Saudi Pharmaceutical Journal (2016) **24**, 104–114

King Saud University

Saudi Pharmaceutical Journal

www.ksu.edu.sa
www.sciencedirect.com

SHORT COMMUNICATION

Synthesis, molecular properties, toxicity and biological evaluation of some new substituted imidazolidine derivatives in search of potent anti-inflammatory agents

Asif Husain [a,*], Aftab Ahmad [b,*], Shah Alam Khan [c], Mohd Asif [d], Rubina Bhutani [a], Fahad A. Al-Abbasi [e]

[a] *Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Hamdard University, New Delhi 110062, India*
[b] *Health Information Technology Department, Jeddah Community College, King Abdulaziz University, P.O. Box-80283, Jeddah 21589, Saudi Arabia*
[c] *Department of Pharmacy, Oman Medical College, Muscat, Oman*
[d] *Department of Pharmacy, Guru Ram Das Institute of Management and Technology, Dehradun 248009, India*
[e] *Department of Biochemistry, Faculty of Science, King Abdulaziz University, Jeddah 21589, Saudi Arabia*

Received 8 January 2015; accepted 20 February 2015
Available online 9 March 2015

KEYWORDS

Imidazolidine;
Indomethacin;
Anti-inflammatory;
Analgesic

Abstract The aim of this study was to design and synthesize pharmaceutical agents containing imidazolidine heterocyclic ring in the hope of developing potent, safe and orally active anti-inflammatory agents. A number of substituted-imidazolidine derivatives (**3a–k**) were synthesized starting from ethylene diamine and aromatic aldehydes. The imidazolidine derivatives (**3a–k**) were investigated for their anticipated anti-inflammatory, and analgesic activity in Wistar albino rats and Swiss albino mice, respectively. Bioactivity score, molecular and pharmacokinetic properties of the imidazolidine derivatives were calculated by online computer software programs viz. Molinspiration and Osiris property explorer. The results of biological testing indicated that among the synthesized compounds only three imidazolidine derivatives namely 4-[1,3-Bis(2,6-

* Corresponding authors. Tel.: +91 11 26059681, +91 11 26059688x5647, +91 989 1116086 (mobile); fax: +91 11 26059686 (A. Husain). Tel.: +966 12 2870026x102, +966 507243943 (mobile); fax: +966 12 2870024 (A. Ahmad).
E-mail addresses: drasifhusain@yahoo.com, ahusain@jamiahamdard.ac.in (A. Husain), abdulsalam@kau.edu.sa, aftab786sa@hotmail.com (A. Ahmad).

Peer review under responsibility of King Saud University.

ELSEVIER Production and hosting by Elsevier

http://dx.doi.org/10.1016/j.jsps.2015.02.008

dichlorobenzyl)-2-imidazolidinyl]phenyl-diethylamine (**3g**), 4-[1,3-Bis(3-hydroxy-4-methoxybenzyl)-2-imidazolidinyl]phenyl-diethylamine (**3i**) and 4-(1,3-Bis(4-methoxybenzyl)-4-methylimidazolidin-2-yl)-phenyl-diethylamine (**3j**) possess promising anti-inflammatory and analgesic actions. Additionally these derivatives displayed superior GI safety profile (low severity index) with respect to the positive control, Indomethacin. All synthesized compounds showed promising bioactivity score for drug targets by Molinspiration software. Almost all the compounds were predicted to have very low toxicity risk by Osiris online software. Compound number (**3i**) emerged as a potential candidate for further research as it obeyed Lipinski's rule of five for drug likeness, exhibited promising biological activity *in-vivo* and showed no risk of toxicity in computer aided screening.

1. Introduction

Inflammation is a normal, natural protective and defense mechanism of the organism to tissue injury caused by various factors which include physical trauma, injurious stimuli, chemical action or microbial infections (Ashley et al., 2012). It is a very common symptom of many chronic diseases such as arthritis, osteoarthritis, inflammatory bowel disease, and chronic asthma which put enormous burden on the economy of the countries. The prevalence of inflammatory diseases is on rise across the world, mostly affecting elderly population (Gautam and Jachak, 2009). Few epidemiological studies conducted elsewhere have also linked inflammation to pathogenesis of stroke, cardiovascular diseases, various types of cancer and to some extent neurodegenerative diseases. Mantovani and Pierotti in 2008 reported that inflammatory reactions and underlying infections are involved in 15–20% of all cancer deaths (Mantovani and Pierotti, 2008).

Nonsteroidal anti-inflammatory drugs (NSAIDs) are the most commonly prescribed medicines for the management and treatment of various inflammatory conditions. These drugs interfere with the production of lipid autacoids known as prostaglandins (PGs), which play an important role in eliciting inflammatory reactions and its sign and symptoms (Ricciotti and Fitzgerald, 2011). NSAIDs block the biosynthesis of PGs primarily by inhibiting the arachidonic acid metabolism via inhibition of several enzymes involved in their synthesis including cyclooxygenase enzyme (COX-1 and COX-2) (Tan et al., 1992). Commonly used NSAIDs exhibit higher selectivity toward COX-1, an enzyme that is involved in the cytoprotection of the gastrointestinal tract (GIT), than COX-2 which is principally responsible to cause inflammation. A majority of the commonly used NSAIDs are non selective inhibitors of both the isozyme forms of cyclooxygenase and thus are associated with undesirable GI effects such as gastric irritation, ulceration, bleeding and renal disorders (Allison et al., 1992; Agnihotri et al., 2010). In order to overcome GI problems, highly selective COX-2 inhibitors (celecoxib, rofecoxib, etc.) were developed and marketed as gastro-protective NSAIDs (Lanza, 1998). However, long term use of some selective COX-2 inhibitors has shown potential limitations including cardiovascular complications, aggravation of ulcers among high-risk patients, delay in healing process of gastroduodenal ulcers, prostacyclin deficiency leading to thrombosis and kidney toxicity (Verrico et al., 2003; Buttgereit et al., 2001). Hence, selective COX-2 inhibitors because of their high cost and undesirable side effects are not the ideal candidates for the treatment/management of various chronic inflammatory disorders and therefore, efforts should be made for the development of new orally active, potent, improved and safer NSAIDs with low or no GI side effects.

Imidazolidines (saturated imidazoles), also known as tetrahydroimidazoles are biologically active nitrogen containing heterocyclic moiety which have been reported to shown wide array of significant bioactivities such as anti-inflammatory, analgesic, α-adrenergic receptor agonist, antimicrobial, antiparasitic, oral hypoglycemic and anticonvulsant activities (Marki et al., 1984; Sharma and Khan, 2001; Caterina et al., 2008; Saczewski et al., 2009; Neves et al., 2010; Robert et al., 2010). They have also been considered as important scaffolds and intermediates for designing and synthesis of medicinal compounds with potential cyclooxygenase-2 (COX-2) inhibition activity (Patel et al., 2004). Several substituted-imidazolidine derivatives have been shown to be potential anti-edema agents in animal models of inflammation. Khan and Chawla, reported them to be promising group of NSAIDs with potential anti-inflammatory activities (Khan and Chawla, 2002).

Literature survey revealed that imidazolidine, a versatile moiety, could be a possible pharmacophore in designing safer anti-inflammatory medicinal agents (Khan and Chawla, 2002; Sharma and Khan, 2001; Khan and Gupta, 2003). In continuation of our work on this moiety (Khan et al., 2012; Husain et al., 2013), it was thought-out to study some new 4-[1,3-bis(substituted-benzyl)-2-imidazolidinyl]phenyl-dialkylamines for their possible *in-vivo* anti-inflammatory plus analgesic actions including gastrointestinal safety (acute ulcerogenicity). Structure–activity relationships of imidazolidine derivatives were also studied to study the effect of various substituents on the biological activity. Oral bioavailability, toxicity potential and pharmacokinetic profile of synthesized compounds were also predicted with the aid of computer programs to select the best candidate(s) among the synthesized compounds for the drug development.

2. Experimental

2.1. Chemistry

Melting points of the synthesized compounds were determined using open capillary tubes on a liquid paraffin bath and are uncorrected. The chemical reactions progress was monitored on silica gel G coated TLC plates in the benzene-ethanol (8:2) solvent system. Compounds on TLC were spotted by exposing to iodine vapors or under UV light. The IR spectra

of the title compounds (in KBr pellets) were recorded on a Thermo Nicolet avatar 330-FT-IR spectrophotometer. ^{1}H and $^{13}CNMR$ spectra of all the prepared compounds were recorded on a Bruker DPX-300 MHz in $CDCl_3$; chemical shift (δ) values are reported in parts per million (*ppm*). The abbreviations used to describe the splitting pattern are as follows: s for singlet; bs for broad singlet, d for doublet; dd for double doublet; t for triplet; m for multiplet. Mass spectra of the synthesized compounds were recorded on LCMS/MS (Perkin–Elmer and LABINDIA, Applied Bio-system). The Perkin–Elmer 240 analyzer was used for elemental analysis of the compounds. The elemental analysis was found within the array of ±0.4% for each element analyzed (C, H, N). We used dry solvents throughout the experiments. Compounds were synthesized as per the reported methods (Husain et al., 2013), and the synthetic protocol is presented in Scheme 1.

