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ORIGINAL PAPER

Synthesis and biological studies of some new acrylic acid ethyl esters of quinolinone

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Abstract A series of new acrylic acid ethyl esters of quinolinones were synthesized from 4-(bromomethyl)-quinolinones and screened for in vitro antimicrobial and in vivo analgesic and anti-inflammatory activities. Most of the compounds with chloro substitution at the C-6 or C-7 position in the quinolinone moiety and a methoxy group in the aryloxy moiety showed potent antibacterial and antifungal activities when compared with non-halogenated quinolinones and the quinolinones bearing a CH_3 at the C-8 position. In a pharmacological evaluation, the halogen substitution at the C-6 or C-7 position in quinolinones was found to enhance both analgesic and anti-inflammatory activities of the molecule when compared with a simple unsubstituted (non-halogenated) quinolinone. The structures of all newly synthesized compounds were characterized by elemental analysis, IR, ^{1}H NMR, ^{13}C NMR, and FAB-MS.

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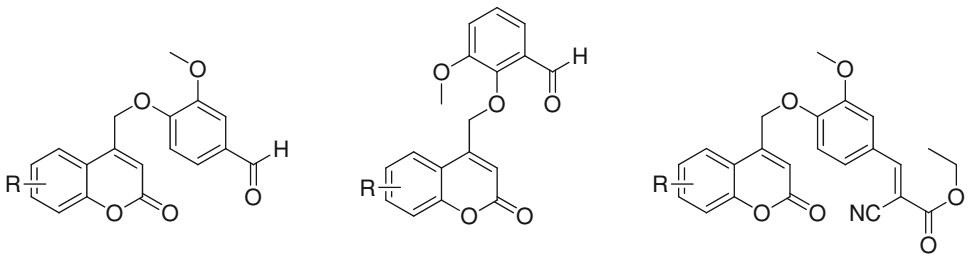
Introduction

Quinolinones belong to the class of lactams which is an indispensable heterocyclic unit to both chemists and biochemists. The natural occurrence and antimicrobial, anti-inflammatory, anti-cancer, anti-HIV, and other miscellaneous properties of this system were recently reviewed [1]. Quinolinone derivatives are metabolized to the corresponding 8-hydroxycoumarins in biological systems and are therefore very good anti-inflammatory and analgesic agents [2]. The triheterocyclic thiazoles synthesized from 4-(aminomethyl)quinolinones and 3-(bromoacetyl)coumarins [3] in our laboratory exhibit promising anti-inflammatory and analgesic activity even after 24 h. We also reported that the introduction of halogens in general and fluorine in particular at position 4 in the aryloxy and arylamino moieties of coumarin and quinolinone [4–6] enhances the antimicrobial as well as analgesic and anti-inflammatory activities. Interest in quinolinones as antibiotics is due to their potent inhibition of bacterial DNA gyrase, which is involved in the growth [7, 8]. Vanillins exhibit antimicrobial properties and have been accepted as safer flavoring agents [9, 10]. In view of this, we reported the analgesic and potent anti-inflammatory activities of the vanillin ethers derived from 4-substituted coumarins (Fig. 1) [11], and novel ethers of a nitrogen analogue of coumarin also known as 1-azacoumarin or quinolinone linked with coumarins [12]. The observed anti-inflammatory activity of quinolinones is due to the generation of a carboxyl group in the biological system [13]. The

Fig. 1 Biologically active coumarin molecules



mechanism of action of the generation of carboxyl group containing anti-inflammatory drugs like indomethacin is based on the interference with prostaglandin synthesis via cyclooxygenase inhibition. Thus the structure of a potential drug should mimic that of arachidonic acid to be viable. Encouraged by our earlier studies, it was thought of interest to synthesize some new aryloxymethyl quinolinones with vanillin and *p*-hydroxybenzaldehyde. They were further converted into corresponding cyanoesters and acetoesters which can act as precursors for generating the carboxylic group and hence are expected to exhibit good anti-inflammatory properties. The present paper reports the synthesis and preliminary biological evaluation of new acrylic acid ethyl esters of quinolinone.

Results and discussion

Syntheses

Various 4-(bromomethyl)quinolinones **1a–1d** were synthesized by the bromination of acetoacetanilides and cyclization of the intermediate ω-bromoacetoacetanilides in sulfuric acid [4, 14]. The various substituted 4-[(4-formylphenoxy)-methyl]quinolinones **2a–2d** were prepared by the reaction of 4-(bromomethyl)quinolinones and *p*-hydroxybenzaldehyde in the presence of anhydrous potassium carbonate and dry ethanol at 100 °C. The corresponding cyanoesters **4a–4d** and acetoesters **6a–6d** were synthesized by the condensation of ethyl cyanoacetate and ethyl acetoacetate in the presence of a catalytic amount of piperidine in dry ethanol (Scheme 1). Similarly, the synthesis of substituted 4-[(4-formyl-2-methoxyphenoxy)methyl]quinolinones **3a–3d** was achieved by the reaction of 4-(bromomethyl)quinolinones and vanillin in the presence of anhydrous potassium carbonate and dry ethanol at 100 °C. The corresponding cyanoesters **5a–5d** and acetoesters **7a–7d** were synthesized by the condensation of ethyl cyanoacetate and ethyl acetoacetate in the presence of a catalytic amount of piperidine in dry ethanol.

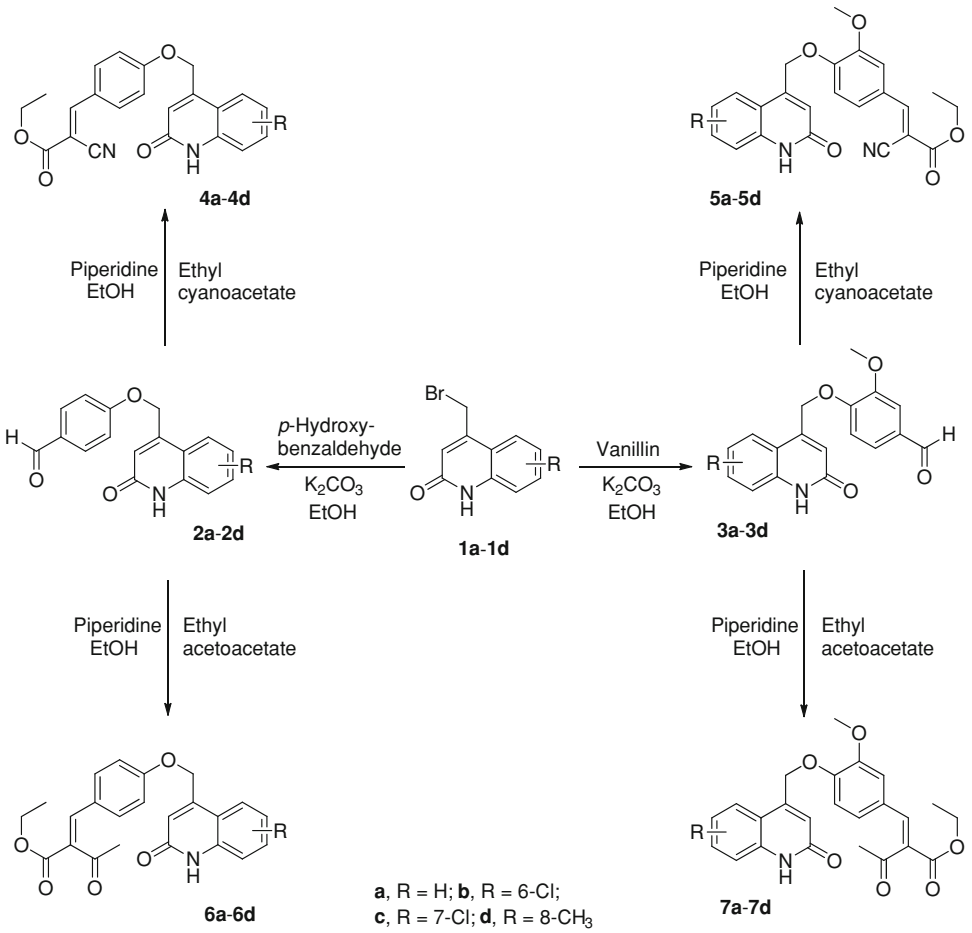
Cyanoesters **4a–4d** and **5a–5d** were also prepared by the condensation of 4-(bromomethyl)quinolinones with 2-cyano-3-(4-hydroxyphenyl)acrylic acid ethyl ester (**8a**) and 2-cyano-3-(4-hydroxy-3-methoxyphenyl)acrylic acid ethyl ester (**8b**) in the presence of anhydrous potassium carbonate and dry ethanol at 100 °C. Similarly the acetoesters **6a–6d** and **7a–7d** were prepared by the condensation of 4-(bromomethyl)quinolinones with 2-acetyl-3-(4-hydroxyphenyl)-acrylic acid ethyl ester (**9a**) and 2-acetyl-3-(4-hydroxy-3-methoxyphenyl)acrylic acid ethyl ester (**9b**) in the presence of anhydrous potassium carbonate and dry ethanol at 100 °C (Scheme 2). This method of preparation is easier than the earlier method (Scheme 1) because the products are obtained in better yield and purity. Synthesis of compounds **2a–2d** and **3a–3d** was carried out (Scheme 1) in order to study their biological activities in comparison with compounds **4a–4d**, **5a–5d**, **6a–6d**, and **7a–7d**.

