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Original article

Nonsteroidal antiinflammatory agents—part 2 antiinflammatory, analgesic and antipyretic activity of some substituted 3-pyrazolin-5-ones and 1,2,4,5,6,7-3*H*-hexahydroindazol-3-ones

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Abstract

As a part of a research project on the synthesis of a number of substituted 1-(pyrimidin-2-yl)-3-pyrazolin-5-ones and 2-(pyrimidin-2-yl)hexahydroindazol-3-ones and as a result of the interesting antiinflammatory, analgesic and antipyretic activities recorded for some of these compounds, some new 3-pyrazolin-5-ones and hexahydroindazol-3-ones linked to substituted imidazolyl, pyrimidyl and tetrahydroquinazolinyl moieties were prepared and evaluated for such activity (Fig. 1). A structure–activity relationship (SAR) comparative study indicated that some compounds from 3-pyrazolin-5-one (**2, 6–8, 10**) and indazolone (**18, 20, 24, 27, 29**) series exhibited pronounced antiinflammatory, analgesic and antipyretic activities relative to indomethacin. Most of these compounds were found to be nearly equipotent in the antiinflammatory screen (ED_{50} = 16.8–19.9 mg/kg) whereas the lead compound, 2-indazolyl-4-pyrimidineacetic acid **24** (Fig. 1), was found to be the most potent among this series (ED_{50} = 9.9 mg/kg). Additionally, the most active compounds were shown to have a large safety margin (ALD_{50} = 3.0 g/kg, po) and devoid of ulcerogenic potentialities when administered orally at a dose of 300 mg/kg.

Keywords: Ethyl arylidenecyanoacetate; Arylidenemalononitrile; Ethoxymethylene-2-phenyl-2-oxazolin-5-one; Dimethyl acetonedicarboxylate; 1-(Imidazol-2-yl and pyrimidin-2-yl)-3-pyrazolin-5-ones synthesis; 2-(Imidazol-2-yl and pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones synthesis; Antiinflammatory activity; Analgesic activity; Antipyretic activity; NSAIDs

1. Introduction

Since the first pyrazolin-5-one was developed by Knorr [1] in 1883, many papers have been reported on the antiinflammatory, analgesic and antipyretic evaluation of several pyrazoles, pyrazolin-3-ones and pyrazolidine-3,5-diones [2–7]. Many of these derivatives such as phenybutazone, febrazone, feclobuzone, mefobutazone, suxibuzone and ramifenazone have found their clinical application as NSAIDs [8–10]. Additionally, several indazoles and indazol-3-ones were reported to possess good antiinflammatory activity [11–15]. Moreover, some indazoles such as bendazac, benzydamine and tetyrhydroindazole are clinically useful as NSAIDs [10]. As a result of our interest on the pyrazolin-5-one and indazol-3-one ring systems, we have described in a previous paper [16] the synthesis of some 1-(pyrimidinyl)-3-pyrazolin-5-ones and 2-(pyrimidinyl)-1,2,4,5,6,7-3*H*-hexahydroindazol-3-ones with different substituents at positions 4 and 5 of the pyrimidinyl moiety. According to this investigation the 5-phenylpyrimidinyl derivative from the 3-pyrazolin-5-one series and the 5-butyl and 5-phenylpyrimidinyl derivatives from indazol-3-one series exhibited interesting antiinflammatory, analgesic and antipyretic activities. For this reason and for further structure–activity relationship (SAR) comparative study, we have now synthesized and tested a new series of substituted pyrazolin-5-ones and 1,2,4,5,6,7-3*H*-hexahydroindazol-3-ones (Fig. 1) linked to different substituted imidazolyl, pyrimidyl and tetrahydroquinazolinyl moieties (Figs. 2 and 3). In fact, most of these compounds were designed as close structural relatives to epirizole (Fig. 1) which is a Japanese drug reported to have a good antiinflammatory activity and proved to

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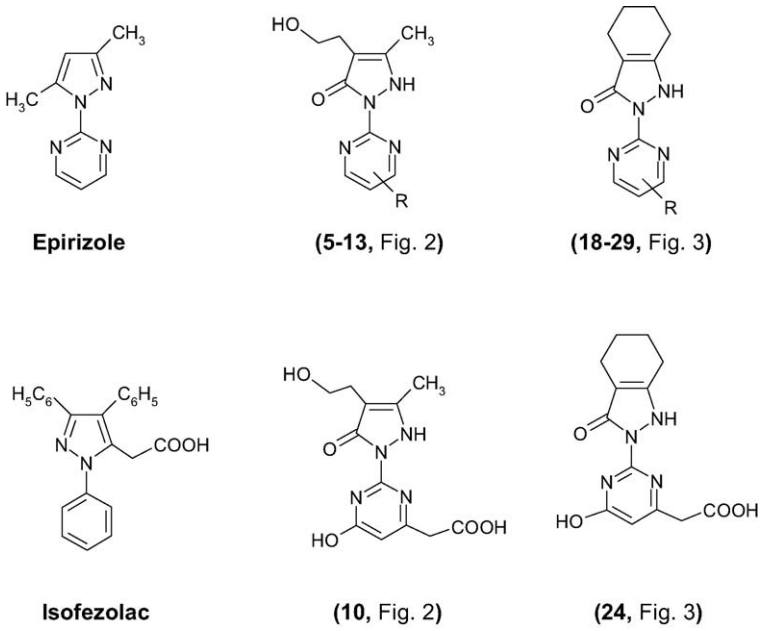
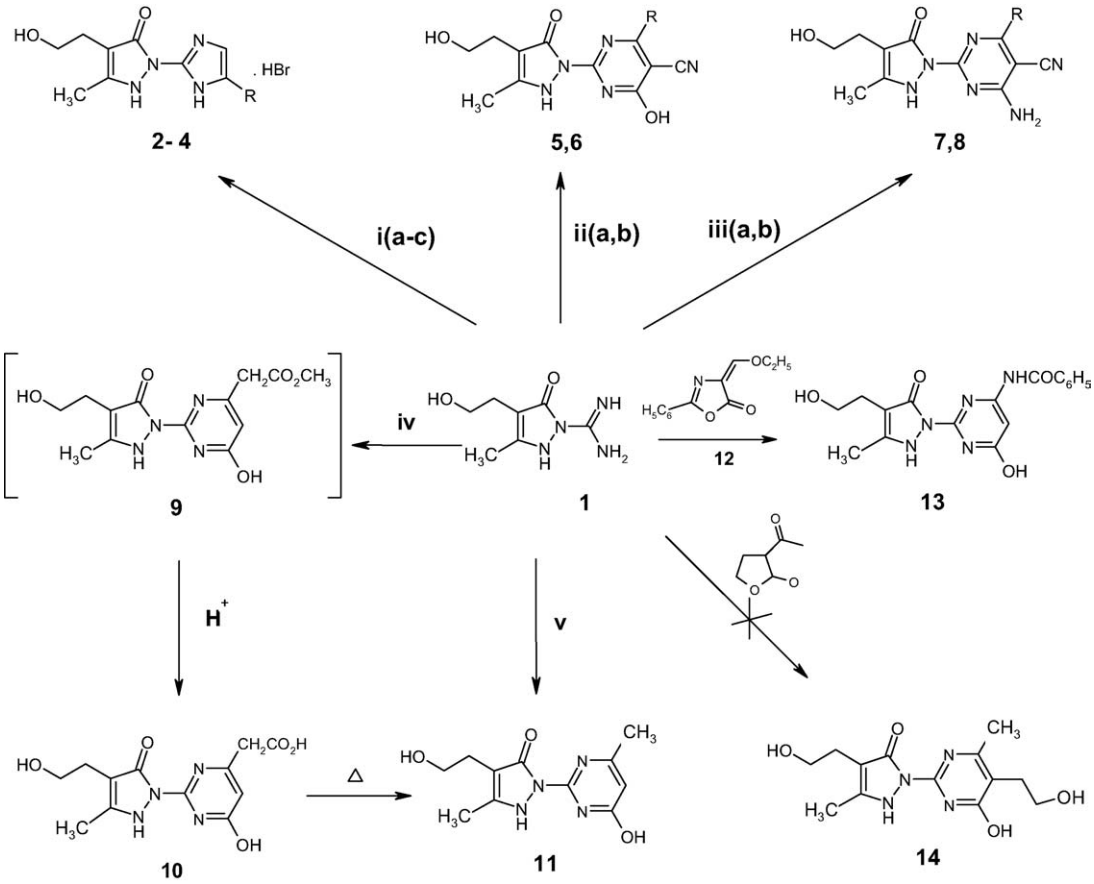


Fig. 1.

