European Journal of Medicinal Chemistry 45 (2010) 5208–5216

Contents lists available at ScienceDirect

European Journal of Medicinal Chemistry

journal homepage: http://www.elsevier.com/locate/ejmech

Original article

Structure activity relationship studies of imidazo[1,2-a]pyrazine derivatives against cancer cell lines

Shailaja Myadaraboina [a], Manjula Alla [a,**], Venkateshwarlu Saddanapu [b], Vittal Rao Bommena [a,*], Anthony Addlagatta [b,***]

[a] Organic Chemistry Division-II, Indian Institute of Chemical Technology, Tarnaka-uppal road, Tarnaka, Hyderabad 500607, India
[b] Center for Chemical Biology, Indian Institute of Chemical Technology, Tarnaka-uppal road, Tarnaka, Hyderabad 500607, India

ARTICLE INFO

Article history:
Received 21 April 2010
Received in revised form
11 August 2010
Accepted 11 August 2010
Available online 18 August 2010

Keywords:
Inhibitor design
Synthesis
Imidazo[1,2-a]pyrazine
Anticancer activity
Structure activity relationship

ABSTRACT

Designed novel imidazo[1,2-a]pyrazine based inhibitors, synthesized by condensing α-aminopyrazines with α-halocarbonyl compounds followed by electrophilic substitutions. Cytotoxic effects on four cancer cell lines evaluated. Based on preliminary data, imidazo[1,2-a]pyrazine template redesigned to improve activity.

1. Introduction

Discovery of new drugs for treatment of cancer has been gaining a great deal of interest mainly due to a universal resistance to conventional single drug chemotherapeutic agents. Multidrug resistance [1] characterized by resistance not only to drugs that are similar structurally and functionally but also cross-resistance to unrelated drugs like doxorubicin, vincristine, vinblastine, colchicine and actinomycin has been documented. Thus, search for novel anticancer agents with diverse chemical structure is need of the hour. Herein, we report the synthesis and evaluation of a series of imidazo[1,2-a]pyrazines as potent anticancer agents.

Imidazo[1,2-a]pyrazines have been gaining attention in drug discovery realm especially as structural analogues of purines [2–4] (Fig. 1). Derivatives of imidazo[1,2-a]pyrazines exhibit various pharmacological activities such as antibacterial [5], anti-inflammatory [6–8], uterine relaxing activity [9], antibronchospastic [10], antiulcer [11], cardiac stimulating [12], antidepressant [13], hypoglycemic activity [14], antiproliferative activity [15], controlling allergic reactions [16], smooth muscle relaxant properties [17] and phosphodiesterase inhibitory activity [18a]. They have also been shown to inhibit the receptor tyrosine kinase EphB4 recently [19].

2. Rationale for designing

The imidazo[1,2-a]pyrazine is an interesting skeleton to design inhibitors with variable substitutions because of its planar shape. While C3, C5 and C6 form one edge of the plane, and the N7, C8 and N1 form the other. The C2 is positioned at the middle where these two edges meet. Note that N1 and N7 are hydrogen bond acceptors. Larger substitutions on C8 may be deleterious on the activity due probably to affecting the hydrogen bonding capacity of N1 and N7. We decided not to modify edge 2. On the edge1, C3 (R_3) and C6 (R_6) are selected for modification. In addition we wanted to explore the role of the substitutions on C2 (R_2) in conjunction with the variation on the face1 on the cytotoxic effects. As a starting set, we have designed and synthesized imidazo[1,2-a]pyrazine derivatives with various modifications (Table 1). The substitution on C6 is limited to only methyl group as it is close to N7, which may have effect on the hydrogen bonding capacity of this molecule. Bulky substitutions were placed on C2 expecting that they may explore the spatial volume of the target receptor on either side. Substitutions on C3

* Corresponding author. Tel.: +91 040 27193150; fax: +91 040 27193382.
** Corresponding author.
*** Corresponding author.
E-mail addresses: manjula@iict.res.in (M. Alla), vittalrao@iict.res.in (V.R. Bommena), anthony@iict.res.in (A. Addlagatta).

0223-5234/$ – see front matter © 2010 Elsevier Masson SAS. All rights reserved.
doi:10.1016/j.ejmech.2010.08.035

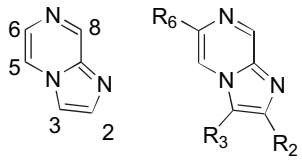


Fig. 1. Schematic diagram of imidazo[1,2-a]pyrazine skeleton. In the left panel, the IUPAC numbering is provided and in the right panel the substitution position is identified.

also include large variations since it is far from hydrogen bond forming atoms (Table 2, entries 1–11).

3. Synthetic chemistry

A series of substituted imidazo[1,2-a]pyrazines have been synthesized with substitutions at 2,3,6-ring positions being varied generating mono-,di-,trisubstituted imidazo[1,2-a]pyrazines possessing functional groups like halo, hydroxymethyl, amine, alkyl, aryl, heteroaryl etc.

A generally established method for synthesis of 3-substituted imidazo[1,2-a]pyrazine derivatives is by condensation of α-halogenocarbonyl compounds with aminopyrazines [20–22]. Accordingly, a series of 3-substituted imidazo[1,2-a]pyrazines were made by condensing 2-aminopyrazine (**1a**) and 2-amino-5-methylpyrazine (**1b**) with 3-bromoacetophenones or halo substituted carbonyls (**2a**–**i**) (Scheme 1). In all reactions, the products i.e, 2-substituted imidazo[1,2-a]pyrazines (**3a**–**i**) were obtained in moderate to good yields. Of the acyl bromides, 4-flourophenacyl bromide was very effective and the corresponding 2-(4-fluorophenyl)-6-methyl imidazo[1,2-a]pyrazine was obtained in high yield (70%). The condensation with heterocyclic acyl bromides was sluggish and low yielding. Amongst the two pyrazines used, 2-amino-5-methylpyrazine (**1b**) was more reactive and corresponding products were obtained in higher yields. All products were completely characterized before proceeding further.

Table 1
Yields and physical properties of Imidazo[1,2-a]pyrazine derivatives synthesized.

