Tropical Journal of Pharmaceutical Research August 2013; 12 (4): 583-589
ISSN: 1596-5996 (print); 1596-9827 (electronic)

Available online at http://www.tjpr.org
http://dx.doi.org/10.4314/tjpr.v12i4.21

Original Research Article

Synthesis, Analgesic and Anti-inflammatory Activities of 3-Ethyl-2-substituted Amino-3H-quinazolin-4-ones

RV Sheorey[1], A Thangathiruppathy[2] and V Alagarsamy[3]*
[1]Department of Pharmacy, Karpagam University, Eachanari Post, Coimbatore – 641 021, [2]Department of Pharmacology, SB College of Pharmacy, Sivakasi – 626 130, Tamilnadu State, [3]Medicinal Chemistry Research Laboratory, MNR College of Pharmacy, Sangareddy, Gr. Hyderabad -502 294, Andrapradesh State, India.

*For correspondence: **Email:** drvalagarsamy@gmail.com; **Tel:** +91 8455-230690; **Fax:** +91 8455-230 555;*

Received: 24 May 2012 *Revised accepted: 10 June 2013*

Abstract

Purpose: *To synthesize a series of novel 3-ethyl-2-substituted amino-quinazolin-4(3H)-ones and evaluate them for their analgesic and anti-inflammatory activities.*
Methods: *The compounds, 3-ethyl-2-substituted amino-quinazolin-4(3H)-ones, were synthesized by reacting the amino group of 3-ethyl-2-hydrazino quinazolin-4(3H)-one with a variety of aldehydes and ketones. The synthesized compounds were characterized by Fourier transform infrared (FTIR), proton-nuclear magnetic resonance spectroscopy (^{1}H-NMR) and mass spectrometry. The purity of the compounds was determined by elemental analysis. Test for analgesic activity was performed by tail-flick technique using Wistar albino mice while anti-inflammatory activity was evaluated by carrageenan-induced paw oedema test in rats. Diclofenac sodium was used as positive control for both analgesic and anti-inflammatory activities.*
Results: *The compound, 3-ethyl-2-(cyclohexylidene-hydrazino)-3H-quinazolin-4-one (AS1), emerged as the active analgesic (activity, 63.89 %) and anti-inflammatory (activity, 60.00 %) compound of the series, and compared well with the reference standard, diclofenac sodium, which exbited analgesic and anti-inflammatory activities of 62.04 and 65.11 %, respectively*
Conclusion: *The compound (AS1) can serve as a lead molecule for further development to a clinically useful novel class of analgesic and anti-inflammatory agents.*

Keywords: *Quinazoline, Analgesic, Anti-inflammatory, Synthesis*

Tropical Journal of Pharmaceutical Research is indexed by Science Citation Index (SciSearch), Scopus, International Pharmaceutical Abstract, Chemical Abstracts, Embase, Index Copernicus, EBSCO, African Index Medicus, JournalSeek, Journal Citation Reports/Science Edition, Directory of Open Access Journals (DOAJ), African Journal Online, Bioline International, Open-J-Gate and Pharmacy Abstracts

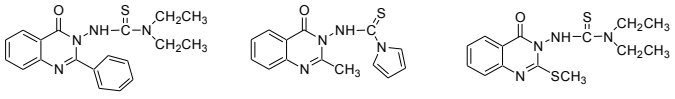
INTRODUCTION

Nonsteroidal anti-inflammatory drugs (NSAIDs) are commonly prescribed for the treatment of acute and chronic inflammation, pain and fever. Most NSAIDs act via inhibition of cyclooxygenase, thus preventing prostaglandin biosynthesis. However, this mechanism of action is also responsible for their main undesirable effects, gastrointestinal (GI) ulceration and, less frequently, nephrotoxicity. Increase in hospitalization and deaths due to GI-related disorders parallels increased use of NSAIDs. Therefore, the discovery of new safer anti-inflammatory drugs represents a challenging goal for such a research area [1-4].

In the course of our going medicinal chemistry research program it was found that quinazolines and condensed quinazolines exhibit potent central nervous system (CNS) activities including analgesic, anti-inflammatory [5] and anticonvulsant behavior [6]. Quinazolin-4(3*H*)-ones with 2,3-substitution are reported to possess significant analgesic, anti-inflammatory [7,8] and anticonvulsant activities [9]. Earlier, we have documented some lead 2-phenyl-3-

substituted quinazolines (Figure 1, I) [10], 2-methyl-3-substituted quinazolines (Figure 1, II) [11], 2-methylthio-3-substituted quinazaolines (Figure 1, III) [12], and 2,3-disubstituted quinazolines [13] that exhibited good analgesic and anti-inflammatory properties.

The present work is an extension of our ongoing efforts towards the development and identification of new molecules for analgesic and anti-inflammatory activities. The objective of the present study was to synthesize a series of 3-ethyl-2-substitutedamino-quinazolin-4(3*H*)-ones and evaluate them for their analgesic and anti-inflammatory activities.



