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Nonsteroidal antiinflammatory agents – Part 1: Antiinflammatory, analgesic and antipyretic activity of some new 1-(pyrimidin-2-yl)-3-pyrazolin-5-ones and 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones☆

El-Sayed A.M. Badawey[a]*, Ibrahim M. El-Ashmawey[b]

[a]*Department of Pharmaceutical Chemistry, Faculty of Pharmacy, University of Alexandria, Alexandria, Egypt*
[b]*Department of Pharmacology, Faculty of Veterinary Medicine, University of Alexandria, Alexandria, Egypt*

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Abstract – In our reinvestigation of the cyclocondensation reaction of aminoguanidine bicarbonate **1** with 2-acetylbutyrolactone **2** and ethyl cyclohexanone-2-carboxylate **6**, we have obtained the respective 1-amidino-3-pyrazolin-5-one derivative **3** and the 2-amidino-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one **7**. These intermediates were utilized for the synthesis of two novel series of 1-(pyrimidin-2-yl)-3-pyrazolin-5-ones and 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones. Selected analogs from both series (15 compounds) were evaluated for their antiinflammatory activity in an acute and subacute model of inflammation. The analgesic and antipyretic activity of the target compounds were also evaluated. A structure–activity relationship (SAR) comparative study indicated that some compounds from both series exhibited excellent antiinflammatory activity, together with good analgesic and antipyretic activity and were found to be more potent than the reference drugs at a dose of 50 mg/kg, po. In consideration of the efficacy of the compounds in these assays, the 5-phenyl derivative **18** from the 1-(pyrimidin-2-yl)pyrazolinone series, the 5-butyl and 5-phenyl derivatives **26**, **27** from the 2-(pyrimidin-2-yl)indazolone series were further studied at graded doses for their acute toxicity (ALD_{50}) and ulcerogenic activity and were shown to have a large safety margin (ALD_{50} > 4.0 g/kg, po) and devoid of ulcerogenic potentialities when administered orally at a dose of 300 mg/kg. © Elsevier, Paris

2-acetylbutyrolactone / β-keto ester / 1-(pyrimidin-2-yl)-3-pyrazolin-5-one synthesis / 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one synthesis / antiinflammatory activity / analgesic activity / antipyretic activity

1. Introduction

The NSAIDs are among the most widely used of all therapeutic agents. They are useful in the treatment of rheumatoid arthritis and some inflammatory diseases. However, long-term use of the NSAIDs has been associated with gastrointestinal ulceration, bleeding and nephrotoxicity [1]. Therefore, investigation of new antiinflammatory agents are still a challenge [2–4]. The currently available NSAIDs belong to different chemical classes [5]. As we are dealing in the present investigation with the synthesis and pharmacological evaluation of some substituted pyrazolinones and their interrelated indazolones (*figures 1–3*) as antiinflammatory, analgesic and antipyretic agents, we have focused in this survey on these two chemical classes. The literature indicated that many pyrazole and indazole derivatives have found their clinical application as NSAIDs. Phenylbutazone (*figure 4*), a prototype of pyrazolidinediones NSAIDs, is a very potent antiinflammatory, however, its use in some countries became restricted to ankylosing spondylitis because of its side effects [1]. Feprazone, the 4-(methylbutenyl)-analog, is comparable to phenylbutazone in efficacy but with fewer side effects on the GI tract [5]. Several related pyrazolidine-3,5-diones, pyrazolin-3-ones and pyrazolin-5-ones are also available as NSAIDs; examples are feclobuzone, mefobutazone, suxibuzone, benzpiperylone, morazone, famprofazone, nifenazon and ramifenazone [6]. In 1969, Daiichi Seiyaku research laboratory started a research project to investigate the antiinflammatory activity of several 1- and 2-(pyrimidin-2-yl)pyrazoles [7, 8]. This led in 1973 to the discovery of epirizole (*figure 4*), a japanese drug which has a good antiinflammatory activity with less toxicity. Interestingly, this product proved to inhibit gastric lesion induced by acidic NSAIDs [5]. With regard to the antiinflammatory