2.2. Synthesis of substituted ethane 1,2 diamines

2.2.1. General procedure for synthesis of N_1,N_2-bis(substituted-benzylidene)ethane-1,2-diamines 1a-j

A mixture of an aromatic aldehyde (0.005 mol), ethylenediamine/1,2-diaminopropane (0.006 mol) and dry benzene (15 mL) with few molecular sieves (4 Å) was refluxed in a Dean–Stark assembly. After the removal of water, the reaction mixture was refluxed for another 6 h to complete the reaction. After successful completion of the reaction, the extra amount of benzene was distilled off to obtain a solid which was crystallized using methanol to give TLC pure compounds **1a–j**.

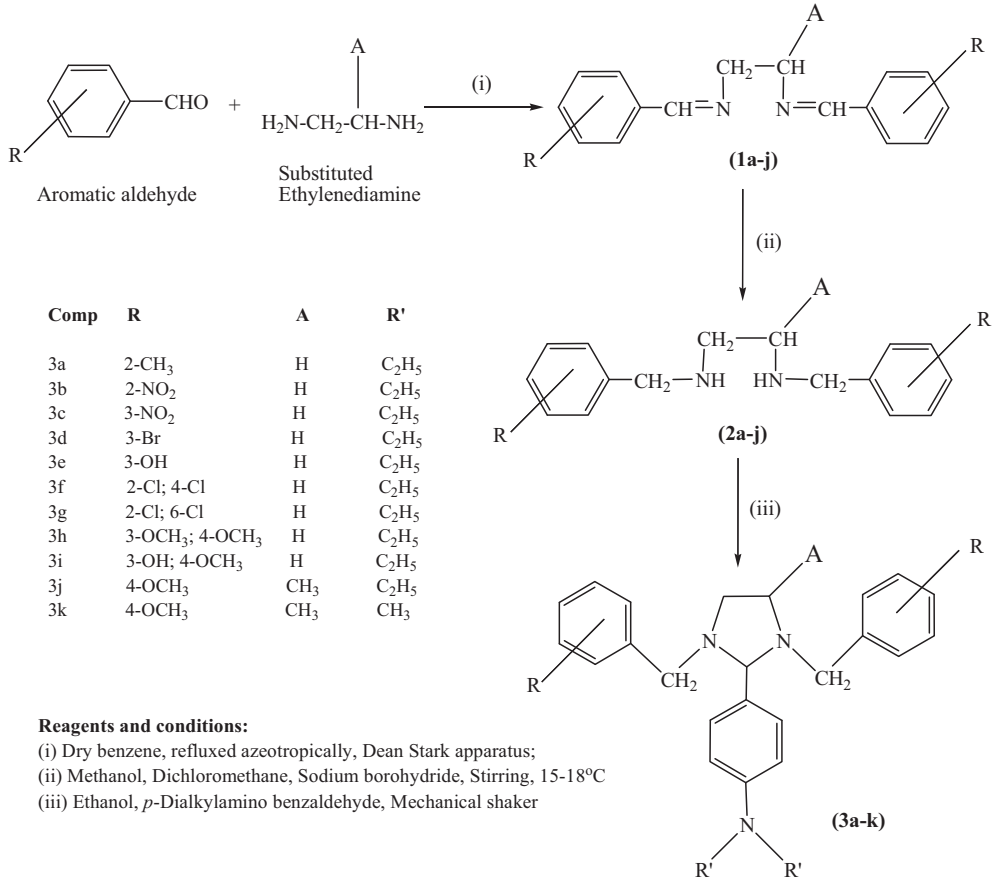
2.2.1.1. N_1,N_2-bis(2-Methylbenzylidene)ethane-1,2-diamine 1a. Yield: 78%, m.p. 122–123 °C. ^{1}H NMR ($CDCl_3$) δ = 2.31 (s, 6H, 2xCH_3), 3.77 (s, 4H, 2xCH_2), 6.73-7.44 (m, 8H, 2xH-3,4,5,6), 8.31 (s, 2H, aldimine protons).

2.2.1.2. N_1,N_2-bis(2-Nitrobenzylidene)ethane-1,2-diamine 1b. Yield: 82%, m.p. 140–142 °C. ^{1}H NMR ($CDCl_3$) δ = 3.96 (s, 4H, 2xCH_2), 7.18–7.89 (m, 8H, 2xH-3,4,5,6), 8.73 (s, 2H, aldimine protons).

2.2.1.3. N_1,N_2-bis(3-Nitrobenzylidene)ethane-1,2-diamine 1c. Yield: 76%, m.p. 131–133 °C. ^{1}H NMR ($CDCl_3$) δ = 3.93 (s, 4H, 2xCH_2), 7.23–8.21 (m, 8H, 2xH-2,4,5,6), 8.68 (s, 2H, aldimine protons).

2.2.1.4. N_1,N_2-bis(3-Bromobenzylidene)ethane-1,2-diamine 1d. Yield: 73%, m.p. 105–106 °C. ^{1}H NMR ($CDCl_3$) δ = 3.99 (s, 4H, 2xCH_2), 7.14–7.98 (m, 8H, 2xH-2,4,5,6), 8.59 (s, 2H, aldimine protons).

2.2.1.5. N_1,N_2-bis(3-Hydroxybenzylidene)ethane-1,2-diamine 1e. Yield: 68%, m.p. 136–138 °C. ^{1}H NMR ($CDCl_3$) δ = 3.86 (s, 4H, 2xCH_2), 6.98–7.56 (m, 8H, 2xH-2,4,5,6), 8.28 (s, 2H, aldimine protons), 10.32 (bs, 1H, OH).



Comp	R	A	R'
3a	2-CH_3	H	C_2H_5
3b	2-NO_2	H	C_2H_5
3c	3-NO_2	H	C_2H_5
3d	3-Br	H	C_2H_5
3e	3-OH	H	C_2H_5
3f	2-Cl; 4-Cl	H	C_2H_5
3g	2-Cl; 6-Cl	H	C_2H_5
3h	3-OCH_3; 4-OCH_3	H	C_2H_5
3i	3-OH; 4-OCH_3	H	C_2H_5
3j	4-OCH_3	CH_3	C_2H_5
3k	4-OCH_3	CH_3	CH_3

Scheme 1 Protocol for synthesis of title compounds (**3a–k**).

2.2.1.6. N_1,N_2-bis(2,4-Dichlorobenzylidene)ethane-1,2-diamine 1f. Yield: 77%, m.p. 119–120 °C. ^{1}HNMR ($CDCl_3$) δ = 3.90 (s, 4H, 2xCH_2), 7.34–7.88 (m, 6H, 2xH-3,5,6), 8.63 (s, 2H, aldimine protons).

2.2.1.7. N_1,N_2-bis(2,6-Dichlorobenzylidene)ethane-1,2-diamine 1g. Yield: 75%, m.p. 140–142 °C. ^{1}H NMR ($CDCl_3$) δ = 3.88 (s, 4H, 2xCH_2), 7.41–7.95 (m, 6H, 2xH-3,4,5), 8.24 (s, 2H, aldimine protons).

2.2.1.8. N_1,N_2-bis(3,4-Dimethoxybenzylidene)ethane-1,2-diamine 1h. Yield: 81%, m.p. 152–153 °C. ^{1}H NMR ($CDCl_3$) δ = 3.84 (s, 16H, 4xOCH_3 + 2xCH_2), 6.82 (d, J = 8 Hz, 2H, 2xH-5), 7.15 (dd, J = 2 Hz, 8 Hz, 2H, H2x6), 7.46 (d, J = 2 Hz, 2H, 2xH-2), 8.26 (s, 2H, aldimine protons).

2.2.1.9. N_1,N_2-bis(3-Hydroxy-4-methoxybenzylidene)ethane-1,2-diamine 1i. Yield: 74%, m.p. 121–124 °C. ^{1}H NMR ($CDCl_3$) δ = 3.82 (s, 6H, 2xOCH_3), 3.95 (s, 4H, 2xCH_2), 6.85 (d, J = 8 Hz, 2H, 2xH5), 7.13 (dd, J = 2 Hz, 8 Hz, 2H, 2xH6), 7.48 (d, J = 2 Hz, 2H, 2xH2), 8.18 (s, 2H, aldimine protons), 10.21 (bs, 1H, OH).

2.2.1.10. N_1,N_2-bis(4-methoxystyryl)propane-1,2-diamine 1j. Yield: 86%, m.p. 85–86 °C. ^{1}H NMR ($CDCl_3$) δ = 1.37 (d, 3H, CH_3), 3.75 (s, 6H, 2xOCH_3), 3.91 (s, 4H, 2xCH_2), 6.86-7.61 (m, 8H, 2xH-2,3,5,6), 8.08 & 8.16 (s, each, 2H, aldimine protons).