Compound	R_6	R_2	R_3	Yield (%)	M.p. (°C)
3a	CH_3	Ph	H	50	123–125
3b	CH_3	4-$C_6H_4CH_3$	H	67	173–175
3c	CH_3	4-C_6H_4F	H	70	138–140
3d	CH_3	*t*-Butyl	H	65	66–68
3e	CH_3	3-Coumarinyl	H	30	108–110
3f	CH_3	1-((2-Nitrophenoxy) methyl)phenyl	H	45	90–92
3g	H	C_6H_5	H	40	100–102
3h	H	4-$C_6H_4CH_3$	H	52	104–106
3i	H	4-C_6H_4F	H	55	92–94
4a	CH_3	Ph	Br	87	105–107
4b	CH_3	4-$C_6H_4CH_3$	Br	86	72–75
4c	CH_3	4-C_6H_4F	Br	74	172–174
4d	CH_3	3-Coumarinyl	Br	70	Liquid
4e	CH_3	*t*-Butyl	Br	68	70–72
4f	H	Ph	Br	57	78–82
4g	H	4-C_6H_4F	Br	65	154–156
4h	H	4-C_6H_4Cl	Br	89	103–105
4i	H	*t*-Butyl	Br	73	85–87
5a	CH_3	Ph	CN	51	162–164
5b	CH_3	4-C_6H_4F	CN	60	187–190
5c	CH_3	1-((2-Nitrophenoxy) methyl)phenyl	CN	46	68–70
5d	H	4-C_6H_4F	CN	50	158–160
5e	H	4-C_6H_4Cl	CN	48	146–148
6a	CH_3	3-Coumarinyl	CH_2OH	45	134–136
6b	CH_3	1-((2-Nitrophenoxy) methyl)phenyl	CH_2OH	48	150–152
7a	CH_3	Ph	Piperidinyl	66	148–150
7b	CH_3	4-C_6H_4F	Morpholinyl	72	140–145
7c	H	4-$C_6H_4CH_3$	Morpholinyl	56	138–140

A series of electrophilic substitutions were performed on 2-substituted imidazo[1,2-a]pyrazines (**3**) to obtain 2,3-disubstituted imidazo[1,2-a]pyrazines (**4**). Since the ring is highly deactivated towards electrophiles, the reactions gave moderate yields of corresponding products. To begin with bromination was attempted and NBS/Ethanol was found to be ideal method [13,23] when compared to direct bromination with molecular bromine (Scheme 1).

The nitrile substitution was achieved by nucleophilc attack on the bromo group of 3-bromo-2-substituted imidazo[1,2-a]pyrazines (**4**) with CuCN in DMF [24] leading to 3-cyano-2-aryl imidazo[1,2-a]pyrazines in moderate yields(**5a**–**e**) (Scheme 1).

Hydroxymethylation was carried out on 2-substituted imidazo [1,2-a]pyrazines (**3**) employing formaldehyde, acetic acid and sodium acetate [25] (Scheme 2). The corresponding 3-hydroxymethyl-2-substituted imidazo[1,2-a]pyrazines (**6a**–**b**) were obtained in moderate yields.

Amination of the 2-substituted imidazo[1,2-a]pyrazines (**3**) occurred at 3-position as shown in Scheme 3 and was found to proceed with better yields compared to hydroxymethylation [26,27]. All newly synthesized compounds were completely characterized by their spectral data (compounds listed in Table 1).

4. Pharmacological screening

All the compounds were characterized by the methods described in Section 5. Cytotoxic effects of these compounds were tested against four different cancer cell lines and their activities are summarized in the Table 2. Interesting results were observed (entries 1–11). Compound **4c** exhibited the most cytotoxic effects on all the four cells lines with best against MCF-7 and SK-N-SH as 13.0 and 13.8 μM, respectively. The next best molecule in this series is **5c** with activities close to 100 μM. All the other compounds displayed much lower efficacies. Compounds **4c** and **5b** are identical except for the substitution on C3, a bromo in the former and the cyano in the later. Altogether, the data point out that bromo substitution is better than the cyano on C3, when the other substituents are identical. Note that *p*-fluorophenyl is the preferred substituent among all the others on C2. The role of methyl group on C6 is not clear, since except for **7c** all the other molecules have it.

To further explore the role of substituents on C2 and C6 while it is either bromo or cyano on the C3, we have synthesized (Table 2, entries 12–23) and evaluated several more derivatives. Surprisingly, removing the p-fluoro substitution on the C3-phenyl group in the compound **4a**, improved the activity by more than ten-fold against all cell lines except in MCF-7, where it is only two-fold improvement. Several derivatives differ by the C6-methyl group. However, no systematic variation has been seen in the activities based on presence or absence of C6-methyl group (Table 2). A fluoro to chloro substitution between **4g** and **4h**, the efficiency of cytotoxicity improves by about eight folds in two cell lines while it remains almost the same in other two (Table 2). Of the four new C3-cyano derivatives only one i.e., **5d** displayed appreciable efficacy against HEPG-2 cell lines.

To summarize, we have designed, synthesized and evaluated imidazo[1,2-a]pyrazine derivatives systematically by varying substitutions on C2, C3 and C6 and established a structure activity relationship. Although, the role of C6 methyl is still not clear, substitution of bromo on C3 and a phenyl on the C2 seems to be important for their activity against cancer cell lines. Further studies are required to identify the target protein, which will enable the design of more specific and efficient molecules.

Table 2
Inhibition values (IC_{50}) of Imidazo[1,2-a]pyrazines on human tumor cell lines.

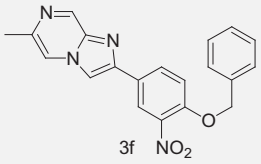
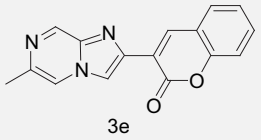
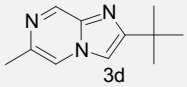
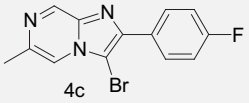
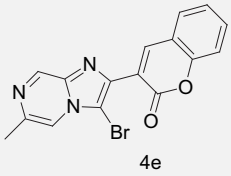
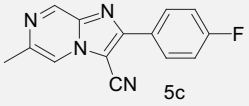
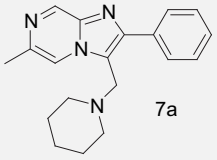
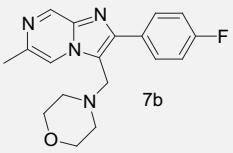
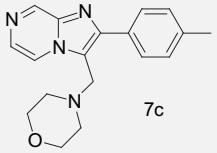
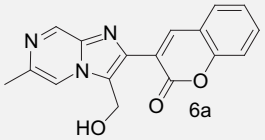
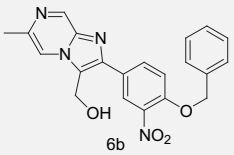
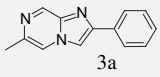
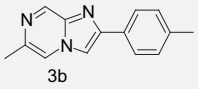
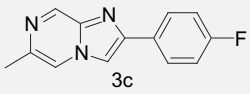
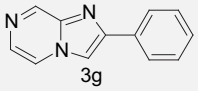
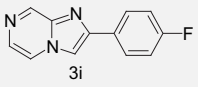
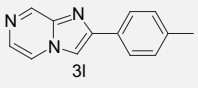
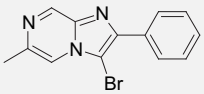
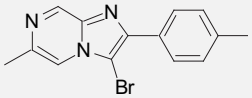
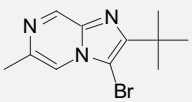
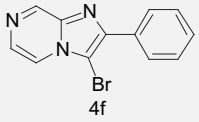
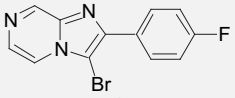
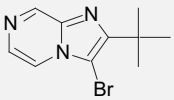
Entry	Compound	IC_{50} values against cancer cell lines (μM)			
		MDAMB-231	SK-N-SH	HepG-2	MCF-7
1	 3f	>1000	154.6	129.8	900.0
2	 3e	N/A	>1000	102.0	>1000
3	 3d	N/A	95.7	992.0	>1000
4	 4c	191.8	13.8	615.0	12.9
5	 4e	>1000	101.6	900.0	730
6	 5c	116.0	103.8	99.5	77.6
7	 7a	107.6	106.7	798.0	98.3
8	 7b	217.5	162.3	871.0	232.8
9	 7c	112.9	126.0	>1000	121.6