☆Dedicated to Prof. S.M. Rida, on the occasion of her 60th birthday

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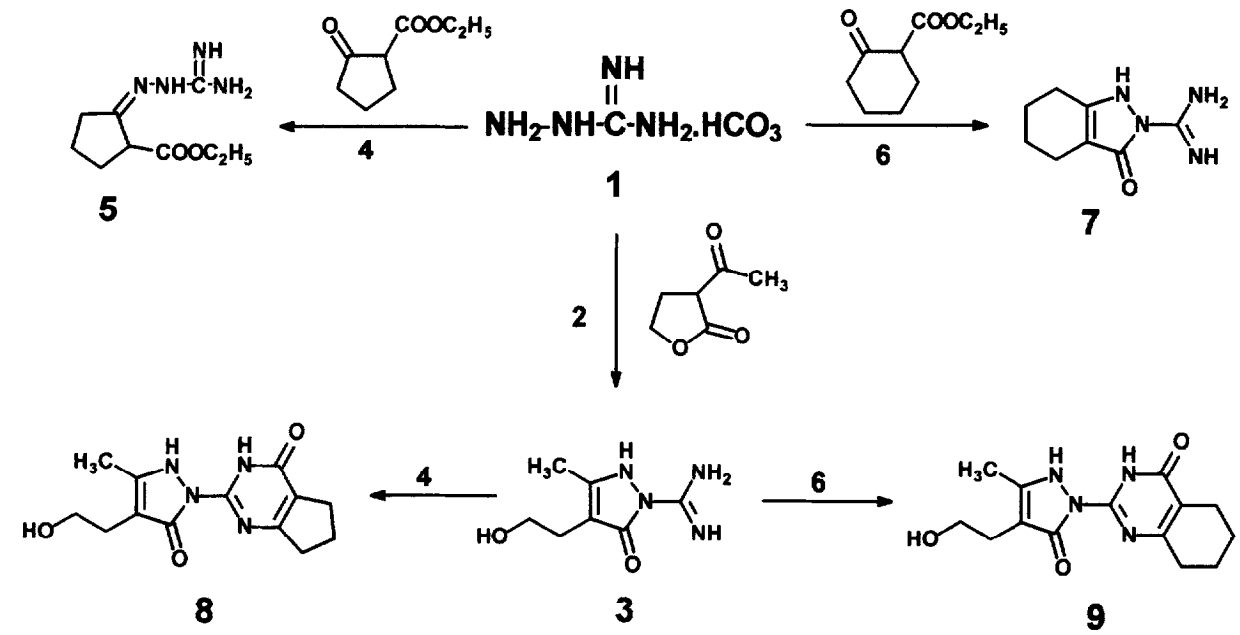
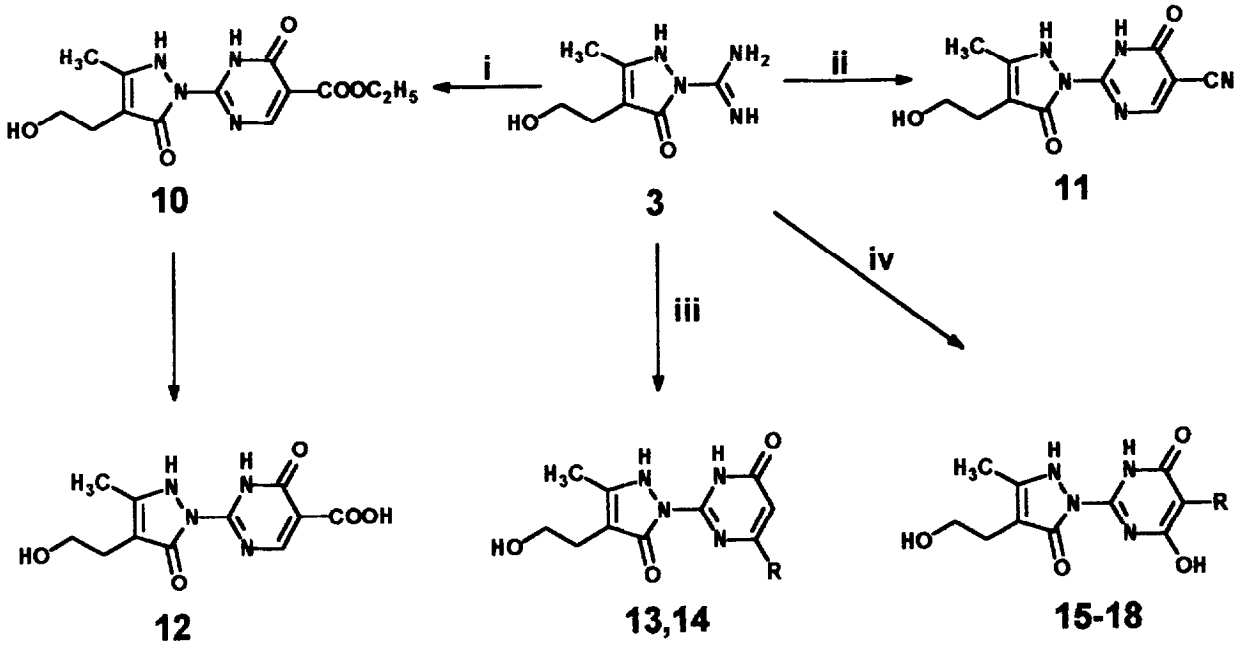


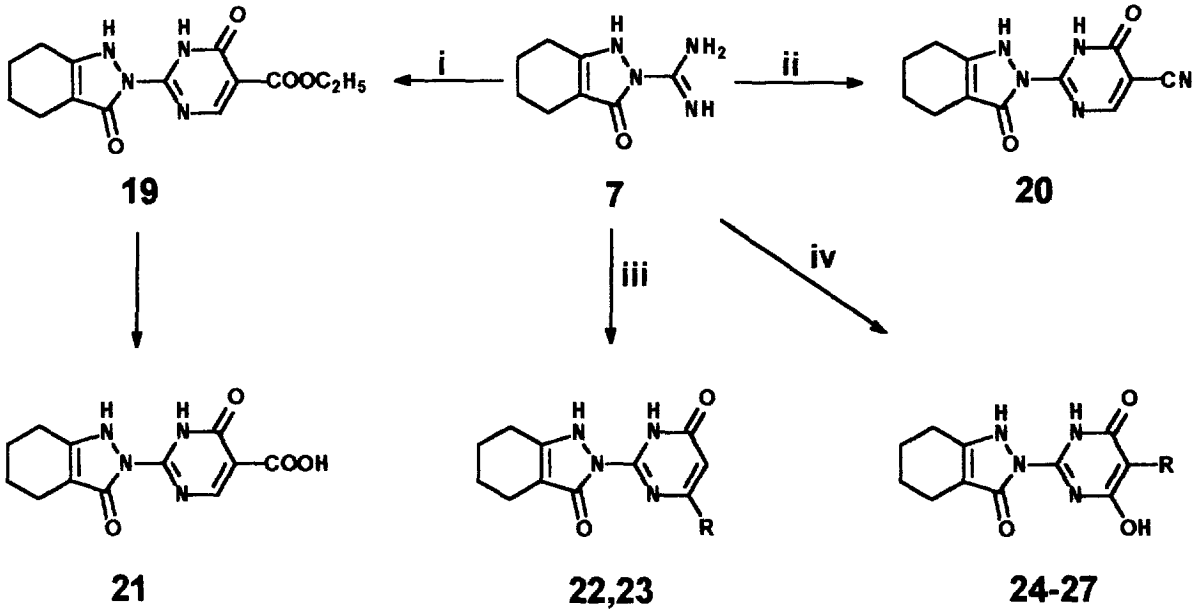
Figure 1.



i = C_2H_5O-CH=C($COOC_2H_5$)$_2$, **ii** = C_2H_5O-CH=C(CN)$COOC_2H_5$, **iii** = R-CO-$CH_2COOC_2H_5$, **iv** = R-CH($COOC_2H_5$)$_2$;

13: R = CH_3, **14**: R = C_6H_5; **15**: R = CH_3, **16**: R = C_2H_5, **17**: R = C_4H_9-n, **18**: R = C_6H_5

Figure 2.



i = C_2H_5O-CH=C($COOC_2H_5$)$_2$, **ii** = C_2H_5O-CH=C(CN)$COOC_2H_5$, **iii** = R-CO-$CH_2COOC_2H_5$, **iv** = R-CH($COOC_2H_5$)$_2$;

22: R = CH_3, **23**: R = C_6H_5; **24**: R = CH_3, **25**: R = C_2H_5, **26** : R = C_4H_9-n, **27**: R = C_6H_5

Figure 3.

