Full Paper

Synthesis of New 2,3-Dihydroquinazolin-4(1*H*)-one Derivatives for Analgesic and Anti-inflammatory Evaluation

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Starting from isatoic anhydrides, several new 2,3-dihydroquinazolin-4(1*H*)-one derivatives bearing chalcone or pyrazole or thiazole moieties at the third position were synthesized. The analgesic and anti-inflammatory activities for most compounds were studied at a dose level of 50 mg/kg via the acetic-acid-induced writhing-response method and carrageenan-induced edema method, respectively. The study showed that the chalcones bearing a 4-chlorophenyl group **4c** or 4-nitrophenyl group **4b** were the most active ones as analgesics. Both chalcone **4c** and *N*-phenyl pyrazole bearing 4-methoxy phenyl group **5b** showed a higher anti-inflammatory activity than celecoxib but still lower than that of diclofenac sodium. Moreover, the chalcone **4c** has nearly the same ulcerogenic index as the selective cyclooxygenase-2 inhibitor celecoxib.

Keywords: Analgesic activities / Anti-inflammatory activities / Dihydroquinazolinone / Synthesis

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Introduction

Quinazolinone derivatives have been found to possess a wide spectrum of activities like antibacterial [1], antifungal [2, 3], anticonvulsant [4, 5] antitumor [6, 7], and they are also H_3 receptor inverse agonists [8].

Some 4(3*H*)-quinazolinone derivatives were reported to have analgesic and anti-inflammatory activities [9] like the well known drug diproqualone which is used primarily for the treatment of inflammatory pain associated with osteoarthritis [10]. It has been reported that substitution of different heterocyclic moieties at the 2- or 3-position of the quinazolinone nucleus modulates the anti-inflammatory activity [11].

Certain chalcones [12–14] and thiazole [15] derivatives were reported to possess analgesic and anti-inflammatory activities. Among the chalcones, 2,4-dichloro-4′-*N*-[*N*′-(4″-methylphenylsulphenyl)urenyl]chalcone (Me-UCH9; Fig. 1) was found to exert anti-inflammatory action [12] through dual inhibition of cyclooxygenase-2 (COX-2) and 5-lipoxygenase activities.

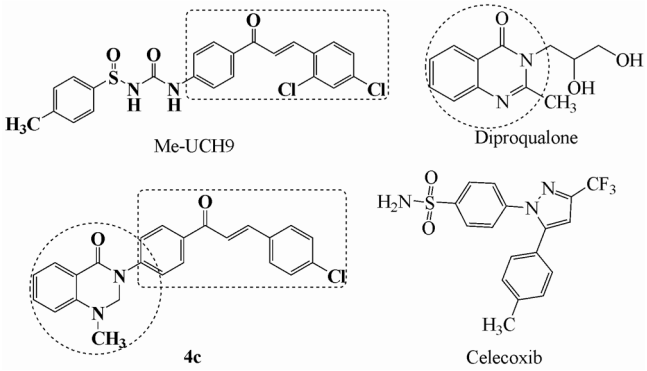

Figure 1. Structures of Me-UCH9, diproqualone, **4c**, and celecoxib.

Moreover, many drugs having a pyrazole moiety are used clinically as analgesic and anti-inflammatory agents such as the selective cyclooxygenase-2 (COX-2) inhibitor celecoxib (Fig. 1) [16].

The use of celecoxib for the treatment of inflammation and pain avoided many side effects of the nonsteroidal anti-inflammatory drugs because of its selectivity as COX-2 inhibitor [17, 18].

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This prompted us to synthesize a new series of 2,3-dihydroquinazolin-4(1*H*)-one derivatives by incorporation of chalcone or thiazole or pyrazole moieties at position 3 of the quinazolinone nucleus which may augment each other in order to produce potent analgesic and anti-inflammatory agents hoping to have lower or no ulcerogenic activity.

Results and discussion

Chemistry

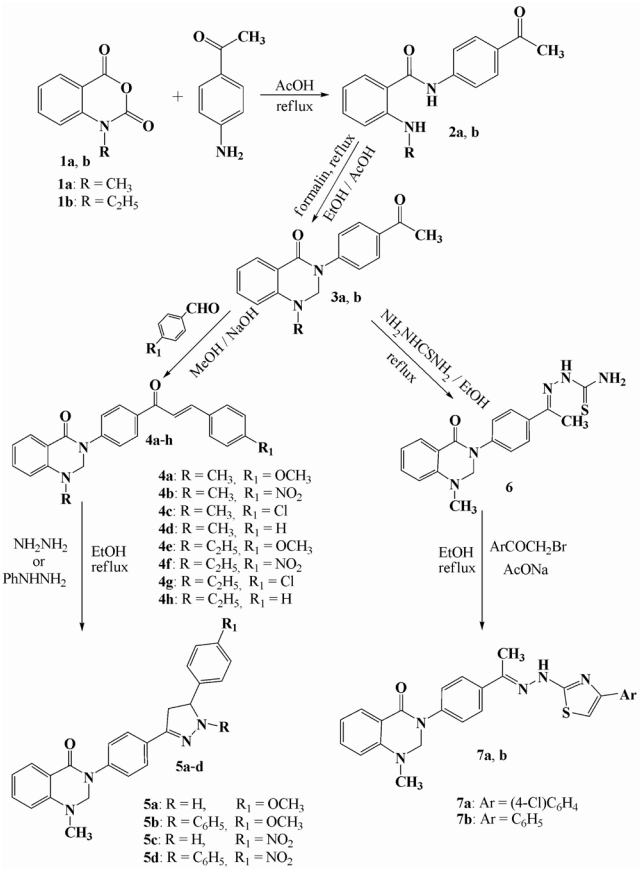
The reaction sequences employed for the synthesis of the target 2,3-dihydroquinazolin-4(1*H*)-one derivatives **3–7** are illustrated in Scheme 1.