All products gave satisfactory analytical and spectroscopic data, which are in full accordance with their assigned structures. The ORTEP diagram of the intermediate compound 4-[(4-formylphenoxy)methyl]quinolinone (**2a**) is shown in Fig. 2 [15].

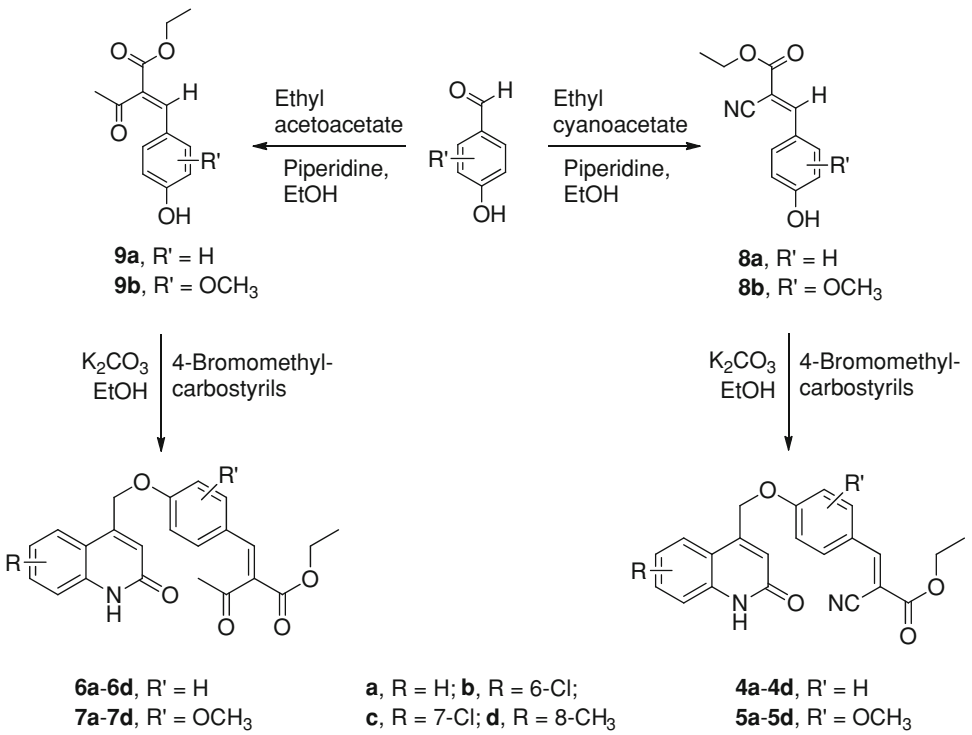
Antimicrobial activity

The antibacterial (against *Escherichia coli* and *Bacillus cirrhosis*) and antifungal (against *Aspergillus niger* and *Rhizoctonia bataticola*) activities of the compounds were tested at three concentrations (25, 50, and 100 μg cm^{-3}) using norfloxacin and griseofulvin as standard antibacterial and antifungal drugs, respectively [16–18]. Tables 1 and 2 list the zones of growth inhibition (in millimeters and as percentage values) obtained against the tested bacteria and fungi. Most of the compounds exhibited pronounced activity against *E. coli* at 50 and 100 μg cm^{-3}, whereas at 25 μg cm^{-3} all compounds are inactive except **2b**, **2c**, **3b**, **3c**, **7b**, and **7c** bearing chlorine functionality in the quinolinone ring system. Conversely, against *B. cirrhosis*, nearly all compounds showed inhibition potency, which ranged from 12 to 83%. In particular, though compounds **2b** and **2c** exhibited marked antibacterial activity against both the bacterial strains, their vanillin analogues **3b** and **3c** exhibited strikingly high potency of up to 72 and 75% inhibition of *E. coli* and similarly 80 and 83% inhibition of *B. cirrhosis* at 100 μg cm^{-3}. On the other hand, modification of the aldehyde moiety in **2a–3d** either by condensation with ethyl acetoacetate or ethyl cyanoacetate (i.e., compounds **4a–7d**) did not show any sign of improved activity against *E. coli* and *B. cirrhosis*. However, compounds **7b** and **7c** exhibited comparable potency of about 46 and 48%

Scheme 1



Scheme 2



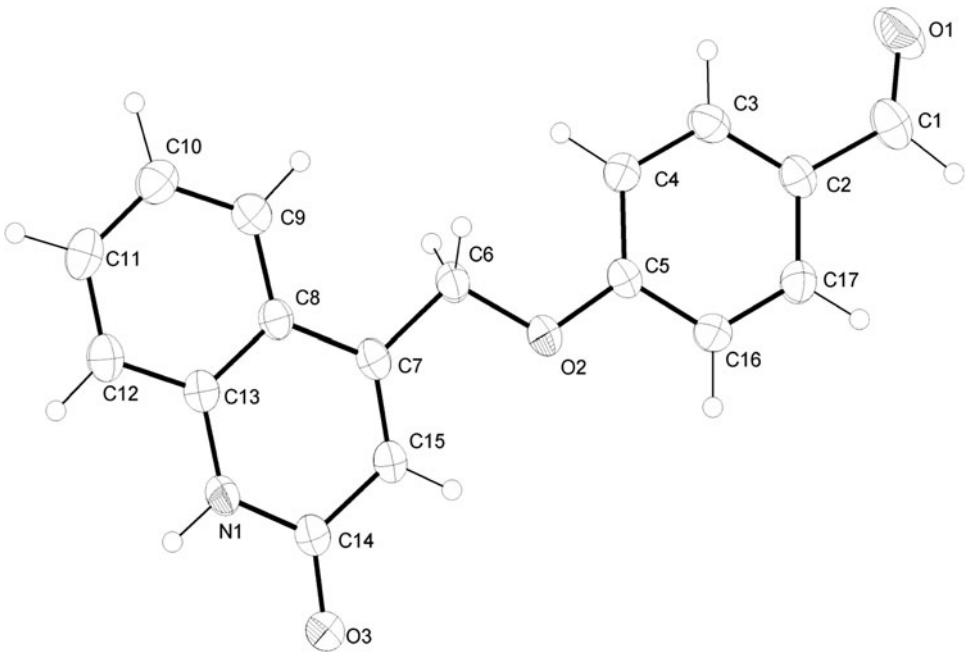


Fig. 2 ORTEP drawing of 4-[(4-formylphenoxy)methyl]quinoline-2(1*H*)-one (**2a**)

