydes in solid state solvent less or minimum solvent conditions as shown in Scheme 1. The structures of the title compounds were determined by IR, ^1H NMR, mass spectrometry, and elemental data. The spectroscopic properties and analytical data were in accord with the proposed structures. The ^1H NMR spectrum for compounds **2a–2i** showed a single peak for the azomethine $-\text{CH}=\text{N}$ proton, which varied from 9.65 up to 9.77 ppm. Such values are very similar to those found in the literature for the azomethine proton. The ^1H NMR spectra of **2a–i** contained multiplet signals due to aromatic protons in the 6.93–7.90 ppm regions. The IR spectra provide valuable information regarding the nature of functionalities present in the compounds. The bands around $1,600\text{--}1,610\text{ cm}^{-1}$ suggested the presence of azomethine linkage in the compounds. The other signals and peaks of ^1H NMR and IR are in complete agreement with the assigned structures, and they are listed in the experimental section.

Scheme 1 Solvent-free synthesis of Antipyrine derivatives



Virtual screenings and molecular properties calculations

Molinspiration calculations (Parvez et al., 2010a)

CLogP (octanol/water partition coefficient) is calculated by the methodology developed by Molinspiration as a sum of fragment-based contributions and correction factors. The method is very robust and is able to process practically all organic and most organometallic molecules. Molecular polar surface area TPSA is calculated based on the methodology as a sum of fragment contributions. O- and N-centered polar fragments are considered. PSA has been shown to be a very good descriptor characterizing drug absorption, including intestinal absorption, bioavailability, Caco-2 permeability and blood–brain barrier penetration. Prediction results of compounds and molecular properties (TPSA, GPCR ligand, and ICM) are valued (Tables 1, 2). Lipophilicity (log P value) and polar surface area (PSA) values are two important predictors of per oral bioavailability of drug molecules (Clark, 1999; Chang et al., 2004). Therefore, we calculated log P and PSA values for compounds **2a–2i** using mol inspiration software programs and compared them to the values obtained for standard market available drugs. For all compounds the calculated log P values were lower than 5, which is the upper limit for drugs to be able to penetrate through biomembranes according to Lipinski's rules. The polar surface area (PSA) is calculated from the surface areas that are occupied by oxygen and nitrogen atoms and by hydrogen atoms attached to them. Thus, the PSA is closely related to the hydrogen bonding potential of a compound (Clark, 1999). Molecules with PSAs of 140 Å or more are expected to exhibit poor intestinal absorption (Clark, 1999). Table 1 show that all the compounds are within this limit with compounds **2b**, **2c**, and **2i** is having minimum comparable values of log P and

PSA. This is also supported by the antibacterial screening data of compounds **2b** and **2i** in terms of maximum zone of inhibitions. It has to be kept in mind that log P and PSA values are only two important, although not sufficient criteria for predicting oral absorption of a drug. To support this contention, note that all the compounds have zero violations of the Rule of 5. The Rule of 5 is a set of parameters devised to aid the screening of potential drug “hits” identified through processes such as high throughput screening (Lipinski et al., 2001). Applying the Rule of 5 increases the probability that a potential chemotherapeutic will have favorable bioavailability. The criteria are as follows (Lipinski et al., 2001): (A) Not more than 5 hydrogen bond donors; (B) Not more than 10 hydrogen bond acceptors; (C) Formula weight less than 500; and (D) Log P less than 5. Two or more violations of the Rule of 5 suggest the probability of problems in bioavailability (Lipinski et al., 2001). All the compounds have zero violations of the Rule of 5. Drug-likeness of compounds **3a–3i** is tabulated in Table 4. Drug-likeness may be defined as a complex balance of various molecular properties and structure features which determine whether particular molecule is similar to the known drugs. These properties, mainly Hydrophobicity, electronic distribution, hydrogen bonding characteristics, molecule size and flexibility and presence of various pharmacophores features influence the behavior of molecule in a living organism, including bioavailability, transport properties, affinity to proteins, reactivity, toxicity, metabolic stability, and many others. Activity of all nine compounds and standard drugs were rigorously analyzed under four criteria of known successful drug activity in the areas of GPCR ligand activity, ion channel modulation, kinase inhibition activity, and nuclear receptor ligand activity. Results are shown for all compounds in Tables 1 and 2 by means of numerical assignment. Likewise all compounds have consistent negative values in all categories and

