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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis of pyrazole-based 1,5-diaryl compounds as potent anti-inflammatory agents

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Abstract Series of 1,5-diaryl pyrazole ester derivatives have been synthesized and found to contain potent inhibitory activity against cyclooxygenase-2 (COX-2) enzyme. The article describes synthesis of the target pyrazole analogs and biological assay using Carrageenan induced rat paw for investigation.

Keywords 1,3-diarylpyrazole ester · COX-2 · Carrageenan induced rat paw edema

Introduction

Various diaryl and triaryl pyrazole and imidazole ring systems have continuously been used for the treatment of pain and inflammation associated with various musculoskeletal disorders, attracted our interest due to their potent biological activities (Tewari and Mishra, 2001; Tewari *et al.,* 2009, 2010a). The mechanism of NSAIDs (non steroidal anti-inflammatory drugs) in reducing inflammatory reactions involves the inhibition of COX enzymes, COX are cyclooxygenase enzymes and catalyses second step of prostaglandin synthesis. The biological studies demonstrated increasing in COX activity in a variety of cells after exposure to endotoxin, pro-inflammatory cytokines, growth factors, hormones, and tumor promoters. This activity required new protein synthesis and was inhibitable by corticosteroids, giving rise to the concept that there might be "consecutive COX activity," further referred to as COX-1, and an "inducible" one, further referred to as COX-2. These two COX isoforms are encoded by different genes, and COX-1 was hypothesized to be "house keeping gene," while COX-2 (Dannhardt and Kiefer, 2001; Almansa *et al.*, 2003) was thought to be involved in inflammation, mitogenesis, and/or specialized signal transduction.

Most NSAIDs act as non-selective inhibitors of cyclooxygenase (COX), by inhibiting the metabolism of arachidonic acid (Insel, 1996) Majority of NSAIDs act non selectively to COX (Dannhardt and Laufer, 2000; Carter, 2000; Talley, 1999; Hla and Neilson, 1992). COX-1 and COX-2, are widely used to treat the signs and symptoms of inflammation, particularly arthritic pain. Anti-inflammatory and analgesic properties of anti pyrene and other pyrazole (Tewari and Mishra, 2001) derivatives have found their clinical application as NSAIDs. Among the highly marketed COX-2 selective NSAID that comprise of the pyrazole nucleus, celecoxib represents a potent anti-inflammatory and analgesic compound. It is considered as a typical model of the diaryl heterocycle template that is known to selectively inhibit the COX-2 enzyme. Some other examples of pyrazole derivatives as NSAIDs are felcobutazone, mefobutazone, morazone, famprofazone, and ramifenazone (Reynold, 1993; Amir and Kumar, 2005; Gursoy *et al.*, 2000; Kumar *et al.*, 2003).

The role of aromatic interactions becomes prominent in drug receptor interactions (Meyer *et al.*, 2003). The importance of trimethylene bridge as synthetic spacer for the detection of intramolecular interactions has been well documented (Leonard, 1979). These points revealed us to

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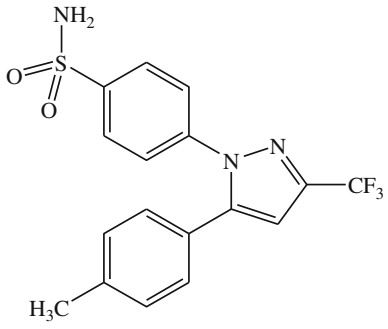
synthesize some pyrazole derivatives using trimethylene spaces; 1,5-diaryl systems.



Celecoxib

Chemistry

All compounds were synthesized by a general experimental procedure involving substitution reactions at room temperature. 3-Methyl-1-phenyl-pyrazol-5-one **1** had been synthesized from the reaction of ethyl acetoacetate and phenyl hydrazine. The reaction of **1** with carbon disulfide followed by methylation gave 4-(bis-methylsulfanyl-methylene)-5-methyl-2-phenyl-2,4-dihydro-pyrazol-3-one (**2a**) and by ethylation gave 4-(bis-ethylsulfanyl-methylene)-5-methyl-2-phenyl-2, 4-dihydro-pyrazol-3-one (**2b**) (Tewari *et al.*, 2009). **2a** and **2b** are the key intermediate in the synthesis of 4-substituted pyrazoles. The base catalyzed alcoholysis of **2a** and **2b** followed by acidification gave pyrazole-ester **3a** and **3b**, respectively, in very good yield. The mass spectrum and elemental analysis are in accordance with the assigned structure. The ^{1}H-NMR spectrum of **3a** had a singlet at δ 2.41 for methyl protons and a singlet at δ 3.92 for methoxy protons and a broad singlet at δ 9.99 for hydroxyl proton. The mass spectrum of **3a** had the base peak at MS (*m*/*z*) M^{+} 233. Similarly, the ^{1}H-NMR and mass spectra of **3b** were found in accordance to the assigned structures. Treatment of **3a** with excess of 1,3-dibromopropane (5 eq.) (Avasthi *et al.*, 1995) in mild basic conditions (anhydrous K_2CO_3) using *N*,*N*-dimethylmethanamide (DMF) as solvent afforded the synthesis of **5a** with quantitative yield and purified by column chromatography. Similarly, **5b** was obtained by treatment of **3b** with excess of 1,3-dibromopropane in basic medium. All products were obtained in quantitative yield with high purity via 100–200 mesh SiO_2, column chromatography with ethyl acetate in chloroform as eluent mixture. Reaction of **3a** and **3b** with 1,3-dibromopropane (0.5 eq) in presence of K_2CO_3 and DMF afforded synthesis of **4a** and **4b**, respectively (Scheme 1).

The base catalyzed nucleophilic substitution of **5a** with 1 eq. of phthalimide in DMF as solvent at room temperature gave **6a** (Tewari *et al.*, 2010b). The pure product was obtained by crystallization with 2% ethyl acetate in hexane. Similarly, **5b** with phthalimide afforded the synthesis of **6b**. The base catalyzed nucleophilic substitution of **5a** and **5b** separately with 1 eq. of benzimidazole and DMF at r.t. gave the synthesis of **7a** and **7b**, respectively (Scheme 2).

The base catalyzed nucleophilic substitution of **5a** and **5b** separately with 1 eq. of pyridone and DMF at r.t. afforded the synthesis of **8a-I**, **8a-II**, **8b-I,** and **8b-II**, respectively (Scheme 3). In the reaction, two products were resulted (N-substituted and O-substituted). Both the products were separated through column chromatography and characterized by NMR spectroscopy. The compound **8a-I** and **8a-II** is confirmed by ^{1}H-NMR and ^{13}C-NMR spectra. In ^{1}H-NMR spectra of **8a-I**, two triplet peaks of methylene group were observed at δ 4.05–4.09 and δ 4.28–4.32 due to the presence of N–CH_2 and O–CH_2, respectively, while in **8a-II**, only one triplet peak of methylene groups was observed at δ 4.36–4.39 due to the same environment (i.e., presence of two O–CH_2 groups). In ^{13}C-NMR spectra of **8a-I,** N–CH_2 group arises at δ 42.4 whereas in **8a-II**, O–CH_2 was observed at δ 63.0 ppm. Similarly, N and O isomers in **8b-I** and **8b-II** was observed by ^{1}H-NMR and ^{13}C-NMR spectroscopy.