Table 2 (*continued*)

Entry	Compound	IC_{50} values against cancer cell lines (μM)			
		MDAMB-231	SK-N-SH	HepG-2	MCF-7
10	 6a	124.5	145.9	853.0	232.6
11	 6b	N/A[a]	91.6	>1000	>1000
12	 3a	>1000	9.6	>1000	>1000
13	 3b	>1000	10.8	>1000	>1000
14	 3c	104.0	159.2	106.6	96.32
15	 3g	>1000	>1000	>1000	960.0
16	 3i	135.9	117.9	737.0	140.8
17	 3l	>1000	>1000	103.9	>1000
18	 4a	15.2	1.0	61.1	6.2
19	 4b	168.9	9.5	85.1	24.9
20	 4d	171.2	123.2	123.0	107.8

(*continued on next page*)

Table 2 (*continued*)

Entry	Compound	IC_{50} values against cancer cell lines (μM)			
		MDAMB-231	SK-N-SH	HepG-2	MCF-7
21	 4f	142.4	91.1	108.7	555.0
22	 4g	101.3	80.6	87.6	81.1
23	 4j	111.3	113. 9	190.0	282.4
24	Doxorubicin	2.3	1.1	0.954	2.4

[a] N/A indicates that the activity could not be measured.

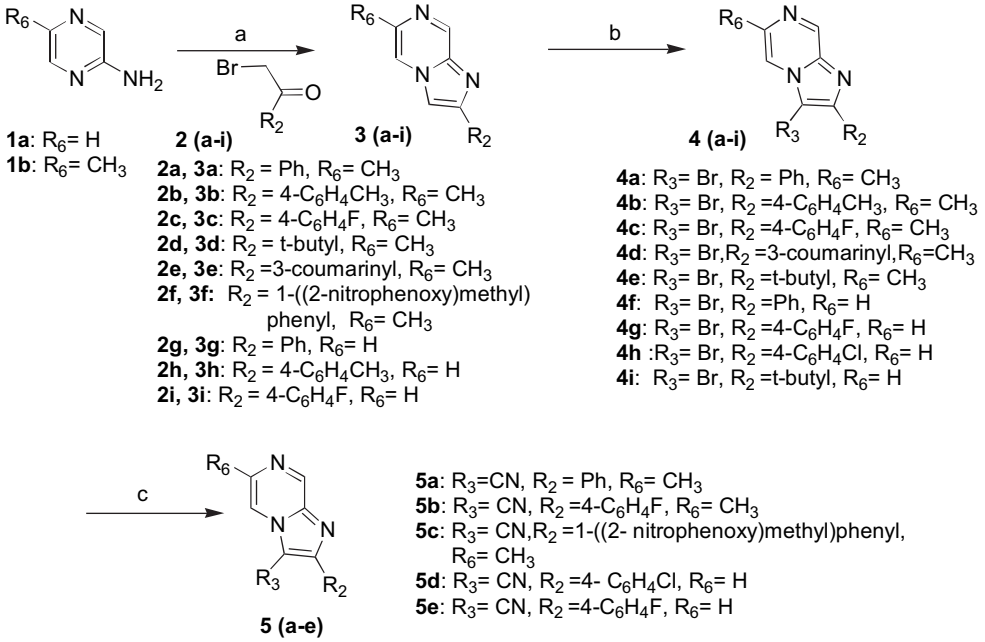
5. Experimental section

5.1. Chemistry

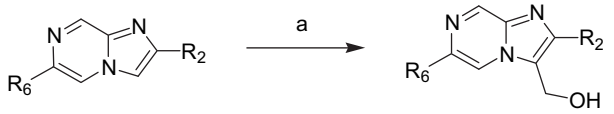
Melting points were determined in open capillary tubes with a Cintex Industrial Corporation melting point apparatus and are uncorrected. IR spectra were obtained on a Perkin-Elmer FTIR spectrophotometer neat or as KBr pellets. ^{1}H NMR spectra were determined on Avance 300 MHz instrument. Chemical shifts are given in δ values referenced to the solvent. Mass spectra were recorded on Waters quadruple mass spectrometry. HRMS was recorded on high resolution QSTAR XL Hybrid/MS system. Analytical thin layer chromatography (TLC) was performed on Merck Silica gel 60 F254 plates. Phenacyl bromide and 4-methylphenacyl bromide purchased from Spectrochem chemicals, India. 3-Nitro-2-benzyloxyphenacyl bromide [29] and 5-methyl-2-amino pyrazine were prepared in the laboratory [30]. Rest of the acyl bromides and 2-aminopyrazine was purchased from Sigma–Aldrich.

5.1.1. General procedure for substituted 2-phenylimidazo[1,2-a] pyrazines (**3a–i**)

To a stirred solution of 2-aminopyrazine (1 mmol) in 30 mL of ethanol, phenacyl bromide (1 mmol) and $NaHCO_3$ (3 mmol) were added and heated to reflux. After completion of the reaction (monitored by TLC), the reaction mixture was cooled to room temperature. The solid was filtered from reaction mixture and



Scheme 1. Synthesis of 2-/2, 3-di substituted imidazo[1,2-a]pyrazine (**3a–3i**, **4a–4i**, **5a–5e**). Reagents and conditions: (a) acyl bromide, $NaHCO_3$, EtOH, reflux; (b) *N*-bromo-succinimide, EtOH, RT; and (c) CuCN, DMF, reflux.



3e:R_2 =3-coumarinyl, R_6= CH_3
3f: R_2 =1-((2-nitrophenoxy)methyl)phenyl, R_6= CH_3

6a: R_2 =3-coumarinyl, R_6= CH_3
6b: R_2 =1-((2-nitrophenoxy)methyl)phenyl, R_6= CH_3

Scheme 2. Synthesis of 3-hydroxymethyl-2-substituted imidazo[1,2-a]pyrazine. Reagents and conditions: (a) formaldehyde, acetic acid, acetic anhydride, 60 °C.

ethanol was removed on rotovapor. The residue was extracted with ethyl acetate and washed with water, dried over anhydrous sodium sulphate, filtered and concentrated. Further purification by column chromatography using neutral alumina (hexane/ethyl acetate, 70/30) gave the desired product.

*5.1.1.1. 6-Methyl-2-phenylimidazo[1,2-a]pyrazine (**3a**).* Yellow solid, Yield: 50%; M.p.123–125 °C; IR (KBr, cm^{-1}): 3414, 3313, 2918, 1417,821, 744; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 9.13 (s, 1H, Ar), 8.21 (s, 1H, Ar), 8.17(s, 1H, Ar), 7.54–7.50 (m, 5H, Ar), 2.16 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 20.62, 108.47, 113.30, 127.45, 128.10, 128.44, 129.42, 131.56, 138.86, 141.73, 152.63; Mass (EI-MS): 209 $[M]^+$; HRMS: Observed value $C_{13}H_{11}N_3$ [M + 1]: 210.1024; Calculated Value: 210.1031.