Table 1 Molinspiration calculations of molecular properties and drug-likeness of compounds (**2a–2i**)

Compd.	Molecular properties calculations						Drug-likeness			
	MW (g/mol)	cLogP	TPSA	OH–NH interact.	N violation	Volume	GPCRL	ICM	KI	NRL
2a	291	2.86	39	0	0	273	–1.05	–1.24	–1.29	–1.27
2b	307	2.80	60	1	0	281	–0.97	–1.27	–1.32	–1.09
2c	307	2.38	60	1	0	281	–0.97	–1.15	–1.21	–1.03
2d	336	2.767	85	0	0	296	–1.22	–1.24	–1.44	–1.14
2e	336	2.79	85	0	0	296	–1.08	–1.22	–1.27	–1.22
2f	326	3.49	39	0	0	286	–1.05	–1.25	–1.32	–1.24
2g	326	3.54	39	0	0	286	–1.00	–1.17	–1.23	–1.29
2h	321	2.91	49	0	0	298	–0.96	–1.21	–1.19	–1.14
2i	281	2.12	52	0	0	254	–1.07	–1.35	–1.54	–2.18

GPCRL GPCR ligand, ICM Ion channel modulator, KI Kinase inhibitor, NRL Nuclear receptor ligand

numerical values conforming and comparable to that of standard drugs used for comparison. Therefore, it is readily seen that all the analogs are expected to have near similar activity to standard drugs used based upon these four rigorous criteria (GPCR ligand, ion channel modulator, kinase inhibitor, and nuclear receptor ligand).

Osiris calculations (Parvez et al., 2010b) Structure based design is now fairly routine but many potential drugs fail to reach the clinic because of ADME-Tox liabilities. One very important class of enzymes, responsible for many ADMET problems, is the cytochromes P450. Inhibition of these or production of unwanted metabolites can result in many adverse drug reactions. Of the most important program, Osiris is already available online. With our recent publication of the drug design combination of various pharmacophore sites by using heterocyclic structure Parvez et al., (2010a, b) it is now possible to predict activity and/or inhibition with increasing success in two targets (bacteria and HIV virus). This is done using a combined electronic/structure docking procedure and an example will be given here. The remarkably well behaved mutagenicity of diverse synthetic molecules classified in data base of CELERON Company of Switzerland can be used to quantify the role played by various organic groups in promoting or interfering with the way a drug can associate with DNA. Toxicity risks (mutagenicity, tumorigenicity, irritation, reproduction) and physicochemical properties (ClogP, solubility, drug-likeness and drug-score) of compounds (**2a–2i**) are calculated by the methodology developed by Osiris as a sum of fragment-based contributions and correction factors (Tables 3, 4). The toxicity risk predictor locates fragments within a molecule which indicate a potential toxicity risk. Toxicity risk alerts are an indication that the drawn structure may be harmful concerning the risk category specified. From the data evaluated in Tables 3 and 4 indicates that, all structures are supposed to be mutagenic when run through the mutagenicity assessment system but as far as irritating and reproductive effects are concerned, all the compounds are at low risk comparable with standard drugs used. The log P value of a compound, which is the logarithm of its partition coefficient between n-octanol and water, is a well-established measure of the