(I) (II) (III)

Figure 1: Some lead molecules of quinazolin-4-one

EXPERIMENTAL

Chemistry

Melting points (mp) were taken in open capillaries on a Thomas Hoover melting point apparatus and are uncorrected. IR spectra of the synthesized compounds were recorded by FT-IR (Shimadzu, Japan) using KBr pellet (ν max in cm^{-1}). The NMR spectra of the synthesized compounds were recorded in deuterated chloroform ($CDCl_3$) (unless specified) with tetramethyl silane (TMS) as internal reference (chemical shift in δ, ppm) using Varian 300 MHz and Bruker 500 MHz (Washington, USA) spectrometers. The mass spectra of the compounds were obtained on Jeol GC mate instrument (Masspec, Japan). Elemental analyses were performed in Perkin-Elmer 2400 CHN elemental analyzer (Waltham, USA). The progress of the reaction was monitored on readymade silica gel plates (Merck) using chloroform : methanol (9:1) as a solvent system. Iodine was used as a developing agent.

3-Ethyl-2-thioxo-2,3-dihydro-1*H*-quinazolin-4-one (4)

A solution of ethylamine (1.31 g, 0.02 mol) in dimethyl sulfoxide (10 mL) was stirred vigorously. To this was added carbon disulphide (1.6 mL) and aqueous sodium hydroxide (1.2 mL of 20 M solution) dropwise during 30 min with stirring. Dimethyl sulphate (2.5 g, 0.02 mol) was then added gradually, keeping the reaction mixture in freezing mixture with stirring which was continued for further 2 h. The reaction mixture was then poured into ice-water and the solid obtained was filtered, washed with water, dried and recrystallized from 95 % ethanol.

Methyl anthranilate (**3**) (1.5 g; 0.01 mol) and the above prepared *N*-(ethyl) methyl dithiocarbamic acid (0.01 mol) were dissolved in ethanol (20 mL). To this anhydrous potassium carbonate (100 mg) was added and refluxed for 23 h. The reaction mixture was cooled in ice and the solid separated was filtered and purified by dissolving in 10 % alcoholic sodium hydroxide solution (2N) and re-precipitating by treatment with 20 % dilute hydrocholoric acid. The solid obtained was filtered, washed with water, and dried. It was recrystallized from 95 % ethanol to afford **(4).**

3-Ethyl-2-methylsulfanyl-3*H*-quinazolin-4-one (5)

The compound, 3-ethyl-2-thioxo-2,3-dihydro-1*H*-quinazolin-4-one (**4**) (2.05 g, 0.01 mol), was dissolved in 40 mL of 2 % ethanol-sodium hydroxide solution (2N). To this, dimethyl sulfate (5.0 mL, 0.01 mol) was added dropwise for 15 min with stirring. The stirring was continued for 1 h, the reaction mixture was then poured into ice water. The solid obtained was filtered, washed with water, dried and recrystallized from 95 % ethanol.

3-Ethyl-2-hydrazino--3*H*-quinazolin-4-one (6)

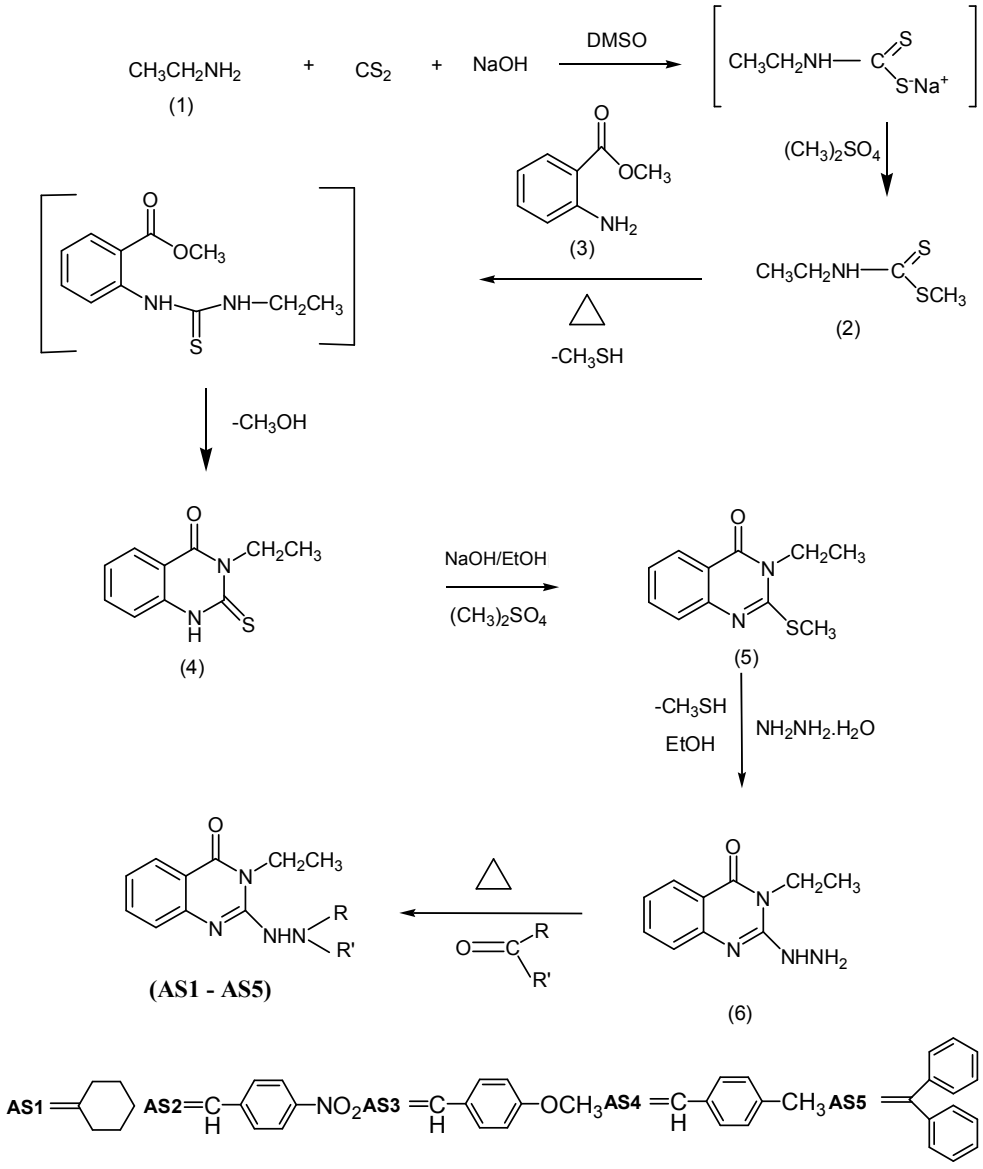
The compound, 3-ethyl-2-methylsulfanyl-3*H*-quinazolin-4-one **(5)** (2.19 g, 0.01 mol), was dissolved in ethanol (95 %, 25 mL). To this, hydrazine hydrate (99 %) (0.5 mL, 0.1 mol) and anhydrous potassium carbonate (100 mg) was added and refluxed for 28 h at inert atmosphere. The reaction mixture was cooled and poured into ice-water. The solid obtained was filtered, washed with water, dried and recrystallized from chloroform : toluene (25:75) mixture.