*5.1.1.2. **6-**Methyl-2-p-tolylimidazo[1,2-a]pyrazine (**3b**).* Light yellow solid, Yield 52%; M.p.173–175 °C; IR (KBr, cm^{-1}): 3418, 2921, 1417, 1108, 825, 741; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ ppm): 8.95 (s, 1H, Ar), 7.80- 7.74 (m, 4H, Ar), 7 .21 (d, 2H, Ar, J = 8.210 Hz), 2.49 (s, 3H, CH_3), 2.40 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz):19.58, 20.15, 107.85, 114.86, 124.94, 128.35, 132.15, 136.94, 137.29, 140.59, 143.87, 152.56; Mass(ESI-MS): 223 $[M]^+$; HRMS: Observed value $C_{14}H_{13}N_3$ [M + 1]: 224.1184: Calculated Value: 224.1187.

*5.1.1.3. (4-Fluorophenyl)-6-methylimidazo[1,2-a]pyrazine (**3c**).* Yellow solid, yield: 58%; M.p.138–140 °C; IR (KBr, cm^{-1}): 3338, 3129, 2925, 1487, 1229, 841, 743; ^{1}H NMR: ($CDCl_3$ + DMSO, 300 MHz, δ): 9.07 (s,1H, Ar), 8.05 (s,1H, Ar), 8.1 (d, 2H, Ar, J = 7.932 Hz), 7.31 (d, 2H, Ar, J = 7.932 Hz), 7.25 (s, 1H, Ar), 2.61 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 20.25, 105.72, 113.60, 113.65, 115.62, 126.90, 127.75, 135.47, 141.26, 152.37, 158.90; Mass (ESI-MS): 228 $[M + 1]^+$; HRMS: observed value $C_{13}H_{10}N_3F$: 228.0948; calculated Value: 228.0937.

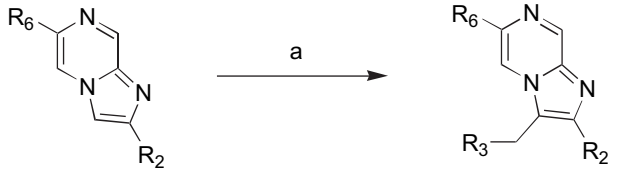
*5.1.1.4. 2-tert-Butyl-6-methylimidazo[1,2-a]pyrazine (**3d**).* Brown solid, Yield: 65%. M.p. 65–68 °C; IR (KBr, cm^{-1}): 3435, 2925, 1744, 816; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.87 (s, 1H, Ar), 7.74 (s, 1H, Ar), 7.29 (s, 1H, Ar), 2.45 (s, 3H, CH_3), 1.37 (s, 9H, t-bu); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 20.51, 29.97, 30.47, 108.37, 115.35, 125.39, 128.85, 137.72, 141.13; Mass (ESI-MS): 190 $[M + 1]^+$; HRMS: Observed value $C_{11}H_{15}N_3$ [M + 1]: 190.1352; Calculated Value: 190.1344.

*5.1.1.5. 3-(6-Methylimidazo[1,2-a]pyrazin-2-yl)-2H-chromen-2-one (**3e**).* brown solid, Yield: 30%. M.p.108–110 °C; IR (KBr, cm^{-1}): 3335, 2924, 1725, 1657, 1025, 874, 447; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ); 8.97 (s, 1H, Ar), 8.81 (s, 1H, Ar), 8.59 (s, 1H, Ar), 7.86 (s, 1H, Ar), 7.82 (d, 1H, Ar, J = 7.62 Hz), 7.32 (t, 1H, Ar, J = 8.34 Hz), 7.19 (d, 2H, Ar, J = 7.62 Hz), 2.39 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 22.34, 108.23, 114.84, 120.87, 122.67, 124.77, 125.86, 126.45, 127.88, 128.47, 128.99, 140.73, 144.12, 146.23, 152.56, 158.96; Mass (ESI-MS): 278 $[M + 1]^+$; HRMS: Observed value $C_{16}H_{11}N_3O_2$ [M + Na]: 300.0752; Calculated Value: 300.0748.

*5.1.1.6. 2-(4-(Benzyloxy)-3-nitrophenyl)-6-methylimidazo[1,2-a]pyrazine (**3f**).* Brown solid, yield 45%; M.p. 90–92 °C; IR (KBr, cm^{-1}): 3440, 2925, 1511, 1340, 1250, 983, 705; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ); 8.47 (s, 1H, Ar), 8.42 (s, 1H, Ar), 8.13 (s, 1H, Ar), 8.09 (s, 1H, Ar), 7.47–7.45 (m, 7H, Ar), 3.60 (s, 2H, OCH_2), 2.44 (s, 3H, CH_3); ^{13}CNMR ($CDCl_3$ + DMSO, 75 MHz): 26.64, 61.03, 109.50, 114.24, 115.08, 122.82, 126.40, 126.66, 128.00, 128.26, 129.31, 131.29, 132.45, 135.01, 141.09, 142.60, 154.88; Mass (ESI-MS): 361 $[M]^+$.

*5.1.1.7. 2-Phenylimidazo[1,2-a]pyrazine (**3g**).* Yellow solid, yield: 40%; M.p.100–102 °C; IR (KBr, cm^{-1}): 3337, 3014, 1658, 730, 432; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.98 (s, 1H, Ar), 8.30 (d, 1H, Ar, J = 3.77 Hz), 8.20 (s, 1H, Ar), 7.96 (d, 2H, J = 7.932 Hz), 7.81 (d, 1H, J = 4.532 Hz), 7.43 (t, 3H, Ar, J = 7.458 Hz); Mass (ESI-MS):196 $[M + 1]^+$.

*5.1.1.8. 2-p-Tolylimidazo[1,2-a]pyrazine (**3h**).* Yellow solid, yield 53%; M.p.104–106 °C; IR (KBr, cm^{-1}): 3336, 3144, 1658, 1531, 811, 510; ^{1}H NMR($CDCl_3$ + DMSO, 300 MHz, δ): 8.95 (s, 1H, Ar), 8.45 (d, 2H, Ar, J = 8.650 Hz), 8.13 (s, 1H, Ar), 7.40 (d, 2H, Ar, J = 8.210 Hz), 7.21 (d, 2H, Ar, J = 8.210 Hz), 2.30 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 20.91, 108.83, 118.52, 125.76, 127.13, 128.96, 129.08, 132.45, 136.89, 141.32, 155.00; Mass (ESI-MS): 210 $[M + 1]^+$; HRMS: observed value $C_{13}H_{11}N_{32}$[M + 1]: 210.1040; calculated value: 210.1031.



3a: R_2 = Ph, R_6= CH_3
3c: R_2 =4-C_6H_4F, R_6= CH_3
3h:R_2 =4-$C_6H_4CH_3$, R_6= H

7a: R_3= piperdinyl R_2 = Ph, R_6= CH_3
7b: R_3=morpholinyl R_2 =4-C_6H_4F, R_6= CH_3
7c: R_3= morpholinyl R_2 =4-$C_6H_4CH_3$, R_6= H

Scheme 3. Synthesis of 3-aminomethyl-2-substituted imidazo[1,2-a]pyrazine. Reagents and conditions: (a) secondary amine, formaldehyde, acetic acid, methanol, reflux.