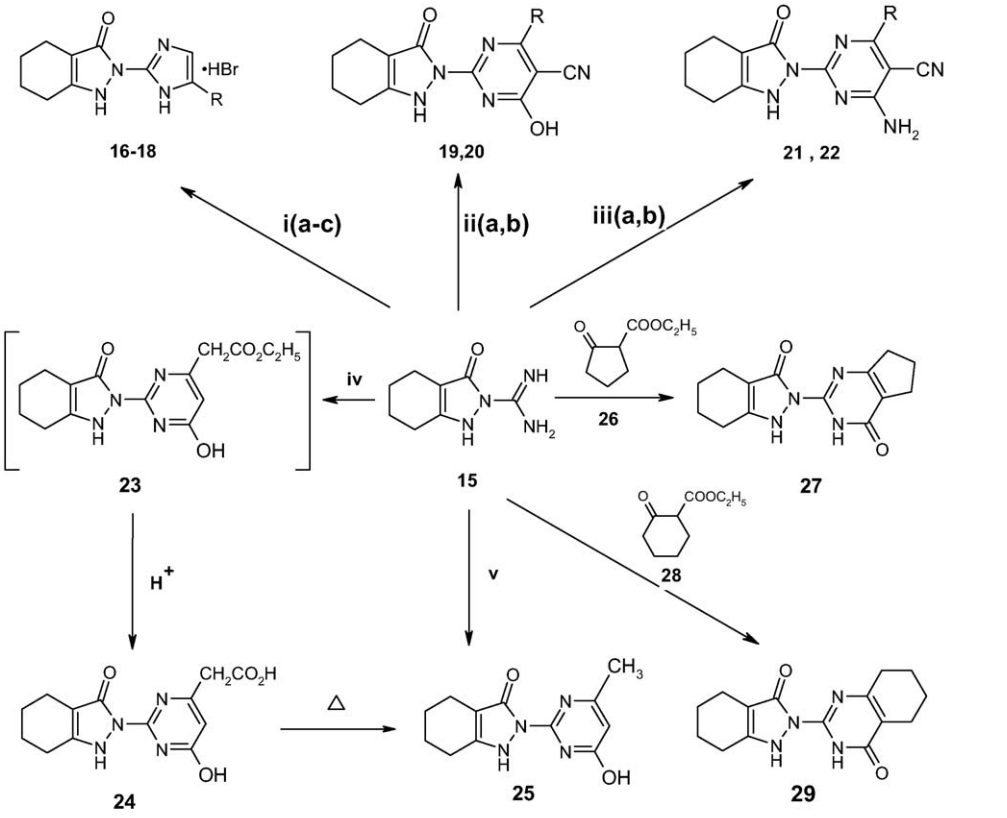


i(a-c) = R-$COCH_2Br$, **ii(a,b)** = R-$CH=C(CN)CO_2C_2H_5$, **iii(a,b)** = R-$CH=C(CN)_2$, **iv** = $CO(CH_2CO_2CH_3)_2$, **v** = $CH_3COCH_2CO_2C_2H_5$

ia: R = C6H5, **ib:** R = C6H4Cl (p), **ic:** R = C6H4CH3 (p), **iia, iiia:** R = C6H5, **iib, iiib:** R = C6H4Cl (p)

2: R = C_6H_5, **3**: R = C_6H_4Cl (*p*), **4**: R = $C_6H_4CH_3$ (*p*), **5,7**: R = C_6H_5, **6,8**: R = C_6H_4Cl (*p*)

Fig. 2.



i(a-c) = R-$COCH_2Br$, **ii(a,b)** = R-$CH=C(CN)CO_2C_2H_5$, **iii(a,b)** = R-$CH=C(CN)_2$, **iv** = $CO(CH_2CO_2CH_3)_2$, **v** = $CH_3COCH_2CO_2C_2H_5$

ia: R = C6H5, **ib:** R = C6H4Cl (p), **ic:** R = C6H4CH3 (p), **iia, iiia:** R = C6H5, **iib, iiib:** R = C6H4Cl (p)

16: R = C6H5; **17:** R = C6H4Cl(p) ; **18:** R = C6H4CH3(p) ; **19,21:** R = C6H5 ; **20,22:** R = C6H4Cl(p)

Fig. 3.

inhibit gastric lesion induced by acidic NSAIDs [9,17,18]. Moreover, some of the target compounds (**10, 24**; Fig. 1) were linked to an acetic acid moiety, a characteristic feature of many NSAIDs such as indomethacin, sulindac, tolmetin, diclofenac, lonazolac and isofezolac (Fig. 1) [9,19].

2. Chemistry

In a previous publication [16] we have described the synthesis of 1-amidino-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one (**1**, Fig. 2) and 2-amidino-1,2,4,5,6,7-hexahydro-3 *H*-indazol-3-one (**15**, Fig. 3). These two intermediates were utilized effectively for the synthesis of several substituted 1-(pyrimidin-2-yl)-3-pyrazolin-5-ones and 2-(pyrimidin-2-yl)-hexahydroindazol-3-ones [16]. In the present investigation the 1-amidinopyrazolinone (**1**) was found to be a useful intermediate for the synthesis of a new series of substituted 1-(imidazol-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-ones (**2–4**; Fig. 2) and 1-(pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-ones (**5–13**; Fig. 2). Similarly, the 2-amidinoindazolone (**15**) was used for the synthesis of some substituted 2-(imidazol-2-yl)-1,2,4,5,6,7-hexahydro-3-*H*-indazol-3-one (**16–18**; Fig. 3), 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones (**19–27**; Fig. 3) and the 2-tetrahydroquinazolinyl derivative (**29**; Fig. 3).

Reacting the 1-amidinopyrazolinone (**1**) with the appropriate substituted phenacyl bromide **i(a–c)** in refluxing ethanol resulted in the respective 1-(4-arylimidazol-2-yl)-3-pyrazolin-5-one hydrobromides (**2–4**). The substituted 1-(5-cyano-6-hydroxy-4-phenylpyrimidin-2-yl)-3-pyrazolin-5-one (**5**, R = C_6H_5) was obtained by reacting **1** with ethyl arylidenecyanoacetate (**iia**, R = C_6H_5) in refluxing ethanol in the presence of piperidine; whereas, the 4-chlorophenyl analog **6** was obtained by reacting **1** with the arylidenecyanoacetate (**iib**, R = *p*-ClC_6H_4) in the presence of sodium ethoxide. On the other hand, the substituted 1-(6-amino-5-cyano-4-phenylpyrimidin-2-yl)-3-pyrazolin-5-one (**7**, R = C_6H_5) was obtained by reacting **1** with the arylidenemalononitrile (**iiia**, R = C_6H_5) in refluxing ethanol in the presence of piperidine; whereas, the 4-chlorophenyl analog (**8**, R = *p*-ClC_6H_4) was obtained by reacting **1** with the arylidenemalononitrile (**iiib**, R = *p*-ClC_6H_4) in the presence of sodium ethoxide. The 4-pyrimidineacetic acid derivative **10** was obtained by condensing **1** with dimethyl acetonedicarboxylate (**iv**) in refluxing ethanol in the presence of potassium carbonate. It is worthy of mentioning that the intermediate methyl ester **9** is quietly unstable and several trials

for its isolation has been attempted but failed. In addition, the TLC and ^{1}H-NMR of the crude product of **10** indicate its contamination with a minute amount of the parent ester **9**, which was converted to **10** during purification. Decarboxylation of the 4-pyrimidineacetic acid **10** in refluxing dimethylformamide yielded the corresponding 4-methylpyrimidinyl derivative **11**. The decarboxylation of **10** was confirmed by ^{1}H-NMR spectral data (see Section 4) and chemically by unequivocal synthesis from the amidinopyrazolinone **1** and ethyl acetoacetate (**v**) [16]. The 4-benzoylaminopyrimidinyl compound **13** was obtained by condensing **1** with ethoxymethylene-2-phenyl-2-oxazolin-5-one (**12**) in refluxing ethanol in the presence of potassium carbonate. Trials to obtain the 5-(2-hydroxyethyl) pyrimidinyl derivative **14** from **1** and 2-acetylbutyrolactone was abortive (Fig. 2).

Fig. 3 illustrates the synthesis of several substituted 2-(imidazol-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one (**16–18**), 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one (**19–27**) and the 2-tetrahydroquinazolinyl derivative (**29**) starting from the 2-amidinoindazolone (**15**).