Table 1 Antibacterial activity of selected compounds (at 25, 50, and 100 μg/cm^3)

Compound	Zone of inhibition											
	E. coli (gram negative)						*B. cirrhosis* (gram positive)					
	100 μg/cm^3		50 μg/cm^3		25 μg/cm^3		100 μg/cm^3		50 μg/cm^3		25 μg/cm^3	
	mm	%	mm	%	mm	%	mm	%	mm	%	mm	%
2a	82	26	–	–	–	–	98	35	45	13	–	–
2b	132	48	86	34	38	12	144	56	92	40	48	21
2c	138	51	92	37	44	17	148	58	94	41	48	21
3a	94	32	–	–	–	–	108	40	48	15	–	–
3b	186	72	121	53	62	30	198	80	134	63	66	35
3c	194	75	127	56	69	35	204	83	138	65	78	44
4a	86	28	–	–	–	–	110	40	60	22	–	–
4b	98	33	42	11	–	–	118	44	62	23	–	–
4c	106	37	48	14	–	–	126	48	74	30	36	12
5a	108	38	46	13	–	–	132	50	80	33	40	15
5b	118	42	60	21	–	–	148	58	92	40	44	18
5c	120	43	64	23	–	–	152	60	98	43	50	23
6a	88	29	–	–	–	–	104	38	50	16	–	–
6b	110	39	46	13	–	–	124	47	70	27	–	–
6c	106	37	40	10	–	–	138	53	82	34	40	15
7a	112	40	60	21	–	–	118	44	58	21	–	–
7b	126	46	84	33	36	11	178	71	124	57	54	26
7c	132	48	90	36	44	17	170	68	118	44	48	21
7d	98	33	46	13	–	–	102	37	48	15	–	–
Std	250	100	210	100	160	100	240	100	200	100	150	100
Ctr	20	–	20	–	20	–	20	–	20	–	20	–

Std standard (norfloxacin, 100% inhibition at each concentration), *Ctr* control (*N*,*N*-dimethylformamide, DMF)

inhibition of *E. coli* at 100 μg cm^{-3} but fairly good activity of 71 and 68% inhibition of *B. cirrhosis* at the same concentration.

The results of antibacterial activities (Table 1) show that compounds **3b** and **3c** showed a very good antibacterial activity of up to 72 and 75% inhibition of *E. coli* and

Table 2 Antifungal activity of the selected compounds (at 25, 50, and 100 μg/cm^3)

Compound	Zone of inhibition											
	A. niger						*R. bataticola*					
	100 μg/cm^3		50 μg/cm^3		25 μg/cm^3		100 μg/cm^3		50 μg/cm^3		25 μg/cm^3	
	mm	%	mm	%	mm	%	mm	%	mm	%	mm	%
2a	62	19	–	–	–	–	102	34	–	–	–	–
2b	136	52	84	35	38	12	160	58	94	37	58	25
2c	142	55	78	32	34	10	164	60	100	40	62	28
3a	98	35	54	18	–	–	110	37	72	26	–	–
3b	194	79	132	62	66	32	206	78	148	64	84	42
3c	206	84	140	66	82	44	214	80	154	67	88	45
4a	72	23	–	–	–	–	118	40	74	27	–	–
4b	100	36	58	21	–	–	132	46	92	36	50	20
4c	110	40	66	25	–	–	136	48	92	36	50	20
5a	98	35	50	16	–	–	100	33	–	–	–	–
5b	136	52	78	32	38	12	126	44	90	35	48	18
5c	140	54	80	33	38	12	138	49	100	40	60	26
6a	60	18	–	–	–	–	110	37	68	25	–	–
6b	112	41	66	25	–	–	142	50	100	40	56	24
6c	108	40	64	24	–	–	148	53	110	45	62	28
7a	98	35	55	17	–	–	100	33	–	–	–	–
7b	162	64	92	40	58	27	212	80	150	65	80	40
7c	170	68	98	43	62	30	218	82	154	67	90	46
7d	110	40	60	22	–	–	136	48	90	35	40	13
Std	240	100	200	100	160	100	260	100	220	100	170	100
Ctr	20	–	20	–	20	–	20	–	20	–	20	–

Std standard (griseofulvin, 100% inhibition at each concentration), *Ctr* control (DMF)

similarly 80 and 83% inhibition of *B. cirrhosis* at 100 μg cm^{-3}. The same compounds were even active at 25 μg cm^{-3} and showed 30, 35, 35, and 44% inhibition of both the bacteria. Compounds **7b** and **7c** showed moderate activity of 46 and 48% inhibition of *E. coli* at 100 μg cm^{-3}, but fairly good activity of 71 and 68% inhibition of *B. cirrhosis* at the same concentration. Most of the tested compounds showed no activity at 25 μg cm^{-3} and moderate activity at 100 μg cm^{-3} for both the bacteria.

Similar to antibacterial activities, the inhibitory profiles of all compounds against fungal strains also registered the same trend at the three studied concentrations. Compounds **2b**, **2c**, **5b**, and **5c** showed a moderate inhibition of 52, 55, 52, and 54%, respectively, at 100 μg cm^{-3} for *A. niger*, and 58, 60, 44, and 49%, respectively, at the same concentration for *R. bataticola* (Table 2). The same compounds showed low activity from 12 to 26% inhibition at 25 μg cm^{-3} for both fungi. Compounds **3b**, **3c**, **7b**, and **7c** showed a very good inhibition of 79, 84, 64, 68% and 78, 80, 80, 82%, respectively, at 100 μg cm^{-3} for both fungi. Compounds **3b** and **3c** showed moderate activity of 32, 44, 42, and 45% even at 25 μg cm^{-3} for both fungi. Similarly compounds **7b** and **7c** showed moderate activity of 40 and 46%, respectively, at 25 μg cm^{-3} for *R. bataticola*. Most of the compounds showed moderate activity for both the fungi at 100 μg cm^{-3} and some of the compounds showed no activity at 25 μg cm^{-3}.

Acute toxicity studies

The acute toxicity studies of the test compounds were performed on albino mice fasted for 24 h [19]. The test compounds were administered orally and intraperitonally (i.p.). The animals were watched for mortality and symptoms until the eighth day. It was found that all compounds possess a good safety profile and no mortality of animals was observed even after 24 h.

Analgesic activity

Abdominal constriction response induced by acetic acid is a sensitive procedure to establish the efficacy of peripherally acting analgesics. Intraperitonial administration of acetic acid causes an increase in the level of PGE2 and PGF 2α [20]. The results of analgesic activity (Tables 3 and 4) indicate that compounds **3a**, **4a**, **6b**, and **7b** showed the highest analgesic activity, more than the standard. Compounds **3b**, **6c**, and **7c** showed more than 50%

Table 3 Anti-inflammatory and analgesic activities of selected compounds

Compound	Edema volume at different time intervals (% of inhibition)							Analgesic activity (number of writhings)
	0.5 h	1 h	2 h	4 h	8 h	12 h	24 h	
Std.	0.63 (3)	0.78 (8)	0.85 (11)	0.65* (35)	0.60* (41)	0.78 (15)	0.95 (13)	19*
Ctr.	0.65	0.85	0.95	1.00	1.01	0.82	0.95	35
3a	0.71 (0)	1.16 (0)	0.85 (11)	0.56* (44)	0.46* (54)	0.51** (38)	0.96 (0)	25*
3b	0.61 (6)	1.01 (0)	0.66* (31)	0.56* (44)	0.50* (50)	0.73 (11)	1.10 (0)	15*
6b	0.75 (0)	1.06 (0)	0.98 (0)	0.40* (60)	0.31* (69)	0.60*** (27)	0.88 (7)	22*
6c	0.73 (0)	1.05 (0)	0.61* (36)	0.38* (62)	0.28* (72)	0.45* (45)	0.98 (0)	13*
7a	0.60 (8)	1.01 (0)	0.55* (42)	0.28* (72)	0.23* (77)	0.50* (39)	0.86 (9)	9*
7b	0.51 (22)	0.78 (8)	0.78 (18)	0.75*** (25)	0.23* (77)	0.56*** (32)	0.86 (9)	24*
7c	0.58 (11)	0.75 (12)	0.75 (21)	0.58* (42)	0.26* (74)	0.45* (45)	0.91 (4)	13*
F value	2.83	38.3	12.0	21.4	27.0	10.6	2.51	86.41