*5.1.1.9. 2-(4-Fluorophenyl)imidazo[1,2-a]pyrazine (**3i**).* Yellow solid, yield: 55%; M.p.92–94 °C; IR(KBr,cm^{-1}): 3360, 2925, 2854, 1144, 840, 741, 516; ^{1}HNMR ($CDCl_3$ + DMSO, 300 MHz, δ): 9.07 (s,1H, Ar), 8.05 (s, 1H, Ar), 8.10 (d, 2H, Ar, J = 8.120 Hz), 7.31 (d, 2H, Ar, J = 8.120 Hz), 7.25 (d, 2H, Ar, J = 7.932 Hz); Mass (ESI-MS): 214 $[M+1]^+$; HRMS: observed value $C_{12}H_8FN_3$ [M + 1]: 214.0783; calculated value 214.0780.

*5.1.2. General procedure for substituted 3-bromo-2-phenylimidazo [1,2-a]pyrazine (**4a**–**i**)*

To a solution of imidazopyrazine (1 mmol) in ethanol, *N*-bromosuccinimide (1.5 mmol) was added and stirred at room temperature. Following the completion of the reaction (monitored by TLC), ethanol was removed from the reaction mixture on rotovapour. Water was added to the residue and extracted with ethyl acetate. Combined organic layers were washed with water, dried over anhydrous sodium sulphate, filtered and concentrated. Further purification by column chromatography using silica gel (hexane/ethyl acetate 90/10) gave a desired product.

*5.1.2.1. 3-Bromo-6-methyl-2-phenylimidazo[1,2-a]pyrazine (**4a**).* Brown solid; yield: 87%; M.p.105–107 °C; IR (KBr, cm^{-1}): 2962, 2926, 2857, 1729, 1156, 1014; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.96 (s, 1H, Ar), 8.0 (d, 2H, Ar, J = 8.309 Hz), 7.89 (s, 1H, Ar), 7.30–7.27 (m, 3H, Ar), 2.61 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 20.60, 93.15, 113.22, 127.47, 128.10, 128.20, 128.49, 131.49, 138.84, 139.33, 141.75; Mass (ESI-MS): 288 $[M]^+$; HRMS: observed value $C_{13}H_{10}BrN_3$: 288.0143; calculated value: 288.0136.

*5.1.2.2. 3-Bromo-6-methyl-2-p-tolylimidazo[1,2-a]pyrazine (**4b**).* Brown solid, yield: 86%; M.p. 72–75 °C; IR (KBr, cm^{-1}): 2922, 2852, 1609, 1450, 1151, 821, 716, 508; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.84 (s, 1H, Ar), 7.94 (d, 2H, Ar, J = 7.806 Hz), 7.91 (s, 1H, Ar), 7.22 (d, 2H, Ar, J = 8.782 Hz), 2.59 (s, 3H, CH_3), 2.43 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 20.79, 28.31, 115.84, 123.89, 126.567, 128.05, 129.047, 129.061, 131.134, 141.482, 142.348, 153.80; Mass (ESI-MS): 302 [M + 1] $^+$; HRMS: observed value $C_{14}H_{12}BrN_3$ [M + 1]: 302.0286; calculated value: 302.0294.

*5.1.2.3. 3-Bromo-2(4-fluorophenyl)-6-methylimidazo[1,2-a]pyrazine (**4c**).* Light yellow solid, yield: 74%; M.p.173–175 °C; IR (KBr, cm^{-1}): 3030, 2923, 1710, 1153, 937, 768, 691; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.82 (s, 1H, Ar), 7.94 (s, 1H, Ar), 7.75 (d, 2H, Ar, J = 7.932 Hz), 7.14 (d, 2H, Ar, J = 7.932 Hz), 2.42 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 29.63, 115.66, 115.99, 116.81, 129.88, 129.98, 130.42, 131.72, 143.33, 144.20; Mass (ESI-MS): 306 $[M]^+$, 308 $[M+2]^+$; HRMS: observed value $C_{13}H_9BrFN_3$: 306.0048; calculated value: 306.0042.

*5.1.2.4. 3-(3-Bromo-6-methylimidazo[1,2-a]pyrazin-2-yl)-2H-chromen-2-one (**4d**).* Liquid, yield: 70%; IR (KBr, cm^{-1}): 3321, 3182, 2924, 1710, 1616, 1462, 1373, 1093; ^{1}H NMR ($CDCl_3$, 300 MHz, δ); 8.97(s, 1H, Ar), 8.81(s, 1H, Ar), 8.59 (s, 1H, Ar), 7.82 (d, 2H, Ar, J = 7.62 Hz), 7.32 (t, 1H, Ar, J = 8.34 Hz), 7.19 (d, 1H, Ar, J = 7.62 Hz), 2.39 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 27.23, 97.20, 114.96, 121.05, 121.57, 122.65, 123.43, 124.67, 126.42, 128.54, 134.43, 141.47, 144.89, 150.65, 152.6, 158.98; Mass (ESI-MS): 357 $[M+1]^+$.

*5.1.2.5. 2-tert-Butyl-3-bromo-6-methylimidazo[1,2-a]pyrazine (**4e**).* Light yellow solid, yield: 80%; M.p. 70 °C; IR (KBr, cm^{-1}): 3381, 2922, 1709, 1453, 1160, 820, 719; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.90 (s, 1H, Ar), 7.94 (s, 1H, Ar), 2.40 (s, 3H, CH_3), 1.34 (s, 9H, t-bu); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 29.07, 33.06, 33.87, 92.57, 115.67, 129.65,132,45, 142.35, 154.01; Mass (ESI-MS): 268 $[M]^+$; HRMS: observed value $C_{11}H_{14}BrN_3$: 268.0444; calculated value: 268.0449.

*5.1.2.6. 3-Bromo-2-phenylimidazo[1,2-a]pyrazine (**4f**).* Light yellow solid, yield: 57%; M.p. 93–95 °C; IR (KBr, cm^{-1}): 3312, 3189, 2925, 1623, 1450, 873, 689, 495; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.99 (s, 1H, Ar), 8.17 (d, 1H, Ar, J = 4.532 Hz), 8.11 (d, 2H, Ar, J = 6.987 Hz), 8.02 (d, 1H, Ar, J = 4.532 Hz), 7.79–7.82 (m, 3H, Ar); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 93.45, 116.34, 127.31, 127.89, 128.35, 129.69, 131.61, 142.41, 143.10; Mass (ESI-MS): 272 $[M]^+$; HRMS: observed value $C_{12}H_8BrN_3$ [M + 1]: 273.9973; calculated value: 273.9979.

*5.1.2.7. 3-Bromo-2-(4-fluorophenyl)imidazo[1,2-a]pyrazine (**4g**).* Light yellow solid, yield: 65%; M.p. 98–100 °C; IR (KBr, cm^{-1}): 3199, 2924, 2853, 1464, 1206, 833,784, 551: ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.82 (s, 1H, Ar), 7.94 (d, 2H, Ar, J = 9.253 Hz), 7.75 (d, 2H, Ar, J = 8.120 Hz), 7.14(d, 2H, Ar, J = 7.932 Hz); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 92.57, 144.18, 143.34, 131.73, 131.76, 130.45, 130.05, 129.90, 115.61, 116.67; Mass (ESI-MS): 291 $[M]^+$, 293 $[M+2]^+$; HRMS: Observed value $C_{12}H_7BrFN_3$; 291.9875; Calculated value: 291.9885.