Biological activity

Materials and methods

Animals

All experiments have been conducted on adult Wistar strain albino rats of either sex, weighing between 150 and 200 g. The animals were obtained from the Central Animal House of the Institute of Medical Sciences, B.H.U. They were kept in colony cages under identical housing conditions at an ambient temperature of 25 ± 2°C and 45–55% relative humidity with a 12 h light–dark cycle in the departmental animal room and fed on standard diet. Animals were acclimatized for a week before use.

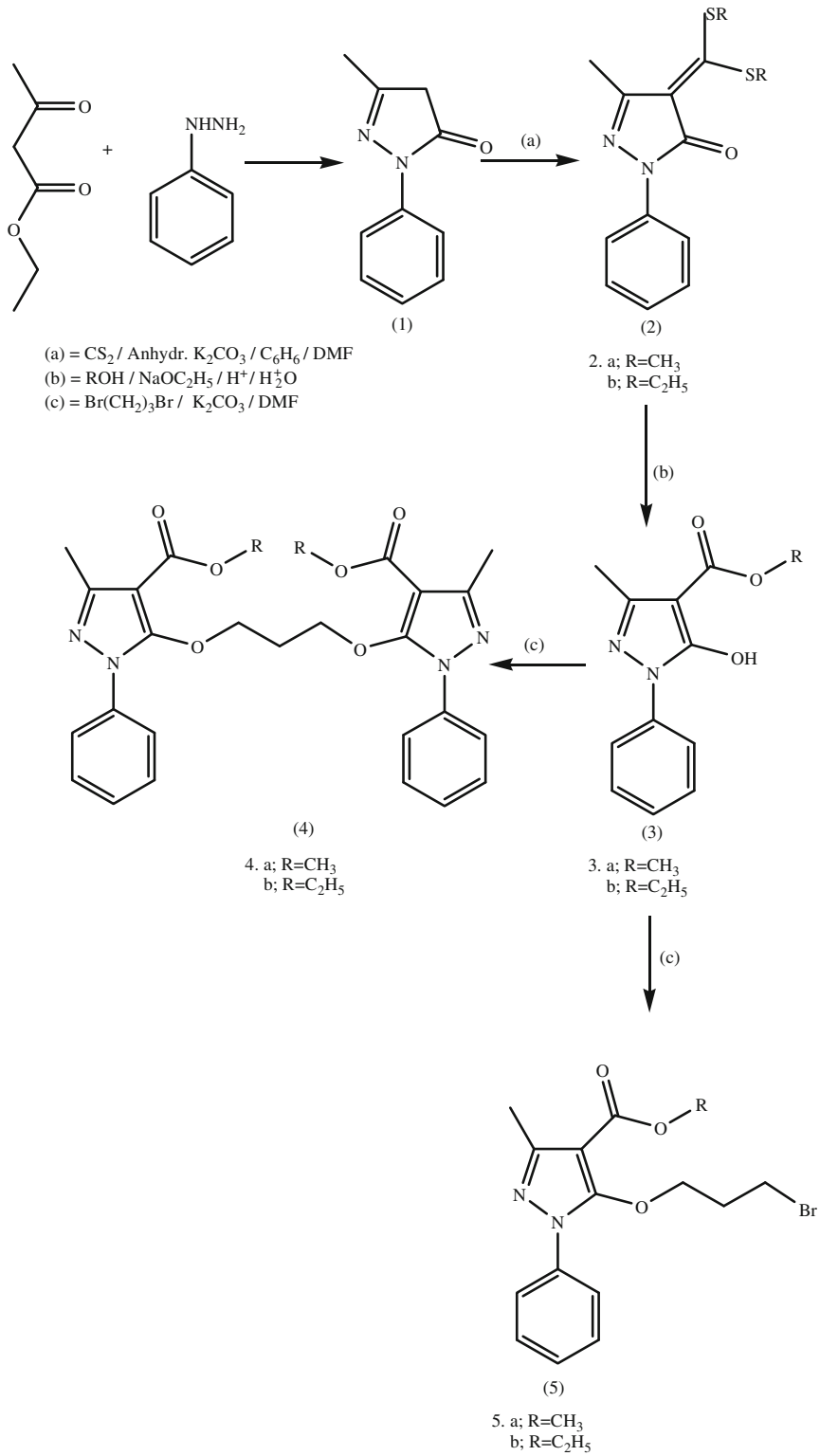
Experimental model of inflammation

Carrageenan induced paw edema (Wintar *et al.*, 1962) was used throughout the investigation.

Standard drugs and solutions

(a) Powder of the pure Carrageenan was used and fresh suspension was prepared in distilled water to make 1% Carrageenan solution; (b) Fresh drug solutions were made by adding 10 mg of drug in 500 mg carboxymethyl cellulose (CMC) and 50 ml distilled water.

