Bioorganic & Medicinal Chemistry 20 (2012) 2199–2207

Contents lists available at SciVerse ScienceDirect

Bioorganic & Medicinal Chemistry

journal homepage: www.elsevier.com/locate/bmc

C–C bond formation at C-2 of a quinoline ring: Synthesis of 2-(1*H*-indol-3-yl) quinoline-3-carbonitrile derivatives as a new class of PDE4 inhibitors

K. Shiva Kumar [a], S. Kiran Kumar [b], B. Yogi Sreenivas [a], Dhilli Rao Gorja [a], Ravikumar Kapavarapu [a], D. Rambabu [a,b], G. Rama Krishna [c], C. Malla Reddy [c], M.V. Basaveswara Rao [d,*], Kishore V.L. Parsa [a], Manojit Pal [a,*]

[a] *Institute of Life Sciences, University of Hyderabad Campus, Gachibowli, Hyderabad 500 046, India*
[b] *Department of Chemistry, K. L. University, Vaddeswaram, Guntur 522 502, AP, India*
[c] *Department of Chemical Sciences, Indian Institute of Science Education and Research, Kolkata 741 252, West Bengal, India*
[d] *Department of Chemistry, Krishna University, Machilipatnam 521 001, AP, India*

ARTICLE INFO

Article history:
Received 18 January 2012
Revised 7 February 2012
Accepted 8 February 2012
Available online 16 February 2012

Keywords:
Quinoline
Indole
$AlCl_3$
X-ray
PDE4

ABSTRACT

A number of 2-(1*H*-indol-3-yl)quinoline-3-carbonitrile derivatives were synthesized via $AlCl_3$-mediated C–C bond forming reaction between 2-chloroquinoline-3-carbonitrile and various indoles. The methodology does not require any N-protection of the indoles employed and provided the corresponding products in good yields. The molecular structure of a representative compound was established unambiguously by single crystal X-ray diffraction and structural elaboration of a compound synthesized has been demonstrated. Many of these compounds synthesized showed PDE4 inhibitory properties in vitro. A brief structure–activity relationship studies within the series along with docking results of a representative compound (EC_{50} ~0.89 μM) is presented.

1. Introduction

Quinoline derivatives in addition to their natural occurrence,[1,2] for example, camptothecin (**A**),[1] and luotonin A (**B**),[2] (Fig. 1) occupy a prominent place in medicinal chemistry due to their diverse pharmacological properties[3–5] such as antimalarial,[3] antitumor,[4] and antibacterial[5] activities. Indoles, on the other hand are considered as privileged structures in the area of drug discovery.[6a] Thus combination of structural features of both in a single scaffold is expected to provide new chemical space that might lead to the identification of novel molecules possessing noteworthy pharmacological properties. For example, 2-(1*H*-indol-3-yl)quinoline derivative **C** (Fig. 1) has been shown to posses promising antibacterial activities.[6b]

In our effort[7a–e] to identify novel inhibitors of PDE4B (phosphodiesterase type 4B) we became interested in exploring the 2-(1*H*-indol-3-yl)quinoline framework for the design of our target molecules (Fig. 2). While indole **D**[7f] and quinoline-based derivatives **E**[7g,h] have been reported individually as potent inhibitor of PDE4 there is no report available on PDE4 inhibitory properties of 2-heteroaryl substituted quinolines including 2-indolyl quinolines. Moreover, the fact that 3-cyano substituted pyridine derivatives **F** (Fig. 2) have shown duel inhibition of PDE4 and IL-23 release[7i] indicated a possible role played by the cyano group in the observed pharmacological activities of these compounds. Taking into consideration of core moiety of **C** (Fig. 1) we therefore designed our target molecules represented by the structure **G** by attaching a cyano group at an appropriate position of the 2-indolyl quinoline framework as shown in Figure 2. Thus the generation of a library of small molecules based on the framework **G** was undertaken. Herein we report the synthesis (Scheme 1) and in vitro pharmacological evaluation of a series of 2-(1*H*-indol-3-yl)quinoline-3-carbonitriles (**3**) as potential inhibitors of PDE4B.

2. Results and discussion

2.1. Chemistry

Introduction of an aryl or indole moiety at C-2 of quinoline ring can be carried out either via Ni-catalyzed reaction of quinoline with arylzinc[8a] or Suzuki coupling of 2-chloroquinoline with arylboronic acids[8b] or reaction of quinoline *N*-oxide with indole.[6b] Alternatively, a quinoline ring can be constructed at C-3 of an indole ring.[6b] While each of these methods has their own merit for the synthesis of specific class of compounds we required a more

* Corresponding authors. Tel.: +91 40 6657 1500; fax: +91 40 6657 1581.
E-mail address: manojitpal@rediffmail.com (M. Pal).

0968-0896/$ - see front matter © 2012 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmc.2012.02.027

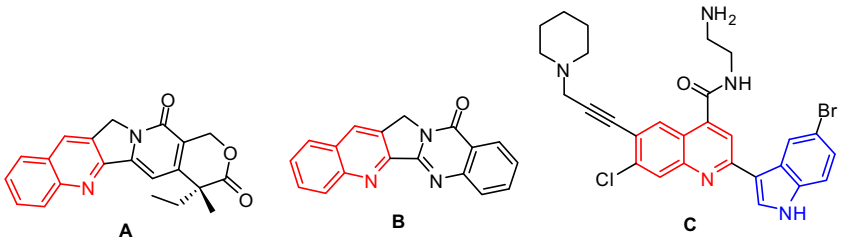


Figure 1. Quinoline-containing alkaloids and biologically active compounds.

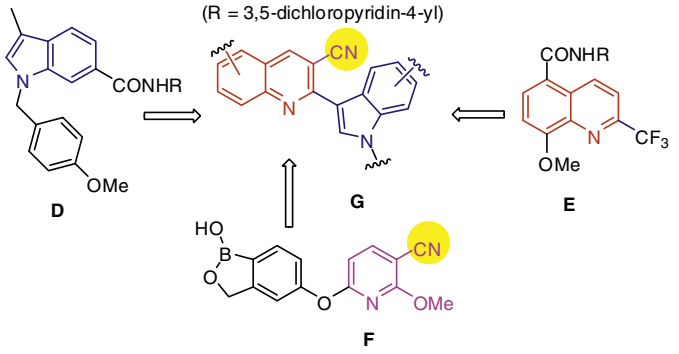
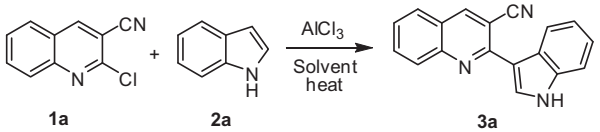


Figure 2. Design of 2-indolyl quinoline based new inhibitors (**G**) of PDE4.

straightforward and inexpensive procedure for the preparation of our desired 2-(1*H*-indol-3-yl)quinolines. Over the years we have been working on the use of $AlCl_3$ as an efficient and inexpensive reagent for the C–C bond forming reactions between heteroaryl chlorides containing –C(Cl)=N– moiety and various arenes or heteroarenes.[9] In further continuation of this research we decided to explore the potential of $AlCl_3$ mediated process in the preparation of our target molecules. Initially, the reaction of 2-chloroquinoline-3-carbonitrile[10] (**1a**) with indole (**2a**) was used to establish the optimum reaction conditions and the results are summarized in Table 1. The reaction was carried out using **1a** (1.0 equiv), **2a** (1.1 equiv) and $AlCl_3$ (1.2 equiv) in a solvent generally at 80 °C (for a lower boiling solvent the reaction was carried out at lower temperature, see Table 1). A number of solvents were investigated including 1,2-dichloroethane (Table 1, entry 1), chloroform (Table 1, entry 2), ethylacetate (Table 1, entry 3), acetonitrile (Table 1, entry 4) and toluene (Table 1, entry 5). The best result was obtained using dichloroethane as a solvent both in terms of product yield and reaction time. While all these reactions were generally carried out using 1.2 equiv of $AlCl_3$ the use of lower quantity of $AlCl_3$, for example, 0.8, 0.5 and 0.2 equiv was also examined. However, the yield of product was decreased significantly in these cases.