3-Ethyl-2-(cyclohexylidene-hydrazino)-3*H*-quinazolin-4-one (AS1)

A mixture of 3-ethyl-2-hydrazino-3*H*-quinazolin-4-one **(6)** (800 mg, 0.004 mol) and cyclohexanone (0.004 mol) in glacial acetic acid (0.25 mL) was refluxed for 30 h. The reaction mixture was poured into ice water and filter. The solid filter cake was recrystallized from ethanol.

3-Ethyl-2-(*N*-(4-nitro-benzylidene-hydrazino))-3*H*-quinazolin-4-one (AS2)

A mixture of 3-ethyl-2-hydrazino-3*H*-quinazolin-4-one **(6)** (800 mg, 0.004 mol) and 4-nitro benzalhedyde (605 mg, 0.004 mol) in glacial



Scheme 1: Synthesis of 3-ethyl-2-substitutedamino-3*H*-quinazolin-4-ones

acetic acid (0.25 mL) was refluxed for 33 h. The reaction mixture was poured into ice water and filter. The solid filter cake was recrystallized from ethanol.

3-Ethyl-2-(*N*-(4-methoxy-benzylidene-hydrazino))-3*H*-quinazolin-4-one (AS3)
A mixture of 3-ethyl-2-hydrazino-3*H*-quinazolin-4-one **(6)** (800 mg, 0.004 mol) and 4-methoxy benzalhedyde (545 mg, 0.004 mol) in glacial acetic acid (0.25 mL) was refluxed for 32 h. The reaction mixture was poured into ice water and filter. The solid filter cake was recrystallized from ethanol.

3-Ethyl-2-(*N*-(4-methyl-benzylidene-hydrazino)-3*H*-quinazolin-4-one (AS4)
A mixture of 3-ethyl-2-hydrazino-3*H*-quinazolin-4-one **(6)** (800 mg, 0.004 mol) and 4-methyl benzalhedyde (480 mg, 0.004 mol) in glacial acetic acid (0.25 mL) was refluxed for 30 h. The reaction mixture was poured into ice water and filter. The solid filter cake was recrystallized from ethanol.

3-Ethyl-2-(*N*-phenyl-benzylidene-hydrazino)-3*H*-quinazolin-4-one (AS5)
A mixture of 3-ethyl-2-hydrazino-3*H*-quinazolin-4-one **(6)** (800 mg, 0.004 mol) and diphenyl ketone (730 mg, 0.004 mol) in glacial acetic acid (0.25 mL) was refluxed for 35 h. The reaction mixture was poured into ice water and filter. The solid filter cake was recrystallized from ethanol.

Animal studies

The synthesized compounds were evaluated for analgesic and anti-inflammatory activities. The test compounds and the standard drugs were administered in the form of a suspension (1 %

carboxy methyl cellulose as a vehicle) by oral route. Each group consisted of six animals. The animals were maintained in colony cages at 25 ± 2 °C, relative humidity of 45 – 55 %, and 12 h/12 h light/dark cycle; they were fed standard animal feed. All the animals were acclimatized for a week before use [14]. The experimental protocol was duly approved by the institutional animal ethical committee (IAEC, F. No.25/559/2010-AWD) constituted by the Committee for the Purpose of Control and Supervision of Experimental Animals (CPCSEA), Ministry of Environment and Forest, New Delhi, India (IAEC, F. No.25/559/2010-AWD).