*2.3. Reduction of N_1,N_2-bis(substituted-benzylidene)ethane-1,2-diamines to get N_1,N_2-bis(substituted-benzyl)ethane-1,2-diamines **2a–j***

The compound ***1a–j*** (0.003 mol) was dissolved in a mixture of methanol (15 mL) and dichloromethane (10 mL). Afterward, a solution of $NaBH_4$ (0.013 mol) in sodium hydroxide solution (2 N; 1 mL) was slowly added with constant shaking. The reaction mixture was stirred for 5 h at the temperature maintained between 15 and 18 °C. Upon completion of reaction, the remaining solvent was removed by distillation. The residue so obtained was diluted with water and finally extracted with ether. Anhydrous sodium sulfate was used to dry the extracted ether layer. The dried layer was filtered and distilled off to get the desired compounds **2a–j**. The products obtained were pale yellow–brown colored viscous oils which were found pure on TLC examination. The compounds were utilized as such for the preparation of final compounds.

2.3.1. N_1,N_2-bis(2-Methylbenzyl)ethane-1,2-diamine 2a

Pale yellow viscous oil, Yield: 62%, ^{1}H NMR ($CDCl_3$) δ = 2.33 (s, 6H, 2xCH_3), 2.85 (s, 4H, 2xCH_2), 3.68 (s, 4H, 2xNCH_2, benzyl), 6.81–7.53 (m, 8H, 2xH-3,4,5,6), 9.31 (bs, 2H, 2xNH).

2.3.2. N_1,N_2-bis(2-Nitrobenzyl)ethane-1,2-diamine 2b

Light brown viscous oil, Yield: 74%, ^{1}H NMR ($CDCl_3$) δ = 3.02 (s, 4H, 2xCH_2), 3.89 (s, 4H, 2xNCH_2, benzyl), 7.65-8.23 (m, 8H, 2xH-3,4,5,6), 9.43 (bs, 2H, 2xNH).

2.3.3. N_1,N_2-bis(3-Nitrobenzyl)ethane-1,2-diamine 2c

Brown viscous oil, Yield: 66%, ^{1}H NMR ($CDCl_3$) δ = 3.02 (s, 4H, 2xCH_2), 3.76 (s, 4H, 2xNCH_2, benzyl), 7.72–8.19 (m, 8H, 2xH-2,4,5,6), 9.46 (bs, 2H, 2xNH).

2.3.4. N_1,N_2-bis(3-Bromobenzyl)ethane-1,2-diamine 2d

Dark yellow viscous oil, Yield: 70%, ^{1}H NMR ($CDCl_3$) δ = 3.03 (s, 4H, 2xCH_2), 3.74 (s, 4H, 2xNCH_2, benzyl), 7.33-7.85 (m, 8H, 2xH-2,4,5,6), 9.13 (bs, 2H, 2xNH).

2.3.5. N_1,N_2-bis(3-Hydroxybenzyl)ethane-1,2-diamine 2e

Pale yellow viscous oil, Yield: 64%, ^{1}H NMR ($CDCl_3$) δ = 2.83 (s, 4H, 2xCH_2), 3.81 (s, 4H, 2xNCH_2, benzyl), 6.83–7.56 (m, 8H, 2xH-2,4,5,6), 9.38 (bs, 2H, 2xNH).

2.3.6. N_1,N_2-bis(2,4-Dichlorobenzyl)ethane-1,2-diamine 2f

Yellow viscous oil, Yield: 72%, ^{1}H NMR ($CDCl_3$) δ = 3.10 (s, 4H, 2xCH_2), 3.75 (s, 4H, 2xNCH_2, benzyl), 7.31–7.94 (m, 6H, 2xH3,5,6), 9.22 (bs, 2H, 2xNH).

2.3.7. N_1,N_2-bis(2,6-Dichlorobenzyl)ethane-1,2-diamine 2g

Dark yellow viscous oil, Yield: 68%, ^{1}H NMR ($CDCl_3$) δ = 3.08 (s, 4H, 2xCH_2), 3.78 (s, 4H, 2xNCH_2, benzyl), 7.29-7.87 (m, 6H, 2xH-3,4,5), 9.17 (bs, 2H, 2xNH).

2.3.8. N_1,N_2-bis(3,4-Dimethoxybenzyl)ethane-1,2-diamine 2h

Pale yellow viscous oil, Yield: 65%, ^{1}H NMR ($CDCl_3$) δ = 2.86 (s, 4H, 2xCH_2), 3.69 (s, 4H, 2xNCH_2, benzyl), 6.96–7.56 (m, 6H, 2xH-2,5,6), 9.25 (bs, 2H, 2xNH).

2.3.9. N_1,N_2-bis(3-Hydroxy-4-methoxybenzyl)ethane-1,2-diamine 2i

Yellow viscous oil, Yield: 67%, ^{1}H NMR ($CDCl_3$) δ = 2.88 (s, 4H, 2xCH_2), 3.73 (s, 4H, 2xNCH_2, benzyl), 6.92-7.58 (m, 6H, 2xH-2,5,6), 9.19 (bs, 2H, 2xNH).

2.3.10. N_1,N_2-bis(4-methoxybenzyl)propane-1,2-diamine 2j

Red viscous oil, Yield: 58%, ^{1}H NMR ($CDCl_3$) δ = 1.29 (m, 3H, CH_3), 3.69 (bs, 3H, CH_2 + CH), 3.78 (s, 4H, 2xNCH_2, benzyl), 6.88-7.54 (m, 8H, 2xH-2,3,5,6), 9.89 (bs, 2H, 2xNH).

*2.4. General procedure for the synthesis of substituted-imidazolidine derivatives **3a–k***

The compound **2a–j** (2 mmol) was dissolved in absolute ethanol (15 mL), and *p*-diethyl/dimethyl-aminobenzaldehyde (2 mmol) was added to it. This reaction mixture was properly shaken for 5 h using mechanical shaker and then kept in a refrigerator for whole night. The reaction mixture was concentrated to reduce to half of its volume and was transferred to ice cold water which gave a solid mass. It was filtered and then crystallized by methanol to get TLC pure compounds ***3a–k***.

*2.4.1. 4-[1,3-Bis(2-methylbenzyl)-2-imidazolidinyl]phenyl-diethylamine **3a***

Yield: 68%, m.p. 118-120 °C. IR (KBr: cm^{-1}): 3110 (C—H), 1583 (C—N), 1605 (C═C).^{1}H NMR ($CDCl_3$) δ = 1.12 (t, 6H, 2x$\underline{CH_3}CH_2$), 2.02 (s, 6H, 2xCH_3), 2.39 & 3.07 (m, each, 2xCH_2, imidazole ring), 3.15 & 3.73 (d, each, 4H, 2xCH_2,

benzyl), 3.27 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.67 (s, 1H, CH, imidazole ring), 6.64 (d, J = 8 Hz, 2H, H3,5, *p*-diethylamino ring), 6.91–7.26 (m, 8H, 2xH3,4,5,6, *o*-tolyl), 7.42 (d, J = 8 Hz, 2H, H-2,6, *p*-diethylamino ring). ^{13}C NMR ($CDCl_3$) δ = 14.71 [$N(CH_2\underline{CH_3})_2$], 22.43 ($CH_3$), 40.86 [$N(\underline{CH_2}CH_3)_2$], 50.25 (C4/5), 56.27 (C6/6'), 88.66 (C2), 112.18 (C9/9'/11/11'), 112.28 (C3''/5''), 126.85 (C7/7'), 127.82 (C1''), 128.21 (C8/8'), 129.34 (C12/12'), 130.38 (C2''/6''), 141.86 (C10/10'), 149.35 (C4''). MS [EI] m/z 427 (M^+). Elemental analysis ($C_{29}H_{37}N_3$); Calcd. C, 81.45; H, 8.72; N, 9.83; found: C, 81.21; H, 8.68; N, 9.76.

2.4.2. 4-[1,3-Bis(2-nitrobenzyl)-2-imidazolidinyl]phenyl-diethylamine **3b**

Yield: 55%, m.p. 164-166 °C. IR (KBr, cm^{-1}): 3114 (C—H), 1589 (C—N), 1604 (C=C). ^{1}H NMR ($CDCl_3$) δ = 1.14 (t, 6H, 2x $\underline{CH_3}CH_2$), 2.38 & 3.12 (m, each, 4H, 2xCH_2, imidazole ring), 3.23 & 3.75 (d, each, 4H, 2xCH_2, benzyl), 3.31 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.70 (s, 1H, CH, imidazole ring), 6.66 (d, J = 8 Hz, 2H, H3,5, *p*-diethylamino ring), 7.03-7.43 (m, 8H, 2H2,3,5,6, *p*-nitrophenyl), 7.46 (d, J = 8 Hz, 2H, H2,6, *p*-diethylamino ring). ^{13}C NMR ($CDCl_3$) δ = 14.53 [$N(CH_2\underline{CH_3})_2$], 40.75 [$N(\underline{CH_2}CH_3)_2$], 50.88 (C4/5), 56.34 (C6/6'), 88.62 (C2), 112.31 (C3''/5''), 111.86 (C11/11'), 112.53 (C9/9'), 126.79 (C7/7'), 127.34 (C1''), 128.98 (C12/12'), 129.12 (C8/8'), 131.03 (C2''/6''), 146.36 (C10/10'), 150.12 (C4''). MS [EI] m/z 489 (M^+), 490. Elemental analysis ($C_{27}H_{31}N_5O_4$); Calcd. C, 66.24; H, 6.38; N, 14.31; found: C, 66.18; H, 6.42; N, 14.26.