compound's Hydrophilicity. Low Hydrophilicity and therefore high log P values may cause poor absorption or permeation. It has been shown for compounds to have a reasonable probability of being well absorb their log P value must not be greater than 5.0. On this basis, all the compounds **2a–2i** is having log P values under the acceptable criteria. Along with this, compound **2b** and **2i** which have shown good antibacterial screening results is having low log p values as compared to other compounds in the series. The aqueous solubility of a compound significantly affects its absorption and distribution characteristics. Typically, a low solubility goes along with a bad absorption and therefore the general aim is to avoid poorly soluble compounds. Our estimated log S value is a unit stripped logarithm (base 10) of a compound's solubility measured in mol/liter. There are more than 80% of the drugs on the market have an (estimated) log S value greater than -4 . In case of compounds **2b** and **2i**, values of log S are low as compared to others in the series. Further, the Table 3 shows drug-likeness of compounds **2a–2i** which is in the comparable zone with that of standard drugs used. We have calculated overall drug-score (DS) for the compounds **2a–2i** and compared with that of standard drugs Ampicillin and streptomycin used as shown in Table 3 and 4. The DS combines drug-likeness, ClogP, logs, molecular weight, and toxicity risks in one handy value that may be used to judge the compound's overall potential to qualify for a drug. The reported compounds **2a–2i** showed moderate to good DS as compared with standard drugs used.

Pharmacology

Antibacterial activity (in vitro) (Parvez et al., 2010a) The antibacterial activity of the series (**2a**)–(**2i**) has been carried out against some strain of bacteria. To determine the antibacterial activity of these agents, Agar cup plate method was used, with Ampicillin and Streptomycin as the reference antibiotics. The prepared compounds were examined against two strains each of Gram-positive and Gram-negative bacteria. The test results, presented in Table 5, suggest that compounds **2a**, **2c**, and **2i** are highly active against two strains each of Gram-positive and Gram-negative bacteria showing the broadest spectrum of

Table 2 Molinspiration calculations of molecular properties and drug-likeness of Ampicillin and Streptomycin

Compd.	Molinspiration calculations						Drug-likeness			
	MW g/mol	cLogP	TPSA	OH–NH interact.	N violation	Volume	GPCRL	ICM	KI	NRL
AMP	349	−0.87	112.73	4	0	299	−0.56	−0.55	−0.90	−0.87
STREP	582	−5.35	336	16	3	497	−0.67	−1.15	−0.76	−1.11

Table 3 Osiris calculations of compounds (**2a–2i**)

Compd.	Toxicity risks				Osiris calculations				
	MUT	TUMO	IRRI	REP	MW	CLP	Log S	DL	D-S
2a	–	–	+	+	291	1.96	–3.24	5.05	0.31
2b	–	–	+	+	307	1.66	–2.94	5.00	0.32
2c	–	–	+	+	307	1.66	–2.94	5.26	0.32
2d	–	–	+	+	336	1.83	–3.70	–2.10	0.16
2e	–	–	+	+	336	1.82	–3.7	–0.11	–0.22
2f	–	–	+	+	325	2.57	–3.98	5.44	0.28
2g	–	–	+	+	325	2.57	–3.98	5.83	0.28
2h	–	–	+	+	321	1.85	–3.25	5.23	0.31
2i	–	–	+	+	281	1.06	–2.92	5.00	0.32

Active risk = –, Non-active risk = +

Table 4 Osiris calculations of Ampicillin and Streptomycin

Compd.	Toxicity risks				Osiris calculations				
	MUT	TUMO	IRRI	REP	MW	CLP	Log S	DL	D-S
AMP	+	+	+	+	349	–0.04	–1.57	10.72	0.91
STREP	+	+	±	+	581	–7.83	–0.96	0.83	0.43

AMP Ampicillin, STREP Streptomycin, MUT Mutagenic, TUMO Tumorigenic, IRR Irritant, REP Reproductive Effective, MW Molecular Weight in g/mol, CLP ClogP, S Solubility, DL Drug-Likeness, D-S Drug-Score

Non-active risk = +, Average = ±

Table 5 Antibacterial activity of compounds (**2a–2i**)