Having the optimized conditions in hand we then examined the reaction of chloro derivative **1** with a range of indoles **2**. The reaction proceeded well irrespective of the substituents present on the indole ring and a variety of 2-(1*H*-indol-3-yl)quinolines were obtained in good to excellent yields (Table 2). Notably, the methodology does not require any N-protection of the indoles employed and provided the corresponding products smoothly (Table 2, entries 1, 3–12). Moreover, due to the easy availability of starting materials and non-usage of expensive catalysts or complex ligands the present methodology may have advantages over the existing processes that involved the use of arylzinc[8a] or arylboronic acid[8b] or quinoline *N*-oxide.[6b] The present methodology therefore is a straightforward process and because of its operational simplicity the methodology appeared to be amenable for scale up synthesis of same or similar class of compounds. All the compounds synthesized were well characterized by spectral (NMR, MS and IR) and analytical data. Additionally, the molecular structure of a representative compound **3a** was established unambiguously by single crystal X-ray diffraction (Fig. 3).[11]

Table 1
Effect of reaction conditions on $AlCl_3$-induced heteroarylation of indole (**2a**) with 2-chloroquinoline-3-carbonitrile (**1a**)[a]



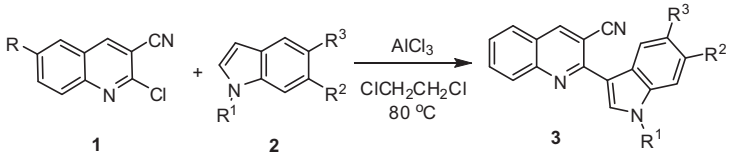
1a 2a 3a

Entry	Solvent	Time (h)	Yield[b] (%)
1	$ClCH_2CH_2Cl$	7	88
2	$CHCl_3$	12	75[c]
3	EtOAc	14	80
4	CH_3CN	12	83
5	Toluene	12	60

[a] All the reactions were carried out using compound **1a** (1.0 equiv), **2a** (1.1 equiv) and $AlCl_3$ (1.2 equiv) in a solvent (5 mL) at 80 °C.
[b] Isolated yield.
[c] The reaction was carried out at 60 °C.

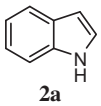
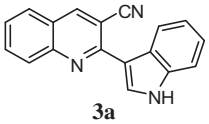
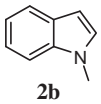
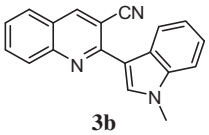
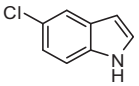
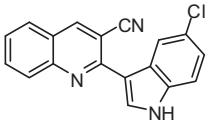
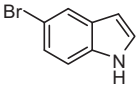
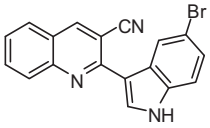
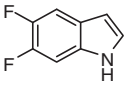
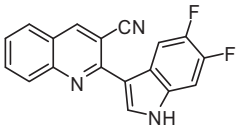
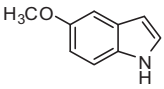
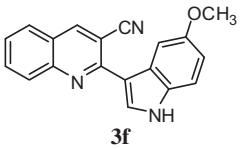
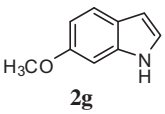
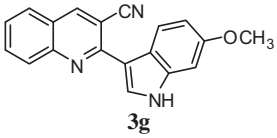
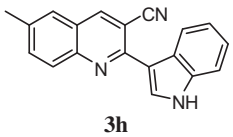
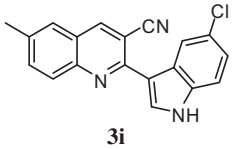
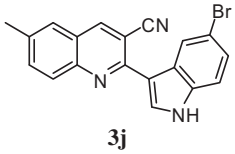
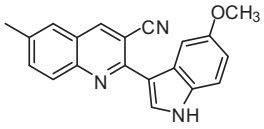
Mechanistically, the reaction proceeds through the complexation of $AlCl_3$ with the ring nitrogen of 2-chloroquinoline-3-carbonitrile followed by nucleophilic attack through C-3 of an indole at the adjacent carbon and finally release of $AlCl_3$ affording the desired product **3** (Scheme 2). In order to understand the role CN group in the present reaction 2-chloroquinoline was reacted with indole (**2a**) under the same reaction conditions, that is, in the presence of $AlCl_3$ in 1,2-dichloroethane at refluxing temperature for 6 h. The reaction proceeded well affording the expected 2-(1*H*-indol-3-yl)quinoline in 79% yield indicating insignificant or no role played by the CN group in the present C–C bond forming reaction.

Having prepared a variety of 2-(1*H*-indol-3-yl)quinoline-3-carbonitriles (**3**) we decided to explore the further structural elaboration of some of the compounds synthesized. Accordingly,



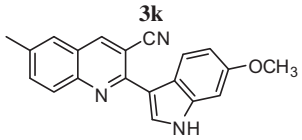
Scheme 1. Synthesis of 2-(1*H*-indol-3-yl)quinoline-3-carbonitriles.

Table 2
Synthesis of 2-(1*H*-indol-3-yl)quinoline derivatives (**3**) via $AlCl_3$-mediated C–C bond forming reaction between 2-chloroquinoline-3-carbonitrile (**1**) and indoles (**2**) (Scheme 1)[a]

Entry	**1**; R=	Reactant (**2**)	Product (**3**)	Time (h)	Yield[b] (%)
1	**1a**; H	 **2a**	 **3a**	6	88
2	**1a**	 **2b**	 **3b**	6	85
3	**1a**	 **2c**	 **3c**	**7**	80
4	**1a**	 **2d**	 **3d**	**7**	82
5	**1a**	 **2e**	 **3e**	6	80
6	**1a**	 **2f**	 **3f**	**7**	75
7	**1a**	 **2g**	 **3g**	**7**	72
8	**1b**; CH_3	**2a**	 **3h**	6	85
9	**1b**	**2c**	 **3i**	7	81
10	**1b**	**2d**	 **3j**	7	80
11	**1b**	**2f**		7	75

(*continued on next page*)

Table 2 (*continued*)

Entry	**1**; R=	Reactant (**2**)	Product (**3**)	Time (h)	Yield[b] (%)
12	**1b**	**2g**	**3k**  **3l**	7	78

[a] All the reactions were carried out using compound **1** (1.0 equiv), an indole **2** (1.1 equiv) and $AlCl_3$ (1.2 equiv) in 1,2-dichloroethane (5 mL) at refluxing temperature.
[b] Isolated yields after column chromatography.