2.4.3. 4-[1,3-Bis(3-nitrobenzyl)-2-imidazolidinyl]phenyl-diethylamine **3c**

Yield: 51%, m.p. 141–142 °C. IR (KBr, cm^{-1}); 3109 (C—H), 1595 (C—N), 1602 (C=C). ^{1}H NMR ($CDCl_3$) δ = 1.15 (t, 6H, 2x$\underline{CH_3}CH_2$), 2.41 & 3.16 (m, each, 4H, 2xCH_2, imidazole ring), 3.19 & 3.81 (d, each, 4H, 2xCH_2, benzyl), 3.33 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.67 (s, 1H, CH, imidazole ring), 6.73 (d, J = 8 Hz, 2H, H3,5, *p*-diethylamino ring), 7.08-7.46 (m, 8H, 2x *m*-chlorophenyl), 7.44 (d, J = 8 Hz, 2H, H2,6, *p*-diethylamino ring). ^{13}C NMR ($CDCl_3$) δ = 14.61 [$N(CH_2\underline{CH_3})_2$], 40.68 [$N(\underline{CH_2}CH_3)_2$], 50.55 (C4/5), 56.41 (C6/6'), 88.37 (C2), 112.84 (C3''/5''), 113.67 (C8/8'), 126.65 (C7/7'), 127.06 (C12/12'), 127.32 (C1''), 128.13 (C9/9'), 129.17 (C11/11'), 130.34 (C2''/6''), 131.21 (C10/10'), 151.16 (C4''). MS [EI] m/z 489 (M^+), 490. Elemental analysis ($C_{27}H_{31}N_5O_4$); Calcd. C, 66.24; H, 6.38; N, 14.31; found: C, 66.31; H, 6.15; N, 14.28.

2.4.4. 4-[1,3-Bis(3-bromobenzyl)-2-imidazolidinyl]phenyl-diethylamine **3d**

Yield: 62%, m.p. 162-164 °C. IR (KBr, cm^{-1}); 3115 (C—H), 1593 (C—N), 1601 (C=C). ^{1}H NMR ($CDCl_3$) δ = 1.13 (t, 6H, 2x$\underline{CH_3}CH_2$), 2.42 & 3.15 (m, each, 4H, 2xCH_2, imidazole ring), 3.18 & 3.69 (d, each, 4H, 2xCH_2, benzyl), 3.31 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.66 (s, 1H, CH, imidazole ring), 6.71 (d, J = 8 Hz, 2H, H3,5, *p*-diethylamino ring), 7.02-7.43 (m, 8H, 2x *m*-bromophenyl), 7.46 (d, J = 8 Hz, 2H, H2,6, *p*-diethylamino ring). ^{13}C NMR ($CDCl_3$) δ = 14.58 [$N(CH_2\underline{CH_3})_2$], 40.72 [$N(\underline{CH_2}CH_3)_2$], 50.58 (C4/5), 56.35 (C6/6'), 87.86 (C2), 112.04 (C3''/5''), 114.25 (C8/8'), 126.84 (C7/7'), 127.13 (C12/12'), 127.15 (C1''), 128.67 (C9/9'), 129.02 (C11/11'), 130.18 (C2''/6''), 133.15 (C10/10'), 150.83 (C4''). MS [EI] m/z 557 (M^+), 555, 558, 559. Elemental analysis ($C_{27}H_{31}Br_2N_3$); Calcd. C, 58.18; H, 5.61; N, 7.54; found: C, 57.86; H, 5.64; N, 7.32.

2.4.5. 4-[1,3-Bis(3-hydroxybenzyl)-2-imidazolidinyl]phenyl-diethylamine **3e**

Yield: 58%, m.p. 144–146 °C. IR (KBr, cm^{-1}); 3353 (OH), 3142 (C—H), 1588 (C—N), 1601 (C=C); ^{1}H NMR ($CDCl_3$) δ = 1.12 (t, 6H, 2x$\underline{CH_3}CH_2$), 2.53 & 3.31 (m, each, 4H, 2xCH_2, imidazole ring), 3.16 & 3.81 (m, each, 4H, 2xCH_2, benzyl), 3.29 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.62 (s, 1H, CH, imidazole ring), 6.65-7.38 (m, 12H, 3x phenyl rings). ^{13}C-NMR ($CDCl_3$) δ = 13.97 [$N(CH_2\underline{CH_3})_2$], 40.08 [$N(\underline{CH_2}CH_3)_2$], 50.26 (C4/5), 56.18 (C6/6'), 88.03 (C2), 112.98 (C3''/5''), 113.81 (C8/8'), 125.78 (C7/7'), 126.35 (C12/12'), 126.71 (C1''), 127.33 (C9/9'), 128.16 (C11/11'), 129.84 (C2''/6''), 132.53 (C10/10'), 149.28 (C4''). MS [EI] m/z 431 (M^+ not observed). Elemental analysis ($C_{27}H_{33}N_3O_2$); Calcd. C, 75.14; H, 7.71; N, 9.74; found: C, 74.96; H, 7.63; N, 9.81.

2.4.6. 4-[1,3-Bis(2,4-dichlorobenzyl)-2-imidazolidinyl]phenyl-diethylamine **3f**

Yield: 70%, m.p. 126-128 °C. IR (KBr, cm^{-1}); 3124 (C—H), 1589 (C—N), 1602 (C=C). ^{1}H NMR ($CDCl_3$) δ = 1.15 (t, 6H, 2x $\underline{CH_3}CH_2$), 2.43 & 3.16 (m, each, 4H, 2xCH_2, imidazole ring), 3.18 & 3.65 (d, each, 4H, 2xCH_2, benzyl), 3.32 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.67 (s, 1H, CH, imidazole ring), 7.11-7.86 (m, 10H, 3x phenyl ring). ^{13}C NMR ($CDCl_3$) δ = 14.63 [$N(CH_2\underline{CH_3})_2$], 40.71 [$N(\underline{CH_2}CH_3)_2$], 50.48 (C4/5), 56.22 (C6/6'), 88.16 (C2), 112.01 (C3''/5''), 113.25 (C11/11'), 114.68 (C9/9'), 116.42 (C8/8'), 125.93 (C7/7'), 126.81 (C1''), 128.97 (C12/12'), 129.85 (C2'/6''), 149.73 (C4''), 153.91 (C10/10'). MS [EI] m/z 537 (M^+), 535, 538, 539. Elemental analysis ($C_{27}H_{29}Cl_4N_3$); Calcd. C, 60.35; H, 5.44; N, 7.82; found: C, 60.08; H, 5.26; N, 7.74.

2.4.7. 4-[1,3-Bis(2,6-dichlorobenzyl)-2-imidazolidinyl]phenyl-diethylamine **3g**

Yield: 64%, m.p. 119–121 °C. IR (KBr, cm^{-1}); 3127 (C—H), 1586 (C—N), 1601 (C=C). ^{1}H NMR ($CDCl_3$) δ = 1.15 (t, 6H, 2x $\underline{CH_3}CH_2$), 2.44 & 3.18 (m, each, 4H, 2xCH_2, imidazole ring), 3.16 & 3.66 (d, each, 4H, 2xCH_2, benzyl), 3.31 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.65 (s, 1H, CH, imidazole ring), 7.06-7.83 (m, 10H, 3x phenyl ring). ^{13}C NMR ($CDCl_3$) δ = 14.63 [$N(CH_2\underline{CH_3})_2$], 40.71 [$N(\underline{CH_2}CH_3)_2$], 50.51 (C4/5), 56.57 (C6/6'), 87.85 (C2), 112.24 (C3''/5''), 113.12 (C11/11'), 114.85 (C9/9'), 115.97 (C8/8'), 125.55 (C7/7'), 126.64 (C1''), 129.71 (C2''/6''), 131.03 (C12/12'), 133.15 (C10/10'), 150.76 (C4''). MS [EI] m/z 537 (M^+), 535, 536, 538, 539. Elemental analysis ($C_{27}H_{29}Cl_4N_3$); Calcd. C, 60.35; H, 5.44; N, 7.82; found: C, 60.08; H, 5.26; N, 7.74.

2.4.8. 4-[1,3-Bis(3,4-dimethoxybenzyl)-2-imidazolidinyl] phenyl-diethylamine **3h**

Yield: 60%, m.p. 132-133 °C. IR (KBr, cm^{-1}); 3111 (C—H), 1590 (C—N), 1600 (C=C), 1070 (C—O). ^{1}H NMR ($CDCl_3$) δ = 1.13 (t, 6H, 2x$\underline{CH_3}CH_2$), 2.39 & 3.16 (m, each, 4H, 2xCH_2, imidazole ring), 3.08 & 3.75 (d, each, 4H, 2xCH_2,

benzyl), 3.31 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.71 (s, 12H, 4xOCH_3), 3.63 (s, 1H, CH, imidazole ring), 6.68-7.46 (m, 10H, 3x phenyl rings). ^{13}C NMR ($CDCl_3$) δ = 14.39 [$N(CH_2\underline{CH_3})_2$], 40.53 [$N(\underline{CH_2}CH_3)_2$], 50.10 (C4/5), 55.63 ($OCH_3$), 56.64 (C6/6\'), 88.57 (C2), 110.86 (C3\"/5\"), 112.57 (C8/8\'), 113.22 (C11/11\'), 120.13 (C12/12\'), 126.85 (C1\"), 129.78 (C2\"/6\"), 130.69 (C7/7\'), 149.25 (C9/9\'), 150.83 (C4\"), 155.63 (C10/10\'). MS [EI] m/z 519 (M^+). Elemental analysis ($C_{31}H_{41}N_3O_4$); Calcd. C, 71.65; H, 7.95; N, 8.09; found: C, 71.38; H, 7.86; N, 8.21.