N-alkyl isatoic anhydrides **1a**, **b** were utilized for the synthesis *N*-(4-acetylphenyl)-2-(alkylamino)benzamide **2a**, **b** via their reaction with *p*-aminoacetophenone in glacial acetic acid. The structure of the novel intermediate **2b** was confirmed using different spectroscopic methods. ^{1}H-NMR revealed the presence of a very characteristic singlet at δ = 2.573 ppm due to methyl group of the acetyl group, in addition to the appearance of two singlet peaks for two NH groups; one of them appeared with the aromatic protons while the other appeared at δ = 10.357 ppm.

In the present work, the key dihydroquinazolinones **3a**, **b** were prepared by cyclization of the benzamide derivatives **2a**, **b** under the effect of formalin through an internal Mannich reaction by heating the reactants at reflux in ethanol containing catalytic amounts of acetic acid. The structure of the novel dihydroquinazolinone **3b** was established using IR and ^{1}H-NMR. The IR spectrum revealed the absence of the absorption bands at ν = 3399 and 3332 cm^{-1} due to NH groups for the starting benzamide intermediate **2b**. Moreover, ^{1}H-NMR showed a characteristic singlet peak at δ = 5.044 ppm for the methylene protons at the 2-position indicating the formation of a quinazoline ring.

The target chalcones 3-(4-(3-(4-substituted phenyl)acryloyl)phenyl)-1-alkyl-2,3-dihydroquinazolin-4(1*H*)-ones **4a–h** were synthesized through a Claisen–Schmidt alkaline condensation of dihydroquinazolinones **3a**, **b** with aromatic aldehyde by stirring the reactants in aqueous methanol containing NaOH solution (50%) at room temperature.

The structure of the novel chalcones **4a–h** were characterized using ^{1}H-NMR spectra which revealed the disappearance of the singlet peak at δ = 2.596 ppm for the methyl group of the acetyl group of starting quinazolinone meanwhile the appearance of a multiplet peak characteristic for the acryloyl protons appeared with the aromatic protons in case of *N*-methyl derivatives **4a–d** while that of *N*-ethyl derivatives **4e–h** appeared at δ = 6.90–7.07 ppm.



Scheme 1. Synthetic pathway for compounds **3–7**.

In the present work, NH-pyrazoles **5a**, **c** and their *N*-phenyl analogs **5b**, **d** were prepared through cyclization of chalcones **4a**, **b** with hydrazine hydrate or phenylhydrazine via heating the reactants at reflux in ethanol.

The structures of the novel NH-pyrazole derivatives **5a**, **c** were confirmed using IR and ^{1}H-NMR. IR measurements revealed the appearance of strong absorption band at ν = 3330 cm^{-1} for the NH group of the NH-pyrazoles **5a**, **c**. Moreover, the ^{1}H-NMR spectrum confirmed the presence of this NH group through the appearance of a broad singlet peak at δ = 10.20–13.50 ppm while their *N*-phenyl derivatives **5b**, **d** did not show such a broad singlet indicating the absence of the NH group.

The formation of the pyrazoles **5a–d** in general was confirmed by the appearance of two multiplet peaks in the range of δ = 2.70–4.10 ppm for the two protons at the 4-position of the pyrazole ring as well as another multiplet peak at the range of δ = 4.78–5.60 ppm due to the proton at the 5-position of the pyrazole ring.

Table 1. Analgesic activity evaluation of the test compounds (**3a**, **4a–d**, **5a–d**) and celecoxib administered in a dose of 50 mg/kg using acetic-acid-induced writhing assay in mice.

Comp. No.	Writhing					Total	% Reduction
	Time interval (min)						
	0–5	5–10	10–15	15–20	20–25		
Cont.	6.2 ± 0.58	31.4 ± 1.81	29.8 ± 2.67	18.2 ± 1.07	14.6 ± 1.03	100.2 ± 2.62	0
Cel.	35.8 ± 0.37§	9.4 ± 1.21§	13.4 ± 0.68§	7 ± 0.32§	5.8 ± 0.8§	35.8 ± 2.85§	64.3
3a	12.6 ± 0.51$^{\S, \#}$	18.6 ± 0.5$^{\S, \#}$	13 ± 0.71$^{\S, \#}$	12 ± 0.71$^{\S, \#}$	8.8 ± 0.86§	64.8 ± 1.77$^{\S, \#}$	35.3$^{\#}$
4a	5.4 ± 1.47$^{\#}$	19.2 ± 2.4$^{\S, \#}$	16.8 ± 1.46$^{\S, \#}$	9.6 ± 1.46§	10.8 ± 2.71$^{\S, \#}$	61.8 ± 7.08$^{\S, \#}$	38.3$^{\#}$
4b	1.2 ± 0.2§	8.4 ± 1.03§	6.4 ± 0.51§	4.2 ± 0.51§	2.4 ± 0.4$^{\S, \#}$	22.6 ± 1.21$^{\S, \#}$	77.4$^{\#}$
4c	1.2 ± 0.37§	3.8 ± 1.02$^{\S, \#}$	2.6 ± 0.4$^{\S, \#}$	1.8 ± 0.4$^{\S, \#}$	1.4 ± 0.4$^{\S, \#}$	10.8 ± 1.74$^{\S, \#}$	89.2$^{\#}$
4d	2.6 ± 0.24§	12.4 ± 1.57§	6.8 ± 1.46§	4.6 ± 1.46§	3.8 ± 1.16§	30.2 ± 5.13§	70
5a	4.6 ± 0.51$^{\#}$	12 ± 0.71§	7.2 ± 0.58§	5.4 ± 0.58§	4 ± 0.45§	33.2 ± 2.5§	67
5b	10.6 ± 1.5$^{\S, \#}$	15 ± 0.43$^{\S, \#}$	11.2 ± 0.37$^{\S, \#}$	8.6 ± 0.37§	8.2 ± 0.37§	53.6 ± 1.36$^{\S, \#}$	46.5$^{\#}$
5c	4.8 ± 1.68$^{\#}$	16.8 ± 0.37$^{\S, \#}$	9.2 ± 0.66$^{\S, \#}$	7.4 ± 0.66§	6.8 ± 0.66§	45 ± 1.38§	55.1
5d	3.2 ± 0.37§	13.4 ± 1.78$^{\S, \#}$	9.2 ± 1.02$^{\S, \#}$	6.6 ± 1.02§	4.8 ± 0.49§	39.2 ± 3.85§	61

Cont. = Control; Cel. = Celecoxib; § $p < 0.05$, compared with the mean value of the vehicle treated group; # $p < 0.05$, compared with the mean value of celecoxib group.