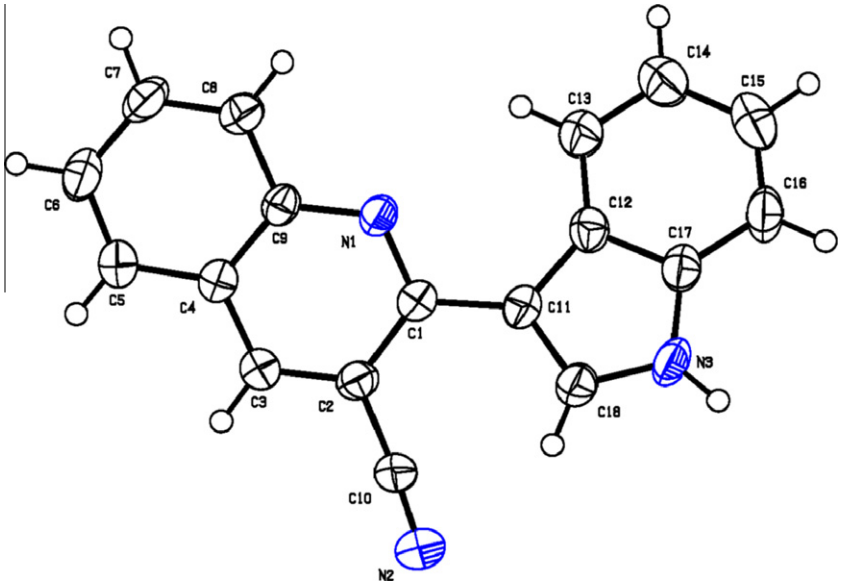
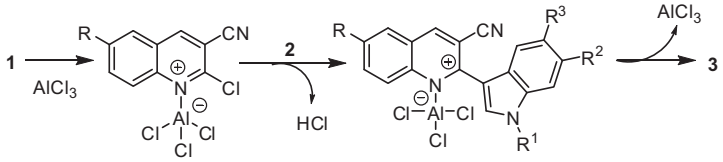
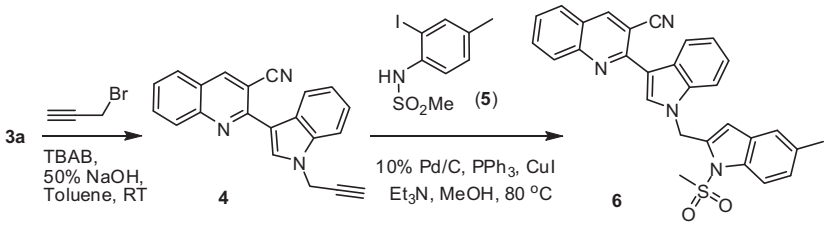


Figure 3. ORTEP representation of the compound **3a** (thermal ellipsoids are drawn at 50% probability level).



Scheme 2. Proposed mechanism of the $AlCl_3$ induced C–C bond forming reaction between **1** and **2**.



Scheme 3. Structural elaboration of compound **3a**.

compound **3a** was converted to 2-(1-(prop-2-ynyl)-1*H*-indol-3-yl)quinoline-3-carbonitrile (**4**) under a similar reaction condition reported earlier.[12] The alkyne **4** was then converted to a 2-substituted indole (**6**) in one pot via a tandem Pd/C mediated coupling-cyclization with the *o*-iodoanilide (**5**) (Scheme 3).[13]

2.2. Pharmacology

Some of the compounds synthesized were tested for their PDE4B inhibitory potential in vitro.[14] In inflammatory and immune cells, the inhibition of cellular responses, including the production and/

or release of proinflammatory mediators, cytokines, and active oxygen species, is associated with elevated levels of cAMP. PDE4, exists in four different isoforms (PDE4A, B, C and D), is specific for the hydrolysis of cAMP to AMP.[7a] Thus, inhibition of PDE4 should result in elevated levels of cAMP in the airway tissues and cells thereby suppressing inflammatory cell function. Indeed, inhibitors of PDE4 have been reported to be beneficial for the treatment of inflammatory and immunological diseases including asthma and chronic obstructive pulmonary disease (COPD). While the dose-limiting side effects for example, nausea and vomiting associated with the first-generation PDE4 inhibitor rolipram[15] were reduced by second-generation inhibitors like cilomilast[16] (Ariflo) and roflumilast, their therapeutic index has delayed market launch so far. Recent studies have indicated that PDE4B subtype is linked to inflammatory cell regulation[17] while the PDE4D subtype is implied in the emetic response.[18] It is therefore desirable to identify potent inhibitors of PDE4B for the potential treatment of asthma and COPD.

In a cell based cAMP reporter assay[14] fold increase of the cAMP level caused by the test compounds over forskolin control was determined. Compounds **3a–l** showed fold increase when tested at 10 μM (Fig. 4). Substituents like Cl (**3c**), Br (**3d**) and F (**3e**) on the indole ring were found to be less effective compared to OMe (**3f**, **3g**, **3k** and **3l**). While OMe was found to be the best among all substituents tested its position was found to be vital (**3f** and **3k** vs **3g** and **3l**). In a dose response study compound **3g** showed dose-dependent fold increase of the cAMP level with an EC_{50} value of 0.89 μM that was comparable to rolipram's EC_{50} of ~0.22 μM in the same assay. Thus, compound **3g** was identified as a promising inhibitor of PDE4B.

To understand the nature of interaction[19] of compound **3g** with PDE4B protein a docking study was performed via the energy minimization and conformational search with the MACROMODEL application in the Schrodinger package. The molecule **3g** was energy minimized for flexibility and then conformational search was performed (see the Section 4). The PDE4B protein (3D3P) crystal structures were retrieved from the protein data bank and a GRID based docking was performed in the present case (see the Section 4). The docking results of **3g** with PDE4 protein (Fig. 5) showed H-boning interaction of the –NH group of indole ring of **3g** with the –C═O group of the ASP392 residue of the PDE4B protein. The Glide score and other parameters of this interaction is shown below

Total glide score = −10.9 K cal/mol; total fraction of the VdW energy in protein–ligand interaction = −5.4 K cal/mol; hydrophobic ensure reward in the interaction = −1.8 K cal/mol; electrostatic rewards = −0.1 K cal/mol; Chemscore H-bond pair term = −0.7 K cal/mol.

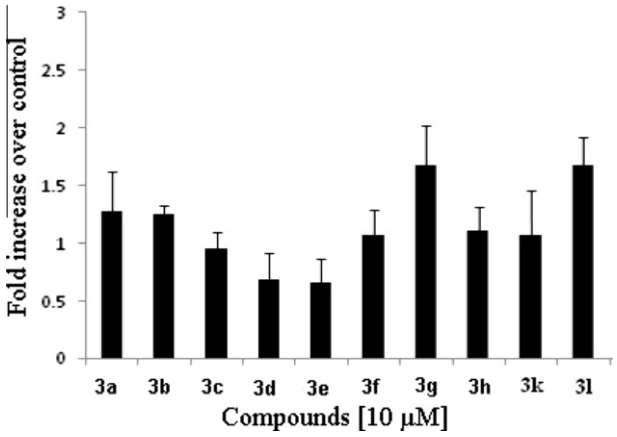


Figure 4. PDE4B HEK 293 cell based reporter screen of **3** (*Y*-axis: fold elevation of cAMP over forskolin control).