Thus reacting **15** with the substituted phenacyl bromide **i**(**a–c**) in refluxing ethanol resulted in the respective 1-(4-aryl-imidazol-2-yl)indazol-3-ones (**16-18**). The substituted 1-(6-hydroxypyrimidin-2-yl)indazol-3-ones (**19, 20**) were obtained by reacting **15** with the respective arylidenecyanoacetate **ii**(**a, b**) in refluxing ethanol in the presence in the presence of sodium ethoxide. On the other hand, the substituted 1-(6-aminopyrimidin-2-yl)indazol-3-one (**21**, R = C_6H_5) was obtained by reacting **15** with the arylidenemalononitrile (**iiia**, R = C_6H_5) in presence of piperidine; whereas, the 4-chlorophenyl analog (**22**, R = *p*-ClC_6H_4) was obtained by reacting **15** with the arylidenemalononitrile (**iiib**, R = *p*-ClC_6H_4) in the presence of sodium ethoxide. The 4-pyrimidineacetic acid derivative **24** was obtained by condensing **15** with dimethyl acetonedicarboxylate (**iv**) in refluxing ethanol in the presence of potassium carbonate. Similar to **9,** the methyl ester **23** is quietly unstable and cannot be isolated. Decarboxylation of the 4-pyrimidineacetic acid **24** in refluxing dimethylformamide yielded the corresponding 4-methylpyrimidinyl derivative **25**. The decarboxylation of **24** was confirmed by ^{1}H-NMR spectral data (see Section 4) and chemically by unequivocal synthesis from the amidinoindazolone (**15**) and ethyl acetoacetate (**v**) [16]. The 2-(cyclopenta[d]pyrimidin-2-yl)-hexahydro-3*H*-indazol-3-one (**27**) was obtained by condensing **15** with ethyl cyclopentanone-2-carboxylate (**26**) in refluxing ethanol in the presence of potassium carbonate; similarly, the 2-(tetrahydroquinazolin-2-yl)-hexahydroindazol-3-one (**29**) was obtained by condensing **15** with ethyl cyclohexanone-2-carboxylate (**28**).

3. Biological results and discussion

3.1. Antiinflammatory (AI) activity (Tables 1 and 2)

To assess the AI of the designed compounds, selected analogs (12 compounds) from both the pyrazolin-5-one and the 3*H*-indazol-3-one series were evaluated by two screening protocols widely used for testing the NSAIDs; namely, the rat dextran-induced paw edema and formaldehyde-induced arthritis screens. The paw edema was employed as a model for acute inflammation, while the formaldehyde-induced arthritis assay was used as a model for sub-acute condition. The ED_{50} of some of the most potent compounds were determined for comparative study with indomethacin as a reference drug.

For the dextran-induced paw model [20], each test compound was dosed orally (at 50 mg/kg) 1 h prior to induction of inflammation by dextran injection, the antiinflammatory activity was then calculated 1–4 h after induction and summarized in Table 1. A comparative study of the antiinflammatory activity of the test compound relative to the reference drug at different time interval indicated the following: after 1 h most of tested compounds from both series showed nearly similar pharmacokinetic profiles as revealed from their potent and rapid onset of action. Surprisingly, after 1 h, these compounds showed inhibition of paw oedema, at a dose of 50 mg/kg, po, higher than indomethacin at a dose of 5 mg/kg, po. The highest activity (after 1 h) was recorded for (**2**, 85%, R = phenyl) from the 1-imidazolylpyrazolinone series and (**18**, 82%, R = *p*-tolyl) from 2-imidazolylindazolone series. On the other hand, the maximum AI activity was recorded for (**8**, 76%, R = 4-choro-phenyl) from the 1-pyrimidinylpyrazolinones and (**27**, 83%, R = fused cyclopentane ring) from the 2-pyrimidinylindazolone series. An outstanding activity (after 1 h) was recorded for the indazol-3-one (**29**, 92%) which is linked to a tetrahydroquinazolinyl moiety at position-2.

Taking the antiinflammatory activity after 3 h as a criteria for comparison, it can be concluded that the 1-imidazolylpyrazolinone (**2**, R = phenyl) displayed a higher inhibition of paw edema (81%) than indomethacin (67%); whereas, the 1-pyrimidinylpyrazolinones (**6–8**) were nearly equipotent to indomethacin. Similarly, the 2-pyrimidinylindazolones (**20, 24, 27**) were nearly effective as indomethacin. Among the 2-imidazolylindazolone series, compound **18** (R = *p*-tolyl) showed an inhibition of paw oedema (69%) comparable to indomethacin (67%); while, the 2-tetrahydroquinazolinylindazolone (**29**) showed a higher inhibition of paw oedema (78%) at a dose of 50 mg/kg, po, than the reference drug (67%) at a dose of 5 mg/kg, po. It is apparent that compound (**24**) is more active (74%) than the 2-pyrazolyl-4-pyrimidineacetic acid (**10**, 69%); and even more effective than indomethacin (67%) after 3 h.

Some of the most active compounds from both series (**2, 8, 10, 16, 24** and **29**) were further tested at 5, 10, 20, 40 and 50 mg/kg body weight in order to determine their ED_{50} values. Most of these compounds were found to be nearly equipotent (ED_{50} = 16.8–19.9 mg/kg, Table 1). Whereas, the lead compound, 2-indazolyl-4-pyrimidineacetic acid **24** (Fig. 1), was found to be the most potent among these series (ED_{50} = 9.9 mg/kg). The ED_{50} of indomethacin was found to be 2.7 mg/kg of body weight.

For the formaldehyde-induced arthritis screen, arthritis was induced by formaldehyde injection in the first and third day and test compounds were administered orally (at 50 mg/kg daily) for 7 days [20]. As a result of inflammation, the level of

Table 1
Antiinflammatory activity of some 1-(imidazol-2-yl and pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-ones **2–10** and 2-(imidazol-2-yl and pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **15–28** in paw edema screen

Compound[a]	Volume of edema [b] (ml ± S.E.)					ED_{50}
	0	1 h	2 h	3 h	4 h	mg/kg
Controls	0.41 ± 0.04	0.62 ± 0.02	0.69 ± 0.02	0.73 ± 0.01	0.75 ± 0.02	–
		(49)	(32)	(22)	(17)	
2	0.41 ± 0.02	0.47 ± 0.04	0.51 ± 0.03	0.49 ± 0.01	0.50 ± 0.01	19.8
		(85)	(76)	(81)	(78)	
3	0.44 ± 0.01	0.53 ± 0.01	0.59 ± 0.01	0.60 ± 0.01	0.61 ± 0.01	–
		(80)	(66)	(64)	(62)	
6	0.45 ± 0.03	0.60 ± 0.03	0.60 ± 0.03	0.59 ± 0.04	0.59 ± 0.02	–
		(67)	(67)	(69)	(69)	
7	0.43 ± 0.01	0.54 ± 0.02	0.58 ± 0.01	0.56 ± 0.02	0.57 ± 0.01	–
		(74)	(65)	(70)	(68)	
8	0.46 ± 0.02	0.57 ± 0.02	058 ± 0.03	0.60 ± 0.03	0.60 ± 0.02	18.9
		(76)	(74)	(70)	(70)	
10	0.45 ± 0.01	0.58 ± 0.01	0.58 ± 0.03	059 ± 0.02	0.62 ± 0.02	18.2
		(71)	(71)	(69)	(62)	
16	0.43 ± 0.01	0.55 ± 0.02	0.58 ± 0.02	0.64 ± 0.02	0.62 ± 0.01	16.8
		(72)	(65)	(51)	(56)	
18	0.49 ± 0.02	0.58 ± 0.03	0.62 ± .04	0.64 ± 0.03	0.60 ± 0.03	–
		(82)	(74)	(69)	(78)	
20	045 ± 0.03	0.56 ± 0.0	0.56 ± 0.01	0.54 ± 0.02	0.57 ± 0.02	–
		(78)	(78)	(80)	(73)	
24	0.43 ± 0.04	0.52 ± 0.04	0.51 ± 0.03	0.54 ± 0.02	0.55 ± 0.03	9.9
		(79)	(81)	(74)	(72)	
27	0.46 ± 0.04	0.54 ± 0.03	0.57 ±	0.60 ± 0.02	0.59 ± 0.02	–
		(83)	(76)	(70)	(72)	
29	0.49 ± 0.03	0.53 ± 0.02	0.60 ± 0.03	0.61 ± 0.03	0.60 ± 0.01	18.9
		(92)	(78)	(76)	(78)	
Indometh-acin	0.42 ± 0.01	0.56 ± 0.01	0.56 ± 0.02	0.56 ± 0.01	0.55 ± 0.02	2.7
		(64)	(67)	(67)	(69)	

[a] Dose levels, po: test compounds (50 mg/kg), indomethacin (5 mg/kg).
[b] In parentheses the percentage of antiinflammatory activity (AI). See experimental section.