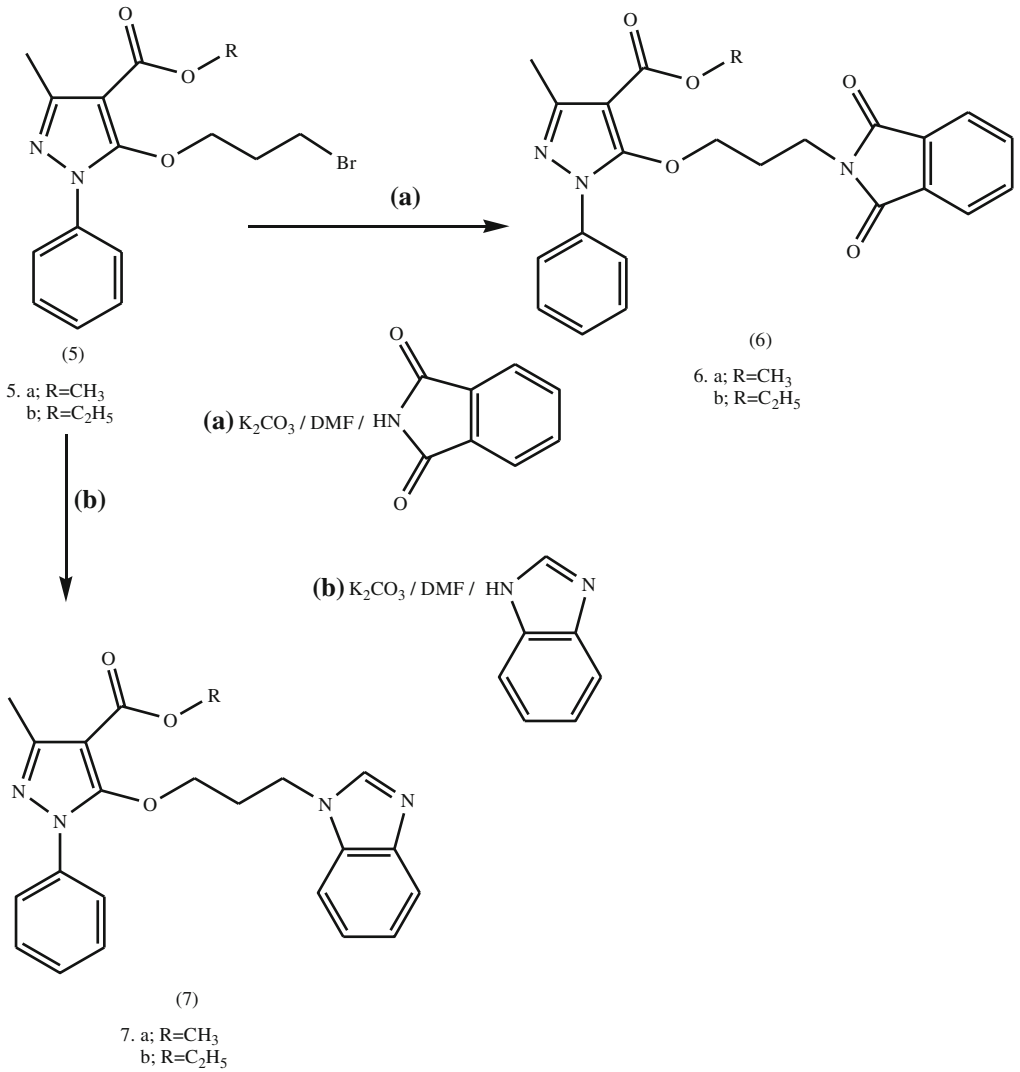
Scheme 1



Experimental inflammation

It was produced by the following method:

(a) Carrageenan induced paw edema in rats: 1% Carrageenan suspension was prepared as a homogeneous solution in distilled water. A volume of 0.1 ml of Carrageenan solution was injected through a 26 gauge needle into the plantar surface of the left hind paw below the plantar aponeurosis. The volume of paw was measured before and at different intervals for 3 h after injection of



Scheme 2

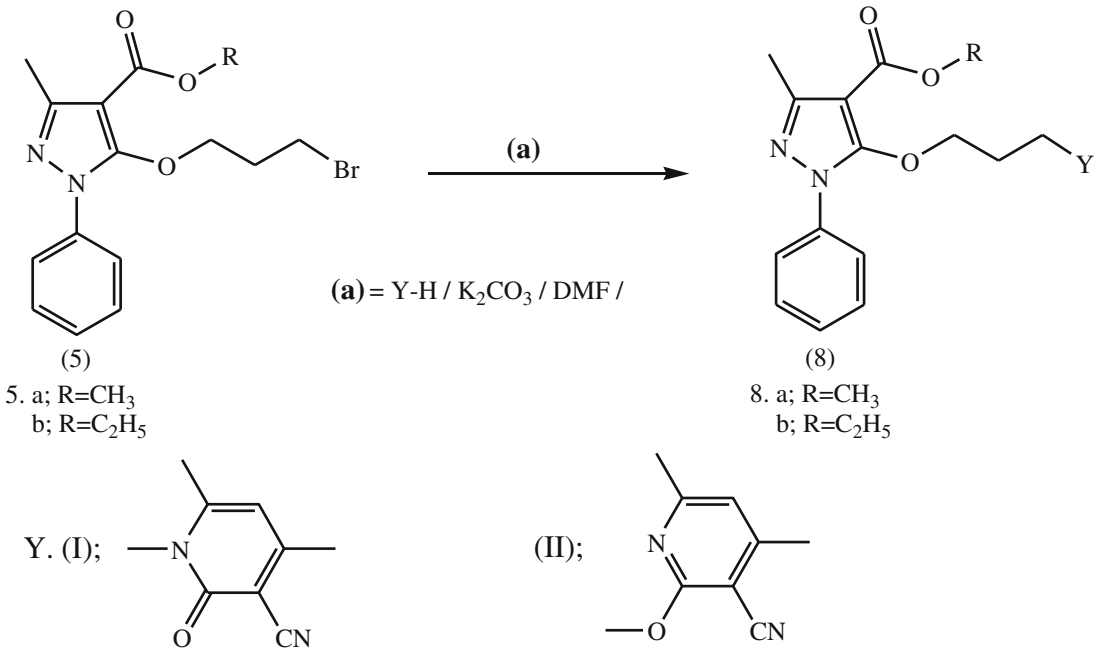
carrageenan. The difference in paw volume before and after administration of the phlogistic agent was taken as the measure of pedal edema. The drugs whose effects have been studied on this particular model were administered as per schedule; (b) Measurement of paw volume: The volume of hind paw of the rats up to the ankle joint was measured plathysmographically by the mercury displacement method. The ankle joint of the rats was marked with a skin marking pencil and the paw was dipped in the mercury, so that the mark on the paw coincides with a prefixed line kept constant on the syringe. The level of the mercury was every time brought to the level of this line by adjusting the height of the displaced mercury. The difference in the paw volume before and after injection of the phlogistic agents was taken as a measure of pedal edema. The change in paw volume was expressed in "ml" of mercury displaced.

Results and observations

In all experiments, Carrageenan was administered into the left hind paw and the paw volume was measured before and at intervals of 30, 90, and 180 min, after Carrageenan injection, as shown in Tables 1 and 2. However, when the rats were reused, Carrageenan injection was given into the right hind paw.

Discussion and conclusion

Carrageenan which is a sulfated polysaccharide, extracted from sea weed has been extensively used to induce inflammation in a number of animal species. The Carrageenan induced rat hind paw edema is now routinely used for the assay of anti-inflammatory agents (Wintar *et al.*, 1962). The reproducibility and the fact that the edema



Scheme 3

Table 1 Percentage edema growth relative to control at different time intervals (Mean ± SEM)

Group	0 min	30 min	90 min	180 min
Control	0.99 ± 0.067	1.27 ± 0.043**	1.36 ± 0.070	1.21 ± 0.072
Nimusilide	1.18 ± 0.064	1.29 ± 0.041	1.25 ± 0.024	1.21 ± 0.021
4a	1.15 ± 0.044	1.34 ± 0.029	1.49 ± 0.025	1.42 ± 0.046
6a	1.17 ± 0.064*	1.25 ± 0.061	1.39 ± 0.063	1.22 ± 0.093
6b	0.98 ± 0.033*	1.11 ± 0.052**	1.18 ± 0.034**	1.01 ± 0.052*
8a-I	0.84 ± 0.056	0.94 ± 0.044***	1.03 ± 0.057**	0.99 ± 0.0.043*
8b-I	0.78 ± 0.050	1.05 ± 0.028	1.16 ± 0.012*	1.04 ± 0.045
8a-II	0.89 ± 0.038	0.81 ± 0.028***	0.92 ± 0.025***	0.99 ± 0.028*
8b-II	0.91 ± 0.086	1.22 ± 0.034	1.18 ± 0.050	1.15 ± 0.022

Results are (Mean ± SEM) of 6 rats in each group. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$

depends entirely on a local inflammatory reaction devoid of antigenic properties, has made Carrageenan most widely employed phlogistic agent in experimental pharmacology (Di Rosa *et al.*, 1971). A good correlation has been shown to exist between the antiphlogistic and anti-inflammatory effects of several drugs (Lombardino *et al.*, 1975).