Compd.	MW (g/mol)	Gram (+) bacteria		Gram (–) bacteria	
		A	B	C	D
2a	368	+++	++	–	–
2b	384	+++	+++	+++	++
2c	384	++	–	+	+
2d	413	++	–	–	+
2e	413	+	–	+	–
2f	402	+++	+++	+++	+++
2g	402	++	–	–	+
2h	398	+	++	+	–
2i	358	+++	+++	+++	+++
AMP	349	+++	++	++	+++
STREP	581	+++	+++	+++	+++

AMP Ampicillin, STREP Streptomycin, A *S.aureus*, B *P.vulgaris*, C *E.coli*, D *P.aeruginosa*. Inactive = – (inhibition zone < 5 mm), slightly active = + (inhibition zone 5–10 mm), moderately active = ++ (inhibition zone 10–15 mm), highly active = +++ (inhibition zone > 15 mm)

antibacterial activity. The rest of the compounds were found to be moderately active, slightly active, or inactive against the tested microorganisms. The results show that the prepared compounds are toxic against the bacteria.

Table 6 MIC of selected compounds

*Compd.	Gram (+) bacteria		Gram (–) bacteria	
	A	B	C	D
2b	50	75	75	100
2f	75	100	100	100
2i	50	75	75	75
AMP	25	25	25	25

AMP Ampicillin, STREP Streptomycin, A *S.aureus*, B *P.vulgaris*, C *E.coli*, D *P.aeruginosa*

* Concentrations were in µg/ml

Minimum inhibitory concentration (MIC) was defined as the lowest compound concentration preventing visible bacterial growth. MICs of selected compounds (activity over 80%) **2b**, **2f**, and **2i** were determined by taking different concentrations of the compound in DMF. The different concentrations were added by using sterilized pipettes to different test tubes containing sterilized broth medium inoculated with test organism. Then all the test tubes were incubated at 37°C for 24 h. Then after incubation period, the presence of growth (turbidity) was observed. It was observed that the MIC of selected compounds was in the range 50–100 µg/ml. This is the MIC of the compounds. Out of three selected compounds, **2i** was shown to have least MIC values as tabulated in Table 6.

Experimental

General

The solvents and reagents used in the synthetic work were of laboratory grade purchased from Qualigens, India and were purified by distillation or crystallization where necessary and their boiling or melting points were compared with the available literature values. Melting points were determined in open capillaries and are uncorrected. ^1H NMR spectra were recorded on a Perkin–Elmer FT-NMR Cryo-magnet Spectrometer 400 MHz (Bruker) instrument using tetramethylsilane (TMS) as an internal standard and CDCl_3 as a solvent. Chemical shifts are given in parts per million (ppm). Infrared spectra were recorded on Shimadzu-IR Prestige 21 in the range 400–4,000 cm^{-1} . Mass spectra were recorded on a Waters Micromass Q-T of Micro spectrometer. The purity of products was checked out on pre-coated TLC plates (Silica gel 60 F254, Merck), visualizing the spots under ultraviolet light and iodine chamber. Elemental analyses were carried out using a Perkin–Elmer, CHNS/O elemental analyzer model 2400.

Antimicrobial screening

The agar cup plate method using Hi-Media agar medium was employed to study the antibacterial activity of **2a–2i** against *S. aureus*, *p. vulgaris*, *P. aeruginosa*, and *E. coli*. Preparation of nutrient broth, subculture, base layer medium, agar medium, and peptone water was done as the standard procedure. 10 mg of the test compound was dissolved in 10 ml of DMF. From this 10 ml of solution was taken and diluted to 100 ml with DMF. Now the concentration of the test compound is 100 $\mu\text{g}/\text{ml}$. These sample solutions were made in suitably labeled sterilized test tubes. The standard drugs used in this testing were Ampicillin and Streptomycin. The concentrations of these drugs were adjusted so as to contain 100 $\mu\text{g}/\text{ml}$. The tests results are shown in Table 5.