Analgesic activity

Test for analgesic activity was performed by tail-flick technique [15,16] using Wistar albino mice (25 - 35 g) of either sex selected by random sampling. Diclofenac sodium, at dose levels of 5, 10 and 20 mg/kg, was administered orally by gastric lavage as reference drug for comparison. The test compounds at three dose levels (5, 10, 20 mg/kg) were administered orally. Reaction time was recorded at 30 min, 1, 2 and 3 h after treatment, and cut-off time was 10 s. Analgesic activity (AA, %) was calculated as in Eq 1.

$$AA\ (\%) = \{(T_2 - T_1)/(10 - T_1)\}100 \quad \ldots\ldots\ldots (1)$$

where T_1 is the reaction time (s) before treatment, and T_2 the reaction time (s) after treatment.

Anti-inflammatory activity

Anti-inflammatory activity was evaluated by carrageenan-induced paw oedema test in Wistar Albino rats [17]. Diclofenac sodium (5, 10 and 20 mg/kg) was administered as a standard drug for comparison. The test compounds were administered orally at three dose levels of 5, 10 and 20 mg/kg. Paw volume was measured using mercury displacement technique with the aid of a plethysmograph immediately before and 30 min, 1, 2 and 3 h after carrageenan injection. Inhibition (I, %) of paw oedema was calculated as in Eq 2.

$$I\ (\%) = 100\{1- (a-x)/(b-y)\} \quad \ldots\ldots\ldots\ldots\ldots (2)$$

where **x** is mean paw volume of the rats before administration of carrageenan and test compounds or reference compound (test group), **a** is the mean paw volume of rats after the administration of carrageenan in the test group (drug-treated), **b** is the mean paw volume of rats after the administration of carrageenan in the control group, **y** is the mean paw volume of rats before administration of carrageenan in the control group.

Statistical analysis

Statistical analysis of the biological activities of the synthesized compounds in the animals was carried out using one-way analysis of variance (ANOVA). In all cases, post-hoc comparisons of the means of individual groups were performed using Tukey's test. A significance level of $p < 0.05$ denote significant difference in all cases. All values are expressed as mean ± standard deviation (SD). GraphPad Prism 3.0 version. (GraphPad GraphPad Software, Inc.11452 El Camino Real, #215, San Diego, CA 92130 USA) was used for the statistical analysis.

RESULTS

The key intermediate 3-ethyl-2-thioxo-2,3-dihydro-1*H*-quinazolin-4-one **4** (Scheme 1) was prepared by reacting ethyl amine (**1)** with carbon disulphide and sodium hydroxide in dimethyl sulphoxide to give sodium dithiocarbamate, which was methylated with dimethyl sulfate to afford the dithiocarbamic acid methyl ester **(2)**. Compound **2** on reflux with methyl anthranilate **(3)** in ethanol yielded the desired 3-ethyl-2-thioxo-2,3-dihydro-1*H*-quinazolin-4-one **(4)** via thiourea intermediate and gave a good yield (85 %). It was confirmed by IR spectra of compound **4** show intense peaks at 3250 cm^{-1} for cyclic thio urea (NH), 1668 cm^{-1} for carbonyl (C=O) and 1220 cm^{-1} for thioxo (C=S) stretching. ^{1}H NMR spectra of **4** showed multiplet around δ 1.0-2.0 and 7.10-7.50 for ethyl (5H) protons and aromatic (4H) protons respectively; and a singlet at δ 10.55 indicating the presence of NH. Data from elemental analysis were in conformity with the assigned structure. Further, the molecular ion recorded in the mass spectra is also in agreement with the molecular weight of the compound.

The IR spectra of compound **5** showed disappearance of NH and C=S stretching signals of cyclic thiourea. It showed a peak for carbonyl (C=O) stretching at 1680 cm^{-1}. The ^{1}H NMR spectra of compound **5** showed singlets due to SCH_3, at δ 2.70, multiplet around 7.20-7.60 for aromatic (4H) protons, respectively. Data from the elemental analyses and molecular ion recorded in the mass spectra further confirmed the assigned structure.

Nucleophilic displacement of methylthio group of **5** with hydrazine hydrate was carried out using ethanol as solvent to afford 3-ethyl-2-hydrazino-3*H*-quinazolin-4-one **6**. The formation of **6** was confirmed by ^{1}H NMR spectra which showed singlets at δ 4.56 and 10.31 due to NH_2 and NH, respectively, a multiplet at δ 1-1.5 and 7.15-7.30

for ethyl (5H) protons and aromatic (4H) protons respectively. The NH and NH_2 signals at 3330, 3205 cm^{-1} are appeared in the IR spectra. It also showed a peak for carbonyl (C=O) at 1670 cm^{-1}. Data from the elemental analyses are in conformity with the assigned structure. Further, the molecular ion recorded in the mass spectra is also in agreement with the molecular weight of the compound.