*5.1.2.8. 3-Bromo-2-(4-chlorophenyl)imidazo[1,2-a]pyrazine (**4h**).* Red colour solid, yield: 89%; M.p. 100–102 °C; IR (KBr, cm^{-1}): 3097, 2981, 1726, 1387, 1175, 756, 665; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.81 (s, 1H, Ar), 7.94 (d, 2H, Ar, J = 9.253 Hz), 7.74 (d, 2H, Ar, J = 8.120 Hz),7.14 (d, 2H, Ar, J = 8.120 Hz); ^{13}CNMR ($CDCl_3$ + DMSO, 75 MHz): 96.45, 108.15, 116.86, 128.35, 128.49, 128.56, 128.63,128.98, 130.21, 130.58; Mass (ESI-MS): 308 $[M]^+$; HRMS: observed value $C_{12}H_7BrClN_3$: 307.9585; calculated value: 307.9590.

*5.1.2.9. 2-tert-Butyl-3-bromoimidazo[1,2-a]pyrazine (**4i**).* Light yellow solid, yield: 73%; M.p. 85–87 °C; IR (KBr, cm^{-1}): 2962, 2926, 1725, 1262, 1013, 800; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.94 (s, 1H, Ar), 8.02 (d, 1H, Ar, J = 5.05 Hz), 7.94 (d, 1H, Ar, J = 4.532 Hz), 1.52 (s, 9H, t-bu); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 28.89, 30.67, 93.54, 115.67,124.78, 129.63, 131.78, 142.35; Mass (ESI-MS): 254 $[M]^+$; HRMS: observed value $C_{10}H_{12}BrN_3$ [M + 1]: 254.0296; calculated value: 254.0294.

*5.1.3. General procedure for substituted 3-cyano-2-phenylimidazo [1,2-a]pyrazine (**5a**–**e**)*

To a stirred mixture 3-bromoimidazopyrazine (1 mmol), cuprous cyanide (1.2 mmol) and DMF (5 mL) were added and refluxed till the reaction was completed (monitored by TLC). After completion of the reaction resulting dark brown solution, while still hot, was poured in to a beaker containing 25% ammonia solution. Toluene was added to the above solution and stirred well until all the lumps have disintegrated. To the cold solution ether was added and filtered through sintered glass funnel. The ether-toluene layer was separated and washed successfully with dilute ammonia solution until the organic layer was colorless. The ether-toluene layer was further washed with diluted HCl followed by water and brine solution, finally dried with anhydrous sodium sulphate, filtered and concentrated. Pure product was obtained by column chromatography using silica gel (hexane/ethyl acetate 85/15).

*5.1.3.1. 6-Methyl-2-phenylimidazo[1,2-a]pyrazine-3-carbonitrile (**5a**).* Brown solid, yield: 54%; M.p. 162–164 °C; IR (KBr, cm^{-1}): 3434, 2923, 2215, 1501, 1241, 777, 699; ^{1}H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 9.18 (s, 1H, Ar), 8.37 (s, 1H, Ar), 8.10 (d, 2H, Ar, J = 8.120 Hz), 7.50–7.40 (m, 3H, Ar), 2.62 (s, 3H, CH_3); ^{13}CNMR: 20.41, 111.05, 115.42, 123.98, 126.80, 128.63, 130.08, 139.57, 141.07,

141.91, 152.81; Mass (ESI-MS): 235 $[M]^+$; HRMS: observed value $C_{14}H_{11}N_4$ $[M+1]$: 235.0977; calculated value: 235.0893.

*5.1.3.2. 2-(4-Fluorophenyl)-6-methylimidazo[1,2-a]pyrazine-3-carbonitrile (**5b**).* Yellow solid, yield: 60%; M.p.187–190 °C; IR (KBr, cm^{-1}): 3426, 2924, 2217, 1608, 1474, 1237, 837; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 9.09 (s, 1H, Ar), 8.37 (brs, 1H, Ar), 8.18 (t, 2H, Ar, $J = 8.687$ Hz), 7.23 (t, 2H, Ar, $J = 8.682$ Hz), 2.62 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 29.38, 115.33, 116.40, 116.14, 121.65, 129.34, 129.52, 142.64, 152.34, 165.77; Mass (ESI-MS): 253 $[M]^+$; HRMS observed value: $C_{14}H_9FN_4$: 253.0883; calculated value: 253.0889.

*5.1.3.3. 2-(4-(Benzyloxy)-3-nitrophenyl)imidazo[1,2-a]pyrazine-3-carbonitrile (**5c**).* Liquid, yield: 46%; IR (KBr, cm^{-1}): 3364, 2926, 2855, 2217, 1739, 1617, 1290, 1173, 731; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.47(s, 1H, Ar), 8.42 (s, 1H, Ar), 8.09 (s, 1H, Ar), 7.47–7.45 (m, 7H, Ar), 3.60 (s, 2H, OCH_2), 2.44 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 26.64, 61.03, 109.50, 114.24, 115.08, 122.82, 123.10,126.40, 126.66, 128.00, 128.26, 129.31, 131.29, 132.45, 135.01, 141.09, 142.60, 154.88; Mass (ESI-MS): 386 $[M+1]^+$.

*5.1.3.4. 2-(4-Chlorophenyl)-6-methylimidazo[1,2-a]pyrazine-3-carbonitrile (**5d**).* Light brown solid, yield: 48%; M.p.146–148 °C; IR (KBr, cm-1): 3097, 2924, 2214, 1473, 1091, 835, 794; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.92 (s, 1H, Ar), 8.60 (s, 1H, Ar), 8.23 (d, 2H, Ar, $J = 7.63$ Hz), 7.82 (s, 1H, Ar), 7.15 (d, 2H, Ar, $J = 7.63$ Hz); ^{13}C NMR: 119.36, 120.87, 122.68, 127.89, 128.01, 129.65, 133.43, 135.55, 142.22, 142. 87; Mass (ESI-MS): 255 $[M+1]^+$.

*5.1.3.5. 2-(4-Fluorophenyl)imidazo[1,2-a]pyrazine-3-carbonitrile (**5e**).* Yellow solid, yield: 50%; M.p. 158–160 °C; IR (KBr, cm^{-1}): 3055, 2924, 2216, 1606, 1478, 1236, 838; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 9.09 (s, 1H, Ar), 8.47 (s, 1H, Ar), 8.18 (d, 2H, Ar, $J = 7.86$ Hz), 7.92 (s, 1H, Ar), 7.23 (d, 2H, Ar, $J = 7.86$ Hz); ^{13}C NMR: 115.20, 125.50, 127.79, 128.73, 128.88, 132.97, 134.65, 136.43, 144.0, 141.82; Mass (ESI-MS): 238 $[M]^+$.