Furthermore, the novel hydrazinecarbothioamide **6** was synthesized via condensation of thiosemicarbazide and the intermediate **3a** by heating the reactants at reflux in ethanol containing a catalytic amount of acetic acid for 2 h. The structure of thioamide **6** was established using IR spectroscopy; it showed the appearance of strong absorption band at ν = 3407, 3270, 3198 cm^{-1} for NH_2 and NH groups as well as ^{1}H-NMR confirmed the presence of such groups through appearance of the corresponding two singlets at δ = 8.34 and 10.216 ppm.

Cyclization of compound **6** was conducted by means of phenacyl bromide derivatives [19] by heating the reactants together at reflux in ethanol in the presence of sodium acetate for 15 h to afford the novel thiazole derivatives **7a**, **b**. Their structures were established using IR and ^{1}H-NMR spectra. The IR showed the disappearance of absorption bands (ν = 3407, 3270 cm^{-1}) and ^{1}H-NMR also revealed the absence of the singlet peak at δ = 10.216 ppm due to the amino group for the starting thioamide **6**.

All the target compounds were characterized by using thin layer chromatography and melting point techniques. Both analytical and spectral data of all compounds are in full agreement with the proposed structures.

Pharmacology

Compounds **3a**, **4a–d**, and **5a–d** were tested for their analgesic activity using the acetic-acid-induced writhing response method [20] in mice in comparison with celecoxib as a reference drug. Moreover, the anti-inflammatory activity for these compounds was determined via carrageenan-induced edema method [21] using celecoxib and diclofenac sodium as reference drugs. Furthermore, the ulcerogenic activity [22] for the compounds was evaluated using celecoxib and indomethacin as reference drugs.

Analgesic activity

The results are listed in Table 1; it was observed that: A) The ranking order of potency for the compounds was: **4c** > **4b** > **4d** > **5a** > celecoxib > **5d**. B) The incorporation of an electron donating group like methoxy group as seen in compound **4a** into the *p*-position of a phenyl group of the parent chalcone **4d** led to a decrease of the analgesic activity. C) In contrast, the chalcone-bearing electron-withdrawing group as observed in compounds **4c** and **4b** (Table 1) showed higher analgesic activity than those containing an electron-donating group **4a** and celecoxib as well. D) The chalcone **4a** bearing the 4-methoxyphenyl group was the least active one as an analgesic but its cyclization to the pyrazole derivatives **5a** and **5b** led to an increase in analgesic activity (Fig. 2). E) The parent compound **3a**; which is chemically a quinazolinone derivative; decreased the abdominal constrictions by 35.3% only, while combining it with other moieties such as chalcones **4a–d** or pyrazoles **5a–d** led to an enhancement of analgesic activity reaching its maximal level (about 89%) in case of the quinazolinone derivative bearing the chalcone moiety **4c** (Fig. 2).

Anti-inflammatory activity

The results which can be drawn from Table 2 are: A) The order of anti-inflammatory activity for the tested compounds was: diclofenac > **4c** > **5b** > celecoxib > **5d**. B) The

Table 2. Effect of the test compounds **3a**, **4a–d**, **5a–d** at a dose of 50 mg/kg on rat hind-paw thickness at different time intervals after induction of edema using carrageenan.