The compounds, 3-ethyl-2-substituted amino-3*H*-quinazolin-4-ones **AS1 - AS5** were obtained by the condensation of amino group of 3-ethyl-2-hydrazino-3*H*-quinazolin-4-one **(6)** with a variety of aldehydes and ketones. Formation of product is indicated by the disappearance of peak due to NH_2 of the starting material in IR and ^{1}H NMR spectrum of all the compounds **AS1 - AS5**. The IR ^{1}H NMR spectra of these compounds showed the presence of peaks due to ($N=CR_1R_2$) carbonyl (C=O), NH and aryl groups. The mass spectra of the the compounds show molecular ion peaks corresponding to their molecular formula. In the mass spectra of compounds **AS1 - AS5,** a common peak at m/z 144 corresponding to quinazolin-4-one moiety appeared. Elemental (C, H, N) analysis satisfactorily confirmed rhe elemental composition and purity of the synthesized compounds.

3-Ethyl-2-thioxo-2,3-dihydro-1*H*-quinazolin-4-one (4)

Yield = 85 %, mp 290-291°C; IR (KBr) cm^{-1}: 3250 (NH), 1668 (C=O), 1220 (C=S); ^{1}H NMR ($CDCl_3$) δ: 0.95-1.05 (t, 3H, $CH_2C\underline{H}_3$),1.39-1.50 (q, 2H, $C\underline{H}_2CH_3$), 7.10-7.50 (m, 4H, ArH), 10.55 (br s, 1H, NH, D_2O Exchangeable); MS (m/z): 206 (M^+); *Anal.* Calcd. for $C_{10}H_{10}N_2OS$: C, 58.22; H, 4.88; N, 13.58. Found: C, 58.25; H, 4.90; N, 13.63.

3-Ethyl-2-methylsulfanyl-3*H*-quinazolin-4-one (5)

Yield = 86 %, mp 173-174 °C; IR (KBr) cm^{-1}: 1680 (C=O), 1605 (C=N); ^{1}H NMR ($CDCl_3$) δ : 0.90-1.10 (t, 3H, $CH_2C\underline{H}_3$), 1.35-1.52 (q, 2H, $C\underline{H}_2CH_3$), 2.70 (s, 3H, SCH_3), 7.20-7.60 (m, 4H ArH); MS (m/z): 220 (M^+); *Anal.* Calcd for $C_{11}H_{12}N_2OS$: C, 59.97; H, 5.49; N, 12.71. Found: C, 59.92; H, 5.51; N, 12.74.

3-Ethyl-2-hydrazino--3*H*-quinazolin-4-one (6)

Yield = 80 %, mp 193-195 °C; IR (KBr) cm^{-1}: 3330, 3205 (NH NH_2), 1670 (C=O), 1610 (C=N); ^{1}H NMR ($CDCl_3$): δ 0.91-1.00 (t, 3H, $CH_2C\underline{H}_3$), 1.30-1.48 (q, 2H, $C\underline{H}_2CH_3$), 4.56 (br s, 2H, NH_2 D_2O Exchangeable), 7.15-7.30 (m, 4H, ArH), 10.31 (br s, 1H, NH D_2O Exchangeable); MS (m/z): 204 (M^+); *Anal.* Calcd for $C_{10}H_{12}N_4O$: C, 58.81; H, 5.92; N, 27.43. Found: C, 58.85; H, 5.90; N, 27.41.

3-Ethyl-2-(cyclohexylidene-hydrazino)-3*H*-quinazolin-4-one (AS1)

Yield = 73 %, mp 224-226 °C; IR (KBr) cm^{-1} : 3250 (NH), 1690 (C=O), 1612 (C=N); ^{1}H-NMR ($CDCl_3$): δ 0.91-1.10 (t, 3H, $CH_2C\underline{H}_3$), 1.29-1.86 (m, 10H, CH_2 cyclohexyl), 1.98-2.03 (q, 2H, $C\underline{H}_2CH_3$), 7.62-7.91 (m, 4H, ArH), 8.90 (br s, 1H, NH, D_2O Exchangeable); MS (m/z): 284 (M^+); *Anal.* Calcd. for $C_{16}H_{20}N_4O$: C, 67.58; H, 7.08; N, 19.70. Found: C, 67.55; H, 7.04; N, 19.73.

3-Ethyl-2-(*N*-(4-nitro-benzylidene-hydrazino))-3*H*-quinazolin-4-one (AS2)

Yield = 71 %, mp 282-283 °C; IR (KBr) cm^{-1} : 3268 (NH), 1679 (C=O), 1612 (C=N); ^{1}H-NMR ($CDCl_3$): δ 1.26-1.32 (t, 3H, $CH_2C\underline{H}_3$), 1.64-1.75 (q, 2H, $C\underline{H}_2CH_3$), 6.12 (s, 1H, CH), 8.02-9.13 (m, 8H, ArH), 9.53 (br s, 1H, NH, D_2O Exchangeable); MS (m/z): 337 (M^+); *Anal.* Calcd. for $C_{17}H_{15}N_5O_3$: C, 60.52; H, 4.48; N, 20.76. Found: C, 60.58; H, 4.53; N, 20.73.