The mediator involved in Carrageenan induced inflammation has been extensively investigated and histamine, serotonin, kinins, and PGs have been implicated (Garcia-Leme, 1978). It has been proposed that histamine and serotonin are responsible for the initial stages of the edema, whereas kinins and PGs are responsible for later stages of the inflammation (Di Rosa *et al.*, 1971; Holsapple *et al.*, 1980). In the present study, sub cutaneous injection of 0.1 ml of 1% Carrageenan into the rat paw produces plasma extravasations and inflammation characterized by increased tissue water and plasma protein exudation with neutrophil extravasations and metabolism of the arachidonic acid by both cyclooxygenase and lipooxygenase enzyme pathway. There are biphasic effects in Carrageenan induced edema. The first phase begins immediately after injection and diminishes in 1 h. The second phase begins at 1 h and remains through 3 h. It is suggested by Kulkarni *et al.* (1986) that the early hyperemia of Carrageenan induced edema results from the release of histamine and serotonin. On the other hand, delayed phase of Carrageenan induced edema results mainly from the potentiating effect of PGs on mediator release, especially of bradykinin. Hydrocortisone and some anti-inflammatory drugs strongly inhibit the second phase of Carrageenan induced edema. However, some anti-inflammatory drugs are effective against both phases

Table 2 Paw edema at different time intervals (ml/rat) (Mean ± SEM)

Group	0 (min)	30 (min)	90 (min)	180 (min)
Control	100 ± 0	130.1 ± 6.54 (30.1)	138.7 ± 4.47 (38.7)	122.3 ± 5.17 (22.3)
Nimusilide	100 ± 0	110.8 ± 2.58 (10.8)	107.90 ± 3.14 (7.9)	102.54 ± 2.52 (2.54)
4a	100 ± 0	115.1 ± 2.88 (15.1)	124.4 ± 3.37* (24.4)	117.9 ± 4.25 (17.9)
6a	100 ± 0	105.9 ± 2.59** (5.9)	104.8 ± 3.98*** (4.8)	98.3 ± 2.54** (1.7)
6b	100 ± 0	138.9 ± 3.56 (38.9)	144.7 ± 6.54 (44.7)	139.3 ± 7.58 (39.3)
8a-I	100 ± 0	117.6 ± 10.02 (17.6)	129.7 ± 3.83* (29.7)	118.5 ± 8.34 (18.5)
8b-I	100 ± 0	122.4 ± 5.60 (22.4)	128.5 ± 4.86 (28.5)	120.4 ± 5.86 (20.4)
8a-II	100 ± 0	116.8 ± 10.02 (16.8)	128.7 ± 3.83* (28.7)	124.9 ± 8.34 (24.9)
8b-II	100 ± 0	121.4 ± 4.53 (21.4)	124.6 ± 3.98** (24.6)	116.8 ± 4.75 (16.8)

$N = 6$ = number of rats in each group. Results in parentheses indicate percentage change from respective control group.
*$p < 0.05$; **$p < 0.01$; ***$p < 0.001$

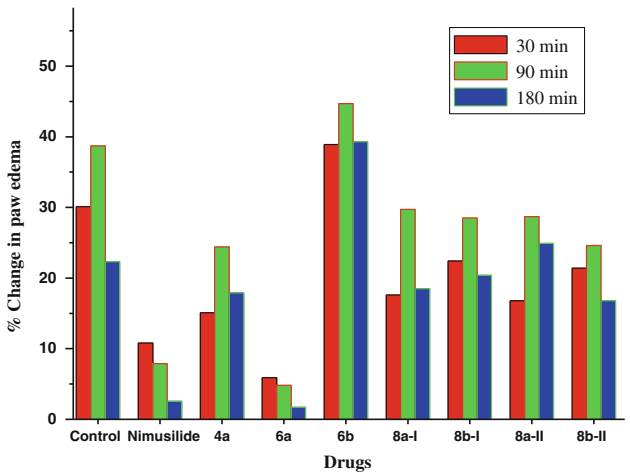


Fig. 1 Effect of different drugs on carrageenan induced rat paw edema

(Kulkarni *et al.*, 1986; Vinegar *et al.*, 1969). According to Insel (1996), steroids and NSAIDs exert their effect by inhibition of inflammation mediator formation.

The results of present study shown that compound **6a** had shown to possess maximum inhibitory effect when compared with control group. It was observed that maximum percentage of paw edema growth in control group at 90 min was 38.7% which was found to decrease up to 4.8% in the group of rats treated with **6a**. It shows very good anti-inflammatory property than Nimusilide (where it was 7.90% at 90 min). **8b-II** have also been found to possess good anti-inflammatory property as the percentage paw edema growth was shown to be only 24.0% when compared with that of control group (where it was 38.7% at 90 min).

Other drugs i.e., **4a**, **8a-I, 8b-I,** and **8a-II** have shown moderate to intermediate effects on inhibitory properties at 90 min, but **6b** has shown no effect as anti-inflammatory agent (Fig. 1).

Therefore, the results indicated that **6a** is potent inhibitor of inflammation, and the anti-inflammatory effect of **6a** on inflammagen induced edema may depend on inhibition of the formation of several inflammation mediators. The structures of **6a** (drug with maximum anti-inflammatory activity) and **6b** (drug with no anti-inflammatory activity) were almost same except that in **6a** ester moiety contained methyl group and in **6b** ester moiety contained ethyl group which being bulkier than methyl group may affect the activity of the particular drug. In conclusion, detailed studies are needed to clarify the mechanism(s) of anti-inflammatory effects of new pyrazole derivatives.

Experimental

All reactions were monitored by thin layer chromatography over silica gel G TLC plates. The melting points were recorded on electrically heated instrument and are uncorrected. NMR spectra were recorded on JEOL AL300 FTNMR spectrometer using TMS as internal reference and chemical shift values are expressed in δ, ppm units. Mass spectra of the compounds were taken with JEOL SX 102/Da-600 mass spectrometer. Analysis was performed on Exeter Analytical Inc. "Model CE-440 CHN Analyzer" instrument.

Preparation of 3-methyl-1-phenyl-pyrazol-5-one (**1**)

In a 250-ml round-bottom flask, ethyl acetoacetate (48 ml, 0.385 mol) was taken and phenyl hydrazine (41.54 ml, 0.385 mol) was added slowly with stirring. The reaction mixture was refluxed on a heating mantle for 2 h. The completion of reaction was checked via TLC. The reaction mixture was cooled and diethyl ether was slowly added with stirring. After 10 min, light yellow colored precipitate appeared which was filtered and washed with ether. Pure white colored 3-methyl-1-phenyl-pyrazol-5-one (**1**) was then recrystallized with hot aqueous ethanol.

Mp = 127°C. Yield = 60.20 g (90%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.20 (s, 3H, CH_3), δ 3.44 (s, 2H, CH_2), δ 7.18–7.87 (m, 5H, Ar–H).

Preparation of 4-(bis-methylsulfanyl-methylene)-5-methyl-2-phenyl-2,4-dihydro-pyrazol-3-one (**2a**)

In a 500-ml round-bottom flask, fitted with guard tube, anhydrous K_2CO_3 (59.48 g, 0.431 mol) was taken, and 20 ml DMF and 40 ml dry benzene were added and stirred in an ice bath for 1 h. In another 500-ml round-bottom flask, fitted with guard tube (**1**) (30.0 g, 0.172 mol) was taken and 20 ml DMF was added. It was stirred for 15 min. Then, carbon disulfide (10.40 ml, 0.172 mol) was added slowly with stirring. It was also stirred for 1 h in an ice bath. Then, the content of first round bottom flask was added to second round bottom flask with stirring and the reaction mixture was stirred for 6 h. With the use of a dropping funnel, methyl iodide (21.46 ml, 0.345 mol) was added very slowly along with 10 ml dry benzene. The addition of methyl iodide was accompanied with the use of ice bath. After complete addition, the reaction was stirred for 6 h. The completion of reaction was checked by TLC. Reaction was worked up. Solvents were removed under pressure through rotary evaporator and the reaction mixture was extracted with $CHCl_3/H_2O$ (200/200 × 2 ml). The $CHCl_3$ layer was dried with anhydrous Na_2SO_4 and filtered. Chloroform was removed and the product was purified via SiO_2-column chromatography. Eluent used was 20% ethyl acetate in hexane.