Table 2
Antiinflammatory activity of some 1-(imidazol-2-yl and pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-ones **2–10** and 2-(imidazol-2-yl and pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **16–29** in formaldehyde-induced arthritis assay

Compound [a]	Level of AST (U/l) ± S.E. [b]	% AI	Level of ALT (U/l) ± S.E. [b]	% AI
Controls without arthritis	55.20 ± 0.50	–	48.8 ± 1.55	–
Controls with arthritis	66.60 ± 2.10	–	57.8 ± 1.18	–
2	59.80 ± 0.87	(60)	51.40 ± 1.35	(71)
3	6o.80 ± 1.42	(51)	51.60 ± 1.31	(69)
6	59.80 ± 0.84	(60)	52.00 ± 1.61	(64)
7	60.00 ± 1.24	(58)	51.4 ± 1.72	(71)
8	60.00 ± 0.96	(58)	51.00 ± 1.85	(76)
10	59.60 ± 1.13	(61)	5o.80 ± 1.16	(78)
16	61.60 ± 1.70	(44)	51.20 ± 1.04	(73)
18	60.00 ± 1.16	(58)	53.80 ± 1.78	(44)
20	61.2 ± 1.04	(47)	52.00 ± 1.44	(64)
24	60.00 ± 1.02	(58)	51.20 ± 2.19	(73)
27	59.60 ± 1.23	(61)	53.20 ± 1.56	(51)
29	59.20 ± 1.09	(65)	50.40 ± 1.92	(82)
Indomethacin	58.40 ± 1.23	(72)	50.60 ± 2.26	(80)

[a] Dose levels, po: test compounds (50 mg/kg), indomethacin (5 mg/kg).
[b] Each value represents the unit/liter for each enzyme together with ± S.E.and inhibition % of the level of both enzymes (% AI) shown in parentheses.

serum transaminase aspartate aminotransferase (AST) and alanine aminotransferase (ALT) enzymes is increased. A decreased level of these enzymes upon administration of the test compounds was taken as an indication of antiinflammatory potentialities [21]. In fact we found in our previous publication that the assessment of AST and ALT enzymes provides a good and simple tool to evaluate the AI activity of the target compounds [16]. Indomethacin (at 5 mg/kg daily) was used as a reference drug and the results are recorded in Table 2. The data indicated that the tetrahydroquinazolinylindazolone **29** (at

50 mg/kg, po) and indomethacin (at 5 mg/kg, po) were nearly equipotent. However, the pyrazolinones (**2, 7, 8, 10**) and indazolones (**16**, **24**) displayed a good antiinflammatory activity but none of them were found to be superior over the reference drug.

A comparative study of the activity of the test compounds in both screen revealed that the pyrazolinones (**2, 6–8, 10**) and indazolones (**18, 20, 24, 27, 29**) were highly active in dextran paw edema screen and less active in formaldehyde induced arthritis test. This would indicate that these compounds are effective in acute inflammation and less active in chronic conditions. The compounds which showed a promising AI in the dextran induced paw edema screen from both series (**2, 6–8, 10, 18, 20, 24, 27, 29**) were further evaluated for their ulcerogenic activity and acute toxicity (ALD_{50}).

3.2. Ulcerogenic activity

The compounds did not show any ulceration or harmful effects on the stomach at a dose of 300 mg/kg po, when administered twice at 2 h interval in fasted rats [22].

3.3. Acute toxicity

The compounds showed a high safety margin when screened at graded doses (100 mg/kg–3.0 g/kg, po) for their acute lethal doses (ALD_{50}). The (ALD_{50}) values were found to be more than 3.0 g/kg.

3.4. Analgesic activity (Table 3)

The analgesic activity was recorded by rat tail withdrawal in response to immersion in water at 55 °C [23]. Indomethacin was used as a reference (5 mg/kg, po). A comparative study of the analgesic activity of the test compound relative to the reference drug (Table 3) at different time interval indicated the following: after 2 h the 2-pyrazolyl-4-pyridineacetic acid (**10**) and the 2-indazolyl-4-pyridineacetic acid (**24**) were found to be less potent than the reference drug; while, all other compounds were found to be superior over the reference drug. However, after 4 h the results revealed a good analgesic activity for the 1-imidazolylpyrazolinones (**2, 3**), 1-pyrimidinylpyrazolinone (**8**), 2-imidazolylindazolones (**16, 18**), 2-pyrimidinylindazolones (**20, 27**) and the tetrahydroquinazolinylindazolone (**29**); these compounds were found to be more potent than the reference drug.

Compounds (**2, 8, 10, 16, 24** and **29**) were further tested for their analgesic activity at 5, 10, 20, 40 and 60 mg/kg body weight in order to determine their ED_{50} values (Table 2). The data revealed that compounds **10** and **24** were nearly equipotent (ED_{50} = 43.3, 47.3 mg/kg,). Whereas, compounds **2** and **8** were more potent (ED_{50} = 20, 23.6 mg/kg). Compounds **16** and **29**

Table 3
Analgesic activity of some 1-(imidazol-2-yl or pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-ones **2–10** and 2-(imidazol-2-yl or pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **16–29**

Compound [a]	Reaction times ± S.E. [b]					ED_{50}
	0	30 min	2 h	3 h	4 h	mg/kg
Controls	2.20 ± 0.20	2.80 ± 0.38	2.80 ± 0.38	2.60 ± 0.41	2.61 ± 0.41	–
2	3.40 ± 0.25	4.00 ± 0.32 (19)	5.60 ± 0.51 (69)	5.20 ± 1.08 (56)	5.40 ± 1.05 (63)	23.6
3	2.60 ± 0.25	4.80 ± 0.67 (85)	4.2 ± 0.38 (62)	4.00 ± 0.45 (54)	4.60 ± 0.69 (77)	–
6	3.00 ± 0.32	3.80 ± 0.60 (27)	4.80 ± 0.38 (60)	4.6 ± 0.52 (53)	4.40 ± 0.83 (47)	–
7	3.00 ± 032	3.8 ± 0.60 (27)	4.8 ± 0.38 (60)	4.6 ± 0.52 (53)	4.40 ± 0.83 (47)	–
8	2.80 ± 0.38	3.2 ± 0.60 (14)	4.80 ± 0.38 (71)	4.8 ± 0.23 (71)	4.60 ± 0.52 (64)	20.0
10	3.2 ± 0.20	4.20 ± 0.60 (31)	4.40 ± 0.41 (38)	3.80 ± 0.60 (19)	4.20 ± 0.61 (31)	43.3
16	3.20 ± 0.20	4.60 ± 0.94 (44)	5.20 ± 0.81 (63)	5.60 ± 0.70 (75)	5.40 ± 0.69 (69)	10.8
18	3.00 ± 0.32	4.40 ± 0.52 (44)	4.60 ± 0.52 (50)	5.20 ± 038 (69)	5.20 ± 0.59 (69)	–
20	2.60 ± 0.25	4.60 ± 0.52 (77	4.60 ± 0.41 (77)	4.60 ± 0.52 (77)	4.40 ± 0.70 (69)	–
24	3.00 ± o.32	3.40 ± 0.25 (13)	4.2 ± 0.50 (40)	4.0 ± 0.72 (33)	4.00 ± 072 (33)	47.3
27	3.00 ± 0.32	4.00 ± 0.71 (33)	4.60 ± 0.52 (47	5.00 ± 0.32 (67)	5.00 ± 0.72 (67)	–
29	2.80 ± 0.38	3.50 ± 0.41 (25)	4.60 ± 0.52 (64)	5.00 ± 0.56 (79)	4.80 ± 0.50 (72)	10.8
Indomethacin	2.60 ± 025	3.20 ± 0.38 (23)	3.80 ± 0.50 (46)	4.40 ± 0.60 (69)	4.20 ± 0.38 (62)	3.0

[a] Dose levels, po: test compounds (50 mg/kg), indomethacin (5 mg/kg).
[b] In parentheses percentage increase of the reaction times calculated in comparison with basal values.

were the most potent among this series (ED_{50} = 10.8 mg/kg). The ED_{50} of indomethacin was found to be 3.0 mg/kg of body weight.