3-Ethyl-2-(*N*-(4-methoxy-benzylidene-hydrazino))-3*H*-quinazolin-4-one (AS3)

Yield = 77 %, mp 268-270 °C; IR (KBr) cm^{-1}: 3305 (NH), 1690 (C=O), 1610 (C=N); ^{1}H-NMR ($CDCl_3$): δ 1.04-1.18 (t, 3H, $CH_2C\underline{H}_3$), 1.42-1.55 (q, 2H, $C\underline{H}_2CH_3$), 3.36 (s, 3H, OCH_3), 6.08 (s, 1H, CH), 7.22-8.31 (m, 8H, ArH), 9.03 (br s, 1H, NH, D_2O Exchangeable); MS (m/z): 322 (M^+); *Anal.* Calcd. for $C_{18}H_{18}N_4O_2$: C, 67.00; H, 5.63; N, 17.38. Found: C, 67.08; H, 5.58; N, 17.40.

3-Ethyl-2-(*N*-(4-methyl-benzylidene-hydrazino)-3*H*-quinazolin-4-one (AS4)

Yield = 74 %, mp 286-288 °C; IR (KBr) cm^{-1} : 3260 (NH), 1682 (C=O), 1605 (C=N); ^{1}H-NMR ($CDCl_3$): δ 0.82-0.96 (t, 3H, $CH_2C\underline{H}_3$), 1.15-1.23 (q, 2H, $C\underline{H}_2CH_3$), 2.15 (s, 3H, CH_3), 6.14 (s, 1H, CH), 7.05-8.01 (m, 8H, ArH), 9.36 (br s, 1H, NH, D_2O Exchangeable); MS (m/z): 306 (M^+); *Anal.* Calcd. for $C_{18}H_{18}N_4O$: C, 70.50; H, 5.92; N, 18.29. Found: C, 70.52; H, 5.93; N, 18.25.

3-Ethyl-2-(*N*-phenyl-benzylidene-hydrazino)-3*H*-quinazolin-4-one (AS5)

Yield = 73 %, mp 267-269 °C; IR (KBr) cm^{-1}: 3316 (NH), 1690 (C=O), 1608 (C=N); ^{1}H-NMR ($CDCl_3$): δ 0.93-1.06 (t, 3H, $CH_2C\underline{H}_3$), 1.26-1.33 (q, 2H, $C\underline{H}_2CH_3$), 6.11 (s, 1H, CH), 7.25-7.45 (m, 4H, ArH), 7.72-8.51 (m, 10H, ArH), 9.51 (br s,

1H, NH, D_2O Exchangeable); MS (m/z): 368 (M^+); *Anal.* Calcd. for $C_{23}H_{20}N_4O$: C, 75.05; H, 5.47; N, 15.20. Found: C, 75.09; H, 5.46; N, 15.23.

The results of analgesic testing indicate that the test compounds exhibited moderate analgesic activity at 30 min of reaction time and an increase in activity at 1 h which reached a peak level at 2 h. Decline in activity was observed at 3 h (Table 1).

Anti-inflammatory activity data (Table 2) indicate that all the test compounds protected rats from carrageenan-induced inflammation moderately at 30 min of reaction time with increased activity at 1 h that reached a peak level at 2 h. Decline in activity was observed at 3 h.

Table 1: Analgesic activity of the synthesized compounds (AS1 - AS5)

Compound code	Dose (mg/kg)	Analgesic activity (%)			
		30 min	*1 h*	*2 h*	*3h*
AS1	5	22.45 ± 2.04^c	28.70 ± 2.81^c	34.72 ± 3.82^c	16.44 ± 4.21^b
	10	37.27 ± 1.65^c	44.91 ± 4.06^c	51.62 ± 3.86^c	31.71 ± 3.33^c
	20	50.23 ± 2.69^c	59.72 ± 3.16^c	63.89 ± 3.34^c	40.05 ± 2.46^c
AS2	5	19.68 ± 2.54^c	23.61 ± 4.36^c	29.63 ± 4.14^c	13.66 ± 1.73^a
	10	29.63 ± 2.99^c	35.45 ± 4.42^c	41.43 ± 3.17^c	26.13 ± 3.96^c
	20	39.35 ± 2.70^c	45.14 ± 1.88^c	46.99 ± 2.54^c	35.42 ± 0.93^c
AS3	5	20.37 ± 2.52^b	26.62 ± 2.22^c	32.41 ± 3.47^c	13.89 ± 5.45^a
	10	34.03 ± 1.98^c	40.05 ± 2.47^c	43.75 ± 3.14^c	27.78 ± 4.66^c
	20	42.13 ± 2.88^c	51.85 ± 4.71^c	56.02 ± 2.28^c	37.96 ± 3.54^c
AS4	5	21.53 ± 1.88^c	29.17 ± 3.72^c	37.04 ± 2.99^c	19.68 ± 2.54^c
	10	35.42 ± 0.93^c	41.20 ± 2.50^c	45.14 ± 1.88^c	28.70 ± 5.93^c
	20	44.21 ± 2.96^c	54.17 ± 3.00^c	59.95 ± 2.46^c	37.73 ± 4.07^c
AS5	5	18.52 ± 2.90^c	24.54 ± 3.26^c	28.47 ± 2.26^c	16.44 ± 2.71^c
	10	31.48 ± 2.62^c	37.04 ± 2.99^c	43.29 ± 5.32^c	32.87 ± 3.88^c
	20	41.20 ± 2.50^c	48.84 ± 2.83^c	54.63 ± 2.65^c	37.04 ± 5.04^c
Control		1.852 ± 1.85^{Ns}	5.787 ± 2.59^{Ns}	7.870 ± 2.50^{Ns}	3.935 ± 2.49^{Ns}
Diclofenac Na	5	21.76 ± 2.20^c	31.48 ± 2.62^c	35.42 ± 4.41^c	17.59 ± 2.52^b
	10	35.18 ± 3.89^c	42.82 ± 5.20^c	48.61 ± 5.83^c	29.17 ± 3.42^c
	20	48.15 ± 4.47^c	58.33 ± 4.40^c	62.04 ± 3.54^c	37.96 ± 3.54^c