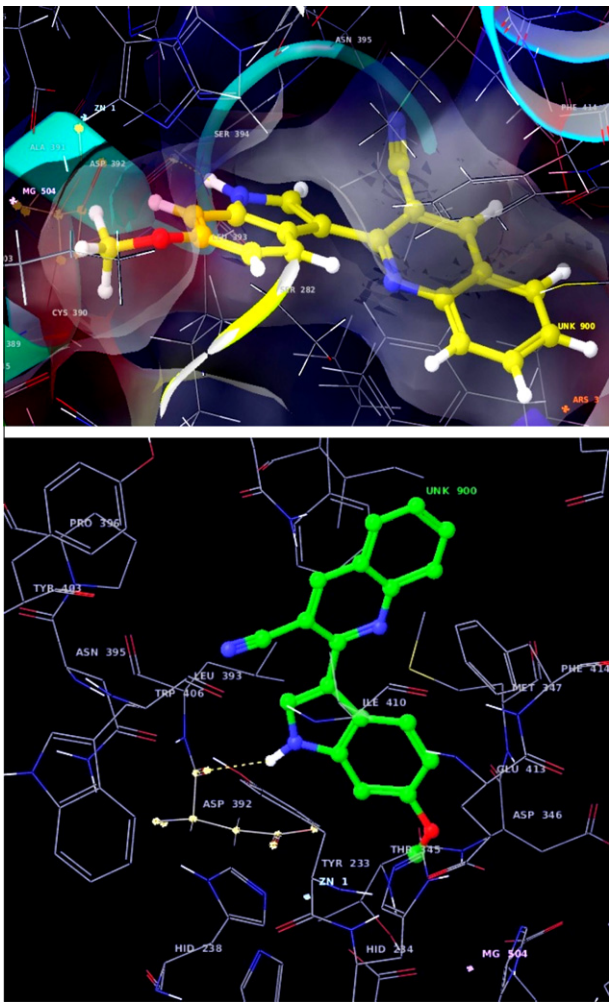


Figure 5. Docking of **3g** at the active site of PDE4B.

Overall, the docking results indicate that the compound **3g** binds well with the PDE4 protein.

3. Conclusions

In conclusion, a number of 2-(1*H*-indol-3-yl)quinoline-3-carbonitrile derivatives were synthesized via $AlCl_3$-mediated C–C bond forming reaction between 2-chloroquinoline-3-carbonitrile and an appropriate indole. The methodology does not require the use of expensive catalysts or complex ligands. All the starting materials used are either commercially available or can be readily made. The methodology appeared to have advantages over the transition-metal-mediated synthesis especially in the large-scale preparation of 2-(1*H*-indol-3-yl)quinoline derivatives. The methodology can be viewed as a useful alternative to the Suzuki reactions as preparation of required boronic acids might be cumbersome. Moreover, the methodology does not require any N-protection of the indoles employed and provided the corresponding products in good yields. The molecular structure of a representative compound was established unambiguously by single crystal X-ray diffraction and structural elaboration of a compound synthesized has been demonstrated. Many of these derivatives showed PDE4B inhibitory properties in vitro and docking studies using the most active compound (EC_{50} ~0.89 μM) is presented. Overall, 2-(1*H*-indol-3-yl)quinoline framework provides a useful basis for the development of novel PDE4 inhibitors for the potential treatment of asthma and COPD.

4. Experimental section

4.1. Chemistry

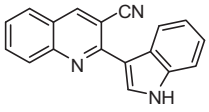
4.1.1. General methods

Unless stated otherwise, reactions were performed under nitrogen atmosphere using oven dried glassware. Reactions were monitored by thin layer chromatography (TLC) on silica gel plates (60 F254), visualizing with ultraviolet light or iodine spray. Flash chromatography was performed on silica gel (230–400 mesh) using distilled hexane, ethyl acetate, dichloromethane. ^{1}H NMR and ^{13}C NMR spectra were determined in $CDCl_3$ or DMSO-d_6 solution by using 400 or 100 MHz spectrometers, respectively. Proton chemical shifts (δ) are relative to tetramethylsilane (TMS, $\delta = 0.00$) as internal standard and expressed in ppm. Spin multiplicities are given as s (singlet), d (doublet), t (triplet) and m (multiplet) as well as b (broad). Coupling constants (J) are given in hertz. Infrared spectra were recorded on a FT-IR spectrometer. Melting points were determined using melting point apparatus and are uncorrected. MS spectra were obtained on a mass spectrometer.

4.1.2. Typical procedure for the synthesis of 2-hetero substituted quinoline-3-carbonitrile (3a–l)

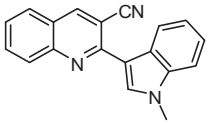
A mixture of 2-chloroquinoline-3-carbonitrile derivative[10] (**1**, 1.0 equiv), an appropriate indole (**2**) (1.1 equiv) and anhydrous $AlCl_3$ (1.2 equiv) in dichloroethane (5 mL) was stirred at 80 °C for time indicated in Table 2 under a nitrogen atmosphere. After completion of the reaction, the mixture was poured into ice-cold water (15 mL), stirred for 10 min and then extracted with ethylacetate (3 × 20 mL). The organic layers were collected, combined, washed with cold water (2 × 20 mL), dried over anhydrous Na_2SO_4 and concentrated under vacuum. The residue obtained was purified by column chromatography using ethylacetate /hexene to afford the desired product.

4.1.3. 2-(1*H*-Indol-3-yl)quinoline-3-carbonitrile (3a)



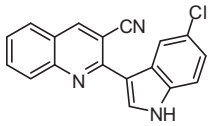
Brown solid; mp 238–240 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.80 (bs, 1H), 9.01 (s, 1H), 8.60 (d, J = 7.2 Hz, 1H), 8.38 (s, 1H), 8.08–7.99 (m, 2H), 7.91–7.88 (m, 1H), 7.63–7.59 (m, 1H), 7.52–7.50 (m, 1H), 7.25–7.17 (m, 2H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 152.9, 148.3, 145.2, 136.6, 133.3, 128.6, 128.4, 127.0, 126.1, 123.6, 122.8, 122.4, 121.0 (2C), 119.2, 113.1, 112.1, 103.3; HPLC: 99.8%, column: X DB C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.05% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/70, 2/70, 9/95, 12/95, 15/70, 18/70; flow rate: 1.0 mL/min; UV 248 nm, retention time 3.90 min; IR (KBr) υ_{max} 3315, 3053, 2226, 1542, 1433 cm^{-1}; HRMS (ESI) calcd for $C_{18}H_{12}N_3$ (M+H)$^+$ 270.1031, found 270.1021.

4.1.4. 2-(1-Methyl-1*H*-indol-3-yl)quinoline-3-carbonitrile (3b)



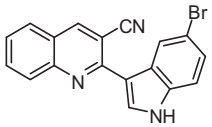
Brown solid; mp 161–163 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.80 (d, J = 6.4 Hz, 1H), 8.55 (s, 1H), 8.29 (s, 1H), 8.17 (d, J = 8.8 Hz, 1H), 7.84–7.79 (m, 2H), 7.56–7.52 (m, 1H), 7.41–7.33 (m, 3H), 3.92 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 153.0, 149.0, 144.2, 137.3, 133.8, 132.6, 129.3, 127.6, 127.1, 126.6, 123.6, 123.0 (2C), 121.5, 119.3, 113.0, 109.4, 103.7, 33.4; IR (KBr) υ_{max} 2961, 2278, 1924, 1521, 1179 cm^{-1}; HRMS (ESI) calcd for $C_{19}H_{14}N_3$ (M+H)$^+$ 284.1188, found 284.1190.

4.1.5. 2-(5-Chloro-1*H*-indol-3-yl)quinoline-3-carbonitrile (3c)



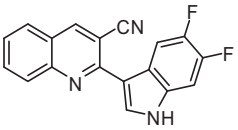
Mp 262–264 °C ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.99 (bs, 1H), 9.07 (s, 1H), 8.62 (d, J = 1.6 Hz, 1H), 8.49 (d, J = 2.8 Hz, 1H), 8.10–8.08 (m, 1H), 8.03–8.01 (m, 1H), 7.94–07.90 (m, 1H), 7.67–7.63 (m, 1H), 7.56–7.54 (m, 1H), 7.27–7.24 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 152.3, 148.1, 145.3, 135.0, 133.3, 129.8, 128.5, 128.4, 127.1 (2C), 125.6, 123.6, 122.7, 121.4, 119.0, 113.6, 112.6, 103.1; IR (KBr) υ_{max} 3331, 2226, 1536, 1435 cm^{-1}; HRMS (ESI) calcd for $C_{18}H_{11}N_3Cl$ (M+H)$^+$ 304.0642, found 304.0632.