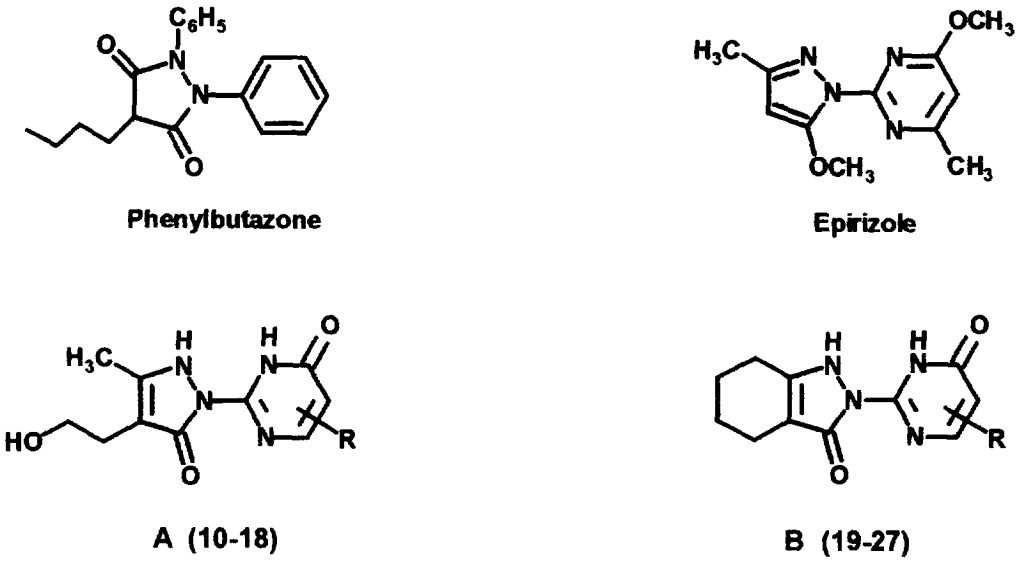


Figure 4.

indazoles, the literature survey indicated that some of them are clinically useful as topical NSAIDs; examples are bendazac, benzydamine and tetrahydroindazole [6]. Additionally, some N-substituted indazoles and indazol-3-ones were reported to possess good antiinflammatory activity [9–12]. Surprisingly, some of these indazoles were proved to exhibit anti-ulcer properties [11, 12].

Encouraged by the fact that the 1-pyrimidinylpyrazole derivative (epirizole, *figure 4*) and some N-substituted indazoles exhibited good antiinflammatory activity with anti-ulcer activity [5, 11, 12], we decided in this work to investigate the possible antiinflammatory, analgesic and antipyretic activity of some related 1-(pyrimidin-2-yl)-3-pyrazolin-5-ones with the general formula **A** (*figures 2* and *4*). For further informations concerning structure–activity relationship (SAR) comparative studies, we have also evaluated the activity of another related series of 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones with the general formula **B** (*figures 3* and *4*)

2. Chemistry

It has been reported that the cyclocondensation of aminoguanidine salts with some β-keto esters such as ethyl acetoacetate and ethyl benzoylacetate in aqueous alcoholic solution yielded the pyrazolin-5-one derivative **I** (*figure 5*) [13, 14]. However, the use of sodium methoxide in this reaction resulted in quite different products [15], namely, 2,3-diaminopyrimidin-4(3*H*)-ones **II** (*figure 5*). On the other hand, the condensation of aminoguanidine bicarbonate **1** with some cyclic β-keto esters such as 2-acetyl-butyrolactone **2**, ethyl cyclopetanone-2-carboxylate **4** and ethylcyclohexanone-2-carboxylate **6** was reported to give the respective 2,3-diaminopyrimidin-4(3*H*)-ones **III–V** (*figure 5*) [16]. According to this procedure [16], compounds **III** and **V** (*figure 5*) were obtained by refluxing **1** and **2** or **6** in *n*-butanol, whereas the cyclopentapyrimidinone **IV** was obtained by reacting **1** with **4** in the presence of sodium methoxide.

As a result of our interest in the use of these cyclic β-keto esters for the synthesis of several heterocycles of biological interest [17–20], we have reinvestigated the condensation of aminoguanidine bicarbonate **1** with 2-acetylbutyrolactone **2**, ethyl cyclopentanone-2-carboxylate **4** and ethyl cyclohexanone-2-carboxylate **6** in refluxing ethanol and in the presence of ammonium acetate (*figure 1*). Interestingly, the reaction of **1** with **2** or **6** afforded the respective 1-amidino-3-pyrazolin-5-one derivative **3** and the 2-amidino-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one **7** which are chemical isomers with both **III** and **V** (*figure 5*) [16]. According to our reaction condition, the formation of

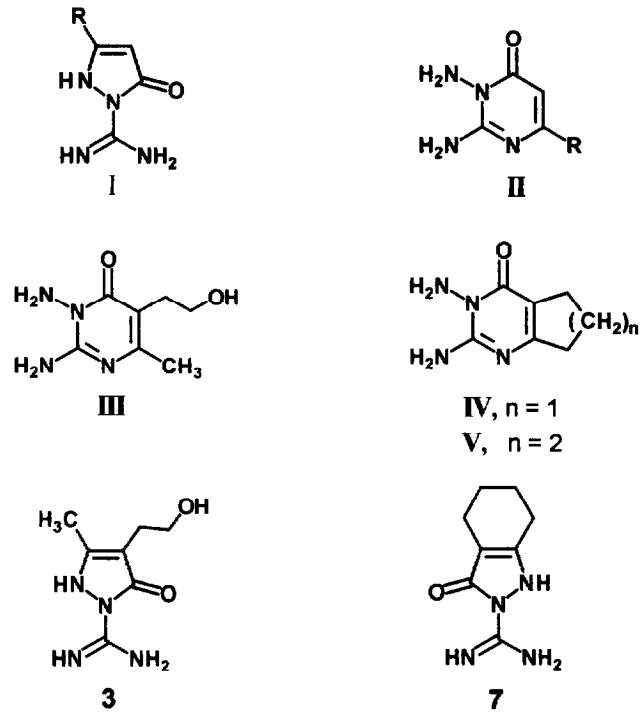


Figure 5.