Comp. No.	Edema thickness (mm)							TI%
	Time (h)							
	1	2	3	4	5	6	24	
Cont.	1.76 ± 0.14	2.9 ± 0.24	3.4 ± 0.19	3 ± 0.25	2.6 ± 0.12	2.2 ± 0.11	0.78 ± 0.051	0
Cel.	1.32 ± 0.15$^{§,\ \&}$	2.02 ± 0.11$^{§,\ \&}$	1.55 ± 0.0709§	1.34 ± 0.0357$^{§,\ \&}$	0.59 ± 0.031$^{§,\ \&}$	1.16 ± 0.0729$^{§,\ \&}$	0.83 ± 0.15$^{\&}$	46.4
Diclo.	1 ± 0.0296§	1.39 ± 0.0206§	1.05 ± 0.10§	0.89 ± 0.0818§	1.16 ± 0.0447§	0.59 ± 0.0666§	0.23 ± 0.0503§	72.3
3a	1.49 ± 0.0386$^{§,\ \&}$	1.88 ± 0.11§	1.05 ± 0.10§	1.78 ± 0.11$^{§,\ \#,\ \&}$	1.37 ± 0.12$^{§,\ \#,\ \&}$	1.37 ± 0.0724$^{§,\ \#,\ \&}$	0.82 ± 0.063$^{\&}$	35.3
4a	1.48 ± 0.0247$^{§,\ \&}$	2.51 ± 0.0307$^{§,\ \#,\ \&}$	2.54 ± 0.0688$^{§,\ \#,\ \&}$	2.15 ± 0.0952$^{§,\ \#,\ \&}$	1.82 ± 0.0777$^{§,\ \#,\ \&}$	1.82 ± 0.05.31$^{§,\ \#,\ \&}$	0.87 ± 0.0691$^{\&}$	14.6
4b	1.32 ± 0.0457$^{§,\ \&}$	1.98 ± 0.1$^{§,\ \&}$	1.81 ± 0.0832$^{§,\ \#,\ \&}$	1.71 ± 0.0826$^{§,\ \#,\ \&}$	1.29 ± 0.0868$^{§,\ \#,\ \&}$	1.29 ± 0.0543$^{§,\ \&}$	1.0 ± 0.0301$^{§,\ \&}$	34.7
4c	1.03 ± 0.0428$^{§,\ \#}$	1.66 ± 0.0398$^{§,\ \#}$	1.24 ± 0.0242$^{§,\ \#}$	1.06 ± 0.0721$^{§,\ \#}$	0.83 ± 0.0238$^{§,\ \#,\ \&}$	0.83 ± 0.0299$^{§,\ \#,\ \&}$	0.44 ± 0.0442$^{§,\ \#,\ \&}$	61.1
4d	1.51 ± 0.0423$^{§,\ \&}$	2.11 ± 0.0613$^{§,\ \&}$	2.09 ± 0.0737$^{§,\ \#,\ \&}$	1.91 ± 0.0598$^{§,\ \#,\ \&}$	1.41 ± 0.0343$^{§,\ \#,\ \&}$	1.41 ± 0.0218$^{§,\ \#,\ \&}$	1.02 ± 0.0711$^{§,\ \&}$	29.1
5a	1.47 ± 0.0452$^{§,\ \&}$	2.28 ± 0.0586$^{§,\ \&}$	2.27 ± 0.0176$^{§,\ \#,\ \&}$	2.12 ± 0.0347$^{§,\ \#,\ \&}$	1.72 ± 02.92$^{§,\ \#,\ \&}$	1.72 ± 0.053$^{§,\ \#,\ \&}$	1.34 ± 0.0486$^{§,\ \#,\ \&}$	16.9
5b	1.13 ± 0.0475$^{§,\ \#}$	1.55 ± 0.0337$^{§,\ \#}$	1.76 ± 0.0386$^{§,\ \&}$	1.49 ± 0.0746$^{§,\ \&}$	1.04 ± 0.0532$^{§,\ \&}$	1.04 ± 0.0466$^{§,\ \&}$	0.6 ± 0.0186$^{\#,\ \&}$	51.5
5c	1.26 ± 0.0141$^{§,\ \&}$	2.50 ± 0.0663$^{§,\ \#,\ \&}$	2.67 ± 0.0717$^{§,\ \#,\ \&}$	2.47 ± 0.0656$^{§,\ \#,\ \&}$	1.81 ± 0.0811$^{§,\ \#,\ \&}$	1.81 ± 0.0706$^{§,\ \#,\ \&}$	1.27 ± 0.0808$^{§,\ \#,\ \&}$	13.8
5d	1.1 ± 0.0223$^{§,\ \#}$	1.54 ± 0.0218$^{§,\ \#}$	1.64 ± 0.0622$^{§,\ \&}$	1.44 ± 0.0287$^{§,\ \&}$	1.22 ± 0.0216$^{§,\ \&}$	1.22 ± 0.0336$^{§,\ \&}$	1.11 ± 0.041$^{§,\ \#,\ \&}$	37.3

Cont. = Control; Cel. = Celecoxib; Diclo. = Diclofenac sodium; TI% = Total inhibition%; § $p < 0.05$ significantly different from control; # $p < 0.05$ significantly different from celecoxib; & $p < 0.05$ significantly different from diclofenac sodium.

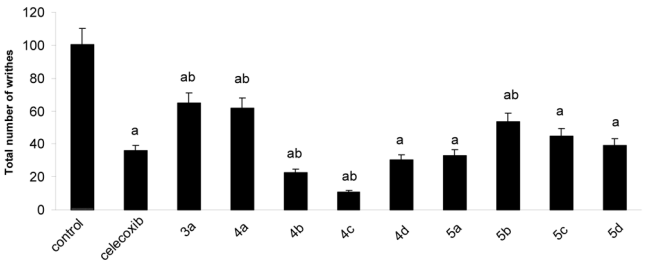


Figure 2. Evaluation of the analgesic activity of the test compounds **3a**, **4a–d**, **5a–d**, and celecoxib at a dose level of 50 mg/kg on acetic-acid-induced constrictions in mice. (a) $p < 0.05$, compared with the mean value of the vehicle-treated group; (b) $p < 0.05$, compared with the mean value of celecoxib group.

chalcone **4a** bearing a 4-methoxyphenyl group showed lower activity than the parent chalcone **4d**. C) The chalcones bearing electron-withdrawing groups (**4b** and **4c**) showed higher anti-inflammatory activity than those containing electron-donating group as seen in compound **4a**. D) The quinazolinone bearing *N*-phenyl pyrazoles (**5b** and **5d**) showed higher anti-inflammatory activity than their corresponding NH-pyrazole analogs (**5a** and **5c**).

Ulcerogenic activity

The ulcerogenic activity for the tested compounds **3a**, **4a–d**, and **5a–d** were determined using celecoxib and indomethacin as reference drugs. It was noted that compounds **3a**, **4a**, **b**, **d**, and **5a**, **c**, **d** showed a zero-ulcer score but the chalcone bearing the 4-chlorophenyl group **4c** and *N*-phenyl pyrazole bearing the 4-methoxyphenyl moiety **5b** exhibited a very slight ulcerogenic activity which is still smaller than that of the reference drug indomethacin (Table 3). It was also observed that the chalcone **4c** has nearly the same ulcerogenic activity as the selective COX-2 inhibitor celecoxib.

Table 3. Ulcerogenic activity of compounds **4c**, **5b**, celecoxib and indomethacin ($n = 5$).

Compounds	Ulcer index
Indomethacin	28.6 ± 0.51
Celecoxib	2.60 ± 0.24§
4c	2.67 ± 0.21$^{§,\ \#}$
5b	5.40 ± 0.4§

§ $p < 0.05$ compared to indomethacin; # $p < 0.05$ compared to celecoxib.