4.1.6. 2-(5-Bromo-1*H*-indol-3-yl)quinoline-3-carbonitrile (3d)



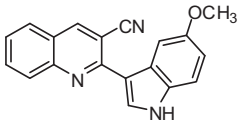
Mp 265–266 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.95 (d, J = 2.0 Hz, 1H), 8.67 (bs, 1H), 8.59 (s, 1H), 8.44 (d, J = 3.2 Hz, 1H), 8.24 (d, J = 7.6 Hz, 1H), 7.90–7.83 (m, 2H), 7.62–7.58 (m, 1H), 7.44–7.41 (m, 1H), 7.37–7.34 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 152.3, 148.1, 145.3, 135.3, 133.4, 129.7, 128.5, 128.4, 127.8, 127.1, 125.3, 124.4, 123.6, 119.0, 114.1, 113.7, 112.5, 103.2; HPLC: 97.2%, column: X Bridge C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.05% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/90, 2/90, 8/98, 10/80, 10/98, 12/90, 15/90; flow rate: 1.0 mL/min; UV 248 nm, retention time 4.08 min; IR (KBr) υ_{max} 2961, 2278, 1924, 1521, 1179 cm^{-1}; HRMS (ESI) calcd for $C_{18}H_{11}N_3Br$ (M+H)$^+$ 348.0113, found 348.0136.

4.1.7. 2-(5,6-Difluoro-1*H*-indol-3-yl)quinoline-3-carbonitrile (3e)



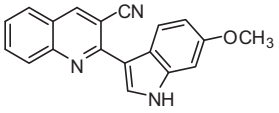
Mp 303–304 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.94 (bs, 1H), 9.03 (s, 1H), 8.55–8.48 (m, 2H), 8.13–8.11 (m, 1H), 8.02–8.00 (m, 1H), 7.93–7.89 (m, 1H), 7.65–7.61 (m, 1H), 7.56–7.52 (m, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 152.4, 148.4, 145.7, 133.7, 132.1, 132.0, 130.2, 128.9 (2C), 128.7, 127.5, 123.9, 119.3, 109.5, 109.3, 103.2, 100.5, 100.3; HPLC: 97.21%, column: X Bridge C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.05% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/90, 2/90, 8/98, 10/80, 10/98, 12/90, 15/90; flow rate: 1.0 mL/min; UV 248 nm, retention time 5.05 min; IR (KBr) υ_{max} 2961, 2278, 1924, 1521, 1179 cm^{-1}; HRMS (ESI) calcd for $C_{18}H_9N_3F_2$ (M+H)$^+$ 306.0828, found 306.0843.

4.1.8. 2-(5-Methoxy-1*H*-indol-3-yl)quinoline-3-carbonitrile (3f)



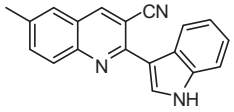
Mp 210–212 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.69 (bs, 1H), 9.03 (s, 1H), 8.39 (d, J = 3.2 Hz, 1H), 8.21 (d, J = 2.4 Hz, 1H), 8.09–8.01 (m, 1H), 7.99–7.92 (m, 1H), 7.90–7.88 (m, 1H), 7.63–7.60 (t, J = 6.8 Hz, 1H), 7.43 (d, J = 8.8, 1H), 6.89 (d, J = 2.0 Hz, 1H), 3.83 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.1, 153.3, 148.6, 145.5, 133.5, 131.8, 129.1, 128.8, 128.7, 127.1 (2C), 123.7, 119.6 (2C) 113.1, 113.0, 104.5, 103.2, 55.6; HPLC: 97.21%, column: X Bridge C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.05% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/90, 2/90, 8/98, 10/80, 10/98, 12/90, 15/90; flow rate: 1.0 mL/min; UV 248 nm, retention time 2.72 min; IR (KBr) υ_{max} 2961, 2278, 1924, 1521, 1179 cm^{-1}; HRMS (ESI) calcd for $C_{19}H_{14}N_3O$ $(M+H)^+$ 300.1137, found 300.1127.

4.1.9. 2-(6-Methoxy-1*H*-indol-3-yl)quinoline-3-carbonitrile (3g)



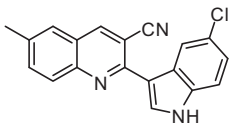
Mp 224–225 °C ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.60 (bs, 1H), 9.02 (s, 1H), 8.52 (d, J = 8.4 Hz, 1H), 8.31 (d, J = 2.8 Hz, 1H), 8.30–8.06 (m, 1H), 8.0–7.98 (m, 1H), 7.91–7.87 (m, 1H), 7.63–7.59 (t, J = 7.4 Hz, 1H), 7.00 (s, 1H), 6.85 (d, J = 8.8 Hz, 1H), 3.38 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.4, 152.7, 148.2, 145.2, 133.1, 128.5 (2C), 128.3, 127.2 (2C), 126.8, 123.4, 123.2, 120.2, 119.2, 113.1, 110.9, 102.9, 55.2; HPLC: 97.1%, column: ZORBAX Eclipse XDB-C18 150 × 4.6 mm 5 μ, mobile phase A: 0.05% Formic acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/70, 2/70, 9/95, 12/95, 15/70, 18/70; flow rate: 1.0 mL/min; UV 240 nm, retention time 2.73 min; IR (KBr) υ_{max} 3375, 2217, 1585, 1428, 1270 cm^{-1}; HRMS (ESI) calcd for $C_{19}H_{14}N_3O$ $(M+H)^+$ 300.1137, found 300.1127.

4.1.10. 2-(1*H*-indol-3-yl)-6-methylquinoline-3-carbonitrile (3h)



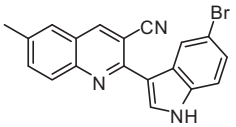
Brown solid; mp 266–268 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.72–8.74 (m, 1H), 8.60 (bs, 1H), 8.49 (s, 1H), 8.37 (d, J = 4 Hz, 1H), 8.10 (d, J = 8.8 Hz, 1H), 7.68 (dd, J = 7.2 and 2 Hz, 1H), 7.59 (s, 1H), 7.46–7.48 (m, 1H), 7.30–7.34 (m, 2H), 2.56 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 152.5, 147.2, 144.7, 136.9, 135.6, 128.7, 128.4, 127.2, 126.4, 123.8, 123.0, 122.7, 121.1, 119.6, 113.4, 112.3 (2C), 103.5, 21.4; HPLC: 99.3%, column: Zorbax XDB C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.05% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/80, 2/80, 9/98, 12/98, 15/80, 18/80; flow rate: 1.0 mL/min; UV 245 nm, retention time 3.29 min; IR (KBr) υ_{max} 3330, 3044, 2232, 1492, 1392 cm^{-1}; HRMS (ESI) calcd for $C_{19}H_{13}N_3$ $(M+H)^+$ 284.1190, found 284.1188.