Minimum inhibitory concentrations of selected compounds were determined by taking different concentrations of the compound in DMF. The different concentrations were added by using sterilized pipettes to different test tubes containing sterilized broth medium inoculated with test organism. Then all the test tubes were incubated at 37°C for 24 h. Then after incubation period, the presence of growth (turbidity) was observed. The results obtained are tabulated in Table 6.

General procedure for the synthesis of azomethines

In a typical solvent less experiment, the aldehyde, namely (0.1 M) furfuraldehyde was added to 4-aminoantipyrine

(0.1 M) contained in a beaker and the mixture was scratched with glass rod (ca. 10 s.), affording an oil (ranging from cream to dark brown in color). On further scratching (ca. 30 s.) the dark yellow solid azomethine was formed Parvez *et al.* 2010b, c. If required, analytically pure product can be obtained by recrystallization (from methanol). Same process was followed for the generation of other azomethines except for compound **2c**, **2d**, **2e**, and **2g** where aldehydes were dissolved in a minimum amount of ethanol followed by the addition of 4-aminoantipyrine. This compound was obtained as light brown solid, Yield: (85%), M.P 195°C. IR (KBr, cm^{-1}): 1650 (C=O), 1600 (C=N), 1034 (N–N). ^1H NMR (CDCl_3): 2.60 (s, 3H, C–CH₃), 3.20 (s, 3H, N–CH₃), 6.33 (d, 1H, C–H, Furan), 6.30 (m, 3H, C–H, Furan), 7.40 (d, 1H, C–H, Furan), 7.12–7.55 (m, 5H, Phenyl H), 9.65 (s, 1H, –CH=N). MS: m/z: 304 (M+23). Anal. calcd. for $\text{C}_{16}\text{H}_{15}\text{N}_3\text{O}_2$: C, 68.31; H, 5.37; N, 14.94; O, 11.37. Found: C, 68.34; H, 5.40; N, 14.96; O, 11.35.

Data

4-((Benzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2a**)

Light yellow solid, Yield: (90%), M.P 162°C. IR (KBr, cm^{-1}): 1660 (C=O), 1610 (C=N), 1040 (N–N). ^1H NMR (CDCl_3): 2.50 (s, 3H, C–CH₃), 3.20 (s, 3H, N–CH₃), 6.98–7.20 (m, 10H, Phenyl), 9.70 (s, 1H, –CH=N). MS: m/z: 291 (M+23). Anal. Calcd. for $\text{C}_{18}\text{H}_{17}\text{N}_3\text{O}$: C, 74.20; H, 5.88; N, 14.42; O, 5.49. Found: C, 74.24; H, 5.35; N, 14.45; O, 5.50.

4-((2-Hydroxybenzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2b**)

Light orange solid, Yield: (92%). M.P 220°C. IR (KBr, cm^{-1}): 1658 (C=O), 1609 (C=N), 3200 (–O–H), N–N (1040). ^1H NMR (CDCl_3): 1.72 (s, 3H, C–CH₃), 2.55 (s, 3H, N–CH₃), 6.65–7.20 (m, 5H, N–Phenyl), 7.35–7.70 (m, 4H, C–Phenyl), 8.25 (s, 1H, –CH=N), 11.45 (s, 1H, –OH). MS: m/z: 325 (M+23). Anal. calcd. for $\text{C}_{18}\text{H}_{17}\text{N}_3\text{O}_2$: C, 70.34; H, 5.58; N, 13.67; O, 10.41. Found: C, 70.35; H, 5.60; N, 13.70; O, 10.45.