the pyrazolinone **3** and indazolone derivative **7**, as the sole products, could be attributed to the presence of ammonium acetate which liberates the aminoguanidine base from its salt and in this case it will react with the β-keto esters as a hydrazine derivative. The structures assigned to compounds **3** and **7** were substantiated by ^{1}H-NMR and ^{13}C-NMR spectral data (see the experimental part). The presence of the amidino function was also confirmed by some chemical reactions (*figures 1–3*). Similarly to **3** and **7**, when aminoguanidine bicarbonate **1** was allowed to react with ethyl cyclopentanone-2-carboxylate **4** under identical reaction conditions, compound **5** was isolated, unexpectedly (*figure 1*). Although the IR and ^{1}H-NMR of this product coincided with structure **5**, we were unable to obtain a satisfactory elemental analysis. Its ^{1}H-NMR spectrum showed the following peaks: δ 1.2 (t) and 4.1 (q) for the C-2 ethyl and 3.4 (t) for the C-2 proton.

The 1-amidino-3-pyrazolin-5-one **3** and 2-amidino-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one **7** were utilized for the synthesis of several 1-(pyrimidin-2-yl)-3-pyrazolin-5-ones and 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones (*figures 1–3*). Reacting **3** with ethyl cyclopetanone-2-carboxylate **4** in refluxing ethanol in the presence of potassium carbo-

nate resulted in the respective 1-(cyclopentapyrimidin-2-yl)pyrazolinone derivative **8**, whereas the 1-(tetrahydroquinazolin-2-yl)pyrazolinone derivative **9** was obtained by reacting **3** with ethyl cyclohexanone-2-carboxylate **6** under similar reaction conditions (*figure 1*). Refluxing **3** and diethyl ethoxymethylenemalonate or ethyl (ethoxymethylene)cyanoacetate with ethanol in the presence of potassium carbonate gave the respective 1-(5-ethoxycarbonylpyrimidin-2-yl)-3-pyrazolin-5-one **10** and the 1-(5-cyanopyrimidin-2-yl) analog **11** (*figure 2*). The carboxylic acid derivative 12 was obtained by alkaline hydrolysis of the parent ester **10**. The 1-(6-substituted-pyrimidin-2-yl)-3-pyrazolin-5-ones **13**, **14** were obtained by condensing **3** with ethyl acetoacetate or ethyl benzoylacetate in the presence of potassium carbonate, whereas the 1-(5-substituted-pyrimidin-2-yl)-3-pyrazolin-5-ones **15–18** were obtained by reacting **3** and the appropriate substituted diethyl malonate in the presence of sodium methoxide.

Figure 3 illustrates the synthesis of several 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **19–27** starting from 2-amidino-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one **7**. Thus refluxing **7** and diethyl ethoxymethylenemalonate or ethyl (ethoxymethylene)-cyanoacetate with ethanol in the presence of potassium carbonate gave the respective 2-(5-ethoxycarbonylpyrimidin-2-yl)hexahydroindazol-3-ones **19** and the 2-(5-cyanopyrimidin-2-yl) analog **20**. Alkaline hydrolysis of **19** gave the carboxylic acid derivative **21**. Analogous to **13**, **14**, the 2-(6-substituted-pyrimidin-2-yl)hexahydroindazol-3-ones **22**, **23** were obtained from **7** and ethyl acetoacetate or ethyl benzoylacetate, respectively. On the other hand, the 2-(5-substituted-pyrimidin-2-yl)-hexahydroindazol-3-ones **24–27** were obtained by refluxing **7** and the appropriate substituted diethyl malonate with methanol in the presence of sodium methoxide.

3. Biological results and discussion

3.1. Antiinflammatory (AI) activity (tables I, II)

To assess the AI activity of the designed compounds, selected analogs (15 compounds) in both 1-(pyrimidin-2-yl)pyrazolin-5-one (*figures 1*, *2*) and their corresponding 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-one (*figure 3*) 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 assay was employed as a model for acute inflammation, while the formaldehyde-induced arthritis assay was used as a model for subacute condition.

For the dextran-induced paw model [21], 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, 2 and 3 h after induction and summarized in *table I*. The data indicated good AI activity for the 1-amidinopyrazolinone (**3**, 38.3%, 1 h), while the 2-amidinoindazolone **7** and the 1-(cyclopentapyrimidinyl)-pyrazolinone **8** were less active. Regarding the 1-(5- and 6-substituted-pyrimidin-2-yl)-3-pyrazolin-5-one series (**10–18**, *figure 4*), the data revealed that a significant AI activity was associated with the 6-phenyl (**14**, 44.74%, 3 h) and 5-phenyl substituents (**18**, 40.43%, 1 h). However, a slight decrease in AI activity was recorded for the 6-methyl- (**13**, 39.47%, 3 h) and 5-butyl derivatives (**17**, 31.9%, 1 h). A marked AI activity was attained with the 5-ethyl carboxylate derivative (**10**, 47.37%, 3 h); however, the activity was declined greatly in the free acid (**12**). The AI profiles of the corresponding 2-(5- and 6-substituted-pyrimidinyl)hexahydro-3*H*-indazolones (**19–27**, *figure 4*) demonstrated weak activity for the ester (**19**) and its corresponding acid (**21**), significant activity for the 6-methyl and the 6-phenyl derivatives (**22**, **23**, 31.90%–38.3%, 1 h) and pronounced activity for 5-butyl and 5-phenyl derivatives (**26** and **27**, 55.4%, 1 h).