4.1.11. 2-(5-Chloro-1*H*-indol-3-yl)-6-methylquinoline-3-carbonitrile (3i)



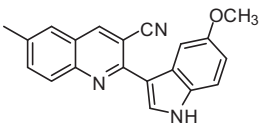
Off white solid; mp 240–242 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 11.99 (bs, 1H), 9.0 (s, 1H), 8.75 (s, 1H), 8.54 (s, 1H), 7.87 (d, J = 7.6 Hz, 1H), 7.80 (d, J = 7.6, 1H), 7.57–7.52 (s, 2H), 7.28–7.25 (m, 1H), 2.82 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 151.5, 147.4, 145.9, 136.4, 135.4, 133.6, 130.2, 127.5, 127.1, 126.7, 126.0, 123.9, 123.0, 122.0, 119.6, 114.0, 113.6, 102.9, 18.4; HPLC: 98.5%, column: X-BRIDGE C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.1% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/70, 2/70, 9/98, 14/98, 15/70, 18/70; flow rate: 1.0 mL/min; UV 225 nm, retention time 7.24 min; IR (KBr) υ_{max} 3336, 2205, 1580, 1494 cm^{-1}; m/z (CI) 318 (M+1, 100%). Elemental analysis found C, 71.68; H, 3.80; N, 13.39 $C_{19}H_{12}ClN_3$ requires C, 71.81; H, 3.81; N, 13.22.

4.1.12. 2-(5-Bromo-1*H*-indol-3-yl)-6-methylquinoline-3-carbonitrile (3j)



Light yellow solid; mp 231–233 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.98 (bs, 1H), 9.0 (s, 1H), 8.93 (s, 1H), 8.51 (s, 1H), 7.85 (d, J = 7.6, 1H), 7.80 (d, J = 7.6, 1H), 7.54–7.50 (m, 2H), 7.38–7.35 (m, 1H), 2.80 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 151.5, 147.4, 145.9, 136.4, 135.6, 133.6, 130.0, 128.1, 127.1, 126.7, 125.5, 125.2, 123.9, 119.6, 114.4, 114.1, 113.5, 102.8, 18.3; HPLC: 98.4%, column: X Bridge C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.1% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/80, 2/80, 9/98, 14/98, 15/80, 18/80; flow rate: 1.0 mL/min; UV 225 nm, retention time 5.50 min; IR (KBr) ν_{max} 3330,, 2225, 1538, 1432 cm^{-1}; m/z (CI) 362 (M+1, 100%). Elemental analysis found C, 63.15; H, 3.30; N, 11.43 $C_{19}H_{12}BrN_3$ requires C, 63.00; H, 3.34; N, 11.60.

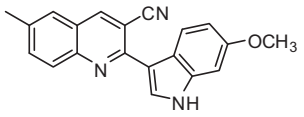
4.1.13. 2-(5-Methoxy-1*H*-indol-3-yl)-6-methylquinoline-3-carbonitrile (3k)



Yellow solid; mp 199–221 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 11.70 (bs, 1H), 9.0 (s, 1H), 8.46 (s, 1H), 8.30 (s, 1H), 7.83 (d, J = 7.6 Hz, 1H), 7.77 (d, J = 7.6, 1H), 7.52–7.41 (m, 2H), 6.88 (d, J = 7.6 Hz, 1H), 3.83 (s, 3H), 2.84 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 155.2, 152.2, 147.5, 145.9, 136.3, 133.4, 131.8, 129.1, 127.0, 126.7 (2C), 123.7, 119.9, 113.6, 113.4, 113.1, 104.2, 102.7, 55.6, 18.6; HPLC: 98.3%, column: X Bridge C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.1% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/50, 2/50, 9/98, 14/98, 16/50, 18/50; flow rate: 1.0 mL/min; UV 220 nm, retention time 8.42 min; IR (KBr) υ_{max} 3330, 3335, 2229, 1585, 1433 cm^{-1}; m/z (CI) 314 (M+1, 100%). Elemental analysis found C,

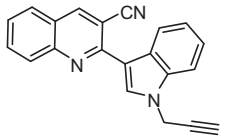
76.45; H, 4.80; N, 13.53 $C_{20}H_{15}N_3O$ requires C, 76.66; H, 4.82; N, 13.41.

4.1.14. 2-(6-Methoxy-1*H*-indol-3-yl)-6-methylquinoline-3-carbonitrile (3l)



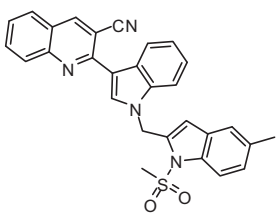
Yellow solid; mp 225–227 °C; 11.59 (bs, 1H), 8.96 (s, 1H), 8.56 (d, J = 7.6, 1H), 8.35 (s, 1H), 7.81 (d, J = 7.6, 1H), 7.74 (d, J = 7.6, 1H), 7.50–7.48 (m, 1H), 7.02 (s, 1H), 6.87 (d, J = 7.6, 1H) 3.80 (s, 3H), 2.79 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.8, 152.0, 147.5, 145.8, 137.8, 136.5, 133.4, 127.7, 126.6 (2C), 123.7, 123.4, 120.5, 119.8, 114.0, 111.3, 102.8, 95.3, 55.6, 18.5; HPLC: 99.3%, column: X Bridge C-18 150 × 4.6 mm 5 μ, mobile phase A: 0.1% Formic Acid in water mobile phase B: CH_3CN, gradient (T/%B): 0/50, 2/50, 9/98, 14/98, 16/50, 18/50; flow rate: 1.0 mL/min; UV 220 nm, retention time 8.41 min; IR (KBr) υ_{max} 3210, 2124, 1486, 1374 cm^{-1}; m/z (CI) 314 (M+1, 100%). Elemental analysis found C, 76.78; H, 4.78; N, 13.27 $C_{20}H_{15}N_3O$ requires C, 76.66; H, 4.82; N, 13.41.

4.1.15. 2-(1-(Prop-2-ynyl)-1*H*-indol-3-yl)quinoline-3-carbonitrile (4)



To a mixture of 2-(1*H*-indol-3-yl)quinoline-3-carbonitrile (**3a**) (2.95 mmol), propargyl bromide (4.42 mmol) and 50 mol% tetrabutylammonium bromide in 2.20 mL of toluene was added 2.20 mL of 50% NaOH drop wise at room temperature. The reaction mixture was then stirred at room temperature for 6 h. Upon completion of the reaction, the reaction mixture was diluted with water and extracted with ethylacetate (3 × 25 mL). The organic layers were collected, combined, dried over anhydrous Na_2SO_4, filtered and concentrated. The residue was purified by silica gel column chromatography to give the desired product as a brown solid; mp 180–182 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.07 (s, 1H), 8.65 (d, J = 7.6 Hz, 1H), 8.49 (s, 1H), 8.11 (d, J = 8.4 Hz, 1H), 8.03 (d, J = 8.0 Hz, 1H), 7.94 (t, J = 7.6 Hz, 1H), 7.64–7.68 (m, 2H), 7.28–7.37 (m, 2H), 5.28 (s, 2H), 2.58 (s, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 152.4, 148.3, 145.4, 136.1, 133.4, 130.7, 128.7, 128.5, 127.3, 126.8, 123.7, 123.2, 122.8, 121.7, 119.0, 113.0, 110.7, 103.4, 78.6, 76.5, 35.9; IR (KBr) υ_{max} 3258, 3049, 2223, 1478 cm^{-1}; m/z (CI) 308 (M+1, 100%). Elemental analysis found C, 82.00; H, 4.20; N, 13.63 $C_{21}H_{13}N_3$ requires C, 82.06; H, 4.26; N, 13.67.