Mp = 46–52°C. Yield = 40.54 g (85%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.52 (s, 3H, CH_3), δ 2.69 (s, 3H, SCH_3), δ 2.76 (s, 3H, SCH_3), δ 7.12–7.99 (m, 5H, Ar–H).

Preparation of 4-(bis-ethylsulfanyl-methylene)-5-methyl-2-phenyl-2, 4-dihydro-pyrazol-3-one (**2b**)

In a 500-ml round-bottom flask, fitted with guard tube, anhydrous K_2CO_3 (59.48 g, 0.431 mol) was taken, and 20 ml DMF and 40 ml dry benzene were added and stirred in an ice bath for 1 h. In another 500-ml round-bottom flask, fitted with guard tube (**1**) (30.0 g, 0.172 mol) was taken and 20 ml DMF was added. It was stirred for 15 min. Then, carbon disulfide (10.40 ml, 0.172 mol) was added slowly with stirring. It was also stirred for 1 h in an ice bath. Then, the content of first round bottom flask was added to second round bottom flask with stirring and the reaction mixture was stirred for 6 h. With the use of a dropping funnel, ethyl iodide (27.67 ml, 0.345 mol) was added very slowly along with 10 ml dry benzene in ice cold condition. After complete addition, the reaction was stirred for 6 h. The completion of reaction was checked by TLC. Reaction was worked up. Solvents were removed under pressure through rotary evaporator and the reaction mixture was extracted with $CHCl_3/H_2O$ (200/200 × 2 ml). The $CHCl_3$ layer was dried with anhydrous Na_2SO_4 and filtered. Chloroform was removed and the product was purified via SiO_2-column chromatography. Eluent used was 20% ethyl acetate in hexane.

Mp = 40–44°C. Yield = 38.85 g (74%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.52 (s, 3H, CH_3), δ 1.31–1.44 (double-t, 6H, $SCH_2C\underline{H}_3$ × 2) (J = 7.5, 7.2, 8.1, 7.5, 7.5 Hz), δ 3.18-3.34 (double-q, 4H, $C\underline{H}_2CH_3$ × 2) (J = 7.5, 7.5, 7.2, 7.5, 7.2, 7.2 Hz), δ 7.12–7.99 (m, 5H, Ar–H).

Preparation of 5-hydroxy-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester (**3a**)

In a 100-ml round-bottom flask, anhydrous $NaOC_2H_5$ (0.47 g, 0.007 mol) was dissolved in 10 ml dry methanol and stirred for 15 min, and then compound (**2a**) (1 g, 0.004 mol) was added and refluxed over an oil bath fitted with water condenser and guard tube at 80°C for 2 h. The completion of reaction was confirmed by TLC. The reaction mixture was worked up. Concentrated HCl (excess till the orange color of reaction mixture turned to light yellow color and the medium becomes acidic) was added slowly drop wise to the reaction mixture, and then excess of water was added with constant shaking. Light orange-yellow precipitate appeared which was filtered and washed with water over buchner-funnel.

Mp = 138–145°C. Yield = 0.8 g (96%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.41 (s, 3H, CH_3), δ 3.92 (s, 3H, OCH_3), δ 7.26–7.79 (m, 5H, Ar–H), δ 9.99 (broad s, 1H, O–H). MS (*m/z*): 233(M^+). Element analysis: % C (calc.) 62.06 (found) 62.53; % N (calc.) 12.06 (found) 12.67; % H (calc.) 5.17 (found) 5.42.

Preparation of 5-hydroxy-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid ethyl ester (**3b**)

The process was exactly same as used for synthesis of compound (**3a**) but the solvent used was dry ethanol for compound (**3b**).

Mp = 95–100°C. Yield = 0.74 g (84%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.38–1.42 (t, 3H, $CH_2C\underline{H}_3$) (J = 5.7, 6.9 Hz), δ 2.41 (s, 3H, CH_3), δ 4.35–4.42 (q, 2H, $C\underline{H}_2CH_3$) (J = 6.9, 5.7, 7.2 Hz), δ 7.27–7.79 (m, 5H, Ar–H), δ 10.08 (broad s, 1H, O–H). MS (*m/z*): 247(M^+). Element analysis: % C (calc.) 63.41 (found) 63.90; % N (calc.) 12.06 (found) 12.50; % H (calc.) 5.69 (found) 5.57.

Preparation of 1,3-di(5-butoxy-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester)-propane (**4a**)

In a 100-ml round-bottom flask, fitted with guard tube, compound (**3a**) (5 g, 0.021 mol) was dissolved in 20 ml DMF, and anhydrous K_2CO_3 (2.97 g, 0.021 mol) was added and stirred at room temperature for 30 min. 1,3-dibromopropane (1.09 ml, 0.011 mol) was added and stirred at room temperature for 24 h. The completion of reaction was checked by TLC and the reaction mixture was worked up as above. Pure (**4a**) was obtained with 10% ethyl acetate in $CHCl_3$. Pure X-ray quality crystals were obtained with 2% ethyl acetate in hexane.

Mp = 58–60°C. Yield = 0.56 g (11%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.94–2.02 (p, 2H, CH_2) (J = 6.3, 6.0, 6.0, 5.7 Hz), δ 2.456 (s, 6H, $CH_3 \times 2$), δ 3.81 (s, 6H, $OCH_3 \times 2$); δ 4.15–4.19 (t, 4H, $OCH_2 \times 2$) (J = 6.0, 6.0 Hz), δ 7.26–7.52 (m, 10H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 15.3, 30.09, 51.1, 72.5, 99.2, 123.3, 127.5, 137.3, 150.9, 155.1, 163.3. MS (*m/z*): 505(M^+). Element analysis: % C (calc.) 64.28 (found) 64.32; % N (calc.) 11.11 (found) 11.18; % H (calc.) 5.55 (found) 5.62.

Preparation of 1,3-di(5-butoxy-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid ethyl ester)-propane (**4b**)

In a 100-ml round-bottom flask, fitted with guard tube, compound (**3b**) (0.46 g, 0.002 mol) was dissolved in 10 ml DMF and anhydrous K_2CO_3 (0.26 g, 0.002 mol) was added. It was stirred at room temperature for 1 h and (**5b**) (0.7 g, 0.002 mol) was added. The reaction mixture was stirred at room temperature for 24 h. Completion of reaction was confirmed via TLC. Reaction mixture was worked up. DMF removed under pressure by rotary evaporator and crude product obtained through extraction with $CHCl_3$/H_2O (20/20 × 2 ml). Organic layer was dried (Na_2SO_4), filtered, removed, and pure (**4b**) was obtained by column chromatography loaded with SiO_2 in chloroform. Eluent used was 15% ethyl acetate in $CHCl_3$. The product obtained was thick yellow oily compound.