3.5. Antipyretic activity (Table 4)

Evaluation of the antipyretic activity [24] at different time interval relative to the indomethacin (Table 4) indicates the following: after 1 h the 1-imidazolylpyrazolinones (**2, 3**), 2-imidazolylindazolone (**18**) and 2-pyrimidinylindazolone (**27**) were found to be nearly similar to indomethacin in potency. On the other hand, after 4 h several compounds were found to be equipotent to indomethacin, particularly the 1-imidazolylpyrazolinone (**2**), 1-pyrimidinylpyrazolinone (**7**), the 1-imidazolylindazolone (**16**, **18**), and 2-tetrahydroquinazolinylindazole (**29**). Taking the activity after 6 h as criteria for comparison, it can be concluded that the 1-imidazolylpyrazolinone (**3**), 1-pyrimidinylpyrazolinone (**7**), 2-imidazolylindazolones (**16, 18**) and 2-tetrahydroquinazolinylindazolone (**29**) were nearly equal in potency to indomethacin. These compounds showed also a similar pharmacokinetic profile relative to indomethacin as indicated by their rapid hypothermic effect after 1 h, which persists for at least 6 h.

At the conclusion of this investigation, the following SAR can be concluded. It is evident that the presence of 4-aryllimidazolyl moiety at position-1 in pyrazolinone series or at position-2 in the indazolone series is essential for AI, analgesic and antipyretic activity. An outstanding AI activity was associated with a 4-phenylimidazolyl moiety in pyrazolinone (**2**) and a 4-tolylimidazolyl moiety in the indazolone (**18**). Additionally, the presence of 4-aryllimidazolyl moiety in both the pyrazolinones (**2, 3**) and indazolones (**16, 18**) promoted an excellent analgesic and antipyretic activity relative to indomethacin.

On the other hand, the presence of 4-arylpyrimidinyl moiety at postion-1 in pyrazolinone series (**6–8**) and at position-2 in the indazolone series (**20, 27**) is associated with AI comparable to indomethacin. The analgesic activity is maintained only with the *p*-chlorophenyl moiety in both the pyrazolinone (**8**) and indazolone (**20**); whereas, the antipyretic activity is retained with a 4-phenyl moiety in the pyrazolinone (**7**). The incorporation of a 4-pyrimidinylacetic (**24**) in the indazolone series is associated mainly with excellent AI and diminished analgesic and antipyretic activity. While, bridging of a tetrahydroquinazoliny (**29**) moiety to the indazolone ring is associated with excellent AI, analgesic and antipyretic activities.

Table 4
Antipyretic activity of some 1-(imidazol-2-yl or pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-ones **2–10** and 2-(imidazol-2-yl or pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **16–29**

Compound[a]	Body temperature ± S.E. [b]				
	0	1 h	2 h	4 h	6 h
Control	38.28 ± 0.10	38.26 ± 0.09 (0.1)	38.18 ± 0.06 (0.1)	38.30 ± 0.05 (0.1)	38.16 ± 0.10 (0.3)
2	37.88 ± 0.13	37.08 ± 0.20 (2.1)	36.96 ± 0.08 (2.4)	36.36 ± 0.15 (4.0)	36.40 ± 0.14 (3.7)
3	38.28 ± 0.13	37.50 ± 0.18 (2.0)	37.14 ± 0.20 (3.0)	36.88 ± 0.32 (3.7)	36.42 ± 0.19 (4.9)
6	37.82 ± 0.17	37.24 ± 0.21 (1.5)	37.06 ± 0.16 (2.0)	36.92 ± 0.10 (2.8)	36.78 ± 0.18 (2.8)
7	38.14 ± 0.15	37.76 ± 0.25 (1.0)	37.54 ± 0.14 (1.6)	36.50 ± 0.08 (4.3)	36.60 ± 0.29 (4.0)
8	37.98 ± 0.06	37.44 ± 0.13 (1.4)	37.02 ± 0.13 (2.5)	36.82 ± 0.11 (3.1)	36.72 ± 0.11 (3.3)
10	37.86 ± 0.18	37.42 ± 0.18 (1.2)	37.04 ± 0.10 (2.2)	36.70 ± 0.27 (3.1)	36.64 ± 0.22 (3.2)
16	38.32 ± 0.10	37.60 ± 0.12 (1.8)	37.10 ± 0.12 (3.1)	36.70 ± 0.21 (4.2)	36.54 ± 0.22 (4.7)
18	38.28 ± 0.10	37.5 ± 0.12 (1.9)	37.00 ± 0.06 (3.3)	36.42 ± 0.10 (4.9)	36.36 ± 0.12 (5.0)
20	38.08 ± 0.19	37.46 ± 0.24 (1.6)	37.18 ± 0.21 (2.4)	37.02 ± 0.26 (2.8)	36.86 ± 0.07 (3.2)
24	38.04 ± 0.14	37.78 ± 0.14 (0.7)	37.36 ± 0.19 (1.9)	37.08 ± 0.15 (2.5)	36.88 ± 0.13 (3.1)
27	38.25 ± 0.07	37.48 ± 0.15 (2.1)	37.22 ± 0.15 (2.6)	36.88 ± 0.21 (3.7)	36.70 ± 0.20 (3.9)
29	37.98 ± 0.15	37.32 ± 0.10 (1.6)	36.80 ± 0.25 (2.9)	36.24 ± 0.29 (4.5)	36.34 ± 0.24 (4.2)
Indomethacin	38.14 ± 0.11	37.40 ± 0.14 (1.9)	36.88 ± 0.08 (3.3)	36.56 ± 0.17 (4.1)	36.44 ± 015 (4.5)

[a] Dose levels, po: test compounds (50 mg/kg), indomethacin (5 mg/kg).
[b] Each value represents the mean ± S.E. with the percentage inhibition shown in parentheses.

4. Experimental protocols

4.1. Chemistry

All melting points were determined in open-glass capillaries on a Gallenkamp melting point apparatus and are uncorrected. The IR spectra were measured in KBr discs on a Perkin-Elmer 1430 spectrophotometer. ^{1}H-NMR spectra were recorded on a Varian Gemini 200 at 200 MHz in DMSO-d_6 using TMS as internal. ^{13}C-NMR spectra were performed on a varian VXR–300 NMR spectrometer at 75 MHz. MS were run on a Finnigan mass spectrometer model SQ/7000 (70 ev.). The microanalyses were performed at the microanalytical unit, Faculty of Science, Cairo University, and the data were within ± 0.4% of the theoretical values

4.1.1. 1-(5-Arylimidazol-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one hydrobromides **2–4** (Table 5)

A mixture of 1-amidino-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one (**1**) [16] (1.84 g, 10 mmol) and the appropriate phenacyl bromide **i(a–c)** (10 mmol) was refluxed in absolute ethanol (20 ml) for 7 h. After cooling, the separated crystalline product was filtered, dried and recrystallized from ethanol. IR ν per cm: 3448–3428, 3240, 2867, 1699–1695, 1648–1646, 1540–1536, 1468, 1440–1432.

^{1}H-NMR (**3**) (δ ppm): 2.35 (s, 3H, C*H*$_3$), 2.4 (t, 2H, –C*H*$_2$–CH$_2$O–), 3,45 (t, 2H, –CH$_2$C*H*$_2$O–), 7.5, 7.84 (two d, 4 Ar–*H*), 7.81(s, H at C-4 of imidazole).

^{1}H-NMR (**4**) (δ ppm): 2.32, 2.37 (two s, 6H, 2C*H*$_3$), 2.4 (t, 2H, –*CH*$_2$CH$_2$O–) 3.45 (t, 2H, CH$_2$*CH*$_2$O–), 7.25, 7.61 (two d, 4 Ar–*H*), 7.66 (s, 1H at C-4 of imidazole).