4.1.16. 2-(1-((1-(Methylsulfonyl)-1*H*-indol-2-yl)methyl)-1*H*-indol-3-yl)quinoline-3-carbonitrile (6)



A mixture of **4** (0.80 mmol), 10% Pd/C (26 mg, 0.025 mmol), PPh_3 (25 mg, 0.10 mmol), CuI (9 mg, 0.045 mmol) and triethylamine (2.42 mmol) in ethanol (5 mL) was stirred at 25 °C for 1 h under nitrogen. The acetylenic compound **5** (1.62 mmol) was added slowly to the mixture with stirring. The reaction mixture was then stirred at 80 °C for the 6 h. The mixture was cooled to rt, diluted with EtOAc (60 mL) and filtered through celite. The filtrate was collected, washed with cold H_2O (2 × 30 mL), dried over Na_2SO_4, filtered and concentrated under vacuum. The residue thus obtained was purified by column chromatography afford the desired product; white solid; mp 205–207 °C; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.78 (m, 1H), 8.57 (s, 1H), 8.40 (s, 1H), 8.22 (d, J = 8.4 Hz, 1H), 7.87–7.82 (m, 3H), 7.58 (t, J = 8.1 Hz, 1H), 7.45–7.43 (m, 1H), 7.37–7.31 (m, 2H), 7.22 (s, 1H), 7.15 (d, J = 7.5 Hz, 1H), 6.25 (s, 1H), 5.77 (s, 2H), 2.95 (s, 3H), 2.39 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 152.4, 148.9, 144.2, 136.8, 135.3, 135.2, 133.6, 132.7, 131.0, 129.3, 129.0, 127.7, 127.2, 127.0, 126.6, 123.8, 123.6, 122.9, 122.0, 121.1, 119.0, 114.0, 113.4, 110.9, 110.2, 104.1, 45.0, 40.6, 21.1; IR (KBr) υ_{max} 2278, 1521, 1179 cm^{-1}; m/z (CI) 491 (M+1, 100%); Elemental analysis found C, 71.05; H, 4.60; N, 11.46 $C_{29}H_{22}N_4O_2S$ requires C, 71.00; H, 4.52; N, 11.42.

4.2. Single crystal X-ray data for compound 3a

Single crystals suitable for X-ray diffraction of **3a** were grown from ethyl acetate and dichloromethane. Single crystals were carefully chosen using a stereo zoom microscope supported by a rotatable polarizing stage. The data was collected at room temperature on Bruker's KAPPA APEX II CCD Duo with graphite monochromated $Mo_{K\alpha}$ radiation (0.71073 Å). The crystals were glued to a thin glass fibre using FOMBLIN immersion oil and mounted on the diffractometer. The intensity data were processed using Broker's suite of data processing programs (SAINT), and absorption corrections were applied using SADABS.[11a] The structure was solved by direct methods and all the non-hydrogen atoms were refined anisotropically while the hydrogen atoms, except hydrogens on N which were refined by picking electron density peaks, fixed in the predetermined positions by Shelxs-97[11b] and Shelxl-97 packages, respectively.

The hydrogen atoms bonded to carbons were positioned geometrically and refined in the riding model approximation with C–H = 0.95 Å, and with U(H) set to 1.2U_{eq}(C). Crystal data of **3a:** Molecular formula = $C_{18}H_{11}N_3$, Formula weight = 269.30, Orthorhombic, $P2_12_12_1$, a = 6.9840 (13) Å, b = 10.983(2) Å, c = 17.125(4) Å, V = 1313.6(5) Å^3, T = 298 K, Z = 4, D_c = 1.367 Mg m^{-3}, μ(Mo Kα) = 0.71073 mm^{-1}, 9989 reflections measured, 2274 independent reflections, 1928 observed reflections [$I > 2.0\sigma(I)$], R$_1$_obs = 0.035, Goodness of fit = 0.835. Crystallographic data (excluding structure factors) for **3a** have been deposited with the Cambridge Crystallographic Data Centre as supplementary publication numbers CCDC 828341.

4.3. Pharmacology

4.3.1. Materials and methods

Cells and reagents: HEK 293 and Sf9 cells were obtained from ATCC (Washington D.C., USA). HEK 293 cells were cultured in DMEM supplemented with 10% fetal bovine serum (Invitrogen Inc., San Diego, CA, USA). Sf9 cells were routinely maintained in Grace's supplemented medium (Invitrogen) with 10% FBS. cAMP was purchased from SISCO Research Laboratories (Mumbai, India). PDE light HTS cAMP phosphodiesterase assay kit was procured from Lonza (Basel, Switzerland).

4.3.2. Evaluation of PDE4 B inhibition by cell based cAMP reporter assay[14]

One day prior to transfection, HEK 293 cells were seeded in p60 cell culture dish (Tarsons Inc.) and were transfected using Lipofectamine 2000, as per the manufacturer's instructions with 1.2 μg of PDE4B expression plasmid and 4.0 μg of pCRELuc plasmid. After 5 h of transfection, medium was aspirated, cells were trypsinized and seeded in 96 well plates at a density of 60,000 cells/well. Plates were incubated overnight in a CO_2 incubator set to 37 °C and 5% CO_2. Twenty four hours post transfection, cells were pre-treated with different compound for 30 minutes, followed by stimulation with 5 μM forskolin for 4 h. Subsequently medium was removed and cells were lysed in reporter lysis buffer (Promega Inc.) for 15 min with gentle rocking at rt. Preliminary screening of the compounds was performed at 30 μM and dose response studies were carried out at eight different concentrations (0.01–60 μM). Luciferase activity in the lysates was measured by a Multilabel plate reader (Perklin Elmer 1420 Multilabel counter). Fold elevation of cAMP over forskolin control was calculated using the following formula and the EC_{50} values were determined using GraphPad Prism. Value obtained with forskolin control is set to one. Inhibition of PDE4B by rolipram was used as reference.

$$\text{Fold activation(FA)} = \frac{\text{Normalized value ofcompound treatment}}{\text{Normalized value of forskolin treatment}}$$

4.4. Docking studies

The docking studies were performed via the energy minimization and conformational search with the MACROMODEL application in the Schrodinger package. The molecules to be docked were energy minimized for flexibility of the molecules and then conformational search was followed. We used OPLS_2005 forcefield and water as implicit solvent. We have followed the PRCG (Polak-Ribier conjugate gradient) method of minimization with 500 iterations with a threshold gradient on 0.05 kJ/mol. The conformational search was based on Montecarlo multiple minimum torsional sampling. The ligands were then finally prepared with LIGPREP application.

The PDE4B protein (3D3P) crystal structures was retrieved from the protein data bank and it was refined with the PROTEIN PREPERATION WIZARD application in which the hydrogens were added and missing side chains and loops were filled with PRIME application. Water molecules were observed within the 5 Å distance and waters were deleted beyond 5 Å from het(hetroatom) groups. Finally, the protein was then optimized and minimized with impref using OPLS_2005 force filed. GRID based docking was performed in the present study.

Acknowledgments

K.S. thanks UGC, New Delhi, India for a Dr. D.S. Kothari Post doctoral fellowship [No. F.4-2/2006(BSR)/13-324/2010(BSR)]. M.P. and K.P. thank DBT, New Delhi, India for financial support (Grant No. BT/PR12829/Med/30/222/2009).

Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmc.2012.02.027.