Data expressed as mean ± SD from six different experiments, each carried out in duplicate; $^a p < 0.05$, $^b p < 0.01$and $^c p < 0.001$ a compared with their respective positive control

Table 2: Anti-inflammatory activity of the synthesized compounds (**AS1 - AS5**)

Compound code	Dose (mg/kg)	Inhibition/Protection (%)			
		30 min	*1 h*	*2 h*	*3h*
AS1	5	26.31 ± 9.98^a	30.16 ± 4.82^b	35.55 ± 3.40^c	20.76 ± 2.10^a
	10	35.18 ± 10.40^b	42.85 ± 8.54^c	46.67 ± 1.46^c	38.59 ± 3.11^b
	20	47.36 ± 9.21^c	57.14 ± 4.64^c	60.00 ± 3.84^c	45.61 ± 3.12^c
AS2	5	12.28 ± 4.00^a	20.23 ± 3.88^a	25.78 ± 1.70^a	16.95 ± 1.37^a
	10	28.06 ± 6.17^b	28.57 ± 4.47^b	32.89 ± 4.06^c	21.92 ± 5.21^a
	20	31.57 ± 6.07^c	38.09 ± 5.53^c	41.33 ± 3.86^c	30.70 ± 2.86^c
AS3	5	21.05 ± 5.08^c	28.57 ± 2.68^c	32.00 ± 2.17^c	19.29 ± 1.28^c
	10	28.07 ± 9.28^b	34.12 ± 4.94^b	36.44 ± 3.38^c	23.97 ± 4.25^a
	20	36.84 ± 6.65^c	42.85 ± 2.53^c	44.00 ± 2.17^c	35.08 ± 2.18^c
AS4	5	15.79 ± 5.92^a	26.19 ± 2.38^c	26.50 ± 3.78^c	17.54 ± 1.17^b
	10	26.31 ± 6.92^b	30.95 ± 6.47^c	32.66 ± 3.89^c	27.19 ± 2.42^b
	20	31.57 ± 4.90^c	38.09 ± 3.88^c	48.00 ± 2.00^c	32.46 ± 1.26^c
AS5	5	16.66 ± 6.28^a	23.80 ± 4.21^c	28.00 ± 1.37^c	17.54 ± 1.01^a
	10	21.05 ± 8.59^a	32.14 ± 5.79^b	41.33 ± 4.35^c	21.32 ± 2.08^a
	20	26.31 ± 7.31^b	40.47 ± 4.12^c	48.00 ± 1.91^c	29.82 ± 3.46^c
Control		1.52 ± 1.85^{Ns}	5.87 ± 2.59^{Ns}	7.87 ± 2.50^{Ns}	3.93 ± 2.49^{Ns}
Diclofenac Na	5	16.66 ± 4.38^a	24.20 ± 5.47^b	30.66 ± 4.68^c	20.17 ± 1.82^b
	10	32.45 ± 9.83^b	36.50 ± 4.33^c	43.78 ± 3.69^c	33.04 ± 2.97^b
	20	41.22 ± 8.74^c	54.36 ± 6.06^c	65.11 ± 4.49^c	47.22 ± 8.06^c

Data expressed as mean ± SD from six different experiments, each carried out in duplicate; $^a p < 0.05$, $^b p < 0.01$and $^c p < 0.001$ a compared with their respective positive control

DISCUSSION

All the synthesized compounds were confirmed by spectral data (IR, NMR and mass spectra). Elemental (C, H, N) analysis satisfactorily confirmed elemental composition and purity of the synthesized compounds.

Compound **AS1** with 1-cyclohexylidene substituent showed good analgesic activity. Replacement of an alkyl chain at the C-2 position with aryl group (compound **AS3, AS4, AS5**) leads to decrease in analgesic activity. Placement of aryl group with electron withdrawing substituent (compounds A**S5**) leads to further decrease of analgesic activity. Compound 3-ethyl-2-(cyclohexylidene-hydrazino)-3*H*-quinazolin-4-one (**AS1**) emerged as the most active analgesic agent and it is moderately more potent than the reference standard, diclofenac sodium. The compound, 3-ethyl-2-(cyclohexylidene-hydrazino)-3*H*-quinazolin-4-one (**AS1**), emerged the most active anti-inflammatory agent. Replacement of an alkyl chain at the C-2 position with aryl group (compound **AS3, AS4, AS5**) leads to decrease in anti-inflammatory activity. Placement of aryl group with electron withdrawing substituent (compound A**S5**) led to further decrease of anti-inflammatory activity.