*5.1.4. General procedure for substituted 3- (imidazo[1,2-a]pyrazin-3-yl)methanol (**6a–b**)*

To a stirred solution of imidazopyrazine (1 mmol) in acetic acid (5.22 mmol), sodium acetate (4.34 mmol) and formaldehyde (8.8 mmol) were added at room temperature. The resulting solution was heated at 60 °c for 24 h. Subsequently, on completion of the reaction (monitored by TLC), reaction mixture was neutralized with saturated $NaHCO_3$ solution and extracted with ethyl acetate. Organic layer was washed with water, dried over anhydrous Na_2SO_4 and filtered. On evaporation of the solvent the crude product obtained was purified by silica gel chromatography (Hexane/ethyl acetate 50/50) to the desired product.

*5.1.4.1. (6-Methyl-2-(3-nitro-4-phenethylphenyl)imidazo[1,2-a]pyrazin-3-yl)methanol (**6a**).* Brown solid, yield: 48%; M.p.: 150–152 °C; IR (KBr, cm^{-1}): 3412, 3019, 2919, 1622, 1509, 1261, 740; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.43 (s, 1H, Ar), 8.39 (s, 1H, Ar), 8.09 (s, 1H, Ar), 7.47–7.45 (m, 7H, Ar), 5.02 (d, 2H, CH_2, $J = 4.267$ Hz), 4.40 (br S, 1H, OH), 3.60 (s, 2H, OCH_2), 2.44 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 26.64, 52.32, 61.03, 109.50, 114.24, 115.08, 122.82, 126.40, 126.66, 128.00, 128.26, 129.31, 131.29, 132.45, 135.01, 141.09, 142.60, 151.20, 154.88; Mass (ESI-MS): 390 $[M]^+$.

*5.1.4.2. 3-(3-(Hydroxymethyl)-6-methylimidazo[1,2-a]pyrazin-2-yl)-2H-chromen-2-one (**6b**).* Dark brown solid, yield: 45%; M.p.136–138 °C; IR (KBr, cm^{-1}): 3322, 2925, 2857,1674, 1592, 1341,1027, 755; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.29 (s, 1H, Ar), 8.19 (s, 1H, Ar), 7.80 (d, 2H, Ar, $J = 6.583$ Hz), 7.20 (d, 1H, Ar, $J = 6.583$ Hz), 7.32 (s, 1H, Ar), 7.09 (t, 1H, Ar, $J = 8.458$ Hz), 5.25 (d, 2H, CH_2, $J = 4.389$ Hz), 4.59 (brs, 1H, OH), 2.29 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 24.18, 51.05, 115.68, 120.50, 122.43, 125.98, 126.48, 127.89,130.92, 134.57, 138.60, 138.68, 139.32, 141.29, 149.78, 153.84, 158.90; Mass (ESI-MS): 308 $[M+1]^+$.

*5.1.5. General procedure for substituted 3-(amino methyl) imidazo [1,2-a]pyrazine (**7a–c**)*

To a stirred solution of imidazopyrazine (1 mmol) and secondary amine (1.2 mmol) in methanol (5 mL) and acetic acid (3 mL), formaldehyde (2 mL) was added and the resulting mixture was refluxed for 8 h. The reaction was monitored by TLC. On disappearance of the starting materials, the solvent was evaporated from the reaction mixture under reduced pressure. The residue was dissolved in ethyl acetate and excess acetic acid quenched with saturated $NaHCO_3$ solution. Organic layer was washed with water, dried over anhydrous Na_2SO_4 and filtered. The residue obtained on evaporation of the solvent was purified by silica gel chromatography (light petroleum ether/ethyl acetate 40/60) to give the desired product.

*5.1.5.1. 6-Methyl-2-phenyl-3-((piperidin-1-yl)methyl)imidazo[1,2-a] pyrazine (**7a**).* Yellow solid, yield: 66%; M.p. 148–150 °C; IR (KBr, cm^{-1}): 3407, 3128, 2922, 1504, 1418, 1112, 815, 732, 695; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 9.07 (s, 1H, Ar), 8.15 (s, 1H, Ar), 7.54–7.50 (m, 5H, Ar), 3.47 (s, 2H, CH_2), 2.45 (t, 4H, $-CH_2$, $J = 7.658$ Hz), 2.16 (s, 3H, CH_3), 1.30 (m, 6H, $-CH_2$); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 20.25, 52.83, 66.53, 81.86, 108.37, 114.45, 115.15, 125.76, 128.18, 128.44, 132.54, 137.93, 141.94, 152.46; Mass (ESI-MS): 307 $[M+1]^+$; HRMS: Observed value $C_{19}H_{22}N_4$ $[M+1]$: 307.1915; Calculated Value: 307.1922.

*5.1.5.2. 2-(4-Fluorophenyl)-6-methyl-3-(morpholinomethyl)imidazo [1,2-a]pyrazine (**7b**).* White solid, yield: 72%; M.p.140–145 °C; IR (KBr, cm^{-1}): 3416, 2922, 2856, 1492, 1113, 861; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.85 (s, 1H, Ar), 8.10 (s, 1H, Ar), 7.77(d, 2H, Ar, $J = 8.458$ Hz), 7.09 (d, 2H, Ar, $J = 8.458$ Hz), 3.87 (s, 2H, CH_2), 3.58 (t, 4H, mor, $J = 7.39$ Hz), 2.49 (t, 4H, mor, $J = 7.39$ Hz), 2.40 (s, 3H, CH_3);^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 21.18, 29.73, 51.70, 53.30, 66.85, 114.86, 115.50, 130.57, 130.68, 138.68, 138.10, 139.32, 142.29, 151.56; Mass (ESI-MS): 327 $[M+1]^+$; HRMS $C_{18}H_{19}FN_4O$ $[M+1]$: observed value: 327.1632; calculated Value: 327.1621.

*5.1.5.3. 3,3-(Morpholinomethyl)-2-p-tolylimidazo[1,2-a]pyrazine (**7c**).* Yellow solid, yield: 56%; m.p. 138–140 °C; IR (KBr, cm^{-1}): 3445, 2936, 1650, 1384, 1113, 638; ^1H NMR ($CDCl_3$ + DMSO, 300 MHz, δ): 8.91 (s, 1H, Ar), 8.20 (d, 1H, Ar, $J = 7.432$ Hz), 7.80 (d, 2H, Ar, $J = 7.176$ Hz), 7.43 (d, 2H, Ar, $J = 7.432$ Hz), 7.38 (d, 1H, Ar, $J = 7.176$ Hz), 3.97 (s, 2H, $-CH_2$), 3.63 (t, 4H, $-CH_2$, $J = 6.23$ Hz), 2.46 (t, 4H, $-CH_2$, $J = 6.23$ Hz), 2.55 (s, 3H, CH_3); ^{13}C NMR ($CDCl_3$ + DMSO, 75 MHz): 23.45, 45.78, 52.83, 66.53, 122.10, 126.45, 128.47, 129.76, 132.27, 134.52, 137.80, 142.98, 144.54, 153.87; Mass (ESI-MS): 309 $[M+1]^+$; HRMS: observed value $C_{18}H_{20}N_4O$ $[M+1]$: 309.1712; calculated value: 309.17150.