Standard error of the mean (SEM) 0.05–0.15. Test compounds were administered at a dose of 100 mg kg^{-1}

Std standard (indomethacin at a dose of 10 mg kg^{-1}), *Ctr* control (carbethoxymethyl cellulose, CMC; 2%)

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ when compared to control

Table 4 Anti-inflammatory and analgesic activities of selected compounds

Compound	Edema volume at different time intervals (% of inhibition)							Analgesic activity (number of writhings)
	0.5 h	1 h	2 h	4 h	8 h	12 h	24 h	
Std.	0.63 (3)	0.78 (8)	0.85 (11)	0.65* (35)	0.60* (41)	0.78 (15)	0.95 (13)	19*
Ctr.	0.65	0.85	0.95	1.00	1.01	0.82	0.95	35
4a	0.65 (0)	0.65 (24)	0.82 (14)	0.50* (50)	0.48* (52)	0.68 (17)	0.75 (21)	17*
4b	0.61 (6)	0.68 (20)	0.73 (23)	0.37* (63)	0.30* (70)	0.66 (20)	0.70*** (26)	10*
4c	0.53 (19)	0.65 (24)	0.70*** (26)	0.52* (48)	0.28* (72)	0.66 (20)	0.70*** (26)	8*
5b	0.43 (34)	0.58* (32)	0.62** (35)	0.40* (60)	0.36* (64)	0.70 (15)	0.65* (8)	9*
5c	0.40 (39)	0.50* (41)	0.42* (56)	0.13* (87)	0.10* (90)	0.52** (37)	0.60** (6)	4*
F value	3.28	5.45	10.4	37.08	34.09	3.05	4.47	192.23

SEM 0.05–0.15. Test compounds were administered at a dose of 100 mg kg^{-1}

Std standard (indomethacin at a dose of 10 mg kg^{-1}), *Ctr* control (2% CMC)

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ when compared to control

inhibition of writhing when compared to the control and the standard group.

Anti-inflammatory activity

The anti-inflammatory activity of the test compounds was assessed by the formalin-induced rat paw edema method [21] and the results are given in Tables 3 and 4. Most of the compounds showed an extremely significant anti-inflammatory activity when compared to the control and the standard group, and their onset of action was much quicker than the standard, because they showed significant anti-inflammatory activity at 2 h. Compound **5c** showed a maximum of 90% inhibition of inflammation at 8 h and maintained activity at 12 and 24 h. The metabolites of this compound may also show some anti-inflammatory activity. Compound **5c** may be more potent than the standard. Similarly compounds with chloro substitution at the C-6 or C-7 position in the quinolinone moiety (**4b**, **4c**, **6b**, **6c**, **7b**, and **7c**) showed 69–77% inhibition of inflammation at 8 h. All compounds showed their highest activity until 8 h and some of the compounds (**4b**, **4c**, **6b**, **7a**, and **7b**) were active even at 12 and 24 h. Hence it can be concluded that these can act as prodrugs. A graphical representation of the anti-inflammatory activity of some of the compounds is given in Figs. 3 and 4.

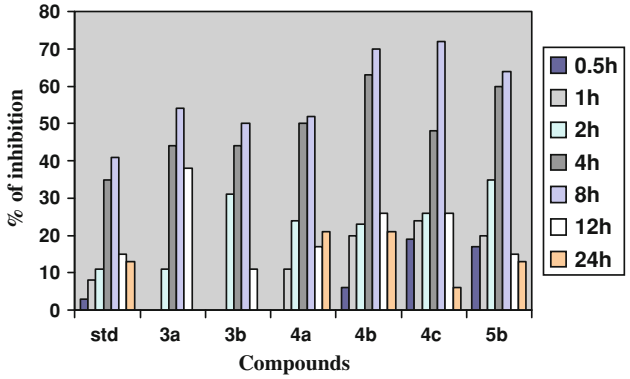


Fig. 3 Anti-inflammatory activity of selected compounds (**3a–5b**) and standard

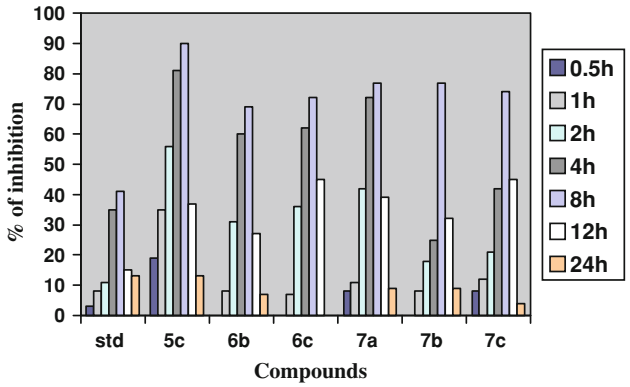


Fig. 4 Anti-inflammatory activity of selected compounds (**5c–7c**) and standard

Conclusion

The present study has shown that the newly synthesized quinolinone compounds **2b**, **2c**, **3b**, **3c**, **5b**, **5c**, **7b**, and **7c** with chloro substitution on quinolinone and a methoxy group in the aryloxy moiety showed potent antibacterial and antifungal activities when compared with unsubstituted quinolinones. The halogen substitution at the C-6 or C-7 position in quinolinones **3a**, **3b**, **4a**, **4b**, **6b**, and **7b** was found to enhance both analgesic and anti-inflammatory activities of the molecule in comparison with non-halogenated molecules. Our findings will be useful for chemists and biochemists conducting further investigations in this field in search of potent antimicrobial, analgesic, and anti-inflammatory agents.

Experimental

The melting points of the products were determined by open capillaries on a Büchi apparatus. The IR spectra were recorded on a Nicolet Impact-410 FT-IR spectrophotometer using KBr pellets. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker AC-300F 300 MHz spectrometer in $CDCl_3$, DMSO-d_6, or a mixture of $CDCl_3$ and CF_3COOH using TMS as an internal standard. D_2O exchange was applied to confirm the assignment of the signals of NH protons. The mass spectra were recorded on an Autospec ESI–MS. The elemental analysis was carried out by using a Heraus CHN rapid analyzer. C, H, and N values of all compounds were well within ±0.4% from the theoretical values. The homogeneity of the compounds was determined by TLC on silica gel 60 F_{254} (Merck) aluminum plates visualized by UV light (254 nm) and iodine vapor. The reagents were all analytical reagent grade or chemically pure. All solvents (including dry ethanol) were dried, deoxygenated, and redistilled before use by standard procedures [22].

General procedure for the synthesis of compounds ***2a–2d***

A mixture of substituted 4-(bromomethyl)quinolinone **1a–1d** (4.00 mmol), 4-hydroxybenzaldehyde (4.00 mmol), and anhydrous potassium carbonate (4.00 mmol) in 20 cm^3 dry ethanol was refluxed on a water bath for 8 h. The separated solid was filtered off, washed with 20% HCl and with excess of cold water, dried, and crystallized from a suitable solvent.