Conclusion

New 2,3-dihydroquinazolin-4(1*H*)-ones combined with either chalcone, pyrazole, or thiazole moieties were synthesized. The analgesic and anti-inflammatory evaluation for most 2,3-dihydroquinazolin-4(1*H*)-ones revealed that the chalcone bearing the 4-chlorophenyl group **4c** was the most active one not only as analgesic but also as anti-inflammatory agent; it possesses nearly the same ulcerogenic index as the selective COX-2 inhibitor celecoxib.

Experimental

Chemistry

Melting points were determined with a Gallenkamp melting point apparatus (Gallencamp, London, UK) and are uncorrected. Infrared (IR) spectra (KBr, cm^{-1}) were recorded on a Bruker Vector, 22FT-IR (Germany) and ^{1}H-NMR spectra were recorded on Varian Mercury-300 (300 MHz) and Varian Gemini 200 MHz spec-

trometers (Varian, Palo Alto, CA, USA) using dimethyl sulfoxide (DMSO)-d_6 as a solvent and tetramethylsilane (TMS) as an internal standard (Chemical shift in δ, ppm). Electron impact mass spectra were determined using a GC/MS Shimadzu QP1000EX (Shimadzu Corporation, Tokyo, Japan) with ionization energy 70 eV. Elemental analyses were determined using the Heraeus (Hanau, Germany) CHNS analyzer and Automatic Elemental Analyser CHN Model 2400 Perkin Elmer (USA) at the Microanalytical Center, Faculty of Science, University of Cairo, Egypt. All the results of the elemental analyses were in an acceptable error range. Thin layer chromatography (TLC) was performed on silica gel G for TLC (Merck, Germany) and spots were visualized by iodine vapors or by irradiation with ultraviolet light (UV; 254 nm). All chemicals were purchased from Sigma Chemical (St. Louis, MO, USA). Intermediates *N*-(4-acetylphenyl)-2-(methylamino)benzamide **2a** and 3-(4-acetylphenyl)-1-methyl-2,3-dihydroquinazolin-4(1H)-one **3a** were prepared according to the reported procedures [23, 24].

*N-(4-Acetylphenyl)-2-(ethylamino)benzamide **2b***

A mixture of *N*-substituted isatoic anhydride **1b** (0.05 mol) and *p*-aminoacetophenone (6.75 g, 0.05 mol) in glacial acetic acid was heated under reflux for 4 h. After cooling, the reaction mixture was poured into cold water (100 mL) and the separated solid was filtered, dried and crystallized from ethanol/H_2O to give the titled compound. Yield: 95%; m.p.: 133°C; IR ν: 3399, 3332 (NH), 3079 (CH, aromatic), 2964 (CH, aliphatic), 1672, 1637 (CO), 1516 (C=C) cm^{-1}; ^{1}H-NMR(200 MHz) δ: 1.203–1.276 (t, 3H, CH_3CH_2), 2.573 (s, 3H, CH_3CO), 3.135–3.241 (q, 2H, CH_2CH_3), 6.639–8.004 (m, 9H, ArH + NH), 10.357 (br s, 1H, NH, exch.) ppm. Anal. calcd. for $C_{17}H_{18}N_2O_2$ (282.14): C, 72.32; H, 6.43; N, 9.92. Found: C, 71.99; H, 6.08; N, 9.77.

*3-(4-Acetylphenyl)-1-ethyl-2,3-dihydroquinazolin-4(1H)-one **3b***

To a solution of the benzamide **2b** (0.01 mol) in ethanol (50 mL), formalin (40%, 1 mL) and few drops of glacial acetic acid were added. The reaction mixture was heated under reflux for 4 h and then concentrated to the half of its volume. After cooling, the separated solid was filtered and crystallized from ethanol. Yield: 86.4%; m.p.: 131–132°C; IR ν: 3041 (CH, aromatic), 2965 (CH, aliphatic), 1664, 1599 (CO), 1500 (C=C) cm^{-1};^{1}H-NMR (200 MHz) δ: 1.107–1.174 (t, 3H, CH_3), 2.596 (s, 3H, CH_3CO), 3.326–3.530 (q, 2H, CH_2CH_3), 5.044 (s, 2H, CH_2N), 6.850–8.023 (m, 8H, ArH) ppm; MS *m*/*z* (rel. int.): 296 [M^+ + 2] (8.3), 295 [M^+ +1] (16.8), 294 [M^+] (62), 293 (100), 119 (30.8), 77 (26.3). Anal. calcd. for $C_{18}H_{18}N_2O_2$ (294.35): C, 73.45; H, 6.16; N, 9.52. Found: C, 73.77; H, 6.02; N, 9.84.

*General procedure for preparation of compounds **4a–h***

To a well stirred solution of compounds **3a**, **b** (0.01 mol) and the appropriate aromatic aldehyde (0.01 mol) in methanol (30 mL), aqueous NaOH solution (50%, 6 mL) was added. The reaction mixture was stirred at room temperature for 48 h. The separated pale yellow solid was filtered, washed with water and then crystallized from suitable solvent.

*3-[4-(3-(4-Methoxyphenyl)acryloyl)phenyl]-1-methyl-2,3-dihydroquinazolin-4 (1H)-one **4a***

Yield: 75%; m.p.: 182–183°C; crystallized from dioxane; IR ν: 3070 (CH, aromatic), 2962 (CH, aliphatic), 1656, 1594 (CO), 1504 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 2.969 (s, 3H, NCH_3), 3.835 (s, 3H, OCH_3), 5.001 (s, 2H, CH_2N), 6.949–8.235 (m, 14H, ArH + acryloyl H) ppm; MS *m*/*z* (rel. int.): 398 [M^+] (46.3), 397 (60), 237 (12.5), 198 (11.3), 161 (11.3), 104 (100). Anal. calcd. for $C_{25}H_{22}N_2O_3$ (398.45): C, 75.36; H, 5.57; N, 7.03. Found: C, 75.60; H, 5.86; N, 7.14.