2.4.9. 4-[1,3-Bis(3-hydroxy-4-methoxybenzyl)-2-imidazolidinyl]phenyl-diethylamine **3i**

Yield: 66%, m.p. 140–142 °C. IR (KBr, cm^{-1}); 3380 (OH), 3120 (C—H), 1592 (C—N), 1602 (C═C), 1064 (C—O). ^{1}H NMR ($CDCl_3$) δ = 1.15 (t, 6H, 2x$\underline{CH_3}CH_2$), 2.36 & 3.15 (m, each, 4H, 2xCH_2, imidazole ring), 3.06 & 3.73 (d, each, 4H, 2xCH_2, benzyl), 3.35 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.73 (s, 6H, 2xOCH_3), 3.62 (s, 1H, CH, imidazole ring), 6.62–7.38 (m, 10H, 3x phenyl rings). ^{13}C NMR ($CDCl_3$) δ = 14.37 [$N(CH_2\underline{CH_3})_2$], 40.48 [$N(\underline{CH_2}CH_3)_2$], 50.15 (C4/5), 55.58 ($OCH_3$), 56.33 (C6/6\'), 89.14 (C2), 110.79 (C3\"/5\"), 111.66 (C8/8\'), 112.56 (C11/11\'), 120.25 (C12/12\'), 127.24 (C1\"), 130.18 (C2\"/6\"), 131.35 (C7/7\'), 149.18 (C9/9\'), 150.45 (C4\"), 156.17 (C10/10\'). MS [EI] m/z 491 (M^+). Elemental analysis ($C_{29}H_{37}N_3O_4$); Calcd. C, 70.85; H, 7.59; N, 8.55; found: C, 70.64; H, 7.38; N, 8.62.

2.4.10. 4-(1,3-Bis(4-methoxybenzyl)-4-methylimidazolidin-2-yl)-phenyl-diethylamine **3j**

Yield: 64%, m.p. 110–112 °C. IR (KBr, cm^{-1}); 3114 (C—H), 1593 (C—N), 1599 (C═C), 1074 (C—O). ^{1}H NMR ($CDCl_3$) δ = 0.98 (d, 3H, CH_3), 1.13 (t, 6H, 2x$\underline{CH_3}CH_2$), 2.46 (m, 2H, CH_2, imidazole ring), 2.89 (m, 1H, CH of imidazole ring carrying methyl group), 3.31 (q, 4H, 2x $CH_3\underline{CH_2}$), 3.01 & 3.75 (d, 4H, 2xCH_2, benzyl), 3.72 (s, 6H, 2xOCH_3), 3.78 (s, 1H, CH, imidazole ring), 6.48-6.82 (m, 8H, 2x *p*-anisyl rings), 6.84 (d, J = 8 Hz, 2H, H3,5, *p*-diethylamino ring), 7.46 (d, J = 8 Hz, 2H, H2,6, *p*-diethylamino ring). ^{13}C NMR ($CDCl_3$) δ = 14.73 [$N(CH_2\underline{CH_3})_2$], 22.46 ($CH_3$), 40.79 [$N(\underline{CH_2}CH_3)_2$], 55.46 (C6\'), 56.26 (C6), 57.48 (C4), 58.12 (C5), 88.85 (C2), 55.36 (OCH_3), 111.83 (C9/9\'/11/11\'), 113.16 (C3\"/5\"), 126.87 (C1\"), 127.63 (C7/7\'), 128.78 (C8/8\'/12/12\'), 129.97 (C2\"/6\"), 150.24 (C4\"), 155.96 (C10/10\'). MS [EI] m/z 473 (M^+). Elemental analysis ($C_{30}H_{39}N_3O_2$); Calcd. C, 76.07; H, 8.30; N, 8.87; found: C, 75.83; H, 8.26; N, 8.65.

2.4.11. 4-(1,3-Bis(4-methoxybenzyl)-4-methylimidazolidin-2-yl)-phenyl-dimethylamine **3k**

Yield: 61%, m.p. 92–94 °C. IR (KBr, cm^{-1}); 3120 (C—H), 1593 (C—N), 1605 (C═C), 1067 (C—O). ^{1}H NMR ($CDCl_3$) δ = 0.95 (d, 3H, CH_3), 2.51 (m, 2H, CH_2, imidazole ring), 2.86 (m, 1H, CH of imidazole ring carrying methyl group), 2.93 (s, 6H, 2xCH_3), 3.03 & 3.66 (d, 4H, 2xCH_2, benzyl), 3.69 (s, 6H, 2xOCH_3), 3.75 (s, 1H, CH, imidazole ring), 6.56–6.75 (m, 6H, 2x *p*-anisyl rings), 6.78 (d, J = 8 Hz, 2H, H3,5, *p*-diethylamino ring), 7.44 (d, J = 8 Hz, 2H, H2,6, *p*-diethylamino ring). ^{13}C NMR ($CDCl_3$) δ = 22.52 (CH_3), 40.15 [$N(CH_3)_2$], 55.14 (OCH_3), 55.53 (C6\'), 56.21 (C6), 57.26 (C4), 59.02 (C5), 89.56 (C2), 112.15 (C9/9\'/11/11\'), 113.24 (C3\"/5\"), 125.58 (C1\"), 126.94 (C7/7\'), 128.05 (C8/8\'/12/12\'), 130.12 (C2\"/6\"), 149.87 (C4\"), 154.52 (C10/10\'). MS [EI] m/z 445 (M^+). Elemental analysis ($C_{28}H_{35}N_3O_2$); Calcd. C, 75.47; H, 7.92; N, 9.43; found: C, 75.40; H, 7.84; N, 9.66.

2.5. Pharmacology

The approval of the protocol of the animal experiments was given by the Institutional Animal Ethics Committee (IAEC) of Jamia Hamdard, New Delhi, India. The synthesized compounds were investigated for their *in-vivo* anti-inflammatory activity by adopting the method of Winter et al.,(1962). Those compounds that showed good anti-inflammatory activity (> 50%) were additionally evaluated for analgesic activity (Seigmund et al., 1957) and ulcerogenic actions (Cioli et al., 1979).

2.5.1. Anti-inflammatory activity

The synthesized compounds were evaluated for their anti-inflammatory activity by adopting carrageenan-induced paw edema method in Wistar albino rats of either sex, weighing 160–200 g. The animals were randomly alienated into twelve different groups comprising six rats in each group. Group I (control) received 0.5% carboxymethyl cellulose (CMC) solution. Group II (standard) received Indomethacin; 20 mg/kg, *p.o.* and groups III–XII received test compounds (20 mg/kg, *p.o.*). The test compounds and standard drugs were administered to all animals and then 30 minute later, an injection of 0.1 mL of 0.1% solution of carrageenan (in sterile 0.9% NaCl solution) was given by subcutaneous route into the sub-plantar region of the right hind paw of each rat including control group. The volume of the animal's paw was determined using glass *Plethysmometer* at 3 h after administration of carrageenan injection. The percentage inhibition of edema was calculated using the formula:

$$\text{Anti} - \text{inflammatory activity}(\%\text{inhibition}) = [(V_c - V_t)/V_c] \times 100 \quad (1)$$

where V_c = The volume of edema of a control group, and V_t = The volume of edema of animals in groups treated with test compounds.

2.5.2. Analgesic activity

The screened compounds which demonstrated good anti-inflammatory activity (> 50%) were further evaluated for their analgesic action by acetic acid induced writhing method (Seigmund et al., 1957). Eight groups of Swiss albino mice (20–30 g) of either sex were made. There were six animals in each group. *Intra-peritoneal* injection of 0.10 mL of 1% solution of aqueous acetic acid was given to induce writhing in animals. These animals were habituated for 30 min before acetic acid injection. The test compounds were administrated orally at a dose of 20 mg/kg for the evaluation of analgesic activity. The control group was given CMC suspension. Second group was given standard drug aspirin and Test compounds suspended in 1.0% CMC at a dose of 20 mg/kg were administered orally to rest of other groups. Intra-peritoneal injection of aqueous acetic acid was given to all mice after one hour of administration of tested drug. Stretching movements comprising of arching of the back, elongation of body and extension of

hind limbs were counted for the duration of 5–15 min due to acetic acid injection. The analgesic activity of the compounds was articulated as percentage protection and calculated using following formula:

$$\text{Analgesic activity}(\%\text{Protection}) = [(W_c - W_t)/W_c] \times 100$$

where W_c = mean number of writhes of control group, and W_t = mean number of writhes of test group.