5.2. Cytotoxicity against four different cancer cell lines

Cellular viability in the presence of test compounds was determined by MTT-microcultured tetrazolium assay following the reported protocol [28]. Two human breast cancer cell lines, MDA-MB-231 (estrogen receptor-negative) and MCF-7 (estrogen receptor-positive), a human neuroblastoma cell line, SK-N-SH, and a human hepatocellular liver carcinoma cell line, Hep G2, are employed in the current study. All the four types of cancer cell lines are seeded to flat bottom 96 (10,000 cells/100 μl) well plate and cultured in the

medium containing 10% serum. Incubated for 24 h in a 5% CO_2 humid chamber so that the cells adhere to the surface. 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) was dissolved in PBS at 5 mg/mL and sterile filtered.

Different concentrations of the compounds were added to the adhered cells. After 48 h, stock MTT solution (10 μl) was added to the culture plate. Cells were further incubated in the CO_2 chamber for 2 h. Following this, media was removed and 100 μl of DMSO was added. Absorbance was measured at 562 nm in a multimode microplate reader (Tecan GENios). The results were represented as percentage of cytotoxity/viability. All the experiments were carried out in duplicates. From the percentage of cytotoxicity the IC_{50} values are calculated and presented in the Table 2.

Acknowledgements

The Ramanujan Fellowship awarded to AA by the Department of Science and Technology, New Delhi supports VS and AA, and MS is supported by fellowship from CSIR.

References

[1] M.M. Gottesman, Cancer Res. 53 (1993) 747.
[2] D.L. Temple Jr., J.P. Yevich, J.D. Catt, D. Owens, C. Hanning, R.R. Covington, R.J. Seidehamel, K.W. Dungan, J. Med. Chem. 23 (1980) 1188–1198.
[3] S.W. Schneller, J.K. Luo, J. Org. Chem. 45 (1980) 4045–4048.
[4] R.A. Glennon, M.E. Rogers, J.D. Smith, M.K.E.J. Said, L. Egle, J. Med. Chem. 24 (1981) 658–661.
[5] Y. Rival, G. Grassy, G. Michel, Chem. Pharm. Bull. 40 (1992) 1170–1176.
[6] E. Abignente, F. Arena, P.De. Caprariis, L. Parente, II Farmaco, Ed. Sci. 30 (1975) 815–822.
[7] E. Abignente, F. Arena, P. De. Caprariis, II Farmaco, Ed. Sci. 36 (1981) 61–80.
[8] M.G. Rimoli, L. Avallone, P. De. Caprariis, E. Luraschi, E. Abignente, W. Filippelli, L. Berrino, F. Rossi. Eur. J. Med. Chem. 32 (1997) 195–203.
[9] O. Vitse, P.A. Bonnet, J. Bompart, H. Viols, G. Subra, J.P. Chapat, G. Grassy, J. Heterocycl. Chem. 34 (1997) 701–707.
[10] P.J. Zimmermann, C. Brehm, W. Buhr, A.M. Palmer, J. Volz, W.A. Simon, Bioorg. Med. Chem. 16 (2008) 536–541.
[11] P.A. Bonnet, A. Michel, F. Laurent, C. Sablayrolles, E. Rechencq, J.C. Mani, M. Boucard, J.P. Chapat, J. Med. Chem. 35 (1992) 3353–3358.
[12] C. Sablayrolles, G.H. Cros, J.C. Milhavet, E. Rechencq, J.P. Chapat, M. Boucard, J.J. Serrano, J.H. McNeill, J. Med. Chem. 27 (1984) 206–212.
[13] W.C. Lumma, W.C. Randall, E.L. Cresson, J.R. Huff, R.D. Hartman, T.F. Lyon, J. Med. Chem. 26 (1983) 357–363.
[14] L.C. Meurer, R.L. Tolman, E.W. Chapin, R. Saperstein, P.P. Vicario, M.M. Zrada, M. MacCoss, J. Med. Chem. 35 (1992) 3845–3857.
[15] K. Zurbonsen, A. Michel, P.A. Bonnet, M.N. Mathieu, C. Chevillard, Gen. Pharmac. 32 (1999) 135–141.
[16] A. Brown, A. Heenderson, C. Lane, M. Lansdell, G. Maw, S. Monaghan, Bioorg. Med. Chem. Lett. 16 (2006) 4697–4700.
[17] A. Michel, F. Lauret, J.P. Chapat, M. Boucard, P.A. Bonnet, Arzeinmittel Forscung 45 (1991) 1288.
[18] (a) Laurent, T. Pocock, K.R.F. Elliot, K. Portet, V. Benezech, O. Vitse, Fundam. Clin. Pharmacol. 10 (1996) 65;
(b) S.J. Cook, K. Archer, A. Martin, K.H. Buchheit, J.R. Fozard, T. Müller, A.J. Miller, K.R.F. Elliot, R.W. Foster, R.C. Small, Br. J. Pharmacol. 114 (1995) 143–151.
[19] S.A. Mitchell, M.D. Danca, P.A. Blomgren, J.W. Darrow, K.S. Currie, J.E. Kropf, S.H. Lee, S.L. Gallion, M. J-Xiong, D.A. Pippin, R.W. DeSimone, D.R. Brittelli, D.C. Eustice, A. Bourret, M. Hill-Dezewi, P.M. Maciejewski, L.L. Elkin, Bioorg. Med. Chem. Lett. 19 (2009) 6991–6995.
[20] P. Guerret, R. Jacquier, G. Maury, J. Heterocycl. Chem. 8 (1971) 643–650.
[21] O. Vitse, F. Laurent, T.M. Pocock, V. Benezech, L. Zanik, K.R.F. Elliott, G. Subra, K. Portet, J. Bompart, J.P. Chapat, R.C. Small, A. Michel, P.A. Bonnet, Bioorg. Med. Chem. 7 (1999) 1059–1065.
[22] V.A. Basiuk, Russ. Chem. Rev. 66 (1997) 187–204.
[23] J. Bradac, Z. Furek, D. Janezic, S. Molan, I. Smerkolj, B. Stanovnik, M. Tisler, B. Vercek, J. Org. Chem. 42 (1977) 4197–4201.
[24] L. Freidman, H. Shechter, J. Org. Chem. 26 (1961) 2522–2524.
[25] J.C. Teulade, P.A. Bonnet, J.N. Rieu, H. Viols, J.P. Chapat, G. Grassay, A. Carpy, J. Heterocycl. Chem. 8 (1971) 643–650.
[26] G. Kolavi, V. Hegde, I. Khan, P. Gadad, Bioorg. Med. Chem. 14 (2006) 3069–3080.
[27] S. Linz, J. Muller, H. Hubner, P. Gmeiner, R. Troschutz, Bioorg. Med. Chem. 17 (2009) 4448–4458.
[28] R.S. Upadhyaya, J.K. Vandavasi, V.N. Rao, S. Vivek, S.S. Dixit, J. Chattopadhyaya, Bioorg. Med. Chem. 17 (2009) 2830–2841.
[29] F. Campos, M.P. Bosch, A. Guerrero, Tetrahedron Assym. 11 (2000) 2705–2718.
[30] W. John, T. Max, A.E. Erickson, J. Am. Chem. Soc. 67 (1945) 802–804.