*3-(4-(3-(4-Nitrophenyl)acryloyl)phenyl)-1-methyl-2,3-dihydroquinazolin-4(1H)-one **4b***

Yield: 93.87%; m.p.: 245°C; crystallized from dioxane; ^{1}H-NMR (200 MHz) δ: 2.989 (s, 3H, NCH_3), 5.038 (s, 2H, CH_2N), 6.953–8.314 (m, 14H, ArH + acryloyl H) ppm. Anal. calcd. for $C_{24}H_{19}N_3O_4$ (413.43): C, 69.72; H, 4.63; N, 10.16. Found: C, 69.73, H, 4.8; N, 9.89.

*3-[4-(3-(4-Chlorophenyl)acryloyl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one **4c***

Yield: 64%; m.p.: 218–219°C; crystallized from ethanol/dioxane (1:1); ^{1}H-NMR (200 MHz) δ: 2.985 (s, 3H, NCH_3), 5.030 (s, 2H, CH_2N), 6.934–8.286 (m, 14H, ArH + acryloyl H) ppm; MS *m*/*z* (rel. int.): 404 [M^+ + 2] (4.8), 403 [M^+ +1] (13.2), 402 [M^+] (13.2), 401 (37.8), 183 (13.4), 133 (26.6), 77 (100). Anal. calcd. for $C_{24}H_{19}ClN_2O_2$ (402.87): C, 71.55; H, 4.75; N, 6.95. Found: C, 71.26; H, 4.99; N, 7.03.

*3-[4-(3-Phenylacryloyl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one **4d***

Yield: 88%; m.p.: 195–196°C; crystallized from dioxane; IR ν: 3070 (CH, aromatic), 2992 (CH, aliphatic), 1656, 1594 (CO), 1504 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 2.989 (s, 3H, NCH_3), 5.032 (s, 2H, CH_2N), 6.934–8.286 (m, 15H, ArH + acryloyl H) ppm. Anal. calcd. for $C_{24}H_{20}N_2O_2$ (368.43): C, 78.24; H, 5.47; N, 7.60. Found: C, 78.28; H, 5.7; N, 7.51.

*3-[4-(3-(4-Methoxyphenyl)acryloyl)phenyl]-1-ethyl-2,3-dihydroquinazolin-4 (1H)-one **4e***

Yield: 45%; m.p.: 205–206°C; crystallized from dioxane; ^{1}H-NMR (200 MHz) δ: 1.177 (brs, 3H, CH_3), 3.536–3.583 (m, 2H, CH_2CH_3), 3.859 (s, 3H, OCH_3), 5.1(s, 2H, CH_2N), 6.919–7.079 (m, 2H, acryloyl H), 7.502–8.248 (m, 12H, ArH) ppm. Anal. calcd. for $C_{26}H_{24}N_2O_3$ (412.48): C, 75.71; H, 5.86; N, 6.79. Found: C, 75.51; H, 6.00; N, 6.83.

*3-[4-(3-(4-Nitrophenyl)acryloyl)phenyl]-1-ethyl-2,3-dihydroquinazolin-4(1H)-one **4f***

Yield: 42%; m.p.: 191–192°C; crystallized from dioxane; IR ν: 3073 (CH, aromatic), 2977 (CH, aliphatic), 1660, 1600 (CO), 1508 (C=C), 1465, 1334 (NO_2) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 1.182 (br s, 3H, CH_3), 3.543–3.579 (m, 2H, CH_2CH_3), 5.115 (s, 2H, CH_2N), 6.921–7.041 (m, 2H, acryloyl H),7.513–8.305 (m, 12H, ArH) ppm. Anal. calcd. for $C_{25}H_{21}N_3O_4$ (427.45): C, 70.25; H, 4.95; N, 9.83. Found: C, 70.13; H, 4.93; N, 9.50.

*3-[4-(3-(4-Chlorophenyl)acryloyl)phenyl]-1-ethyl-2,3-dihydroquinazolin-4(1H)-one **4g***

Yield: 41%; m.p.: 180°C; crystallized from dioxane/DMF (4:1); IR ν: 3054 (CH, aromatic), 2975 (CH, aliphatic), 1656, 1598 (CO), 1496 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 1.175 (s, 3H, CH_3), 3.545–3.579 (m, 2H, CH_2CH_3), 5.120 (s, 2H, CH_2N), 6.917–7.038 (m, 2H, acryloyl H), 7.504–8.314 (m, 12H, ArH) ppm. Anal. calcd. for $C_{25}H_{21}ClN_2O_2$ (416.5): C, 72.02; H, 5.04; N, 6.72. Found: C, 72.81; H, 5.29; N, 7.05.

3-[4-(3-Phenylacryloyl)phenyl]-1-ethyl-2,3-dihydroquinazolin-4(1H)-one ***4h***

Yield: 41%; m.p.: 167–168°C; crystallized from dioxane; IR ν: 3052 (CH, aromatic), 2967 (CH, aliphatic), 1656, 1598 (CO), 1500 (C=C) cm^{-1}; ^{1}H-NMR (300 MHz) δ: 1.129–1.153 (t, 3H, CH_3), 3.515–3.567 (q, 2H, CH_2CH_3), 5.078 (s, 2H, CH_2N), 6.893–7.003 (m, 2H, acryloyl H), 7.451–8.235 (m, 13H, ArH) ppm; MS *m/z* (rel. intensity): 382 [M^+] (22.9), 381 (36.2), 119 (72.4), 77 (100). Anal. calcd. for $C_{25}H_{22}N_2O_2$ (382.45): C, 78.51; H, 5.80; N, 7.32. Found: C, 78.75; H, 5.61; N, 7.50.

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

To a suspension of compound **4a** or **4b** (0.001 mol) in ethanol, hydrazine hydrate (99%, 0.1 mL, 0.002 mol) or phenylhydrazine (0.001 mol) was added. The reaction mixture was refluxed for 12 h and then concentrated. After cooling, the obtained crystalline product was filtered and recrystallized from the proper solvent.