2.5.3. Acute ulcerogenic activity

Acute ulcerogenesis test was performed according to the method as described by Cioli et al. (1979). Albino rats weighing about 160–200 g were randomly divided into eight different groups having six animals in each group. The evaluation of ulcerogenic activity was carried out after *oral* administration of test compounds or indomethacin at the dose of 60 mg/kg which is 3 times of anti-inflammatory dose. The rats of control group received oral administration of vehicle as a suspension of 1% CMC. Food was stopped 24 h before administration of the test compounds, while water was continuously given. The animals were fed with normal diet after the drug treatment for the duration of 17 h and then animals were sacrificed. The rats' stomach was isolated and opened alongside the greater curvature. The opened stomach was properly washed with distilled water and cleaned smoothly by sinking into normal saline. The damage to the mucosal was scrutinized with the help of a 10× magnifying glass. The following scoring system was used to assess the mucosal damage for each stomach: 0.5: redness, 1.0: spot ulcers, 1.5: hemorrhagic streaks, 2.0: ulcers > 3 but < 5, 3.0: ulcers > 5. The severity index of gastric mucosal damage was calculated by subtracting the mean score of control group from mean score of each treated group or severity index of gastric mucosal damage = the mean score of each treated group minus the mean score of control group. There was no mucosal damage in the control group with severity index of 0.00.

2.6. Calculation of pharmacokinetic parameters and toxicity potential

Chemical structures and SMILES notations of the title compounds were obtained by using ACD labs Chemsketch version 12.0. SMILES notations of the imidazolidine derivatives were then fed in the online Molinspiration software version 2011.06 (www.molinspiration.com) to calculate various molecular properties and to predict bioactivity score for drug targets including enzymes and nuclear receptors, kinase inhibitors, GPCR ligands, and ion channel modulators. Molecular properties such as partition coefficient (Log P), Topological polar surface area (TPSA), hydrogen bond donors and acceptors, rotatable bonds, number of atoms, molecular weight, and violations of Lipinski's rule of five were calculated to evaluate the drug likeness of the synthesized compounds (Lipinski et al., 1997). The bioactivity score and drug likeness properties of the imidazolidine derivatives were compared with the standard drugs indomethacin and aspirin. The absorption percentage (% Ab) was also calculated by the reported method of Zhao et al. (2002) by using the following formula:

$$\%\text{Ab} = 109 - [0.345 \times \text{TPSA}]$$

Osiris property explorer (www.organicchemistry.org/prog/peo/), an online cheminformatics tool, was used to determine pharmacokinetic parameters such as toxicity potential, solubility and overall drug-likeness of imidazolidine derivatives. The results of virtual screening are valued and color coded either green or red for properties such as effect on reproductive system, irritant effect and tumorigenicity. Properties shown in red indicate high risks of undesired effects while a green color shows drug-conform behavior, compatibility and safety *in-vivo*.

2.7. Statistical analysis

All data generated from the animal experiments were calculated as mean ± SEM. The one-way ANOVA followed by Dunnett's multiple comparison test was employed to find out the statistical differences between the treatments and the standard.

3. Results and discussion

3.1. Chemistry

The title compounds, 4-[1,3-bis(substituted-benzyl)-2-imidazolidinyl]phenyl-dialkylamines (**3a–k**), were successfully synthesized from N_1,N_2-bis(substituted-benzyl)ethane-1,2-diamines (**2a–j**) which in turn were prepared by the reduction of N_1,N_2-bis(substituted-benzylidene)ethane-1,2-diamines (**1a–j**). Different steps of synthesis have been shown in the Scheme 1. In the initial step, appropriate aromatic aldehydes were reacted azeotropically with ethylenediamine/1,2-diaminopropane to obtain N_1,N_2-bis(substituted-benzylidene)ethane-1,2-diamines (**1a–j**). In the next step, the reduction of the compounds (**1a–j**) in the presence of sodium borohydride furnished N_1,N_2-bis(substituted-benzyl)ethane-1,2-diamines (**2a–j**). These compounds were oily (viscous) in nature and pale yellow to brown in color. In the final step, the compound **2a–j** was condensed with *p*-diethyl/dimethyl-aminobenzaldehyde to obtain the desired imidazolidines (**3a–k**).

The structures of the prepared compounds were confirmed with the help of spectral and microanalysis data results. The IR spectra of imidazolidine derivatives showed absorption bands (cm^{-1}) at 3108–3142 (C—H) and 1583–1595 (C—N). In general, ^{1}H NMR spectra (PMR) of the desired compounds revealed the signals of aromatic, diethyl/dimethyl, methylenes of imidazolidines ring and benzylic methylene protons. Triplet at around δ 1.1 and quartet at δ 3.2 revealed the presence of —N(CH_2CH_3)$_2$ group. The multiplets at around δ 2.4 & 3.1 and a singlet at around δ 3.6 accounted for 2xCH_2 and CH of imidazolidines ring. Two doublets each at around δ 3.2 and 3.7 indicated the presence of two benzylic methylenes. The three phenyl rings protons are shown in the region of δ 6.5–7.8. An analysis of spectra indicates that the imidazolidines ring methylene protons as well as the benzylic methylene protons are non-equivalent and showed geminal coupling in PMR. ^{13}C NMR spectral data of the imidazolidine derivatives revealed peaks around δ 14.2 for [N($CH_2\underline{CH}_3$)$_2$] and δ 40.6 for [N($\underline{CH}_2CH_3$)$_2$]. Other peaks were shown at appropriate δ-values. Mass spectra of the compounds showed molecular ion peaks (M^+) in reasonable intensities. The

Table 1 Anti-inflammatory, analgesic and ulcerogenic activities of the compounds (**3a–k**).

Compound	Anti-inflammatory activity % Inhibition[a] (After 3 h)	Analgesic activity % Protection[a]	Ulcerogenic effect (Severity index[b])
3a	34.18 ± 3.97**	nt	nt
3b	42.46 ± 2.93**	nt	nt
3c	45.43 ± 3.54**	nt	nt
3d	38.85 ± 1.97**	nt	nt
3e	29.72 ± 2.01**	nt	nt
3f	53.92 ± 2.14*	40.94 ± 2.05**	0.58 ± 0.23
3g	57.53 ± 2.89	51.44 ± 2.73**	0.91 ± 0.30*
3h	52.22 ± 3.36**	36.23 ± 1.55**	0.67 ± 0.16
3i	64.11 ± 2.19	56.88 ± 3.04**	1.08 ± 0.24*
3j	61.35 ± 3.18	45.65 ± 3.32**	0.75 ± 0.21
3k	54.35 ± 3.00*	42.39 ± 3.21**	0.42 ± 0.15
Control	–	–	0.00 ± 0.0
Standard 1†	67.09 ± 2.74	nt	2.08 ± 0.39**
Standard 2†	nt	59.78 ± 2.55	nt

* $p < 0.05$.
** $p < 0.01$.
† Standard 1 = Indomethacin; †Standard 2 = Aspirin; nt = not tested.
[a] Relative to the standard and data were analyzed by one-way ANOVA followed by Dunnett's multiple comparison test for $n = 6$.
[b] Relative to control and data were analyzed by one-way ANOVA followed by Dunnett's multiple comparison test for $n = 6$.

molecular ion or other related ions formed the appropriate isotopic abundances because of chlorine atom(s) presence. Microanalysis data were found in the range of ±0.4% for the theoretical numbers of the analyzed elements (C, H, N).

3.2. Pharmacokinetic parameters and toxicity potential

Lipinski's rule of five is commonly used by pharmaceutical chemists in drug design and development to predict oral bioavailability of potential lead or drug molecules. According to Lipinski's "rule of five", a candidate molecule will likely to be orally active, if: i) the molecular weight is under 500, ii) the calculated octanol/water partition coefficient (Log P) < 5, iii) there were fewer than 5 hydrogen bond donors (OH and NH groups) and, iv) there are less than ten hydrogen bond acceptors (notably N and O) (Lipinski et al., 1997). The molecular properties of imidazolidine derivatives (**3a–k**) were calculated by using Molinspiration cheminformatics software and are presented in Table 2. Compound no **3e** and **3i** did not violate any of the Lipinski's rule of five, however one violation was observed for five compounds (**3a**, **3b**, **3c**, **3h** and **3j**). All other compounds showed two violations and are expected to be orally inactive.

Molecular hydrophobicity or lipophilicity is indicated by Log P or partition coefficient. Log P values of all the title compounds except **3e**, **3h** and **3i** were found to be more than 5 and are in clear violation of Lipinski's rule of five, suggesting poor permeability across cell membrane. Log P values of indomethacin and aspirin, the standard drugs were found to be well under 5 justifying their oral use. Molecular weight of six imidazolidine derivatives was found to be less than 500 and thus these molecules are anticipated to be easily transported, diffused and absorbed as compared to large molecules. Number of hydrogen bond acceptors (O and N atoms) and number of hydrogen bond donors (NH and OH) in the synthesized compounds **3a-k** were in accordance with the Lipinski's rule of five i.e. less than 10 and 5 respectively. It can be predicted that among ten synthesized imidazolidine derivatives, only compounds **3e** and **3i** are likely to be orally active as they obeyed Lipinski's rule of five.