4-[(1,2-Dihydro-2-oxoquinolin-4-yl)methoxy]-benzaldehyde (**2a**, $C_{17}H_{13}NO_3$)
Yield: 87%; colorless crystals (acetic acid); m.p.: 250–252 °C; IR (KBr):$\bar{\nu}$ = 3,431 (N–H stretching), 1,688 (C=O stretching, aldehyde), 1,669 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 5.50 (s, 2H, C4-CH_2), 6.60 (s, 1H, C3-H of quinolinone), 7.20–7.92 (m, 9H, Ar–H), 9.92 (s, 1H, CHO), 11.80 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 162.50, 160.44, 158.28, 156.34, 154.28, 136.89, 133.48, 132.80, 127.08, 124.60, 119.40, 116.30, 116.21, 114.47, 114.30, 109.10, 65.98 ppm; FAB-MS: m/z = 280 (M + H).

4-[(6-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-benzaldehyde (**2b**, $C_{17}H_{12}ClNO_3$)
Yield: 82%; colorless crystals (ethanol); m.p.: 198–200 °C; IR (KBr):$\bar{\nu}$ = 3,438 (N–H stretching), 1,680 (C=O stretching, aldehyde), 1,658 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 5.45 (s, 2H, C4-CH_2), 6.68 (s, 1H, C3-H of quinolinone), 7.10–7.90 (m, 8H, Ar–H), 9.90 (s, 1H, CHO), 11.78 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 163.56, 161.84, 158.28, 157.34, 153.28, 134.89, 133.48, 132.80, 128.08, 127.60, 119.40, 116.30, 115.21, 114.47, 114.30, 108.10, 64.98 ppm; FAB-MS: m/z = 315 (M + H).

4-[(7-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-benzaldehyde (**2c**, $C_{17}H_{12}ClNO_3$)
Yield: 78%; colorless crystals (ethanol + dioxan); m.p.: 210–212 °C; IR (KBr):$\bar{\nu}$ = 3,440 (N–H stretching), 1,689 (C=O stretching, aldehyde), 1,665 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 5.52 (s, 2H, C4-CH_2), 6.65 (s, 1H, C3-H of quinolinone), 7.08–7.88 (m, 8H, Ar–H), 9.94 (s, 1H, CHO), 11.80 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 162.58, 160.90, 158.68, 156.34, 155.28, 136.89, 134.48, 133.80, 128.08, 127.60, 119.40, 117.30, 116.21, 115.47, 114.30, 109.10, 66.98 ppm; FAB-MS: *m/z* = 315 (M + H).

4-[(1,2-Dihydro-8-methyl-2-oxoquinolin-4-yl)methoxy]-benzaldehyde (**2d**, $C_{18}H_{15}NO_3$)
Yield: 88%; colorless crystals (ethanol + dioxan); m.p.: 212–214 °C; IR (KBr):$\bar{\nu}$ = 3,434 (N–H stretching), 1,692 (C=O stretching, aldehyde), 1,670 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 2.50 (s, 3H, C8-CH_3 of quinolinone), 5.55 (s, 2H, C4-CH_2), 6.65 (s, 1H, C3-H of quinolinone), 7.20–7.94 (m, 8H, Ar–H), 9.90 (s, 1H, CHO), 11.81 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 164.58, 161.90, 159.68, 157.34, 156.28, 137.89, 135.48, 134.80, 129.08, 128.60, 118.40, 117.30, 116.21, 115.47, 114.30, 108.10, 65.90, 18.20 ppm; FAB-MS: *m/z* = 295 (M + H).

General procedure for the preparation of compounds ***3a–3d***

A mixture of substituted 4-(bromomethyl)quinolinone **1a**–**1d** (4.00 mmol), vanillin (4.00 mmol), and anhydrous potassium carbonate (4.00 mmol) in 20 cm^3 dry ethanol was refluxed on a water bath for 8 h. The separated solid was filtered off, washed with 20% HCl and with excess of cold water, dried, and crystallized from a suitable solvent.

4-[(1,2-Dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxybenzaldehyde (**3a**, $C_{18}H_{15}NO_4$)
Yield: 77%; colorless crystals (ethanol); m.p.: 208–210 °C; IR (KBr):$\bar{\nu}$ = 3,443 (N–H stretching), 1,680 (C=O stretching, aldehyde), 1,659 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 3.74 (s, 3H, OCH_3), 5.25 (s, 2H, C4-CH_2), 6.59 (s, 1H, C3-H of quinolinone), 6.84–7.47 (m, 7H, Ar–H), 9.64 (s, 1H, CHO), 11.48 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 163.56, 161.40, 158.68, 156.34, 154.28, 136.89, 133.48, 132.86, 128.18, 125.65, 119.40, 116.30, 116.21, 114.47, 114.30, 108.10, 65.98, 55.02 ppm; FAB-MS: *m/z* = 311 (M + H).

4-[(6-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxybenzaldehyde (**3b**, $C_{18}H_{14}ClNO_4$)
Yield: 72%; colorless crystals (ethanol); m.p.: 216–218 °C; IR (KBr):$\bar{\nu}$ = 3,440 (N–H stretching), 1,688 (C=O stretching, aldehyde), 1,662 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 3.96 (s, 3H, OCH_3), 5.38 (s, 2H, C4-CH_2), 6.60 (s, 1H, C3-H of quinolinone), 6.80–7.50 (m, 6H, Ar–H), 9.80 (s, 1H, CHO), 11.52 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 163.50, 160.94, 158.18, 157.24, 154.48, 134.89, 133.48, 132.80, 128.08, 127.60, 119.40, 117.30, 116.21, 114.47, 113.30, 108.10, 65.78, 54.60 ppm; FAB-MS: *m/z* = 346 (M + H).

4-[(7-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxybenzaldehyde (**3c**, $C_{18}H_{14}ClNO_4$)
Yield: 73%; colorless crystals (ethanol + dioxan); m.p.: 215–217 °C; IR (KBr):$\bar{\nu}$ = 3,440 (N–H stretching), 1,689 (C=O stretching, aldehyde), 1,665 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 5.52 (s, 2H, C4-CH_2), 6.65 (s, 1H, C3-H of quinolinone), 7.08–7.88 (m, 8H, Ar–H), 9.94 (s, 1H, CHO), 11.80 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 162.58, 160.90, 158.68, 156.34, 155.28, 136.89, 134.48, 133.80, 128.08, 127.60, 119.40, 117.30, 116.21, 115.47, 114.30, 109.10, 66.98 ppm; FAB-MS: *m/z* = 346 (M + H).

4-[(1,2-Dihydro-8-methyl-2-oxoquinolin-4-yl)methoxy]-3-methoxybenzaldehyde (**3d**, $C_{19}H_{17}NO_4$)
Yield: 80%; colorless crystals (ethanol + dioxan); m.p.: 220–222 °C; IR (KBr):$\bar{\nu}$ = 3,434 (N–H stretching), 1,662 (C=O stretching, aldehyde), 1,658 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 2.45 (s, 3H, C8-CH_3 of quinolinone), 3.96 (s, 3H, OCH_3), 5.48 (s, 2H, C4-CH_2), 6.60 (s, 1H, C3-H of quinolinone), 6.80–7.81 (m, 7H, Ar–H), 9.80 (s, 1H, CHO), 11.82 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 163.18, 161.90, 158.58, 157.34, 156.28, 137.89, 136.48, 134.80, 129.08, 128.66, 119.40, 117.30, 116.21, 115.47, 114.30, 109.10, 65.90, 55.06, 20.10 ppm; FAB-MS: *m/z* = 326 (M + H).

General procedure for the preparation of compounds ***4a–4d***

A mixture of substituted 4-[(4-formylphenoxy)methyl] quinolin-2(1*H*)-one **2a**–**2d** (4.0 mmol), ethyl cyanoactate (4.0 mmol), and a catalytic amount of piperidine in 20 cm^3 ethanol was stirred for 6 h at room temperature and left overnight. The resulting yellow liquid was poured onto crushed ice. The yellow solid obtained was filtered off, washed with excess of cold water, dried, and crystallized from a suitable solvent.