3-[4-(5-(4-Methoxyphenyl)-4, 5-dihydro-1H-pyrazol-3-yl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one ***5a***

Yield: 87%; m.p.: 175°C; crystallized from ethanol; IR ν: 3330 (NH), 3045 (CH, aromatic), 2956 (CH, aliphatic), 1660 (CO), 1598 (C=N), 1508 (C=C) cm^{-1}; ^{1}H-NMR (300 MHz) δ: 2.882–2.888 (m, 1H, C_4-pyrazole H), 2.917 (s, 3H, NCH_3), 3.383 (m, 1H, C_4-pyrazole H), 3.739 (s, 3H, OCH_3), 4.781 (m, 1H, C_5-pyrazole H), 4.832 (s, 2H, CH_2N), 6.887–7.852 (m, 12H, ArH), 10.20 (s, 1H, NH, exch.) ppm; MS *m/z* (rel. int.): 414 [M^+ + 2] (4.7), 413 [M^+ + 1] (15), 412 [M^+] (57.5), 279 (15), 206 (10.2),145 (19.7), 104 (100). Anal. calcd. for $C_{25}H_{24}N_4O_2$ (412.48): C, 72.80; H, 5.86; N, 13.58. Found: C, 72.36; H, 5.95; N, 13.66.

3-[4-(5-(4-Methoxyphenyl)-1-phenyl-4,5-dihydro-1H-pyrazol-3-yl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one ***5b***

Yield: 42%; m.p.: 198–200°C; crystallized from dioxane; IR ν: 3039 (CH, aromatic), 2997 (CH, aliphatic), 1652 (CO), 1596 (C=N), 1498 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 2.947 (s, 3H, NCH_3), 3.047–3.165 (dd, 1H, C_4 pyrazole H), 3.716 (s, 3H, OCH_3), 3.829–3.974 (dd, 1H, C_4-pyrazole), 4.927 (s, 2H, CH_2N), 5.406–5.496 (dd, 1H, C_5-pyrazole H), 6.887–7.814 (m, 17H, ArH) ppm. Anal. calcd. for $C_{31}H_{28}N_4O_2$ (488.58): C, 76.21; H, 5.78; N, 11.47. Found: C, 76.14; H, 5.54; N, 11.47.

3-[4-(5-(4-Nitrophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one ***5c***

Yield: 50%; m.p.: 308–309°C; crystallized from dioxane/DMF (1:1); ^{1}H-NMR (300 MHz) δ: 2.759 (s, 1H, C_4-pyrazole H), 2.983 (s, 3H, NCH_3), 3.333–3.55 (m, 1H, C_4-pyrazole H), 4.921 (br s, 1H, C_5-pyrazole H), 4.964 (s, 2H, CH_2N), 6.917–8.107 (m, 12H, ArH), 13.57 (br s, 1H, NH, exch.) ppm. Anal. calcd. for $C_{24}H_{21}N_5O_3$ (427.46): C, 67.44; H, 4.95; N, 16.38. Found: C, 67.70; H, 4.56; N, 16.24.

3-[4-(5-(4-Nitrophenyl)-1-phenyl-4,5-dihydro-1H-pyrazol-3-yl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one ***5d***

Yield: 41%; m.p.: 233°C; crystallized from dioxane; IR ν: 2945 (CH, aliphatic), 1662 (CO), 1598 (C=N), 1497 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 2.970 (s, 3H, NCH_3), 3.256–3.284 (m, 1H, C_4-pyrazole H), 3.951–4.103 (m, 1H, C_4-pyrazole H), 4.942 (s, 2H, CH_2N), 5.60–5.80 (m, 1H, C_5-pyrazole H), 6.782–8.269 (m, 17H, ArH) ppm; MS *m/z* (rel. int.): 503 [M^+] (100), 358 (56.1), 281 (12.2), 251 (12.2), 236 (39), 57 (22). Anal. calcd. for $C_{30}H_{25}N_5O_3$ (503.55): C, 71.56; H, 5.00; N, 13.91. Found: C, 71.53; H, 4.72; N, 13.64.

2-[1-(4-(1-Methyl-4-oxo-1,2-dihydroquinazolin-3(4H)yl)phenyl)ethylidene]hydrazinecarbothioamide ***6***

A mixture of compound **3a** (0.01 mol) and thiosemicarbazide (0.01 mol) in ethanol (20 mL) containing ten drops of glacial acetic acid was heated under reflux for 2 h. The separated solid was filtered, washed, dried, and recrystallized from dioxane/DMF mixture (5:1).

Yield: 84%; m.p.: 240°C; IR ν: 3407, 3270, 3198 (NH_2, NH), 3154 (CH, aromatic), 2965 (CH, aliphatic), 1659 (CO), 1593 (C=N), 1495 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 2.34 (s, 3H, CH_3), 2.955 (s, 3H, NCH_3), 4.938 (s, 2H, CH_2N), 6.904–8.034 (m, 8H, ArH), 8.34 (s, 1H, NH exch.), 10.216 (br s, 2H, NH_2, exch.) ppm; MS *m/z* (rel. int.): 355 [M^+ + 2] (12.8), 353 [M^+] (19.1), 279 (100), 237 (14.9), 194 (19.1), 161 (29.8), 108 (17), 55 (29.8). Anal. calcd. for $C_{18}H_{19}N_5O_5$ (353.44): C, 61.17; H, 5.42; N, 19.81. Found: C, 61.01; H, 5.61; N, 19.51.

General procedure for preparation of compounds ***7a, b***

A suspension of compound **6** (0.001 mol) and the appropriate phenacyl bromide (0.0015 mol) in ethanol (20 mL) was heated under reflux for 3 h. Then, anhydrous sodium acetate (0.0015 mol) was added and the reaction mixture was heated again under reflux for 12 h. It was cooled and poured into cold water (100 mL). The separated solid was filtered, dried, and recrystallized from a suitable solvent.