Topological polar surface area is very much correlated with the hydrogen bonding of a molecule and is a very good

Table 2 Drug likeness score for the synthesized imidazolidine derivatives **(3a–k).**

Compound	miLog P[a]	TPSA[b]	*n* Atoms	*n* ON[c]	*n* OHNH[d]	*n* violation	*n* rotb[e]	% ABS[f]	MW[g]
3a	6.256	9.71	32	3	0	1	8	105.65	427.64
3b	5.276	101.36	36	9	0	1	10	74.03	489.58
3c	5.324	101.36	36	9	0	1	10	74.03	489.58
3d	7.025	9.71	32	3	0	2	8	105.65	557.37
3e	4.448	50.17	32	5	2	0	8	91.69	431.58
3f	8.022	9.71	34	3	0	2	8	105.65	537.36
3g	7.974	9.71	34	3	0	2	8	105.65	537.36
3h	4.748	46.65	38	7	0	1	12	92.96	519.67
3i	4.133	68.64	36	7	2	0	10	85.32	491.63
3j	5.898	28.18	35	5	0	1	10	99.28	473.66
3k	5.146	28.18	33	5	0	1	8	99.28	445.61
Indomethacin	3.986	68.54	25	5	1	0	4	85.35	357.79
Aspirin	1.434	63.60	13	4	1	0	3	87.057	180.16

[a] Logarithm of partition coefficient between *n*-octanol and water (miLogP).
[b] Topological polar surface area (TPSA).
[c] Number of hydrogen bond acceptors (*n*-ON).
[d] Number of hydrogen bond donors (*n*-OHNH).
[e] Number of rotatable bonds (*n*-rotb).
[f] Percentage of absorption (%ABS).
[g] Molecular weight (MW).

indicator of the bioavailability of drug molecule. TPSA of imidazolidine derivatives was observed in the range of 9.71–101.36 A^0 and is well below the limit of 160 A^0. The percentages of absorption for title compounds calculated from TPSA ranged between 74.03 and 105.65% and indicated good oral bioavailability.

The bioactivity scores of title compounds for drug targets were also predicted by Molinspiration and are presented in Table 3. A molecule having bioactivity score more than 0.00 is most likely to exhibit considerable biological activities, while values −0.50 to 0.00 are expected to be moderately active and if score is less than −0.50 it is presumed to be inactive. The results clearly reveal that the physiological actions of imidazolidine derivatives might involve multiple mechanisms and could be due to the interactions with GPCR ligands, nuclear receptor ligands, and inhibit protease and other enzymes. The bioactivity score of compounds is suggestive of moderate interaction with all drug targets. The most promising compounds as per the bioactivity scores were identified to be **3e**, **3i** and **3k** which are predicted to act by more than three mechanisms (Table 3). The identified compounds showed better bioactivity score than aspirin for all drug targets.

Results of toxicity risks and drug score assessment of synthesized imidazolidine derivatives were envisaged by Osiris property explorer and are presented in Table 4. This online program predicts on the basis of functional group similarity of the investigated compound with the extensively in vitro – and *in-vivo* studied compounds present in its database. The results are color coded such as red, green and yellow (Actelion's property explorer, 2001). Green color suggests low toxic potential, yellow means mild toxicity and red color indicates high probability of toxicity. The results clearly indicated that all compounds except **3k** would be safe and expected to show low or no toxicity regarding tumorigenicity, irritant effect and effect on reproductive system.

3.3. Pharmacology

Rat paw edema induced by carrageenan is a more frequently employed experimental animal model to study the *in-vivo* anti-inflammatory activity of natural or synthetic medicinal agents (Franzotti et al., 2000; Gupta et al., 2006). Edema induced by carrageenan involved release of various inflammatory mediators but it is predominantly due to release of prostaglandins, bradykinins and other related autacoids causing migration of macrophages and polymorphonuclear neutrophils to the site of inflammation (Fernandes et al., 2007). The results of *in-vivo* anti-inflammatory presented in Table 1 indicated that compounds **3g**, **3i** and **3j** possess good anti-inflammatory action which showed 57.53%, 64.11% and 61.35% inhibition, respectively. The activity of these compounds was comparable to inhibition produced by the

Table 3 Bioactivity score of the synthesized compounds (**3a-k**) according to Molinspiration cheminformatics software.

Compound	GPCR ligand	Ion channel modulator	Kinase inhibitor	Nuclear receptor ligand	Protease inhibitor	Enzyme inhibitor
3a	−0.10	−0.35	−0.26	−0.30	−0.18	−0.22
3b	−0.16	−0.31	−0.31	−0.33	−0.20	−0.22
3c	−0.18	−0.34	−0.31	−0.32	−0.22	−0.24
3d	−0.17	−0.36	−0.29	−0.39	−0.24	−0.24
3e	−0.05	−0.27	−0.21	−0.17	−0.13	−0.14
3f	−0.09	−0.30	−0.25	−0.28	−0.18	−0.23
3g	−0.07	−0.29	−0.21	−0.32	−0.16	−0.22
3h	−0.11	−0.42	−0.23	−0.28	−0.18	−0.20
3i	−0.09	−0.33	−0.20	−0.23	−0.18	−0.16
3j	−0.06	−0.37	−0.27	−0.17	−0.09	−0.19
3k	−0.07	−0.37	−0.27	−0.17	−0.06	−0.17
Indomethacin	0.24	−0.31	−0.11	0.42	−0.11	0.30
Aspirin	−0.76	−0.32	−1.06	−0.44	−0.82	−0.28

Table 4 Drug-likeness/scores and toxicity calculations of imidazolidine derivatives (**3a-k**) based on Osiris property explorer.

Compound	Solubility	Drug-likeness	Drug score	Tumorigenic	Reproductive effect	Irritant effect
3a	−4.56	−3.18	0.12	Yellow	Green	Green
3b	−4.79	−10.3	0.13	Yellow	Green	Green
3c	−4.79	−8.31	0.13	Yellow	Green	Green
3d	−5.54	−6.27	.07	Yellow	Green	Green
3e	−3.28	−2.96	0.17	Yellow	Green	Green
3f	−6.81	−1.97	0.06	Yellow	Green	Green
3g	−6.81	−2.6	0.06	Yellow	Green	Green
3h	−3.94	−1.16	0.15	Yellow	Green	Green
3i	−3.31	−2.07	0.16	Yellow	Green	Green
3j	−4.28	−1.69	0.14	Yellow	Green	Green
3k	−3.68	3.23	0.37	Red	Green	Green
Indomethacin	−5.4	9.41	0.56	Green	Green	Green
Aspirin	−1.93	−0.48	0.14	Red	Red	Green

standard drug, Indomethacin (67.09%). Three more compounds, **3f**, **3h** and **3k**, showed significant anti-inflammatory activity with 53.92%, 52.22% and 54.35% inhibition, respectively. Other compounds displayed modest to low activity with % inhibition in the range of 29.72–45.43%. It can be summarized that the anti-inflammatory activity of the synthesized compounds may be due to the inhibition of inflammatory mediators release and possibly due to the inhibition of cyclooxygenase synthesis similar to indomethacin. The results showed that disubstituted phenyl rings attached to the imidazolidine ring (**3f**, **3g**, **3h** & **3i**) exhibited improved activity as compared to that of the mono-substituted phenyl rings. The induction of methyl group at 4th position of the imidazolidine ring (**3j** & **3k**) enhanced the anti-inflammatory action. The occurrence of nitro group (**3b** & **3c**) to the mono-substituted phenyl rings on the imidazolidine ring demonstrated significant activity. Similar pattern of activity was also observed in analgesic screening of the compounds.

Compounds (**3f**, **3g**, **3h**, **3i**, **3j** & **3k**) which showed good anti-inflammatory activity (> 50% inhibition) were also evaluated for their analgesic activity in acetic acid induced writhing model in mice. The writhing induced by acetic acid is a sensitive method and has been used since long time as a screening tool to assess analgesics acting on periphery (Hunskaar and Hole, 1987). The inflammatory pain is primarily due to excitation of pain nerve ending by the release of endogenous substances (Kulkarni and Jain, 2005).

Two compounds, **3g** & **3i**, showed good analgesic action with 51.44% and 56.88% protection, respectively, and their analgesic activity was almost at par with standard drug, Aspirin, which showed 59.78% protection. Rest of the compounds illustrated analgesic activity in the range of 36.23–45.65% protection (**Table 1**). A significant difference ($p < 0.01$) was found in the analgesic activity of all tested compounds.

In ulcerogenic test, the tested compounds (**3f**, **3g**, **3h**, **3i**, **3j** & **3k**) showed reduced ulcerogenic activity (as per severity index), ranging from 1.08 to 0.42, while the standard drug indomethacin demonstrated high severity index of 2.08 (Table 1). The results demonstrated that the all the tested compounds were low in their GIT toxicity and are better tolerated than indomethacin. Compound **3k** showed the least sign of GI toxicity while compound **3i** was found to be the most toxic in synthesized compounds which showed almost double severity index as compared to **3k**. A significant difference in ulcerogenic activity was also observed for compounds **3g** and **3i** ($p < 0.05$). Compound **3i** emerged as the most effective anti-inflammatory and analgesic agent but with slightly high ulcerogenic index with respect to synthesized compounds. Compound **3j** exhibited better anti-inflammatory and ulcerogenic activity but showed weaker analgesic activity in comparison with **3g**.