It is apparent from these results that aryl substitution at C-5 or C-6 of the pyrimidinyl moiety in the pyrazolinone series promoted good AI activity (**14**, **18**), while alkyl substitution resulted in less active compounds (**13**, **17**). The highest activity in this series was recorded for the 5-ethyl carboxylate derivative **10** but it was less potent than the reference drug, whereas in the indazolone series the highest AI activity was associated with alkyl or aryl substituents only at C-5 (**26**, **27**). These compounds were the most active among both series and were more potent than the reference drug. The effects of absorption and biotransformation could be recognized in both series, for instance the maximum AI activity was recorded after 3 h for most of the pyrazolinones (e.g. **10–14**), while their corresponding indazolones (**19–23**) were characterized by rapid action since their maximum effects were attained after 1 h. However, the presence of 5-substituted barbituric acid residue in both series (**17**, **18**; **26**, **27**) promoted a rapid effect.

For the formaldehyde-induced arthritis screen, arthritis was induced by formaldehyde injection in the first and third day and the test compounds were administered orally (at 50 mg/kg daily) for seven days [21]. As a result of inflammation, the level of serum transaminase aspartate aminotransferase (AST) and alanine aminotransferase (ALT) enzymes is increased and a decreased level of these enzymes upon administration of the test compounds will reflect their anti-

Table I. Antiinflammatory activity of some 1-(2-pyrimidinyl)-3-pyrazolin-5-ones **8–18** and 2-(pyrimidin-2-yl)-hexahydro-3*H*-indazol-3-ones **19–27** in paw edema screen.

Compound	Volume of edema[a] (mL ± S.E.)			
	0	1 h	2 h	3 h
Controls	0.36 ± 0.02	0.83 ± 0.01	0.73 ± 0.02	0.74 ± 0.02
3	0.43 ± 0.01	0.72 ± 0.02 (38.30)	0.70 ± 0.01 (27.03)	0.68 ± 0.02 (34.21)
7	0.38 ± 0.04	0.83 ± 0.01 (04.30)	0.77 ± 0.01 (–5.40)	0.71 ± 0.03 (13.23)
8	0.36 ± 0.01	0.75 ± 0.01 (17.02)	0.71 ± 0.03 (05.41)	0.70 ± 0.03 (10.53)
10	0.46 ± 0.04	0.83 ± 0.04 (21.30)	0.73 ± 0.01 (27.03)	0.66 ± 0.03 (47.37)
12	0.39 ± 0.02	0.80 ± 0.03 (12.80)	0.74 ± 0.02 (05.41)	0.66 ± 0.01 (28.95)
13	0.38 ± 0.01	0.71 ± 0.02 (29.80)	0.65 ± 0.01 (27.03)	0.61 ± 0.01 (39.47)
14	0.43 ± 0.02	0.92 ± 0.04 (–4.30)	0.75 ± 0.01 (13.51)	0.64 ± 0.02 (44.74)
17	0.39 ± 0.03	0.71 ± 0.03 (31.90)	0.74 ± 0.01 (05.41)	0.76 ± 0.05 (02.63)
18	0.40 ± 0.02	0.68 ± 0.02 (40.43)	0.71 ± 0.04 (16.22)	0.68 ± 0.02 (26.32)
19	0.42 ± 0.01	0.80 ± 0.02 (19.20)	0.80 ± 0.02 (–2.70)	0.79 ± 0.03 (02.63)
21	0.41 ± 0.01	0.78 ± 0.04 (21.30)	0.74 ± 0.05 (10.81)	0.75 ± 0.04 (10.53)
22	0.42 ± 0.01	0.71 ± 0.03 (38.30)	0.75 ± 0.02 (10.81)	0.75 ± 0.01 (13.23)
23	0.40 ± 0.01	0.72 ± 0.02 (31.90)	0.80 ± 0.02 (–8.12)	0.79 ± 0.03 (–2.63)
26	0.47 ± 0.01	0.68 ± 0.08 (55.32)	0.71 ± 0.01 (35.14)	0.67 ± 0.01 (47.37)
27	0.42 ± 0.04	0.63 ± 0.03 (55.32)	0.72 ± 0.02 (18.92)	0.68 ± 0.02 (31.58)
Indomethacin	0.39 ± 0.01	0.61 ± 0.01 (53.20)	0.59 ± 0.01 (46.00)	0.57 ± 0.01 (52.63)

[a]In parentheses the percentage of antiinflammatory activity (AI); see the experimental part.

inflammatory potentialities. Accordingly, we found that the assessment of the level of AST and ALT enzymes provides a good and simple tool to measure the AI activity of the target compounds [22]. Indomethacin (at 5 mg/kg daily) and acetylsalicylic acid (ASA) (at 50 mg/kg daily) were used as reference drugs and the results are recorded in *table II*. The data indicated that the 1-amidinopyrazolinone **3** and the 2-amidinoindazolone **7** were equipotent as aspirin, while the 1-(cyclopentapyrimidinyl) pyrazolinone **8** was nearly similar to indomethacin in potency. In the 1-(5- and 6-substituted-pyrimidin-2-yl)-3-pyrazolin-5-one series **10–18**, good AI activity was observed with the 6-methyl (**13**), 6-phenyl (**14**) and 5-phenyl (**18**) substituents while a weak activity was associated with the 5-butyl substituent (**17**). The AI profiles of the related 2-(5- and 6- substituted-pyrimidin-2-yl)indazol-3-one series **19–27** demonstrated weak activity for the 6-methyl and 6-phenyl compounds **22**, **23** and excellent activity for the 5-butyl and 5-phenyl derivatives **26**, **27**. Unlike the paw edema screen, the 5-carboxylic acid derivatives in both series (**12**, **21**) revealed good activity, while their parent ethyl esters (**10**, **19**) were less active. Several compounds from both series (e.g. **13**, **14**, **18**, **21**, **26** and **27**) were found to be more potent than indomethacin and acetylsalicylic acid. However, the most potent compounds were the 5-phenyl derivative **18** from the pyrazolinone series and the 5-butyl and 5-phenyl derivatives **26**, **27** from the indazolone series. Again it is obvious that the AI activity was greatly enhanced by the presence of a 5-substituted-barbituric acid moiety in both series (e.g. **18**, **26**, **27**). For this reason, these compounds 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 p.o., when administered twice at 2 h interval in fasted rats. At lower dose levels aspirin and phenylbutazone were reported to cause serious gastric ulcers [23].