4-((4-Hydroxybenzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2c**)

Light orange solid, Yield: (90%). M.P 190°C. IR (KBr, cm^{-1}): 1658 (C=O), 1609 (C=N), 3200 (–O–H), N–N (1040). ^1H NMR (CDCl_3): 1.72 (s, 3H, C–CH₃), 2.55 (s, 3H, N–CH₃), 6.65–7.20 (m, 5H, N–Phenyl), 7.35–7.70 (m,

4H, C–Phenyl), 8.25 (s, 1H, –CH=N), 11.45 (s, 1H, –OH). MS: *m/z*: 325 (M+23). Anal. calcd. for C₁₈H₁₇N₃O₂: C, 70.34; H, 5.58; N, 13.67; O, 10.41. Found: C, 70.35; H, 5.60; N, 13.70; O, 10.45.

4-((2-Nitrobenzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2d**)

Orange solid, Yield: (88%). M.P 158°C. IR (KBr, cm⁻¹): 1660 (C=O), 1614 (C=N), 1040 (N–N). ¹H NMR (CDCl₃): 2.45 (s, 3H, C–CH₃), 3.18 (s, 3H, N–CH₃), 7.30–7.90 (m, 10H, Aromatic H), 9.85 (s, 1H, –CH=N). Mass (*m/z*): [M+23]; 359 (100%). Anal. calcd. for C₁₈H₁₆N₄O₃: C, 64.28; H, 4.79; N, 16.66; O, 14.27. Found: C, 64.30; H, 4.82; N, 16.70; O, 14.30.

4-((3-Nitrobenzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2e**)

Orange Solid, Yield: (91%). M.P 162°C. IR (KBr, cm⁻¹): 1658 (C=O), 1615 (C=N), 1040 (N–N). ¹H NMR (CDCl₃): 2.40 (s, 3H, C–CH₃), 3.15 (s, 3H, N–CH₃), 7.30–7.89 (m, 10H, Aromatic H), 9.85 (s, 1H, –CH=N). Mass (*m/z*): [M+23]; 359 (100%). Anal. calcd. for C₁₈H₁₆N₄O₃: C, 64.28; H, 4.79; N, 16.66; O, 14.27. Found: C, 64.34; H, 4.85; N, 16.75; O, 14.35.

4-((2-Chlorobenzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2f**)

Yellow solid, Yield: (89%). M.P 165°C. IR (KBr, cm⁻¹): 1655 (C=O), 1605 (C=N), N–N (1038). ¹H NMR (CDCl₃): 2.47 (s, 3H, C–CH₃), 3.18 (s, 3H, N–CH₃), 6.97–7.82 (m, 9H, Phenyl), 9.77 (s, 1H, –CH=N). MS: *m/z*: 348 (M+23). Anal. Calcd. for C₁₈H₁₆ClN₃O: C, 66.36; H, 4.95; N, 12.90; O, 4.91. Found: C, 66.43; H, 4.98; N, 12.99; O, 4.97.

4-((4-Chlorobenzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2g**)

Light orange solid, Yield: (92%). M.P 230°C. IR (KBr, cm⁻¹): 1652 (C=O), 1608 (C=N), 1038 (N–N). ¹H NMR (CDCl₃): 2.45 (s, 3H, C–CH₃), 3.15 (s, 3H, N–CH₃), 6.95–7.85 (m, 9H, Phenyl), 9.70 (s, 1H, –CH=N). MS: *m/z*: 348 (M+23). Anal. Calcd. for C₁₈H₁₆ClN₃O: C, 66.36; H, 4.95; N, 12.90; O, 4.91. Found: C, 66.40; H, 4.99; N, 12.98; O, 4.98.

4-((4-Methoxybenzylideneamino-2-yl) methyleneamino)-1,2-dihydro-2,3-dimethyl-1-phenylpyrazol-5-one (**2h**)

Yellow solid, Yield: (94%). IR (KBr, cm⁻¹): 1660 (C=O), 1610 (C=N), 3200 (O–CH₃), N–N (1045). ¹H NMR

(CDCl₃): 2.50 (s, 3H, C–CH₃), 3.13 (s, 3H, N–CH₃), 3.85 (s, 3H, O–CH₃), 6.93–7.87 (m, 9H, Phenyl), 9.69 (s, 1H, –CH=N). MS: *m/z*: 344 (M+23). Anal. Calcd. for C₁₉H₁₉N₃O₂: C, 71.01; H, 5.96; N, 13.08; O, 9.96. Found: C, 71.04; H, 5.99; N, 13.10; O, 9.98.