4.1.2. 1-(5-Cyano-6-hydroxy- 4-phenylpyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one **5** (Table 5)

The title compound was prepared by refluxing a mixture of **1** (1.84 g, 10 mmol), ethyl benzylidenecyanoacetate (**iia**, R = C_6H_5) (2.0 g, 10 mmol) and piperidine (1 ml) in absolute ethanol (20 ml) for 18 h. The reaction mixture was concentrated, after cooling the separated product was filtered, dried and crystallized from ethanol. IR ν per cm: 3252–2822, 2210, 1638, 1597, 1511. ^{1}H-NMR (δ ppm): 2.03 (s, 3H, C*H*$_3$), 2.65 (t, 2H, –C*H*$_2$CH$_2$O–), 3.75 (t, 2H, –CH$_2$C*H*$_2$*O*–), 7.49–7.56 (m, 3 Ar–*H*), 7.82–7.84 (d, 2 Ar–*H*), 11.83 (bs, 1H, NH). MS m/z (%): 339 [(0.41), *M* + 2]; 280 (100); 251 (98.17); 225 (37.19); 171 (18.02); 128 (37.34); 104 (44.79); 55 (40.88).

4.1.3. 1-[4-(4-chlorophenyl)-5-cyano-6-hydroxypyrimidin-2-yl]-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one **6** (Table 5)

To an ethanolic solution of sodium ethoxide (sodium metal 0.25 g, 11 mmol in absolute ethanol 20 ml) was added ethyl *p*-chlorobenzylidene cyanoacetate (**iib**, R = *p*-Cl C_6H_4) (2.37 g, 10 mmol) and **1** (1.84 g, 10 mmol). The reaction mixture was heated under reflux for 24 h and the solvent was evaporated under reduced pressure. The residue was dissolved in water and neutralized with 2 N hydrochloric acid. The separated product was filtered, washed with water, dried and crystallized from ethanol. IR ν per cm: 3415, 3241, 2213, 1663, 1588, 1536, 1471. ^{1}H-NMR (δ ppm): 2.25 (s, 3H, C*H*$_3$), 2.36 (t, 2H, –C*H*$_2$CH$_2$O–), 2.45 (t, 2H, –CH$_2$*CH*$_2$O–), 7.68–8.07 (two d, 4 Ar–*H*). ^{13}C-NMR (δ ppm): 14.45 (CH$_3$); 25.47 (CH$_2$); 60.10 (CH$_2$O); 116.08 (CN); 168.42 (C=O); 129.12, 130.33 (C-2,6 and C-3,5 of the phenyl ring, respectively); 101.2, 159.59 (C-3 and C-4 of pyrazolone, respectively); 134.18, 137.28 (C-1 and C-4 of phenyl, respectively) 92.99, 148.15, 154.58, 165.89 (C-4,5,2,6 of pyrimidine, respectively).

4.1.4. 1-(6-Amino-5-cyano-4-phenylpyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one **7** (Table 5)

The title compound was prepared as described for **5** from **1** (1.84 g, 10 mmol), benzylidene malononitrile (**iiia**, R = C_6H_5) (1.54 g, 10 mmol) and piperidine. IR ν per cm: 3479, 3276–2924, 2206, 1662, 1637, 1551, 1484, 1449. ^{1}H-NMR (δ ppm): 2.09 (s, 3H, CH$_3$), 2.34 (t, 2H, –C*H*$_2$CH$_2$O–), 3.44 (t, 2H, CH$_2$C*H*$_2$*O*–), 4.67 (bs, 1H, *H*OCH$_2$CH$_2$), 7.57–7.62 (m, 3 Ar–*H*), 7.88 (d, 2 Ar–*H*), 8.08 (bs, 2H, NH$_2$). MS m/z (%): 336 [(0.84), M$^+$]; 305 (100); 277 (1.24); 251 (8.91); 195 (41.43); 169 (9.89); 153 (18.48); 104 (20.47); 53 (14.95).

4.1.5. 1-[6-Amino4-(4-chlorophenyl)-5-cyanopyrimidin-2-yl]-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one **8** (Table 5)

It was prepared as described for **6** from **1** (1.84 g, 10 mmol) and *p*-chlorobenzylidene malononitril (**iiib**, R = *p*-C 1 C_6H_4) (1.89 g, 10 mmol) in ethanolic sodium ethoxide. IR ν per cm: 3410, 3082–2825, 2201, 1656, 1597, 1538, 1493, 1417. ^{1}H-NMR (δ ppm): 1.89 (s, 3H, C*H*$_3$), 3.25 (t, 2H, -*CH*$_2$CH$_2$O–), 3.45 (t, 2H, CH$_2$C*H*$_2$O–), 7.2 (bs, 2H, NH$_2$), 7.59, 7.95 (two d, 4 Ar–*H*). ^{13}C-NMR (δ ppm): 13.87 (CH$_3$);

Table 5
Experimental data of 1-(Imidazol-2-yl and pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-ones **2–13**

Compound	R	M.P. (°C)	Yield (%)	Formula [a]
2	C_6H_5–	278–280	43	$C_{15}H_{16}N_4O_2$.HBr
3	C_6H_4Cl (*p*)	240–241	35	$C_{15}H_{15}N_4O_2Cl$.HBr
4	$C_6H_4CH_3$ (*p*)	293–294	36	$C_{16}H_{18}N_4O_2$.HBr
5	C_6H_5–	310–311	27	$C_{17}H_{15}N_5O_3$
6	C_6H_4Cl (*p*)	204–205	75	$C_{17}H_{14}N_5O_3Cl$
7	C_6H_5–	262–263	33	$C_{17}H_{16}N_6O_2$
8	C_6H_4Cl (*p*)	> 350	32	$C_{17}H_{15}N_6O_2$
10	–	111–112	35	$C_{12}H_{14}N_4O_5$
11	–	230–232	64	$C_{11}H_{14}N_4O_3$
13	–	265–267	67	$C_{17}H_{17}N_5O_4$

[a] (C, H, N) microanalytical data are within ± 0.4% of the theoretical values.

27.80 (CH_2–O); 63.14 (CH_2); 117.41 (CN); 168.16 (C=O); 128.76, 130.79 (C-2, 6 and C-3, 5 of phenyl, respectively); 92.68, 165.31 (C-3 and C-4 of pyrazolinone, respectively); 135.84, 135.93 (C-1 and C-4 of phenyl, respectively); 79.89, 152.90, 157.40, 165.49 (C-4,5,2,6 of pyrimidine, respectively). MS m/z (%): 371 [(0.25), M^+]; 294 (5.00); 280 (14.34); 213 (12.50); 172 (19.40); 149 (40.07); 119 (10.88); 55 (100).

4.1.6. 2-[4-(2-Hydroxyethyl)-3-methyl-5-oxo-3-pyrazolin-1-yl]-6-hydroxy-4-pyrimidineacetic acid **10** (Table 5)

A mixture of **1** (1.84, 10 mmol), dimethyl acetonedicarboxylate (**iv**) (1.7 g, 10 mmol) and anhydrous potassium carbonate (0.7 g, 5 mmol) was refluxed in absolute ethanol (20 ml) for 48 h. After cooling the separated potassium salt was filtered, dissolved in ice-cooled water, acidified while stirring with a cold solution of 2 N hydrochloric acid to pH 2–3. The separated white crystalline product was filtered, purified by dissolving in cold 5 N sodium hydroxide and filtered from any turbidity. The filtrate was neutralized by drop-wise addition of cold 5 N hydrochloric acid, the product was filtered, washed with water and dried. IR ν per cm: 3381, 2931, 2881, 1638, 1562, and 1433. ^{1}H-NMR (δ ppm): 2.17 (s, 3H, C*H*$_3$), 2.22 (s, 2H, C*H*$_2$CO$_2$H), 2.32 (t, 2H, –C*H*$_2$CH$_2$O–), 3.42 (t, 2H, –CH$_2$C*H*$_2$O), 4.63 (t,1H, –CH$_2$CH$_2$O*H*), 6.12 (s,1H at C-5 of pyrimidine).

4.1.7. 1-(4-Methyl–6-hydroxypyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one **11** (Table 5)

The title compound was obtained by heating **10** (0.2 g) in dimethylformamide for 1 h. The separated product was filtered and crystallized from DMF.

IR ν per cm: 3263, 3074–2886, 1673, 1642, 1588, 1562. ^{1}H -NMR (δ ppm): 2.17, 2.21 (two s, 2 C*H*$_3$), 2.32 (t, 2H, –*CH*$_2$CH$_2$O–), 3.43 (t, 2H, –CH$_2$*CH*$_2$O–), 4.6 (bs, 1H, –CH$_2$CH$_2$O*H*), 6.11 (s, 1H at C-5 of pyrimidine).