3.3. Acute toxicity (ALD_{50})

The compounds showed a high safety margin when screened at graded doses (0.1–4.0 g/kg, p.o.) for their acute lethal doses (ALD_{50}). The ALD_{50} were found to be > 4.0 g/kg.

Table II. Antiinflammatory activity of some 1-(2-pyrimidinyl)-3-pyrazolin-5-ones **8–18** and 2-(pyrimidin-2-yl)hexahydro-3*H*-indazol-3-ones **19–27** in formaldehyde-induced arthritis assay.

Compound	Level of AST		Level of ALT	
	(u/L) ± S.E.[a]	% AI	(u/L) ± S.E.[a]	% AI
Controls without arthritis	60.22 ± 2.50	–	54.22 ± 3.50	–
Controls with arthritis	71.13 ± 5.41	–	71.13 ± 4.03	–
3	66.01 ± 4.22	(46.93)	61.22 ± 5.10	(58.60)
7	65.00 ± 4.00	(56.19)	63.47 ± 3.82	(45.30)
8	65.21 ± 4.01	(54.26)	60.24 ± 3.93	(64.40)
10	67.23 ± 3.50	(35.75)	61.57 ± 6.24	(56.53)
12	66.21 ± 3.50	(45.10)	57.97 ± 3.57	(77.82)
13	63.20 ± 6.21	(72.69)	58.72 ± 5.14	(73.39)
14	64.10 ± 5.21	(64.44)	59.22 ± 2.44	(70.43)
17	68.21 ± 5.00	(26.76)	64.44 ± 4.92	(39.56)
18	61.01 ± 5.50	(92.76)	56.34 ± 6.21	(87.46)
19	70.01 ± 2.40	(10.27)	66.97 ± 2.56	(24.60)
21	63.20 ± 6.22	(72.69)	59.22 ± 4.00	(70.43)
22	68.40 ± 5.50	(25.02)	62.51 ± 4.33	(50.98)
23	69.30 ± 6.20	(16.77)	64.17 ± 5.21	(41.16)
26	62.31 ± 5.30	(80.84)	57.42 ± 6.23	(81.08)
27	63.20 ± 6.20	(72.69)	57.42 ± 3.20	(81.08)
Indomethacin	64.32 ± 3.26	(62.42)	60.32 ± 3.52	(63.93)
Acetylsalicylic acid	66.21 ± 3.23	(45.10)	62.51 ± 3.21	(50.98)

[a]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.

3.4. Analgesic activity (table III)

The analgesic activity was recorded by the hot-plate method [24]. Indomethacin was used as a reference (5 mg/kg, p.o.) and the data are summarized in *table III*. It indicated that the 2-amidinohexahydroindazolone **7** and the cyclopentapyrimidinylpyrazolinone **8** have moderate activity, whereas the 1-amidinopyrazolinone **3** is less active. The results obtained by the 1-(pyrimidin-2-yl)pyrazolinone series **10–18** indicated that a good analgesic activity was obtained when the pyrimidinyl moiety is substituted with 5-carboxylic acid (**12**), 5-butyl (**17**), 5-phenyl (**18**) substituents and a weak activity was exhibited by the 5-ethyl carboxylate (**10**), 6-methyl (**13**) and 6-phenyl (**14**) substituted derivatives. On the other hand, most of the related 2-(pyrimidin-2-yl)indazolones **19–27** demonstrated a moderate to good analgesic activity and the highest activity was recorded for the 5-ethyl carboxylate (**19**) and 5-butyl (**26**) derivatives. Although the compounds in both series showed a variable degree of analgesic activity, they have similar pharmacokinetic profiles as their maximum effects appeared after 2 h. At this time interval, several compounds were found to be more potent than the reference drugs, particularly, those which possess a 5-substituted-barbituric acid structure such as the 5-butyl (**17**) and 5-phenyl (**18**) compounds from the pyrazolinone series and the 5-butyl (**26**) and the 5-phenyl (**27**) compounds from the indazolone series.

3.5. Hypothermic and antipyretic activity (table IV)

The compounds were tested for their hypothermic activity and proved to possess moderate hypothermic activity relative to indomethacin. They were further evaluated for their antipyretic activity [24], using indomethacin as reference drug (*table IV*). The data indicated that most of the pyrazolinones which have a 5-substituted pyrimidinyl moiety exhibited good antipyretic activity, particularly the 5-ethyl carboxylate (**10**), 5-carboxylic acid (**12**), 5-butyl (**17**) and 5-phenyl (**18**) derivatives. On the other hand, moderate antipyretic activity was displayed by most of the 2-(pyrimidin-2-yl)hexahydroindazolones **19–27**.

In conclusion, this study revealed that a number of the tested pyrazolinones and indazolones exhibited good antiinflammatory and analgesic activity. Furthermore, most of the tested pyrazolinones were more active as antipyretics than the corresponding indazolones. In both series the promising activity was always noticeable among the derivatives which carry a 5-substituted pyrimidinyl moiety. The absence of ulcerogenic potentialities and the high safety margin recorded for some of this type of compounds (**18**, **26**, **27**) makes it worthy to investigate the activity of some related new derivatives. This will be the subject of our next publication.

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 recorded on a Perkin-Elmer spectrophotometer using samples in potassium bromide discs. The ^{1}H-NMR spectra were recorded on a Varian Gemini 200 at 200 MHz using (DMSO-d_6). The ^{13}C-NMR (360 MHz) spectra acquisition was performed on a Bruker AM 360 instrument. Microanalyses were performed on a Carlo Erba 1106 analyzer and are within ±0.4 of the theoretical percentages.