Conclusions

The functionalized compounds **2a–2i** can easily be prepared. The compounds were varied to possess a broad range of lipophilicity character, revealed by Log P values less than 5. All compounds were determined to express zero violations to the Rule of 5, hence an indication of favorable bioavailability based on drug-likeness. The considerable number of hydrogen donor/acceptor atoms incurred significant hydrophilic character into the majority of these drugs (supported by low CLP values). Comparing relative activity scores of Ampicillin and streptomycin to those of standard drugs utilizing four drug classes (GPCR ligand, ion channel modulator (kinase inhibitor, and nuclear receptor ligand) showed all compounds are very highly correlated with expected similar bio-activity. In general, these compounds typically could form the highly interesting combined two or more pharmacophore sites in one molecule. It has been suggested that some functional groups along with heterocyclic system present in these compounds displayed role of biological activity that may be responsible for the increase of hydrophobic character and liposolubility of the molecules. This in turn enhances activity of the compounds and biological absorbance, so as, all the Synthesized compounds have good antibacterial properties with compounds **2b** and **2i** shows highest screening points with **2i** is shown to have least MIC values. These results prompt several pertinent observations (Pal *et al.*, 2007; Parvez *et al.*, 2010b): (i) This type of derivatives can furnish an interesting model for studying the interaction of antibiotics with viral target because the possible charge modification of substituent and O/N/S of pharmacophore groups; (ii) The future flexible pharmacophore site (s) geometric conformation enables us to prepare molecules for multi-therapeutic materials with high selectivity.

Acknowledgments Authors are thankful to UGC [F-33-301/2007(SR)], New Delhi for financial assistance to carry out this study. We are grateful to SAIF Punjab University, Chandigarh for the help in undertaking NMR, Mass spectra and Dr. Reddy's Laboratory for undertaking IR spectra. Special thanks are due to the Head, Department of Microbiology, Rashtrasant Tukadoji Maharaj Nagpur University, Nagpur for antimicrobial screening. Prof. Jyotsna Meshram would like to thank in particularly Dr. Johann Gasteiger for the online molecular properties calculations.