CONCLUSION

The synthesized compounds exhibited promising analgesic and anti-inflammatory activities. Among them, compound 3-ethyl-2-(cyclohexylidene-hydrazino)-3*H*-quinazolin-4-one (**AS1**) is the most active compound of the series and is as potent as the reference standard (diclofenac sodium). Hence, this compound is a potential lead molecule for a clinically useful novel class of analgesic and anti-inflammatory agents.

REFERENCES

1. *van Ryn J, Botting RM. New insights into the mode of action of anti- inflammatory drugs. Inflamm Res 1995; 44: 1-10.*
2. *van Ryn J, Trummlitz G, Pairet M. COX-2 selectivity and inflammatory processes. Curr Med Chem 2000; 7: 1145-1161.*
3. *van Ryn J. Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. Nature: New Biology 1971; 231: 232-235.*
4. *Beuck M. Nonsteroidal Antiinflammatory Drugs: A New Generation of Cyclooxygenase Inhibitors Angew Chem Int Ed 1999; 38: 631–633.*
5. *Alagarsamy V, Revathi R, Vijayakumar S, Ramseshu KV. Synthesis and pharmacological investigation of some novel 2,3-disubstituted quinazolin-4(3H)-ones as analgesic and antiinflammatory agents. Pharmazie 2003; 58: 233-236.*
6. *Alagarsamy V, Thangathirupathy A, Mandal SC, Rajasekaran S, Vijayakumar S, Revathi R, Anburaj J, Arunkumar S, Rajesh S. Pharmacological evaluation of 2-substituted (1,3,4) thiadiazolo quinazolines. Indian J Pharm Sci 2006; 68: 108-111.*
7. *Abdel-Rahman AE, Bakhite EA, Al-Taifi EA. Synthesis and antimicrobial testing of some new S-substituted-thiopyridines, thienopyridines, pyridothienopyrimidines and pyridothienotriazines. Pharmazie 2003; 58: 372-377.*
8. *Chambhare, RV, Khadse BG, Bobde AS, Bahekar RH. Synthesis and preliminary evaluation of some N-[5-(2-furanyl)-2-methyl-4-oxo-4H-thieno[2,3-d]pyrimidin-3-yl]-carboxamide and 3-substituted-5-(2-furanyl)-2-methyl-3H-thieno[2,3-d]pyrimidin-4-ones as antimicrobial agents. Eur J Med Chem 2003; 38: 89-100.*
9. *Santagati NA, Caruso A, Cutuli VMC, Caccamo F. Synthesis and pharmacological evaluation of thieno[2,3-d]pyrimidin-2,4-dione and 5H-pyrimido [5,4-b]indol-2,4-dione derivatives. Il Farmaco 1995; 50: 689-695.*
10. *Alagarsamy V, Solomon VR, Vanikavitha G, Paluchamy V, Chandran MR, Sujin AA, Thangathiruppathy A, Amuthalakshmi S, Revathi R. Synthesis, analgesic, anti-inflammatory and antibacterial activities of some novel 2-phenyl-3-substituted quinazolin-4(3H) ones. Biol Pharm Bull 2002; 25: 1432-1435.*
11. *Alagarsamy V, Murugananthan G, Venkateshperumal R. Synthesis, analgesic, anti-inflammatory and antibacterial activities of some novel 2-methyl-3-substituted quinazolin-4-(3H)-ones. Biol Pharm Bull 2003; 26: 1711-1714.*
12. *Alagarsamy V, Rajesh R, Meena R, Vijaykumar S, Ramseshu KV, Duraianandakumar T. Synthesis, Analgesic, Anti-inflammatory and Antibacterial Activities of some Novel 2-Methylthio-3-Substituted Quinazolin-4(3H)-ones. Biol Pharm Bull 2004; 27: 652-656.*
13. *Alagarsamy V, Muthukumar V, Pavalarani N, Vasanthanathan P, Revathi R. Synthesis, analgesic and anti-inflammatory activities of some novel 2,3-disubstituted quinazolin-4(3H)-ones. Biol Pharm Bull 2003; 26: 557-559.*
14. *Committee for the purpose of control and supervision on experiment on animals. CPCSEA guidelines for laboratory animal facility, Indian J Pharmacol 2003; 35: 257-274.*
15. *Kulkarni SK. Heat and other physiological stress-induced analgesia: Catecholamine mediated and naloxone reversible response. Life Sciences 1980; 27:185-188.*
16. *Amour RE, Smith DL. A method for determinating loss of pain sensation. J Pharm Exp Therap 1941; 72: 74-78.*
17. *Winter CA, Risely EA, Nuss GW. Carregeenin induced oedema in hind paw of the rat as assay for Antiinflammatory drugs. Exp Biol Med 1962; 111: 544-547.*