4.1.1. 1-Amidino-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one 3

A mixture of aminoguanidine bicarbonate **1** (13.61 g, 100 mmol), 2-acetylbutyrolactone **2** (10.8 mL, 100 mmol) and ammonium acetate (15.42 g, 200 mmol) was refluxed in absolute ethanol (150 mL) for 15 h. After cooling, the product was filtered, washed with ethanol and dried; yield: 14.0 g (76%); m.p. 196–197 °C (EtOH). IR ν cm^{-1} 3500–2000, 1690, 1610, 1450. ^{1}H-NMR: δ 1.9 (s, C*H*$_3$), 2.25 (t, –C*H*$_2$CH$_2$O), 3.4 (t, –CH$_2$C*H*$_2$O), 5.0 (t, O*H*), 7.9 (bs, N*H*$_2$). ^{13}C-NMR: δ 13.0 (CH$_3$), 26.0 (CH$_2$), 61.0 (CH$_2$O), 91.0 (C-3), 153.0 (C-amidino), 155.0 (C-4); 165.0 (C=O). Anal. $C_7H_{12}N_4O_2$ (C, H, N).

4.1.2. Ethyl N-guanidinocyclopentanimine-2-carboxylate 5

A mixture of aminoguanidine bicarbonate **1** (13.61 g, 100 mmol), ethyl cyclopentanone-2-carboxylate **4** (15.0 mL, 100 mmol) and ammonium acetate (15.42 g, 200 mmol) was refluxed in absolute ethanol (150 mL) for 15 h. Excess solvent was removed under vacuum, the residue dissolved in water and the product was extracted with chloroform. The chloroformic extract was dried over anhydrous sodium sulfate, filtered and evaporated under vacuum. The remaining oily product was treated with acetone to give a white crystalline product; yield: 13.0 g; m.p. 107–108 °C (acetonitrile). IR ν cm^{-1}: 3400, 3100–2500, 1730, 1680, 1600, 1550. ^{1}H-NMR: δ 1.2 (t, C*H*$_3$CH$_2$), 1.6–2.4 (m, 3 C*H*$_2$), 3.4 (t, C*H* at C-2), 4.1 (q, CH$_3$–C*H*$_2$), 7.5 (bs, N*H*$_2$ of guanidino).

4.1.3. 2-Amidino-1,2,4,5,6,7-hexahydro-3H-indazol-3-one 7

It was prepared as described for **3**, from aminoguanidine bicarbonate **1** (13.61 g, 100 mmol), ethyl cyclohexanone-2-carboxylate **6** (16.0 mL, 100 mmol) and ammonium acetate (15.42 g, 200 mmol); yield: 7.5 g (41.6%); m.p. 241–242 °C (EtOH). IR ν cm^{-1}: 3600–2200, 1670, 1630, 1550. ^{1}H-NMR: δ 1.6 [m, –CH$_2$(C*H*$_2$)$_2$CH$_2$–], 2.15 and 2.4 [2 t, –C*H*$_2$(CH$_2$)$_2$C*H*$_2$–], 7.8 (bs, N*H*$_2$). Anal. $C_8H_{12}N_4O$ (C, H, N).

4.1.4. 1-(6,7-Dihydro-4-oxo-3H,5H-cyclopenta[d]pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one 8

A solution of **3** (1.84 g, 10 mmol) and ethyl cyclopentanone-2-carboxylate **4** (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. The separated sodium salt of the title compound was filtered, dissolved in water, acidified with hydrochloric acid (pH 3–4) to obtain a white product of **8**; yield 0.6 g (21.7%), m.p. 230–232 °C (DMF). IR ν cm^{-1}: 3300, 3200–2000, 1670, 1600, 1580, 1550. ^{1}H-NMR: δ 2.0 (m, –CH$_2$C*H*$_2$CH$_2$–), 2.2 (s, C*H*$_3$), 2.35 (t, –C*H*$_2$CH$_2$OH), 2.65 and 2.8 (2 t, –C*H*$_2$CH$_2$C*H*$_2$–), 3.5 (t, –CH$_2$C*H*$_2$OH), 4.6 (bs, OH). Anal. $C_{13}H_{16}N_4O_3$ (C, H, N).

Table III. Analgesic activity of some 1-(2-pyrimidinyl)-3-pyrazolin-5-ones **8–18** and 2-(pyrimidin-2-yl)-hexahydro-3*H*-indazol-3-ones **19–27**.

Compound	Reaction times[a] ± S.E.				
	0	30 min	2 h	3 h	4 h
Controls	7.2 ± 0.2	7.4 ± 0.5	7.2 ± 0.4	7.6 ± 0.2	7.3 ± 0.2
3	6.9 ± 0.4	8.2 ± 0.2 (18.8)	8.3 ± 0.1 (20.29)	8.3 ± 0.5 (20.29)	7.8 ± 0.6 (13.04)
7	7.3 ± 0.2	9.5 ± 0.1 (30.14)	10.9 ± 0.1 (49.31)	9.2 ± 0.2 (26.03)	8.6 ± 0.2 (17.81)
8	7.6 ± 0.4	9.6 ± 0.2 (26.32)	11.2 ± 0.4 (47.37)	10.2 ± 0.3 (34.21)	9.8 ± 0.3 (28.95)
10	7.6 ± 0.2	9.3 ± 0.2 (22.37)	10.2 ± 0.4 (34.21)	9.6 ± 0.2 (26.32)	8.6 ± 0.1 (13.16)
12	6.8 ± 0.1	9.8 ± 0.4 (44.12)	10.3 ± 0.2 (51.47)	8.6 ± 0.1 (26.47)	8.2 ± 0.3 (20.59)
13	7.5 ± 0.2	8.9 ± 0.2 (18.67)	9.2 ± 0.6 (22.67)	9.0 ± 0.2 (20.00)	8.8 ± 0.4 (17.33)
14	7.6 ± 0.4	9.5 ± 0.2 (25.3)	9.9 ± 0.4 (30.26)	9.7 ± 0.3 (27.63)	8.2 ± 0.2 (07.89)
17	7.4 ± 0.2	9.7 ± 0.7 (31.08)	12.4 ± 0.5 (67.57)	9.6 ± 0.2 (29.73)	9.3 ± 0.2 (25.68)
18	6.9 ± 0.4	8.7 ± 0.5 (26.08)	11.8 ± 0.6 (71.01)	10.4 ± 0.3 (50.72)	9.8 ± 0.6 (42.03)
19	7.3 ± 0.2	9.7 ± 0.4 (32.88)	11.9 ± 0.5 (63.01)	9.3 ± 0.6 (27.40)	8.9 ± 0.1 (21.92)
21	8.1 ± 0.6	9.3 ± 0.6 (14.81)	12.1 ± 0.2 (49.38)	10.3 ± 0.4 (27.16)	9.6 ± 0.2 (18.52)
22	7.6 ± 0.1	9.7 ± 0.2 (27.63)	11.2 ± 0.4 (47.37)	9.3 ± 0.6 (22.37)	9.0 ± 0.5 (18.42)
23	6.8 ± 0.3	8.9 ± 0.2 (30.88)	10.5 ± 0.5 (54.41)	9.2 ± 0.2 (35.29)	8.6 ± 0.1 (26.47)
26	7.2 ± 0.6	8.9 ± 0.2 (23.61)	11.9 ± 0.4 (65.28)	9.5 ± 0.2 (31.94)	9.2 ± 0.4 (27.78)
27	7.5 ± 0.2	9.6 ± 0.8 (28.00)	11.6 ± 0.3 (54.67)	10.2 ± 0.2 (36.00)	9.2 ± 0.2 (22.67)
Indomethacin[b]	7.4 ± 0.5	11.4 ± 0.4 (54.05)	11.9 ± 0.2 (60.81)	11.8 ± 0.6 (59.46)	11.2 ± 0.4 (51.35)