2-Cyano-3-[4-[(1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-phenyl]-2-propenoic acid ethyl ester (**4a**, $C_{22}H_{18}N_2O_4$)
Yield: 86%; colorless crystals (acetic acid); m.p.: 264–266 °C; IR (KBr):$\bar{\nu}$ = 3,431 (N–H stretching), 2,221

(C≡N stretching), 1,722 (C=O stretching, ester), 1,659 (C=O stretching, amide) cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): $\delta = 1.40–1.45$ (t, 3H, CH_3-ester), 4.32–4.38 (q, 2H, CH_2-ester), 5.51 (s, 2H, C4-CH_2), 7.19–8.10 (m, 9H, Ar–H), 7.70 (s, 1H, =CH ethylenic), 8.44 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 166.56$, 162.40, 158.68, 156.34, 154.28, 136.89, 133.48, 132.86, 130.30, 129.32, 128.18, 126.33, 125.65, 119.40, 116.30, 116.21, 114.47, 114.30, 108.10, 65.98, 63.02, 15.30 ppm; FAB-MS: $m/z = 375$ (M + H).

2-Cyano-3-[4-[(6-chloro-1,2-dihydro-2-oxoquinolin-4-yl)-methoxy]phenyl]-2-propenoic acid ethyl ester
(**4b**, $C_{22}H_{17}ClN_2O_4$)
Yield: 81%; colorless crystals (acetic acid); m.p.: 290–292 °C; IR (KBr):$\bar{\nu} = 3{,}413$ (N–H stretching), 2,221 (C≡N stretching), 1,727 (C=O stretching, ester), 1,659 (C=O stretching, amide) cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): $\delta = 1.42–1.47$ (t, 3H, CH_3-ester), 4.43–4.50 (q, 2H, CH_2-ester), 5.58 (s, 2H, C4-CH_2), 7.20–8.10 (m, 8H, Ar–H), 7.69 (s, 1H, =CH ethylenic), 8.40 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 165.55$, 161.42, 159.60, 155.38, 154.28, 136.89, 133.48, 132.86, 130.39, 129.32, 128.68, 127.33, 125.65, 118.40, 116.30, 116.22, 114.47, 114.30, 109.10, 66.18, 62.12, 15.28 ppm; FAB-MS: $m/z =$ 409 (M + H).

2-Cyano-3-[4-[(7-chloro-1,2-dihydro-2-oxoquinolin-4-yl)-methoxy]phenyl]-2-propenoic acid ethyl ester
(**4c**, $C_{22}H_{17}ClN_2O_4$)
Yield: 82%; colorless crystals (acetic acid); m.p.: 299–300 °C; IR (KBr):$\bar{\nu} = 3{,}420$ (N–H stretching), 2,226 (C≡N stretching), 1,725 (C=O stretching, ester), 1,662 (C=O stretching, amide) cm^{-1}; ^1H NMR (300 MHz, DMSO-d_6): $\delta = 1.41–1.48$ (t, 3H, CH_3-ester), 4.45–4.52 (q, 2H, CH_2-ester), 5.50 (s, 2H, C4-CH_2), 6.68–8.08 (m, 8H, Ar–H), 8.20 (s, 1H, =CH ethylenic), 11.80 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): $\delta = 166.15$, 162.52, 159.60, 154.38, 154.18, 136.89, 133.48, 132.86, 130.39, 129.32, 128.68, 127.33, 125.65, 119.40, 117.30, 116.20, 114.47, 114.00, 109.10, 65.98, 62.32, 16.08 ppm; FAB-MS: $m/z = 409$ (M + H).

2-Cyano-3-[4-[(1,2-dihydro-8-methyl-2-oxoquinolin-4-yl)-methoxy]phenyl]-2-propenoic acid ethyl ester
(**4d**, $C_{23}H_{20}N_2O_4$)
Yield: 84%; colorless crystals (acetic acid); m.p.: 278–280 °C; IR (KBr):$\bar{\nu} = 3{,}434$ (N–H stretching), 2,228 (C≡N stretching), 1,728 (C=O stretching, ester), 1,659 (C=O stretching, amide) cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): $\delta = 1.42–1.48$ (t, 3H, CH_3-ester), 2.48 (s, 3H, C8-CH_3 of quinolinone), 4.40–4.48 (q, 2H, CH_2-ester), 5.52 (s, 2H, C4-CH_2), 7.10–8.02 (m, 8H, Ar–H), 7.72 (s, 1H, =CH ethylenic), 8.48 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): $\delta = 166.10$, 162.45, 158.60, 155.18, 154.28, 136.89, 133.48, 132.86, 130.34, 129.32, 128.68, 127.33, 125.65, 118.40, 116.35, 116.20, 114.40, 114.15, 108.10, 65.28, 60.82, 16.18 ppm; FAB-MS: $m/z = 390$ (M + H).

General procedure for the preparation of compounds ***5a–5d***

A mixture of substituted 4-[(4-formyl-2-methoxyphen oxy)methyl]quinolin-2(1*H*)-one **3a–3d** (4.0 mmol), ethyl cyanoactate (4.0 mmol), and a catalytic amount of piperidine in 20 cm^3 ethanol was stirred for 6 h at room temperature and left overnight. The resulting yellow liquid was poured onto crushed ice. The yellow solid obtained was filtered off, washed with excess of cold water, dried, and crystallized from a suitable solvent.

2-Cyano-3-[4-[(1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxyphenyl]-2-propenoic acid ethyl ester
(**5a**, $C_{23}H_{20}N_2O_5$)
Yield: 80%; colorless crystals (ethanol + dioxan); m.p.: 230–232 °C; IR (KBr):$\bar{\nu} = 3{,}431$ (N–H stretching), 2,209 (C≡N stretching), 1,715 (C=O stretching, ester), 1,661 (C=O stretching, amide) cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$ + CF_3COOH): $\delta = 1.40–1.45$ (t, 3H, CH_3-ester), 4.38–4.48 (q, 2H, CH_2-ester), 5.54 (s, 2H, C4-CH_2), 7.02–8.26 (m, 8H, Ar–H), 7.76 (s, 1H, =CH ethylenic), 12.45 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$ + CF_3COOH): $\delta = 168.56$, 164.40, 159.68, 157.34, 155.28, 138.89, 135.48, 133.86, 131.30, 129.32, 128.18, 126.40, 125.65, 120.40, 116.30, 116.21, 115.47, 114.30, 109.19, 66.18, 62.20, 55.08, 17.33 ppm; FAB-MS: $m/z = 405$ (M + H).

2-Cyano-3-[4-[(6-chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxyphenyl]-2-propenoic acid ethyl ester (**5b**, $C_{23}H_{19}ClN_2O_5$)
Yield: 78%; colorless crystals (acetic acid); m.p.: 240–242 °C; IR (KBr):$\bar{\nu} = 3{,}422$ (N–H stretching), 2,227 (C≡N stretching), 1,715 (C=O stretching, ester), 1,672 (C=O stretching, amide) cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$ + CF_3COOH): $\delta = 1.42–1.48$ (t, 3H, CH_3-ester), 4.36–4.45 (q, 2H, CH_2-ester), 3.78 (s, 3H, OCH_3), 5.50 (s, 2H, C4-CH_2), 7.02–8.10 (m, 7H, Ar–H), 7.78 (s, 1H, = CH ethylenic), 12.48 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$ + CF_3COOH): $\delta = 167.70$, 165.47, 158.65, 156.34, 154.28, 139.89, 134.48, 133.86, 130.30, 129.32, 128.18, 126.40, 125.55, 120.40, 117.30, 116.21, 115.47, 114.30, 108.11, 67.18, 61.20, 54.80, 18.13 ppm; FAB-MS: $m/z = 440$ (M + H).