4. Conclusions

The present work reports the synthesis, spectral characterization and biological activities including anti-inflammatory, analgesic and ulcerogenic activities of newly synthesized series of imidazolidine derivatives (**3a–k**). Anti-inflammatory and analgesic activity results revealed that three compounds, 4-[1,3-Bis(2,6-dichlorobenzyl)-2-imidazolidinyl]phenyl-diethylamine (**3g**), 4-[1,3-Bis(3-hydroxy-4-methoxybenzyl)-2-imidazolidinyl]phenyl-diethylamine (**3i**), and 4-(1,3-Bis(4-methoxybenzyl)-4-methylimidazolidin-2-yl)-phenyl-diethylamine (**3j**), were good in their bio-actions. Moreover, these new derivatives were also low in their ulcerogenic action (GIT toxicity) which is a common unwanted effect of all commonly prescribed NSAIDs. Thus it could be concluded that compounds containing imidazolidine ring possess significant anti-inflammatory and analgesic activities and these could lead to drug discovery or lead molecule for anti-inflammatory therapy. However, further detailed investigations such as *in-vivo* pharmacokinetic profile, toxicity, mechanism(s), are needed to evaluate their potential of developing into the therapeutic agents.

Acknowledgement

The authors are thankful to UGC, Govt. of India, New Delhi for providing financial assistance.

References

Actelion's property explorer, 2001, Thomas Sander, Actelion's Pharmaceuticals Ltd., Gewerbestrasse 16, 4123 Allschwil, Switzerland.

Agnihotri, S., Wakode, S., Agnihotri, A., 2010. An overview on anti-inflammatory properties and chemo-profiles of plants used in traditional medicine. Indian J. Nat. Products Resour. 1 (2), 150–167.

Allison, M.C., Howatson, A.G., Torrance, C.J., Lee, F.D., Russell, R.I.G., 1992. Gastrointestinal damage associated with the use of nonsteroidal antiinflammatory drugs. N. Engl. J. Med. 327 (11), 749–754.

Ashley, N.T., Weil, Z.M., Nelson, R.J., 2012. Inflammation: mechanisms, costs and natural variation. Annu. Rev. Ecol. Evol. Syst. 43, 385–406.

Buttgereit, F., Burmester, G., Simon, L.S., 2001. Gastrointestestinal toxic side effects of non-steroidal anti-inflammatory drugs and cyclooxygenase-2-specific inhibitors. Am. J. Med. 110, 135–195.

Caterina, M.C., Perillo, I.A., Boiani, L., Pezaroglo, H., Cerecetto, H., Gonzalez, M., Salerno, A., 2008. Imidazolidines as new anti-*Trypanosoma cruzi* agents: biological evaluation and structure-activity relationships. Bioorg. Med. Chem. 16 (5), 2226–2234.

Cioli, V., Putzolu, S., Rossi, V., Barcellona, P.S., Corradino, C., 1979. The role of direct tissue contact in the production of gastrointestinal ulcer by anti-inflammatory drugs in rats. Toxicol. Appl. Pharmacol. 50, 283–289.

Fernandes, E.S., Passos, G.F., Medeiros, R., 2007. Anti-inflammatory effects of compounds alpha-humulene and (-)-trans-caryophyllene isolated from the essential oil of *Cordia verbenacea*. Eur. J. Pharmcol. 569, 228–236.

Franzotti, E.M., Santos, C.V., Rodrigues, H.M., Mourao, R.H., Andrade, M.R., Antoniolli, A.R., 2000. Anti-inflammatory, analgesic activity and acute toxicity of *Sida cordifolia* L. (*Malva branca*). J. Ethnopharmacol. 72, 273–277.

Gautam, R., Jachak, S.M., 2009. Recent developments in anti-inflammatory natural products. Med. Res. Rev. 29 (5), 767–820.

Gupta, M., Mazumder, U.K., Gomathi, P., Selvan, V.T., 2006. Antiinflammatory evaluation of leaves of *Plumeria acuminate*. BMC Comp. Alter. Med. 6, 36.

Hunskaar, S., Hole, K., 1987. The formalin test in mice: dissociation between inflammatory and non-inflammatory pain. Pain 30, 103–114.

Husain, A., Bhutani, R., Kumar, D., Shin, D.S., 2013. Synthesis and biological evaluation of novel substituted-Imidazolidine derivatives. J. Kor. Chem. Soc. 57 (2), 227–233.

Khan, M.S., Chawla, G., 2002. Tetrahydroimidazoles-A promising group of expected NSAIDS-their synthesis and anti-inflammatory activity. Indian J. Chem. 41B, 653–663.

Khan, M.S., Gupta, M., 2003. Synthesis and evaluation of anti-inflammatory and analgesic activity of some new 1,3-diphenyl-2-aryltetrahydroimidazoles. Indian J. Chem. 42B, 2086–2090.

Khan, M., Husain, A., Sharma, S., Rashid, M., 2012. Microbiological evaluation of 4-substituted-imidazolidine derivatives. Indian J. Pharm. Sci. 74, 80–83.

Kulkarni, S.K., Jain, N.K., 2005. Coxibs: the new super aspirins or unsafe pain killers? Indian J. Pharmacol. 37, 86–89.

Lanza, F.L., 1998. A guideline for the treatment and prevention of NSAID-induced ulcers. Am. J. Gastroenterol. 93, 2037–2046.

Lipinski, C.A., Lombardo, F., Dominy, B.W., Feeney, P.J., 1997. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. Adv. Drug. Deliver Rev. 23, 4–25.

Mantovani, A., Pierotti, M.A., 2008. Cancer and inflammation: a complex relationship. Cancer Lett. 267, 180–181.

Marki, F., Buch, O., Delini-Stula, A., Kraetz, J., Petermann, H., Radeka, E., Schweizer, A., Thomann, P., Truog, A., 1984. Sulfonyliminoimidazolidines, a new class of oral hypoglycemic agents, Toxicity and general pharmacology of 1-[p-[2-(crotonylamino)-ethyl]-phenylsulfonyl]-3-cyclohexy l-2-imino- imidazolidine (CGP 11 112. Arzneimittelforschung 34 (3), 247–252.

Neves, J.K., filter your current search Botelho, SP., filter your current search Melo, CM., filter your current search Pereira, VR., filter your current search Lima, MC., filter your current search Pitta, IR., filter your current search Albuquerque, MC., filter your current search Galdino, Sl., 2010. Biological and immunological activity of new imidazolidines against adult worms of Schistosoma mansoni. filter your current search Parasitol. Res. 107(3), 531–538.

Patel, M.V., Bell, R., Majest, S., Henry, R., Kolasa, T., 2004. Synthesis of 4,5-Diaryl-1H-pyrazole-3-ol derivatives as potential COX-2 inhibitors. J. Org. Chem. 69, 7058–7065.

Ricciotti, E., Fitzgerald, G.A., 2011. Prostaglandins and Inflammation. Arterioscler. Thromb. Vasc. Biol. 31 (5), 986–1000.

Robert, J.G., Daryl, S.W., Paul, J.B., Elena, F., Anton, D.M., Shilina, A.R., Sac-Pham, T., 2010. Synthesis and biological activity of a series of tetrasubstituted-imidazoles as P2X 7 antagonists. Bioorg. Med. Chem. Lett. 20 (16), 4951–4954.

Saczewski, J., Hudson, A.L., Rybczynska, A., 2009. 2-[(aryl-methoxy)imino]imidazolidines with potential biological activities. Acta Poloniae Pharmaceutia-Drug Research. 66 (6), 671–679.

Seigmund, E., Cadmus, R., Lu, G., 1957. A method for evaluating both non-narcotic and narcotic analgesics. Proc. Soc. Exp. Biol. 95, 729–731.

Sharma, V., Khan, M.S., 2001. Synthesis of novel tetrahydroimidazole derivatives and studies for their biological properties. Eur. J. Med. Chem. 36, 651–658.

Tan, T.M., Chen, Y., Kong, K.H., Bai, J., Li, Y., Lim, S.G., 1992. NSAID, ulcer and prostaglandins. J. Rhematol. 19, 68–73.

Verrico, M.M., Weber, R.J., McKaveney, T.P., Ansani, N.T., Towers, A.L., 2003. Adverse drug events involving COX-2 inhibitors. Ann. Pharmacother. 37 (9), 1203–1213.

Winter, C.A., Risley, E.A., Nuss, G.W., 1962. Carrageenin-induced edema in hind paw of the rat as an assay for anti-inflammatory drugs. Proc. Soc. Exp. Biol. 111, 544–547.

Zhao, Y., Abraham, M.H., Le, J., Hersey, A., Luscombe, C.N., Beck, G., Sherborne, B., Cooper, I., 2002. Rate-limited steps of human oral absorption and QSAR studies. Pharm. Res. 19 (10), 1446–1457.