References and notes

1. (a) Rigby, J. H.; Danca, D. M. *Tetrahedron Lett.* **1997**, *38*, 4969; (b) Leue, S.; Miao, W.; Kanazawa, A.; Génisson, Y.; Garc¸on, S.; Greene, A. E. *J. Chem. Soc. Perkin Trans. 1* **2001**, 2903; (c) Comins, D. L.; Nolan, J. M. *Org. Lett.* **2001**, *3*, 1611.
2. Toyota, M.; Komori, C.; Ihara, M. *Heterocycles* **2002**, *56*, 101.
3. (a) Samosorn, S.; Bremner, J. B.; Ball, A.; Lewis, K. *Bioorg. Med. Chem.* **2006**, *14*, 857; (b) Foley, M.; Tilley, L. *Pharmacol. Ther.* **1998**, *79*, 55.
4. (a) Myers, A. G.; Tom, N. J.; Fraley, M. E.; Cohen, S. B.; Madar, D. J. *J. Am. Chem. Soc.* **1997**, *119*, 6072; (b) Comins, D. L.; Hong, H.; Saha, J. K.; Jianhua, G. *J. Org Chem.* **1994**, *59*, 5120; (c) Shen, W.; Coburn, C. A.; Bornmann, W. G.; Danishefsky, S. J. *J. Org. Chem.* **1993**, *58*, 611.
5. Yearick, K.; Ekoue-Kovi, K.; Iwaniuk, D. P.; Natarajan, J. K.; Alumasa, J.; DeDios, A. C.; Roepe, P. D.; Wolf, C. *J. Med. Chem.* **1995**, *2008*, 51.
6. For a review, see: (a) de Sa' Alves, F. R.; Barreiro, E. J.; Fraga, C. A. M. *Mini. Rev. Med. Chem.* **2009**, *9*, 782; (b) Hoemam, M. Z.; Kumaravel, G.; Xie, R. L.; Rossi, R. F.; Meyer, S.; Sidhu, A.; Cunny, G. D.; Hauske, J. R. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 2675.
7. (a) Kodimuthali, A.; Jabaris, S. S. L.; Pal, M. *J. Med. Chem.* **2008**, *51*, 5471; (b) Reddy, G. R.; Reddy, T. R.; Joseph, S. C.; Reddy, K. S.; Reddy, L. S.; Kumar, P. M.; Krishna, G. R.; Reddy, C. M.; Rambabu, D.; Kapavarapu, R.; Lakshmi, C.; Meda, T.; Priya, K. K.; Parsa, K. V. L.; Pal, M. *Chem. Commun.* **2011**, *47*, 7779; (c) Kodimuthali, A.; Gupta, R.; Parsa, K. V. L.; Prasunamba, P. L.; Pal, M. *Lett. Drug Des. Disc.* **2010**, *7*, 402; (d) Kumar, P. M.; Kumar, K. S.; Mohakhud, P. K.; Mukkanti, K.; Kapavarapu, R.; Parsa, K. V. L.; Pal, M. *Chem. Commun.* **2012**, *48*, 431; (e) Pal, S.; Durgadas, S.; Nallapati, S. B.; Mukkanti, K.; Kapavarapu, R.; Lakshmi, C.; Parsa, K. V. L.; Pal, M. *Bioorg. Med. Chem. Lett.* **2011**, *21*, 6573; (f) Hulme, C.; Moriarty, K.; Miller, B.; Mathew, R.; Ramanjulu, M.; Cox, P.; Souness, J.; Page, K. M.; Uhl, J.; Travis, J.; Huang, F.-C.; Labaudiniere, R.; Djuric, S. W. *Bioorg. Med. Chem. Lett.* **1867**, *1998*, 8; (g) Buckley, G. M.; Cooper, N.; Dyke, H. J.; Galleway, F.; Gowers, L.; Haughan, A. F.; Kendall, H. J.; Lowe, C.; Maxey, R.; Montana, J. G.; Naylor, R.; Oxford, J.; Peake, J. C.; Picken, C. L.; Runcie, K. A.; Sabin, V.; Sharpe, A.; Warneck, J. B. H. *Bioorg. Med. Chem. Lett.* **2002**, *12*, 1613; (h) Billah, M.; Buckley, G. M.; Cooper, N.; Dyke, H. J.; Egan, R.; Ganguly, A.; Gowers, L.; Haughan, A. F.; Kendall, H. J.; Lowe, C.; Minnicozzi, M.; Montana, J. G.; Oxford, J.; Peake, J. C.; Picken, C. L.; Piwinski, J. J.; Naylor, R.; Sabin, V.; Shih, N.-Y.; Warneck, J. B. H. *Bioorg. Med. Chem. Lett.* **2002**, *12*, 1617; (i) Akama, T.; Antunes, J.; Freund, Y.; Kimura, R.; Dong, C.; Sanders, V.; Zhang, Y-K.; Graves, G.; Lu, X.; Sharma, R.; Sales, M.; Nieman, J.; Singh, R.; Ding, C.; Plattner, J. Structure–Activity Studies of Novel Oxaborole Dual Inhibitors of PDE4 and IL-23 Release. For a poster, see: http://www.anacor.com/pdf/Anacor-Akama0509.pdf.
8. (a) Tobisu, M.; Hyodo, I.; Chatani, N. *J. Am. Chem. Soc.* **2009**, *131*, 12070; (b) Fleckenstein, C. A.; Plenio, H. *Chem. Eur. J.* **2008**, *14*, 4267.
9. (a) Pal, M.; Batchu, V. R.; Parasuraman, K.; Yeleswarapu, K. R. *J. Org. Chem.* **2003**, *68*, 6806; (b) Pal, M.; Batchu, V. R.; Dager, I.; Swamy, N. K.; Padakanti, S. *J. Org. Chem.* **2005**, *70*, 2376; (c) Kodimuthali, A.; Nishad, T. C.; Prasunamba, P. L.; Pal, M. *Tetrahedron Lett.* **2009**, *50*, 354; (d) Kodimuthali, A.; Chary, B. C.; Prasunamba, P. L.; Pal, M. *Tetrahedron Lett.* **2009**, *50*, 1618; (e) Kumar, K. S.; Chamakuri, S.; Vishweshwar, P.; Iqbal, J.; Pal, M. *Tetrahedron Lett.* **2010**, *51*, 3269.
10. Upadhyay, S.; Chandra, A.; Singh, R. M. *Indian J. Chem.* **2009**, *48B*, 152.
11. (a) Bruker SADABS V2008–1, Bruker AXS.: Madison, WI, USA (2008).; (b) Sheldrick, G. M. SHELX93, Program for Crystal Structure Determination, University of Göttingen (1997).
12. Damodiran, M.; Muralidharan, D.; Perumal, P. T. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 3611.
13. Pal, M.; Subramanian, V.; Batchu, V. R.; Dager, I. *Synlett* **2004**, 1965.
14. Wang, P.; Myers, J. G.; Wu, P.; Cheewatrakoolpong, B.; Egan, R. W.; Billah, M. M. *Biochem. Biophys. Res. Commun.* **1997**, *234*, 320.
15. Cheung, W. Y. *Biochemistry* **1967**, *6*, 1079.
16. Dastidar, S. G.; Rajagopal, D.; Ray, A. *Curr. Opin. Invest. Drugs* **2007**, *85*, 364.
17. Jin, S.-L. C.; Lan, L.; Zoudilova, M.; Conti, M. *J. Immun.* **2005**, *175*, 1523.
18. Robichaud, A.; Stamatiou, P. B.; Jin, S.-L. C.; Lachance, N.; MacDonald, D.; Laliberte, F.; Liu, S.; Huang, Z.; Conti, M.; Chan, C.-C. *J. Clin. Invest.* **2002**, *110*, 1045.
19. For in depth computational studies on PDE4 inhibitors, see: (a) Chakraborti, A. K.; Gopalakrishnan, B.; Sobhia, M. E.; Malde, A. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 2473; (b) Chakraborti, A. K.; Gopalakrishnan, B.; Sobhia, M. E.; Malde, A. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 1403.