2-Cyano-3-[4-[(7-chloro-1,2-dihydro-2-oxoquinolin-4-yl)-methoxy]-3-methoxyphenyl]-2-propenoic acid ethyl ester
(**5c**, $C_{23}H_{19}ClN_2O_5$)
Yield: 80%; colorless crystals (acetic acid); m.p.: 280–282 °C; IR (KBr):$\bar{\nu}$ = 3,428 (N–H stretching), 2,229 (C≡N stretching), 1,712 (C=O stretching, ester), 1,665 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.40–1.48 (t, 3H, CH_3-ester), 4.38–4.45 (q, 2H, CH_2-ester), 3.78 (s, 3H, OCH_3), 5.52 (s, 2H, C4-CH_2), 6.88–8.02 (m, 7H, Ar–H), 8.20 (s, 1H, =CH ethylenic), 11.78 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ = 168.78, 165.40, 157.69, 156.40, 154.30, 138.88, 134.48, 133.86, 130.30, 129.20, 128.20, 126.40, 125.55, 121.48, 118.33, 116.20, 115.47, 114.30, 107.70, 66.80, 62.40, 55.87, 17.33 ppm; FAB-MS: *m/z* = 440 (M + H).

2-Cyano-3-[4-[(1,2-dihydro-8-methyl-2-oxoquinolin-4-yl)-methoxy]-3-methoxyphenyl]-2-propenoic acid ethyl ester
(**5d**, $C_{24}H_{22}N_2O_5$)
Yield: 80%; colorless crystals (acetic acid); m.p.: 258–260 °C; IR (KBr):$\bar{\nu}$ = 3,434 (N–H stretching), 2,210 (C≡N stretching), 1,710 (C=O stretching, ester), 1,659 (C=O stretching, amide) cm^{-1};^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.42–1.49 (t, 3H, CH_3-ester), 2.48 (s, 3H, C8-CH_3 of quinolinone), 4.40–4.46 (q, 2H, CH_2-ester), 3.82 (s, 3H, OCH_3), 5.58 (s, 2H, C4-CH_2), 6.90–8.20 (m, 7H, Ar–H), 8.28 (s, 1H, =CH ethylenic), 11.80 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ = 167.79, 164.48, 158.61, 157.44, 155.36, 138.88, 135.48, 133.46, 130.20, 129.20, 128.45, 127.40, 124.55, 122.48, 118.33, 116.20, 115.47, 114.30, 108.60, 66.70, 63.40, 56.57, 20.30, 18.33 ppm; FAB-MS: *m/z* = 419 (M + H).

General procedure for the preparation of compounds ***6a–6d***

A mixture of substituted 4-[(4-formylphenoxy)methyl]quinolin-2(1*H*)-one **2a–2d** (4.0 mmol), ethyl acetoacetate (4.0 mmol), and a catalytic amount of piperidine in 20 cm^3 ethanol was stirred for 6 h at room temperature and left overnight. The resulting yellow liquid was poured to crushed ice. The yellow colored solid obtained was filtered off, washed with excess of cold water, dried, and crystallized from a suitable solvent.

2-[4-[(1,2-Dihydro-2-oxoquinolin-4-yl)methoxy]phenyl-methylene]-3-oxobutanoic acid ethyl ester
(**6a**, $C_{23}H_{21}NO_5$)
Yield: 80%; colorless crystals (acetic acid); m.p.: 288–290 °C; IR (KBr):$\bar{\nu}$ = 3,431 (N–H stretching), 1,720 (C=O stretching, ester), 1,680 (C=O stretching, ester), 1,658 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 1.45–1.48 (t, 3H, CH_3-ester), 2.45 (s, 3H, CH_3-acetyl), 4.41–4.50 (q, 2H, CH_2-ester), 5.65 (s, 2H, C4-CH_2), 7.20–8.10 (m, 9H, Ar–H), 7.76 (s, 1H, =CH ethylenic), 8.46 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 167.56, 163.40, 159.68, 157.34, 155.18, 138.89, 135.48, 132.86, 131.30, 129.32, 128.18, 126.40, 125.66, 120.40, 116.30, 116.20, 115.47, 114.30, 108.20, 67.18, 63.20, 24.48, 15.33 ppm; FAB-MS: *m/z* = 392 (M + H).

2-[4-[(6-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-phenylmethylene]-3-oxobutanoic acid ethyl ester
(**6b**, $C_{23}H_{20}ClNO_5$)
Yield: 76%; colorless crystals (acetic acid); m.p.: 230–232 °C; IR (KBr):$\bar{\nu}$ = 3,413 (N–H stretching), 1,722 (C=O stretching, ester), 1,692 (C=O stretching, ester), 1,660 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 1.45–1.50 (t, 3H, CH_3-ester), 2.48 (s, 3H, CH_3-acetyl), 4.43–4.50 (q, 2H, CH_2-ester), 5.38 (s, 2H, C4-CH_2), 7.18–8.15 (m, 8H, Ar–H), 7.80 (s, 1H, =CH ethylenic), 8.40 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 166.59, 162.70, 158.88, 156.44, 155.18, 138.89, 136.48, 133.88, 130.20, 129.32, 128.18, 126.40, 125.66, 120.40, 117.40, 116.20, 115.47, 114.30, 109.22, 66.88, 62.10, 23.58, 16.13 ppm; FAB-MS: *m/z* = 426 (M + H).

2-[4-[(7-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-phenylmethylene]-3-oxobutanoic acid ethyl ester
(**6c**, $C_{23}H_{20}ClNO_5$)
Yield: 72%; colorless crystals (acetic acid); m.p.: 220–222 °C; IR (KBr):$\bar{\nu}$ = 3,420 (N–H stretching), 1,725 (C=O stretching, ester), 1,678 (C=O stretching, ester), 1,662 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.38–1.47 (t, 3H, CH_3-ester), 2.50 (s, 3H, CH_3-acetyl), 4.40–4.48 (q, 2H, CH_2-ester), 5.52 (s, 2H, C4-CH_2), 6.68–7.98 (m, 8H, Ar–H), 8.10 (s, 1H, =CH ethylenic), 11.78 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ = 167.19, 163.40, 159.18, 156.44, 155.18, 137.89, 136.48, 135.88, 131.25, 129.32, 128.38, 126.40, 125.66, 122.40, 117.40, 116.20, 115.47, 114.30, 107.20, 67.58, 63.15, 24.58, 15.53 ppm; FAB-MS: *m/z* = 426 (M + H).

2-[4-[(1,2-Dihydro-8-methyl-2-oxoquinolin-4-yl)methoxy]-phenylmethylene]-3-oxobutanoic acid ethyl ester
(**6d**, $C_{24}H_{23}NO_5$)
Yield: 78%; colorless crystals (acetic acid); m.p.: 260–262 °C; IR (KBr):$\bar{\nu}$ = 3,434 (N–H stretching), 1,724 (C=O stretching, ester), 1,688 (C=O stretching, ester), 1,655 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 1.45–1.49 (t, 3H, CH_3-ester), 2.45 (s, 3H, CH_3-acetyl), 2.52 (s, 3H, C8-CH_3 of quinolinone),

4.42–4.50 (q, 2H, CH_2-ester), 5.54 (s, 2H, C4-CH_2), 7.10–8.10 (m, 8H, Ar–H), 7.78 (s, 1H, =CH ethylenic), 8.48 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, $CDCl_3$): δ = 166.19, 163.50, 157.48, 155.14, 154.10, 138.89, 136.48, 133.88, 131.20, 129.32, 128.18, 126.40, 125.66, 121.40, 118.40, 116.20, 115.17, 114.30, 108.20, 67.18, 63.60, 25.18, 20.12, 15.17 ppm; FAB-MS: m/z = 406 (M + H).

General procedure for the preparation of compounds ***7a–7d***

A mixture of substituted 4-[(4-formyl-2-methoxyphenoxy)-methyl]quinolin-2(1*H*)-one **3a–3d** (4.0 mmol), ethyl aceto-acetate (4.0 mmol), and a catalytic amount of piperidine in 20 cm^3 ethanol was stirred for 6 h at room temperature and left overnight. The resulting yellow liquid was poured onto crushed ice. The yellow solid obtained was filtered off, washed with excess of cold water, dried, and crystallized from a suitable solvent.