Yield = 0.5 g (49%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.32–1.37 (t, 3H, $CH_2C\underline{H}_3$) (J = 6.9, 7.2 Hz), δ 1.96–2.00 (t, 2H, CH_2) (J = 6.0, 6.3 Hz), δ 2.46 (t, 6H, $CH_3 \times 2$), δ 4.15–4.19 (t, 4H, $OCH_2 \times 2$) (J = 6.0, 6.3 Hz), δ 4.25–4.32 (q, 4H, $OC\underline{H}_2CH_3 \times 2$) ($J$ = 7.2, 6.9, 7.2 Hz), δ 7.26–7.51 (m, 10H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 14.2, 15.2, 29.9, 59.7, 72.3, 99.2, 123.1, 127.3, 128.7, 137.2, 150.7, 154.8, 162.7.

Preparation of 5-(3-bromo-propoxy)-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester (**5a**)

Anhydrous K_2CO_3 (1.78 g, 0.013 mol) and compound (**3a**) (3 g, 0.013 mol) were added in 20 ml DMF in a 100-ml round-bottom flask, fitted with guard tube, and stirred for 30 min. 1,3-dibromopropane (1.31 ml, 0.064 mol) was added and stirred at room temperature for 30 h. The completion of reaction was checked by TLC and the reaction mixture was worked up. Pure (**5a**) was obtained by column loaded with SiO_2 in $CHCl_3$. Eluent used was pure chloroform. Fluorescent green colored oily liquid.

Yield = 3.32 g (72%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.14–2.22 (p, 2H, CH_2) (J = 6.0, 6.0, 6.0, 6.0 Hz), δ 2.47 (s, 3H, CH_3), δ 3.37–3.41 (t, 2H, CH_2) (J = 6.3, 6.3 Hz), δ 3.87 (s, 3H, OCH_3), δ 4.29–4.34 (t, 2H, CH_2) (J = 5.7, 5.7 Hz), δ 7.26–7.62 (m, 5H, Ar–H).

Preparation of 5-(3-bromo-propoxy)-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid ethyl ester (**5b**)

In a 100-ml round-bottom flask, fitted with guard tube, compound (**3b**) (3 g, 0.012 mol) and K_2CO_3 (1.68 g, 0.012 mol) were stirred in 20 ml DMF for 20 min and 1, 3-dibromopropane (1.23 ml, 0.060 mol) was added. It was stirred for 30 h at room temperature and the completion of reaction was confirmed by TLC. The reaction mixture was worked up and the product (**5b**) was purified via column chromatography loaded with silica in chloroform.

Mp = 45–47°C. Yield = 3.15 g (70%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.37–1.41 (t, 3H, $OCH_2C\underline{H}_3$) (J = 6.9, 7.5 Hz), δ 2.16–2.22 (p, 2H, CH_2) (J = 6.0, 6.3, 6.0 Hz), δ 2.47 (s, 6H, $CH_3 \times 2$), δ 3.35–3.39 (t, 2H, CH_2) (J = 6.3, 6.6 Hz), δ 4.30–4.37 (m, 4H, CH_2, $OC\underline{H}_2CH_3$), δ 7.26–7.62 (m, 5H, Ar–H).

Preparation of 5-[3-(1,3-dioxo-1,3-dihydro-isoindol-2-yl)-propoxy]-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester (**6a**)

Phthalimide (0.20 g, 0.001 mol) and anhydrous K_2CO_3 (0.19 g, 0.001 mol) were stirred in 15 ml DMF for 10 min and (**5a**) (0.5 g, 0.001 mol) was added. The reaction was stirred at r. t. for 24 h and the completion of reaction was checked by TLC. After completion of reaction, solvent was removed via rota vapor and the product was extracted with

$CHCl_3/H_2O$ (20/20 × 2 ml). The $CHCl_3$ layer was dried (Na_2SO_4), filtered, evaporated, and pure (**6a**) was obtained from 3% ethyl acetate in hexane mixture.

Mp = 74°C. Yield = 0.22 g (37%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.02–2.11 (p, 2H, CH_2) (J = 6.6, 6.9, 6.9, 6.6 Hz), δ 2.46 (s, 3H, CH_3), δ 3.70–3.75 (t, 2H, NCH_2) (J = 6.9, 7.5 Hz), δ 3.82 (s, 3H, OCH_3), δ 4.23–4.27 (t, 2H, OCH_2) (J = 6.6, 6.3 Hz), δ 7.26–7.84 (m, 9H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 15.3, 28.9, 34.8, 51.0, 73.7, 99.3, 123.2, 127.5, 129.0, 132.0, 133.0, 137.5, 151.0, 155.1, 163.4, 168.1. MS (*m/z*): 420(M^+). Element analysis: % C (calc.) 65.87 (found) 66.26; % N (calc.) 10.02 (found) 10.15; % H (calc.) 5.01 (found) 5.14.

Preparation of 5-[3-(1,3-dioxo-1,3-dihydro-isoindol-2-yl)-propoxy]-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid ethyl ester (**6b**)

Phthalimide (0.20 g, 0.001 mol) and anhydrous K_2CO_3 (0.19 g, 0.001 mol) were stirred in 15 ml DMF for 20 min and (**5b**) (0.5 g, 0.001 mol) was added. The reaction was stirred at r. t. for 24 h and the completion reaction was checked by TLC. After completion of reaction, solvent was removed via rota vapor and the product was extracted with $CHCl_3/H_2O$ (30/30 × 2 ml). The $CHCl_3$ layer was dried (Na_2SO_4), filtered, evaporated, and pure (**6b**) was obtained from 3% ethyl acetate in hexane mixture.

Mp = 78–80°C. Yield = 0.30 g (51%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.33–1.38 (t, 3H, $CH_2C\underline{H}_3$) (J = 6.9, 7.2 Hz), δ 2.04–2.08 (t, 2H, CH_2) (J = 6.9, 6.9 Hz), δ 2.46 (s, 3H, CH_3), δ 3.69–3.74 (t, 2H, NCH_2) (J = 7.2, 7.2 Hz), δ 4.25–4.32 (q, 4H, OCH_2, $OC\underline{H}_2CH_3$) (J = 7.5, 6.9, 6.6 Hz), δ 7.26–7.84 (m, 9H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 14.3, 15.3, 28.8, 34.7, 59.8, 73.4, 99.5, 123.3, 127.4, 129.0, 132.0, 133.8, 137.4, 150.9, 155.0, 162.9, 168.0. MS (*m/z*): 434(M^+). Element analysis: % C (calc.) 66.51 (found) 67.34; % N (calc.) 9.69 (found) 9.76; % H (calc.) 5.31 (found) 5.40.

Preparation of 5-(3-benzimidazol-1-yl-propoxy)-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester (**7a**)

Benzimidazole (0.49 g, 0.004 mol) and anhydrous K_2CO_3 (0.57 g, 0.004 mol) were stirred in 20 ml DMF for 30 min at r. t. and (**5a**) (1.48 g, 0.004 mol) was added slowly with stirring. The reaction was stirred at room temperature for 30 h and the completion of reaction was confirmed via TLC. DMF was removed via rotary evaporator and the product was extracted with $CHCl_3/H_2O$ (50/50 × 2 ml). The $CHCl_3$ layer was dried (Na_2SO_4), filtered, evaporated, and pure (**7a**) was obtained as oily liquid through column loaded with silica in chloroform. Eluent used was 5% ethyl acetate in chloroform.