4.1.8. 1-(4-Benzoylamino-6-hydroxypyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one **13** (Table 5)

A mixture of **1** (0.55 g, 3 mmol) and ethoxymethylene-2-phenyl-2-oxazolin-5-one (**12**) (0.65 g, 3 mmol) was refluxed in ethanol (20 ml) in presence of anhydrous potassium carbonate (0.42 g, 1.5 mmol) for 3 h. The separated product was filtered, stirred in water and neutralized with 2 N hydrochloric acid to pH 3–4. IR ν per cm: 3380, 3140–2880, 1660, 1620, 1540, 1510, and 1490. ^{1}H-NMR (δ ppm): 2.25 (s, 3H, *CH*$_3$), 2.40 (bs, 2H, –C*H*$_2$CH$_2$O–), 3.55 (t, 2H, –CH$_2$C*H*$_2$O–), 4.65 (t,1H, –CH$_2$CH$_2$O*H*), 7.47–7.70 (m, 3 Ar–*H*), 7.95 (d, 2 Ar–*H*), 8.65 (s, 1H at C-5 of pyrimidine), 9.3 (bs,1H, CON*H*C$_6$H$_5$).

4.1.9. 2-(5-Arylimidazol-2-yl)-1,2,4,5,6,7-hexahydro-3-H-indazol-3-one hydrobromides **16–18** (Table 6)

These compounds were prepared as described for **2–4** from 2-amidino-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one **15** (1.8 g, 10 mmol) and the appropriate phenacyl bromide **i(a–c)** (10 mmol). The separated crystalline products were filtered, dried and recrystallized from ethanol. IR ν per cm: 3480–3360, 3227–2798, 1698–1693, 1552–1540, 1485–1475, 1449–1448, 1411–1410.

^{1}H-NMR (**16**) δ ppm: 1.70 (m, 4H, –CH$_2$(C*H*$_2$)$_2$CH$_2$–), 2.25, 2.70 (two t, 4H –C*H*$_2$(CH$_2$)$_2$C*H*$_2$–), 7.30–7.45 (m, 3 Ar-*H*), 7.63 (s,1H, C-5 of imidazole), 7.73 (d, 2 Ar–*H*), 8.50 (bs, 1H, NH–Imidazol).

^{1}H-NMR (**18**) δ ppm: 1.67 (m, 4H, –CH$_2$(C*H*$_2$)$_2$CH$_2$–), 2.23, 2.68 (two t, 4H, –C*H*$_2$(CH$_2$)$_2$C*H*$_2$–), 2.32 (s, 3H, C*H*$_3$), 7.58,7.60 (two d, 4 Ar–*H*), 8.63 (s,1H, NH– of imidazole).

4.1.10. 2-[4-Aryl-5-cyano-6-hydroxypyrimidin-2-yl]-1,2,4,5,6,7-hexahydro-3H-indazol-3-ones **19, 20** (Table 6)

They were prepared as described for **6** from **15** (1.8 g, 10 mmol) and the appropriate ethyl arylidenecyanoacetate **ii** (**a**, **b**) (10 mmol) in ethanolic sodium ethoxide. The separated products were filtered, dried and crystallized from ethanol. IR ν per cm: 3443–3441, 3217, 2856, 2216–2215, 1656, 1592–1587, 1536, 1487.

^{1}H-NMR (**19**) δ ppm: 1.73 (m, 4H, –CH$_2$(C*H*$_2$)$_2$CH$_2$–), 2.00, 2.56 (two t, –C*H*$_2$(CH$_2$)$_2$C*H*$_2$–), 7.58–7.64 (m, 3 Ar–*H*), 8.03 (d, 2 Ar–*H*).

^{1}H-NMR (**20**) δ ppm: 1.69 (m, 4H, –CH$_2$(C*H*$_2$)$_2$CH$_2$–), 2.19, 2.56 (two t, –C*H*$_2$(CH$_2$)$_2$C*H*$_2$–), 7.69, 8.06 (dd, 4 Ar–*H*).

4.1.11. 2-(6-Amino-5-cyano-4-phenylpyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one **21** (Table 6)

The title compound was prepared as described for **5** from **15** (1.8 g, 10 mmol) and benzylidene malononitrile (**iiia,** R = C$_6$H$_5$) (10 mmol) in the presence of piperidine. The separated product was filtered, dried and crystallized from ethanol. IR ν per cm: 3485, 3279–2962, 2209, 1663, 1627, 1552, 1515, 1551, 1430.^{1}H-NMR (δ ppm): 1.64–170 (m, 4H, –CH$_2$(C*H*$_2$)$_2$CH$_2$–), 2.11, 2.31 (two t, –C*H*$_2$(CH$_2$)$_2$C*H*$_2$–), 7.56–7.60 (m, 3 Ar–*H*), 7.90 (d, 2 Ar–*H*), 8.78 (s, 2H, NH$_2$), 12.01 (s, 1H, NH).

4.1.12. 2-[6-Amino-4-(4-chlorophenyl)-5-cyanopyrimidin-2-yl]-1,2,4,5,6,7-hexahydro-3H-indazol-3-one **22** (Table 6)

This compound was prepared as described for **6** from **15** (1.8 g, 10 mmol) and *p*-chlorobenzylidine malononitrile (**iiib,** R = *p*-ClC$_6$H$_4$) (10 mmol) in ethanolic sodium ethoxide. The

Table 6
Experimental data of 2-(Imidazol-2-yl and pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **16–29**

Compound	R	M.P. (°C)	Yield (%)	Formula [a]
16	C$_6$H$_5$–	282–283	48	C$_{16}$H$_{16}$N$_4$O.HBr
17	C$_6$H$_4$Cl (*p*)	240–241	31	C$_{16}$H$_{15}$N$_4$OCl.HBr
18	C$_6$H$_4$CH$_3$ (*p*)	292–293	34	C$_{17}$H$_{18}$N$_4$O.HBr
19	C$_6$H$_5$–	263–264	35	C$_{18}$H$_{15}$N$_5$O2
20	C$_6$H$_4$Cl (*p*)	280–281	50	C$_{18}$H$_{14}$N$_5$O$_2$Cl
21	C$_6$H$_5$–	281–282	20	C$_{18}$H$_{16}$N$_6$O
22	C$_6$H$_4$Cl (*p*)	> 350	32	C$_{18}$H$_{15}$N$_6$OCl
24	–	130–131	35	C$_{13}$H$_{13}$N$_4$O$_4$
25	–	220–222	61	C$_{12}$H$_{14}$N$_4$O$_2$
27	–	245–246	83	C$_{14}$H$_{16}$N$_4$O$_2$
29	–	217–218	10	C$_{15}$H$_{18}$N$_4$O2

[a] (C, H, N) microanalytical data are within ± .4% of the theoretical values.

separated product was filtered, dried and crystallized from DMF. IR ν per cm: 3411, 3081–2888, 2201, 1657, 1593, 1538, 1493, 1415.

4.1.13. 6-Hydroxy-2-(3-oxo-1,2,4,5,6,7-hexahydro-3H-indazol-2-yl)-4-pyrimidine-acetic acid **24** (Table 6)

It was prepared as described for **10** from **15** (1.8 g, 10 mmol), dimethyl acetonedicarboxylate (**iv**) (1.7 g, 10 mmol) and anhydrous potassium carbonate (0.7 g, 5 mmol). ^{1}H-NMR (δ ppm): 1.26 (m, 4H, –CH_2(CH_2)$_2$–CH_2–), 2.16, 2.49 (two t, 4H, –CH_2(CH_2)$_2$$CH_2$–), 3.51 (s, 2H, CH_2CO_2H), 6.09 (s, 1H at C-5 of pyrimidine).

4.1.14. 2-(4-Hydroxy-6-methylpyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one **25** (Table 6)

The title compound was obtained by heating **24** (0.2 g) in dimethylformamide for 1 h. The separated product was filtered and recrystallized from DMF. IR ν per cm: 3303, 2928–2842, 1662, 1591, 1500, 1500, 1419. ^{1}H-NMR (δ ppm): 1.66 (m, 4H, –CH_2(CH_2)$_2$$CH_2$–), 2.16, 2.49 (two t, –$CH_2$($CH_2$)$_2$$CH_2$–), 2.20 (s, 3H, CH_3), 5.97(s, 1H at C-5 of pyrimidine), 12.50 (s, 1H, NH).