2-[[4-[(1,2-Dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxyphenyl]methylene]-3-oxobutanoic acid ethyl ester (**7a**, $C_{24}H_{23}NO_6$)

Yield: 83%; colorless crystals (ethanol + dioxan); m.p.: 192–194 °C; IR (KBr):$\bar{\nu}$ = 3,430 (N–H stretching), 1,724 (C=O stretching, ester), 1,660 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.40–1.48 (t, 3H, CH_3-ester), 2.42 (s, 3H, CH_3-acetyl), 4.40–4.47 (q, 2H, CH_2-ester), 3.78 (s, 3H, OCH_3), 5.50 (s, 2H, C4-CH_2), 6.69–7.90 (m, 8H, Ar–H), 8.15 (s, 1H, =CH ethylenic), 11.78 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ = 168.56, 164.40, 158.68, 157.34, 155.18, 138.89, 135.48, 133.86, 131.30, 129.32, 128.18, 126.40, 125.66, 120.40, 116.30, 116.20, 115.47, 114.30, 109.20, 67.18, 62.20, 54.28, 24.48, 18.33 ppm; FAB-MS: m/z = 422 (M + H).

2-[[4-[(6-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxyphenyl]methylene]-3-oxobutanoic acid ethyl ester (**7b**, $C_{24}H_{22}ClNO_6$)

Yield: 78%; colorless crystals (acetic acid); m.p.: 240–242 °C; IR (KBr):$\bar{\nu}$ = 3,434 (N–H stretching), 1,720 (C=O stretching, ester), 1,680 (C=O stretching, ester), 1,658 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.42–1.50 (t, 3H, CH_3-ester), 2.48 (s, 3H, CH_3-acetyl), 4.40–4.48 (q, 2H, CH_2-ester), 3.80 (s, 3H, OCH_3), 5.48 (s, 2H, C4-CH_2), 6.80–8.00 (m, 7H, Ar–H), 8.08 (s, 1H, =CH ethylenic), 11.65 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ = 168.60, 164.44, 158.61, 157.34, 156.28, 138.89, 135.48, 133.86, 131.30, 129.22, 128.18, 127.10, 125.66, 120.40, 116.50, 116.20, 115.47, 114.30, 108.20, 68.10, 63.30, 55.18, 25.18, 18.30 ppm; FAB-MS: m/z = 456 (M + H).

2-[[4-[(7-Chloro-1,2-dihydro-2-oxoquinolin-4-yl)methoxy]-3-methoxyphenyl]methylene]-3-oxobutanoic acid ethyl ester (**7c**, $C_{24}H_{22}ClNO_6$)

Yield: 80%; colorless crystals (acetic acid); m.p.: 277–279 °C; IR (KBr):$\bar{\nu}$ = 3,433 (N–H stretching), 1,724 (C=O stretching, ester), 1,690 (C=O stretching, ester), 1,663 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.38–1.42 (t, 3H, CH_3-ester), 2.45 (s, 3H, CH_3-acetyl), 4.40–4.48 (q, 2H, CH_2-ester), 3.88 (s, 3H, OCH_3), 5.68 (s, 2H, C4-CH_2), 6.78–8.03 (m, 7H, Ar–H), 8.10 (s, 1H, =CH ethylenic), 11.81 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ = 167.90, 163.84, 158.81, 157.74, 156.78, 139.19, 135.48, 133.86, 130.30, 129.22, 128.18, 127.10, 125.66, 120.40, 117.50, 116.80, 115.47, 114.30, 109.20, 67.10, 62.30, 54.18, 24.18, 17.40 ppm; FAB-MS: m/z = 456 (M + H).

2-[[4-[(1,2-Dihydro-8-methyl-2-oxoquinolin-4-yl)methoxy]-3-methoxyphenyl]methylene]-3-oxobutanoic acid ethyl ester (**7d**, $C_{25}H_{25}NO_6$)

Yield: 82%; colorless crystals (acetic acid); m.p.: 250–252 °C; IR (KBr): $\bar{\nu}$ = 3,442 (N–H stretching), 1,722 (C=O stretching, ester), 1,682 (C=O stretching, ester), 1,662 (C=O stretching, amide) cm^{-1}; ^{1}H NMR (300 MHz, DMSO-d_6): δ = 1.41–1.50 (t, 3H, CH_3-ester), 2.40 (s, 3H, CH_3-acetyl), 2.51 (s, 3H, C8-CH_3 of quinolinone), 4.40–4.48 (q, 2H, CH_2-ester), 3.84 (s, 3H, OCH_3), 5.52 (s, 2H, C4-CH_2), 6.75–8.10 (m, 7H, Ar–H), 8.15 (s, 1H, =CH ethylenic), 11.72 (s, 1H, NH) ppm; ^{13}C NMR (75 MHz, DMSO-d_6): δ = 168.10, 162.14, 158.81, 157.74, 156.78, 139.19, 135.44, 133.66, 130.30, 129.22, 128.18, 127.10, 125.66, 120.40, 118.50, 116.80, 115.47, 114.30, 108.27, 68.11, 63.44, 54.18, 24.10, 20.43, 18.30 ppm; FAB-MS: m/z = 436 (M + H).

Antimicrobial assay

The in vitro antimicrobial activity of the test compounds was examined against the two bacterial microorganisms *E. coli* (gram negative) and *B. cirrhosis* (gram positive) and the two fungal microorganisms *A. niger* and *R. bataticola*. DMF was used as a solvent control, and the reference drugs used were norfloxacin and griseofulvin. The tests were carried out by the cup-plate method [16–18], at a concentration of 100, 50, and 25 μg cm^{-3}. The zone of inhibition was measured in millimeters after 48 h of incubation at 37 °C. The percentage inhibition of test compounds was calculated by relating the zone of inhibition of the test compound ($ZOI_{test\ compound}$) to those of the standard ($ZOI_{standard}$, taken as 100%) and control ($ZOI_{control}$) as follows: % inhibition = ($ZOI_{test\ compound} - ZOI_{control}$)/($ZOI_{standard} - ZOI_{control}$) × 100%.

Acute toxicity

Groups of six albino mice weighing 20–25 g were fasted overnight and treated per orally and i.p. with the test compounds [19]. The dose was varied from 1,000 to 100 mg kg^{-1} body weights. The animals were observed for 24 h for any signs of acute toxicity such as increased or decreased motor activity, tremors, convulsion, sedation, lacrimation, etc. No mortality of the animals was observed even after 24 h. Hence the LD_{50} cutoff value of the test compounds was fixed as 1,000 mg kg^{-1}, so that 100 mg kg^{-1}, i.e., 1/10 of the cutoff value, was taken as the screening dose for the evaluation of anti-inflammatory activity.

Analgesic activity

The analgesic activity of the test compounds was carried out in vivo by using an abdominal constriction test induced by 0.6% acetic acid (0.1 cm^3/10 g) in mice [20]. Albino mice of both sexes (18–22 g) were used. Compounds were administered orally (10 mg kg^{-1}) as a suspension in 5% carbethoxymethyl cellulose (vehicle). Indomethacin (10 mg kg^{-1}) was used as the standard drug under the same conditions.

Anti-inflammatory activity

The anti-inflammatory activity of the test compounds was assessed by the formalin-induced rat paw edema inhibition method according to Winter et al. [21], by employing 3.5% of formalin as the phlogistic agent. All test compounds were administered orally as suspensions in 2% CMC, 30 min before the injection of the phlogistic agent, at a dose of 100 mg kg^{-1} body weight. Indomethacin was used as a standard at a dose of 10 mg kg^{-1} body weight. A group of six Sprague Dawley rats of either sex were used in each experiment. Plain CMC (2%) served as control. The paw edema volume was measured with the help of a plethysmograph by the mercury displacement method at 0 h (immediately after injection of formalin), 1, 2, 3, 4, and 5 h.

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