Yield = 0.70 g (43%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.19–2.21 (p, 2H, CH_2) (J = 4.8 Hz), δ 2.47 (s, 3H, CH_3), δ 3.85 (s, 3H, OCH_3), δ 4.16–4.71 (double-t, 4H, OCH_2, NCH_2) (J = 6.3, 6.9, 4.5, 5.4, 5.4, 7.2 Hz), δ 7.52 (s, 1H, CH), δ 7.27–8.03 (m, 9H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 14.6, 29.0, 40.5, 50.3, 71.7, 98.7, 108.9, 119.3, 121.6, 122.2, 122.8, 127.1, 128.4, 132.7, 136.6, 142.4, 142.8, 150.0, 154.2, 162.4.

Preparation of 5-(3-benzimidazol-1-yl-propoxy)-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid ethyl ester (**7b**)

In a 100-ml round-bottom flask, fitted with guard tube, benzimidazole (0.16 g, 0.001 mol) and anhydrous K_2CO_3 (0.19 g, 0.001 mol) were dissolved and stirred for 30 min followed by addition of (**5b**) (0.5 g, 0.001 mol). The reaction was stirred at r. t. for 48 h. Completion of reaction was confirmed via TLC and the reaction was worked up. DMF was removed using rotary evaporator and the product was extracted with $CHCl_3/H_2O$ (20/20 × 2 ml). The $CHCl_3$ layer was dried (Na_2SO_4), filtered, evaporated, and pure (**7b**) was obtained as oily liquid.

Yield = 0.02 g (36%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.34–1.39 (t, 3H, $OCH_2C\underline{H}_3$) (J = 6.9, 6.9 Hz), δ 2.18–2.22 (t, 2H, CH_2) (J = 5.7, 5.4 Hz), δ 2.47 (s, 3H, CH_3), δ 4.08–4.36 (m, 6H, CH_3 × 3), δ 7.26–7.78 (m, 9H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 14.2, 15.3, 29.5, 41.2, 59.8, 72.1, 99.5, 109.3, 120.1, 122.0, 122.7, 127.8, 129.0, 133.2, 137.2, 142.9, 150.7, 154.7, 162.7.

Preparation of 5-[3-(3-cyano-4,6-dimethyl-2-oxo-2H-pyridin-1-yl)-propoxy]-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester (8a-I) and 5-[3-(3-cyano-4,6-dimethyl-pyridin-2-yloxy)-propoxy]-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester (**8a-II**)

In a 100-ml round-bottom flask, fitted with guard tube, pyridone (0.42 g, 0.003 mol) and anhydrous K_2CO_3 (0.38 g, 0.003 mol) were dissolved in 20 ml DMF for 30 min and (**5a**) (1.0 g, 0.003 mol) was added slowly with stirring. The reaction was stirred at r. t. for 48 h. The completion of reaction was confirmed via TLC. Reaction was worked up. Solvent was removed via rotary evaporator and the product mixture was extracted with $CHCl_3/H_2O$ (60/60 × 2 ml). The $CHCl_3$ layer was dried (Na_2SO_4), filtered, and evaporated. The product mixture was crystallized with 5% ethyl acetate in hexane and recrystallized

with 2% ethyl acetate in hexane to obtain pure **(8a-I)** and **(8a-II)**.

(8a-I): Mp = 110–112°C. Yield = 0.20 g (17%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.04–2.11 (p, 2H, CH_2) (J = 5.4, 5.7, 7.5 Hz, δ 2.32–2.36 (d, 6H, $CH_3 \times 2$) (J = 12.9 Hz), δ 2.46 (s, 3H, CH_3), δ 3.85 (s, 3H, OCH_3), δ 4.05–4.09 (t, 2H, CH_2) (J = 7.5, 7.8 Hz), δ 4.28–4.32 (t, 2H, CH_2) (J = 5.4, 5.4 Hz), δ 5.98 (s, 1H, CH), δ 7.26–7.59 (m, 5H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 15.3, 20.6, 28.3, 42.4, 51.1, 73.5, 99.3, 101.4, 109.4, 115.3, 123.5, 127.8, 129.1, 137.3, 150.4, 150.8, 155.0, 157.9, 160.8, 163.3. MS (*m/z*): 421(M^+). Element analysis: % C (calc.) 65.71 (found) 65.31; % N (calc.) 13.33 (found) 13.50; % H (calc.) 5.71 (found) 5.88.

(8a-II): Mp = 66–70°C. Yield = 0.06 g (5%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 2.13–2.17 (p, 2H, CH_2), δ 2.43 (s, 6H, $CH_3 \times 2$), δ 2.46 (s, 3H, CH_3), δ 3.86 (s, 3H, OCH_3), δ 4.36–4.38 (q, 2H, OCH_2) (J = 4.2 Hz), δ 4.38–4.39 (t, 4H, $OCH_2 \times 2$) (J = 4.2 Hz), δ 6.68 (s, 1H, CH), δ 7.21–7.60 (m, 5H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 15.2, 20.8, 24.5, 28.3, 29.1, 51.1, 63.0, 72.5, 93.9, 99.5, 109.4, 114.7, 123.6, 127.3, 128.8, 137.4, 151.0, 154.2, 160.5, 163.4, 163.6. MS (*m/z*): 421(M^+). Element analysis: % C (calc.) 65.71 (found) 65.53; % N (calc.) 13.33 (found) 13.01; % H (calc.) 5.71 (found) 5.42.

Preparation of 5-[3-(3-cyano-4,6-dimethyl-2-oxo-2H-pyridin-1-yl)-propoxy]-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid ethyl ester (8b-I) and 5-[3-(3-cyano-4,6-dimethyl-pyridin-1-yloxy)-propoxy]-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid ethyl ester (**8b-II**)

In a 100-ml round-bottom flask, fitted with guard tube, pyridone (0.21 g, 0.001 mol) and anhydrous K_2CO_3 (0.38 g, 0.003 mol) were dissolved in 10 ml DMF for 1 h and (**5b**) (0.5 g, 0.001 mol) was added slowly with stirring. The reaction was stirred at r. t. for 24 h. The completion of reaction was confirmed via TLC. Reaction was worked up. Solvent was removed via rotary evaporator and the product mixture was extracted with $CHCl_3/H_2O$ (20/20 × 2 ml). The $CHCl_3$ layer was dried (Na_2SO_4), filtered, and evaporated. Column was loaded with SiO_2 in $CHCl_3$ and (**8b-II**) was eluted first out of the column with pure chloroform, and (**8b-I**) was eluted with 10% ethyl acetate in chloroform.