4.1.15. 2-(6,7-Dihydro-4-oxo-3H,5H-cyclopenta[d]pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one **27** (Table 6)

A solution of **15** (1.8 g, 10 mmol) and ethyl cyclopentanone-2-carboxylate **26** (1.5 ml, 10 mmol) in absolute ethanol (40 ml) was refluxed in the presence of anhydrous potassium carbonate (0.7 g, 5 mmol) for 5 h. After cooling the separated potassium salt of the title compound was filtered, dissolved in water and acidified with 2 N hydrochloric acid to pH (3,4). The obtained white product was filtered, washed with water, dried and crystallized from acetonitril. IR ν per cm: 3288, 2936, 1647, 1596, 1570, 1555, 1536, 1502. ^{1}H-NMR (δ ppm): 1.67 (m, 4H, –CH_2(CH_2)$_2$$CH_2$), 1.99, 2.16 (two t, –C$H_2$($CH_2$)$_2$$CH_2$–), 2.49 (m, 2H, –$CH_2CH_2CH_2$–), 2.61, 2.78 (two t, 4H, –$CH_2CH_2CH_2$–), 12.5 (s, 1H, NH).

4.1.16. 2-(5,6,7,8-Tetrahydro-4-oxo-3H-quinazolin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one **29** (Table 6)

It was prepared as described for **27** from **15** (1.8 g, 10 mmol) and ethyl cyclohexanone-2-carboxylate (**28**) (1.6 ml, 10 mmol). IR ν per cm: 3307, 2939, 2852, 1645, 1600, 1537, 1519, 1501. ^{1}H-NMR (δ ppm): 1.66, 1.72 (two m, 8H, two –CH_2(CH_2)$_2$$CH_2$–), 2.17, 2.32, 2.50, 2.53 (four t, 8H, two –CH_2(CH_2)$_2$$CH_2$–), 12.42 (bs, 1H, NH).

4.2. Pharmacological evaluation

Albino rats of both sexes (pregnant females excluded), weighing 200–250 g (unless otherwise specified), in groups of five rats were used to test the following pharmacological activities.

4.2.1. Antiinflammatory activity (Tables 1, 2)

4.2.1.1. Dextran-induced paw edema in rats (Table 1). A solution of dextran (6% w/v) in 0.9% sodium chloride saline, 0.1 ml, was injected into the subplanter region of the left hind paw 1 h after the oral administration of the test compound (at a dose level of 50 mg/kg). The paw volume was then measured and re-measured again 1, 2, 3 and 4 h after administration of dextran. One group of five rats was kept as control and one group received the standard drug indomethacin (at a dose of 5 mg/kg) [20]. The percentage antiinflammatory activity was calculated by the formula % antiinflammatory = $(1 - dt/dc) \times 100$, where dt = difference in paw volume in drug treated groups; dc = difference in paw volume in control treated group.

4.2.1.2. Determination of ED_{50} (Table 1). Some of the most active compounds from both series (**2, 8, 10, 16, 24** and **29**) were further tested at 5, 10, 20, 40 and 50 mg/kg body weight and the ED_{50} was determined by measuring the inhibition of edema volume 3 h after the dextran injection.

4.2.1.3. Determination of serum transaminases in arthritic rats (Table 2). Formladehyde (2% v/v) solution, 0.1 ml, was injected in the first and third day into the left hind paw just beneath the planter aponeurosis to induce arthritis. The test compounds were administered daily orally for 7 days and serum was obtained on the eighth day [20]. The levels of both serum AST and ALT were measured according to the method described by Reitman and Frankel [21].

4.2.2. Ulcerogenic activity

Groups of five rats each weighing 180–200 g and fasted for 24 h were used. Drugs were given orally at a dose of 300 mg/kg po, and administered twice at 2 h interval. Rats were killed by ether inhalation 6 h after the first dose. Their stomachs were removed, opened along the greater curvature and examined for the presence of gastric ulcers or hyperemia [22].

4.2.3. Acute toxicity

Groups of five rats each were fasted for 24 h prior to the administration of the tested compounds. The compounds were screened at graded doses (0.1–3.0 g/kg, po) for their acute lethal doses (ALD_{50}) and the mortalities were recorded at each dose level after 24 h.

4.2.4. Analgesic activity (Table 3)

Analgesic activity was determined using tail withdrawal response to immersion of rat tail in water at 55 °C [23]. The test compounds were administered orally at a dose of 50 mg/kg and indomethacin was used as a reference drug (5 mg/kg). The recorded values were the average of five administrations ± S.E. and the percentage increase of the reaction time was calculated in comparison with the basal values. The analgesic activity for compounds (**2, 8, 10, 16, 24** and **29**) were determined at 5, 10, 20, 40 and 60 mg/kg body weight in order to determine their ED_{50} values (Table 2).

4.2.5. Antipyretic activity (Table 4)

The antipyretic activity of the test compounds on the feverish body temperature was determined following a reported procedure [24]. Groups of five fasted rats (24 h) were injected subcutaneously with brewer's yeast in physiological saline at a dose of 150 mg/kg of body weight. After 17 h the initial body temperature was measured and the test compounds were administered orally at a dose of 50 mg/kg. The body temperature was recorded after 1, 2, 4 and 6 h from the administration of the test compound. The percentage increase of the activity was calculated in comparison with the basal values and indomethacin was used as a reference drug (5 mg/kg).

References

[1] L. Knorr, Chem. Ber. 16 (1883) 2597.
[2] I. Matsuuro, Togashi, S. Matsubara, et al., Japan Kokai 74, 43, 985, Chem. Abstr. 81 (1974) 49699w.
[3] L.F. Lee, S.R. Bertenshaw, P.C.T. Int. Appl. WO 95 15, 315, Chem. Abstr. 123 (1995) 340111g.
[4] M. Matsuo, O.T. Jsujik, N. Konishi, Eur Pat. Appl. Ep 554, 829, Chem. Abstr. 120 (1994) 8589r.
[5] S. Sharma, V.K. Srivastava, A. Kumar, Eur. J. Med. Chem. 37 (2002) 689–697.
[6] B. Maggio, G. Daidone, D. Raffa, et al., Eur. J. Med. Chem. 36 (2001) 737–742.
[7] E. Bansal, V.K. Srivastava, A. Kumar, Eur. J. Med. Chem. 36 (2001) 81–92.
[8] H.P. Rang, M.M. Dale, J.M. Ritter, Pharmacology, Churchill Livingstone, 1995 (pp.246–255).
[9] E. Cullen, J. Pharm. Sci. 73 (1984) 579–589.
[10] J.F.F. Reynold, Martindale, the Extra Pharmacopeia, 30th ed, Pharmaceutical Press, London, 1993 (pp. 1–36).
[11] P. Bruneau, C. Delvare, M.P. Edwards, R.M. McMillan, J. Med. Chem. 34 (1991) 1028–1036.
[12] Asahi Chemical Industry Co, Jpn. Kokai Tokkyo J.P. 58, 159, 471, Chem. Abstr. 101 (1984) 55095u.
[13] Asahi Chemical Industry Co, Jpn. Kokai Tokkyo J.P. 58, 159, 473, Chem. Abstr. 100 (1984) 85688m.
[14] E. Tse, L. Buttner, Y. Huang, I.H. Hall, Arch. Pharm. Pharm. Med. Chem. 329 (1996) 35–40.
[15] S. Schenone, D. Bruno, A. Ranise, IL Farmaco 58 (2003) 845–849.
[16] E.A.M. Badawey, I.M. El-Ashmawey, Eur. J. Med. Chem. 33 (1998) 349–361.
[17] T. Naito, T. Yoshikarawa, S.I. Kitahara, N.E. Aoki, Chem. Pharm. Bull. (Tokyo) 17 (1969) 1467–1478.
[18] Y. Oshima, T. Akimoto, W. Tsukada, T. YamasAKI, Zigmond, K. Yamaguchi, H. Kojima, Chem. Pharm. Bull. (Tokyo) 17 (1969) 1492–1497.
[19] O.W. Foye, T.L. Lemke, D.A. Williams, Principle of medicinal chemistry, 4th edition, Lea & Febiger Book, 1995 (pp. 554).
[20] G.B. Singh, S. Singh, S. Bani, B.D. Gupta, S.K. Banerjee, J. Pharm. Pharmacol. 44 (1992) 456–458.
[21] S. Reitman, S. Frankel, Am. J. Clin. Path. 26 (1957) 56–60.
[22] G. Daidone, B. Maggio, D. Raffa, Eur. J. Med. Chem. 29 (1994) 707–711.
[23] P.A.J. Janssen, D.N. Kellett, J.E. Davis, Arz. Forsch. 13 (1963) 502–507.
[24] F. Manna, F. Chimenti, A. Bolasco, Eur. J. Med. Chem. 27 (1992) 633–639.