3-[4-(1-(2-(4-((4-Chlorophenyl)thiazol-2-yl)hydrazono)-ethyl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one ***7a***

Yield: 66%; m.p.: 283–284°C; crystallized from DMF/H_2O (1:1); IR ν: 3216 (NH), 3061 (CH, aromatic), 2986 (CH, aliphatic), 1648 (CO), 1559 (C=N), 1498 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 2.350 (s, 3H, CH_3), 2.942 (s, 3H, NCH_3), 4.927 (s, 2H, CH_2N), 6.889–7.927 (m, 13H, ArH + C_5-thiazole H), 11.310 (s, 1H, NH, exch.) ppm; MS *m/z* (rel. int.): 489 [M^+ + 2] (23.8), 488 [M^+ + 1] (30.1), 487 [M^+] (59.1), 278 (96.4), 210 (42.7), 168 (35), 105 (100). Anal. calcd. for $C_{26}H_{22}ClN_5OS$ (488.00): C, 63.99; H, 4.54; N, 14.35. Found: C, 64.01; H, 4.26; N, 14.04.

3-[4-(1-(2-(4-Phenylthiazol-2-yl)hydrazono)ethyl)phenyl]-1-methyl-2,3-dihydroquinazolin-4(1H)-one ***7b***

Yield: 71%; m.p.: 205°C; crystallized from dioxane/DMF (4:1); IR ν: 3213 (NH), 3062 (CH, aromatic), 2957 (CH, aliphatic), 1644 (CO), 1557 (C=N), 1503 (C=C) cm^{-1}; ^{1}H-NMR (200 MHz) δ: 2.342 (s, 3H, CH_3), 2.964 (s, 3H, NCH_3), 4.94 (s, 2H, CH_2N), 6.942–7.996 (m, 14H, ArH + C_5-thiazole H), 11.310 (s, 1H, NH, exch.) ppm. Anal. calcd. for $C_{26}H_{23}N_5OS$ (453.56): C, 68.85; H, 5.11; N, 15.44. Found: C, 68.74; H, 4.97; N, 14.72.

Pharmacology

Animals

Male albino rats and male mice, weighing 150–200 g and 20–25 g each, respectively, were used. All experimental animals were provided from the Faculty of Veterinary Medicine, Zagazig University, Egypt. All animals were held under standard laboratory

conditions in the animal house (temperature 27°C) with a 12/12 light-dark cycle. Animals were fed laboratory diet and water *ad libitum*. All experiments were carried out using five animals per group. The animal experiments were performed in accordance with international guidelines.

Drugs

Carrageenan (carrageenan kappa-type III) and all other reagents were purchased from Sigma Chemical Co. (St. Louis, MO, USA). The test compounds **3a**, **4a–d**, **5a–d**, and reference drugs indomethacin, celecoxib, or diclofenac sodium were used in the following assays.

Analgesic-activity evaluation using the acetic-acid-induced writhing response method

The test was carried out using the previously described technique by Elisabetsky *et al*. [20]. Mice were divided into eleven groups each consisting of five mice and were injected intraperitoneally (i.p.) with 0.1 mL/10 g body weight of 0.6% acetic acid solution in saline 1 h after the oral administration of the test compounds **3a**, **4a–d**, **5a–d** at a dose of 50 mg/kg. The frequency of writhing was recorded within 25 min from the injection of acetic acid. Celecoxib was administrated to one group of mice at a dose level of 50 mg/kg as a positive control. One group of mice was left as a control.

Anti-inflammatory activity evaluation using the carrageenan-induced edema method

The effects of test compounds **3a**, **4a–d**, and **5a–d** on rat-paw edema induced by carrageenan were studied as described by Winter *et al.* [21]. The compounds and celecoxib were tested at a dose of 50 mg/kg orally while diclofenac sodium was tested at a dose of 4 mg/kg. Test compounds and celecoxib were suspended in gum acacia (7%), while diclofenac sodium was dissolved in hot distilled water. The diameter of the right paw of each animal was determined using a micrometer. The control group received only the corresponding vehicle. Thirty minutes later, paw edema was induced by subcutaneous injection of 0.1 mL carrageenan (0.1%) into the subplantar surface of the right hind paw of all animals. The paw diameter was measured 1, 2, 3, 4, 6, and 24 h after carrageenan injection and is recorded in Table 2.

Statistics

Since the time course of the effect was followed, it was possible to use the cumulative anti-inflammatory effect during the whole observation period as the area under the curve (AUC). Because the AUC represents the integrated anti-inflammatory effect (variation of paw diameter) during the observation period, it then includes both the maximal response and the duration of action. The AUC relating variation of edema to time was obtained using the trapezoidal rule [25]. Total inhibition (TI,%) was obtained for each group and then recorded using the following ratio:

$$TI\,(\%) = [\text{AUC control} - \text{AUC treat}] \times 100/\text{AUC control} \quad (1)$$

Data were expressed as mean ± standard error of mean (SEM) of five animals.

Ulcerogenic activity

All tested compounds **3a**, **4a–d**, and **5a–d** were investigated for their ulcerogenic activity using indomethacin as a reference drug [22]. Male albino rats weighing 150–200 g were fasted for 12 h prior to drug administration. Water was supplied *ad libitum*. The animals were divided into 12 equal groups (each of five). The first group received 7% gum acacia (suspending vehicle) orally once a day and was left as a control, whereas the second and third group received indomethacin and celecoxib at a dose of 18 and 50 mg/kg/day orally, respectively. Groups four to twelve received the test compounds **3a**, **4a–d**, and **5a–d** at 50 mg/kg/day. The test compounds were administered once a day for three successive days. The animals were killed by an overdose of ether six hours after the last dose. The stomachs were removed, opened along the greater curvature, and examined for ulceration. The number and diameter of discrete areas of damage in the glandular mucosa were scored (Table 3). The ulcer score was calculated according to the 1-to-10 scoring system of Valcavi *et al*. [22].

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