(8b-I): Mp = 78–82°C. Yield = 0.25 g (42%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.35–1.40 (t, 3H, $OCH_2C\underline{H}_3$) (J = 7.2, 6.9 Hz), δ 2.03–2.05 (m, 2H, CH_2) (J = 5.4 Hz), δ 2.30 (s, 3H, CH_3), δ 2.35 (s, 3H, CH_3), δ 2.46 (s, 3H, CH_3), δ 4.03–4.33 (t, 2H, $OC\underline{H}_2CH_3$) (J = 7.2, 6.6 Hz), δ 4.29–4.33 (t, 4H, $CH_2 \times 2$) (J = 6.9, 3.6 Hz), δ 5.97 (s, 1H, CH), δ 7.26–7.69 (m, 5H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 14.3, 15.4, 20.6, 28.2, 42.4, 59.9, 73.4, 99.5, 101.3, 109.4, 115.3, 123.5, 127.8, 129.0, 137.3, 150.4, 150.8, 154.9, 157.9, 160.7, 162.8. MS (*m/z*): 434(M^+). Element analysis: % C (calc.) 66.51 (found) 66.02; % N (calc.) 9.69 (found) 9.03; % H (calc.) 5.31 (found) 5.30.

(8b-II): Mp = 54–55°C. Yield = 0.10 g (17%). ^{1}H NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 1.36–1.41 (t, 3H, $OCH_2C\underline{H}_3$) (J = 6.9, 7.2 Hz), δ 2.13–2.17 (m, 2H, CH_2) (J = 6.0, 6.6 Hz), δ 2.42 (s, 6H, $CH_3 \times 2$), δ 2.46 (s, 3H, CH_3), δ 4.32–4.41 (m, 6H, $CH_2 \times 3$, $OC\underline{H}_2CH_3$, $OCH_2 \times 2$), δ 6.67 (s, 1H, CH), δ 7.19–7.59 (m, 5H, Ar–H). ^{13}C NMR (300 MHz, 25°C, $Si(CH_3)_4$, $CDCl_3$): δ 14.4, 15.2, 19.9, 24.4, 29.0, 59.9, 62.9, 72.4, 93.8, 114.6, 117.4, 123.4, 127.2, 128.7, 137.4, 151.0, 154.1, 155.0, 160.4, 162.9, 163.5. MS (*m/z*): 434(M^+). Element analysis: % C (calc.) 66.51 (found) 66.33; % N (calc.) 9.69 (found) 9.51; % H (calc.) 5.31 (found) 5.71.

References

Almansa C, Alfon J, De Arriba A, Cavalcanti F, Escamilla I, Gomez L, Miralles oliva AR, Bartolli J, Carceller E, Merlos M, Garcia-Rafenell J (2003) Synthesis and Structure-activity relationship of a new series of COX-2 selective inhibitors: 1,5-diarylimidazoles. J Med Chem 46:3463–3475

Amir M, Kumar S (2005) Synthesis and anti-inflammatory, analgesic, ulcerogenic and lipid Peroxidation activities of 3,5-dimethyl pyrazoles, 3-methyl pyrazol-5-ones and 3,5-disubstituted pyrazolines. Indian J Chem 44B:2532–2537

Avasthi K, Chandra T, Bhakuni DS (1995) A convenient and general synthesis of 1, w-bis (4,6-dimethyl-thio-1H-pyrazolo [3,4-d] pyrimidin-l-yl) alkanes and their regioisomers. Indian J Chem 34B:944–949

Carter JS (2000) Inhibitors of cyclooxygenase-2: November 1999–April 2000. Expert Opin Ther Pathol 10:1011–1020

Dannhardt G, Kiefer W (2001) Cyclooxygenase inhibitors—current status and future prospects. European J Med Chem 36:109–126

Dannhardt G, Laufer S (2000) Structural approaches to explain the selectivity of COX-2 inhibitors: Is there a common pharmacophore? Curr Med Chem 7:1101–1112

Di Rosa M, Giroud JP, Willoughby DA (1971) Studies of the mediators of the acute inflammatory response induced in rats in different sites by carrageenan and turpentine. J Pathol 104:15–29

Garcia-Leme J (1978) Bradykininsystem. In: Vane JR, Ferreira SH (eds) Handbook of experimental pharmacology, vol 50/I. Springer, Berlin, p 464

Gursoy A, Demirayak S, Capan G, Erol K, Vural K (2000) Synthesis and preliminary evaluation of new 5-pyrazolinone derivatives as analgesic agents. Euro J Med Chem 35:359–364

Hla T, Neilson K (1992) Human cyclooxygenase-2 cDNA. Proc Nati Acad Sci USA 89:7384–7388

Holsapple MP, Schnur M, Yim GKW (1980) Pharmocological modulation of edema mediated by prostaglandin, serotonin and histamine. Agents and Actions 10:368–373

Insel PA (1996) Analgesic, antipyretic and anti-inflammatory agents and drugs employed in the treatment of gout. In: Hardman JG,

Limbird LE (eds) Goodman & Gilman's the pharmacological basis of therapeutics. McGraw-Hill, New York, pp 617–657

Kulkarni SK, Mehta AK, Kunchandy J (1986) Anti-inflammatory actions of clonidine, guanfacine and B-HT 920 against various inflammagen-induced acute paw oedema in rats. Arch Int Pharmacodyn 279:324–334

Kumar A, Sharma S, Bajaj K, Bansal D, Sharma S, Archana K, Saxena KK, Lata S, Gupta B, Srivastava VK (2003) Synthesis and anti-inflammatory, analgesic, ulcerogenic and cyclooxygenase activities of novelquinasolinyl-pyrazolines. Indian J Chem 42:1979–1984

Leonard NJ (1979) Stacking interaction between tryptophan and uracil in a synthetic model compound. Acc Chem Res 12:423–429

Lombardino JG, Otterness KK, Wiseman EH (1975) Acidic anti-inflammatory agents—correlations of some physical, pharmacological and clinical data. Arzneimittel-Forschung 25:1629–1635

Meyer EA, Castellano RK, Diederich F (2003) Interactions with aromatic rings in chemical and biological recognition. Angew Chem Int Ed Eng 42:1210–1250

Reynold JEF (1993) The extra pharmacopoeia. In: Reynold JEF (ed) Martindale, 30th edn. Pharmaceutical Press, London, p 1

Talley JJ (1999) Selective inhibitors of cyclooxygenase-2 (COX-2). Prog Med Chem 36:201–234

Tewari AK, Mishra A (2001) Synthesis and anti-inflammatory activities of N4,N5-disubstituted-3-methyl-1H-pyrazolo[3,4-c]pyridazines. Bioorganic Med Chem 9:715–718

Tewari AK, Dubey R, Mishra A (2009) 2-substituted-8-methyl-3, 6-dihydroimidazo [4,5-c] pyrazolo[3,4-e]pyridazine as an Anti-inflammatory agent. Med Chem Res (2011) 20:16–22

Tewari AK, Srivastava P, Singh VP, Singh A, Goel RK, Gopimohan C (2010a) Novel anti-inflammatory agents based on pyrazole based dimeric compounds; design, synthesis, docking and in vivo activity. Chem Pharm Bull Japan 58:634–638

Tewari AK, Srivastava P, Singh VP, Puerta C, Valerga P (2010b) Condensed phase conformational isomerisation of 5-[3-(1, 3-Dioxo-1,3-dihydro-isoindol-2-yl)-propoxy]-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester. Arkivoc (ix): 127–136

Vinegar R, Schreiber W, Hugo R (1969) Biphasic development of Carrageenan edema in rats. J Pharmacol Exp Ther 166:96–103

Wintar CA, Risley EA, Nuss GW (1962) Carragenin-induced edema in hind paw of the rat as an assay for anti-inflammatory drugs. Proc Soc Exp Biol 111:544