References

- Adams JP (2000) Nitro and related compounds. *J Chem Soc Perkin Trans 1*:3075–3695
- Bey P, Vevert JP (1977) Synthesis of alpha-alkyl and alphafluoroalkylised methyl-alpha-amino acids. *Tetrahedron Lett* 18:1455–1458
- Bezaz B, Zervas LJ (1961) On the Peptides of L-Lysine. *J Am Chem Soc* 83:719–722
- Chang LCW, Spanjersberg RF, von Frijtag Drabbe K, unzel JK, Mulder-Krieger T, van den Hout G, Beukers MW, Brussee J, IJzerman AP (2004) 2,4,6-Trisubstituted pyrimidines as a new class of selective adenosine A₁ receptor antagonists. *J Med Chem* 47:6529–6540
- Clark DE (1999) Rapid calculation of polar molecular surface area and its application to the prediction of transport phenomena. *J Pharm Sci* 88:807–814
- Goder K (1985) Zur Einführung synthetischer Schlafmittel in die Medizin. *M, vol. 18*. Bern, New York
- Higuchi M, Yamamoto K (1999) Novel cyclic molecules: selective synthesis of cyclic phenylazomethines. *Org Lett* 1:1881–1883
- Jarrahpour A, Motamedifar M, Pakshir K, Hadi N, Zarei M (2004) Synthesis of novel azo schiff bases and their antibacterial and antifungal activities. *Molecules* 9:815–824
- Jayaselli J, Cheemala JMS, Rani DPG, Pal S (2008) Derivatization of enolic OH of piroxicam: a comparative study on esters and sulfonates. *J Braz Chem Soc* 19:509–515
- Kawai H, Nakai H, Suga M, Yuki S, Watanabe T, Saito KI (1997) Effects of a novel free radical scavenger, MCI-186, on ischemic brain damage in the rat distal middle cerebral artery occlusion model. *J Pharmacol Exp Ther* 281:921–927
- Kees KL, Fitzgerald JJ, Steiner KE, Mattes JF, Mihan B, Tosi T, Mondoro D, McCalebr ML (1996) New potent antihyperglycemic agents in db/db mice: synthesis and structure–activity relationship studies of (4-substituted benzyl)(trifluoromethyl)pyrazoles and pyrazolones. *J Med Chem* 39:3920–3928
- Kimata A, Nakagawa H, Ohyama R, Fukuuchi T, Ohta S, Suzuki T, Miyata N (2007) New potent antihyperglycemic agents in db/db mice: synthesis and structure–activity relationship studies of (4-substituted benzyl)(trifluoromethyl)pyrazoles and pyrazolones. *J Med Chem* 50:5053–5056
- Layer RW (1963) The chemistry of imines. *Chem Rev* 63:489–510
- Lipinski CA, Lombardo F, Dominy BW, Feeney PJ (2001) Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Adv Drug Deliv Rev* 46:3–26
- Lucas RA, Dickel DF, Dziemian MJ, Hensle BL, McPhillarnay HB (1960) Some hypotensive amino steroid glycosides. *J Am Chem Soc* 82:5688–5693
- Macho V, Kralik M, Hudec J (2004) One stage preparation of Schiff's bases from nitroarenes, aldehydes and carbon monoxide at presence of water. *J Mol Catal A Chem* 209:69–73
- Pal S, Bindu P, Venna PR, Dubey PK (2007) Novel naproxen derivatives: lewis acid/transition-metal free synthesis via C–C bond forming reaction. *Lett Org Chem* 4:292–296
- Parvez A, Jyotsna M, Tiwari V (2010a) Towards a novel approach to bis-β-lactam synthesis using Vilsmeier reagent as an efficient entity via Staudinger cycloaddition reaction. *J Heterocyclic Chem (in Press)*
- Parvez A, Jyotsna M, Tiwari V (2010b) Zeolite as an efficient and recyclable activation surface for the synthesis of Bis-thiazolidinones: theoretical screening owing to. *Experimental Biology* 3(3):195–200
- Parvez A, Pagadala R, Jyotsna M (2010c) Exploring microwave synthesis for co-ordination: synthesis, spectral characterization and comparative study of transition metal complexes with binuclear core derived from 4-amino-2, 3-dimethyl-1-phenyl-3-pyrazolin-5-one. *J coord chem* 63(2):323–329
- Pericherla S, Mareddy J, Geetha R, Gollapudi PV, Pal S (2007) Chemical modifications of nimesulide. *J Braz Chem Soc* 18:384–389
- Tarafder MTH, Jin KT, Crouse KA, Ali AM, Yamin BM, Fun HK (2002) Coordination chemistry and bioactivity of Ni²⁺, Cu²⁺, Cd²⁺ and Zn²⁺ complexes containing bidentate NS Schiff bases derived from S-benzylidithiocarbamate and the X-ray crystal structure of bis[S-benzyl-β-N-(5-methyl-2-furylmethylene) dithiocarbamate]cadmium(II). *Polyhedron* 21(25–26):2547–2554
- Watanabe T, Yuki S, Egawa M (1994) Nishi H. Protective effects of MCI-186 on cerebral ischemia: possible involvement of free radical scavenging and antioxidant actions. *J Pharmacol Exp Ther* 268:1597–1604