[a]In parentheses percentage increase of the reaction times calculated in comparison with basal values.
[b]Phenylbutazone and acetylsalicylic acid showed (50%) after 30 min [24].

Table IV. Antipyretic activity of some 1-(2-pyrimidinyl)-3-pyrazolin-5-ones **8–18** and 2-(pyrimidin-2-yl)-hexahydro-3*H*-indazol-3-ones **19–27**.

Compound	Body temperature[a] ± S.E.				
	0	1 h	2 h	4 h	6 h
Controls	38.3 ± 0.1	38.2 ± 0.1	38.2 ± 0.3	38.0 ± 0.1	38.1 ± 0.1
7	38.7 ± 0.1	37.3 ± 0.3 (3.6)	36.3 ± 0.3 (6.2)	36.4 ± 0.2 (5.9)	36.6 ± 0.1 (5.4)
3	38.2 ± 0.1	37.0 ± 0.2 (3.1)	37.2 ± 0.1 (2.6)	36.8 ± 0.1 (3.6)	36.8 ± 0.2 (3.6)
8	37.7 ± 0.2	37.4 ± 0.1 (0.8)	37.1 ± 0.1 (1.6)	36.8 ± 0.4 (2.4)	36.6 ± 0.3 (2.9)
10	37.9 ± 0.4	36.8 ± 0.3 (2.9)	36.2 ± 0.2 (4.5)	36.3 ± 0.1 (4.2)	36.1 ± 0.2 (4.7)
12	38.5 ± 0.1	37.5 ± 0.2 (2.6)	36.5 ± 0.1 (5.2)	36.5 ± 0.2 (5.2)	36.7 ± 0.3 (4.7)
13	37.9 ± 0.1	37.3 ± 0.2 (1.6)	37.1 ± 0.4 (2.1)	36.9 ± 0.1 (2.6)	36.9 ± 0.2 (2.6)
14	38.5 ± 0.2	37.6 ± 0.2 (2.3)	37.1 ± 0.1 (3.6)	36.9 ± 0.2 (4.2)	36.9 ± 0.2 (4.2)
17	37.9 ± 0.3	37.0 ± 0.2 (2.4)	36.8 ± 0.1 (2.9)	37.0 ± 0.2 (2.4)	36.9 ± 0.2 (2.6)
18	37.9 ± 0.2	37.7 ± 0.2 (0.5)	36.7 ± 0.1 (3.2)	36.3 ± 0.3 (4.2)	36.2 ± 0.1 (4.5)
19	37.8 ± 0.2	37.2 ± 0.2 (1.6)	37.1 ± 0.1 (1.9)	37.2 ± 0.3 (1.6)	37.3 ± 0.1 (1.3)
21	38.0 ± 0.1	37.6 ± 0.3 (1.1)	37.0 ± 0.2 (2.6)	36.9 ± 0.1 (2.9)	37.1 ± 0.2 (2.4)
22	37.6 ± 0.3	36.6 ± 0.2 (2.7)	36.9 ± 0.1 (1.9)	36.9 ± 0.2 (1.9)	36.5 ± 0.2 (2.9)
23	38.2 ± 0.1	37.9 ± 0.2 (0.8)	37.8 ± 0.3 (1.0)	37.6 ± 0.2 (1.6)	37.5 ± 0.2 (1.8)
26	37.9 ± 0.1	37.6 ± 0.4 (0.8)	36.6 ± 0.2 (3.4)	37.0 ± 0.2 (2.4)	37.3 ± 0.3 (1.6)
27	37.6 ± 0.3	37.1 ± 0.1 (1.3)	36.2 ± 0.2 (3.7)	36.6 ± 0.1 (2.7)	37.0 ± 0.2 (1.6)
Indomethacin	37.7 ± 0.4	36.7 ± 0.2 (2.7)	36.6 ± 0.1 (3.0)	36.5 ± 0.2 (3.2)	36.3 ± 0.1 (3.7)

[a]Each value represents the mean ± S.E. with the percentage inhibition shown in parentheses.

4.1.5. 1-(5,6,7,8-Tetrahydro-4-oxo-3H-quinazolin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one ***9***

It was prepared as described for **8**, from **3** (1.84 g, 10 mmol) and ethyl cyclohexanone-2-carboxylate **6** (1.6 mL, 10 mmol); yield 1.7 g (58.6%), m.p. 188–190 °C (DMF); IR ν cm^{-1}: 3400, 3300–2200, 1660, 1600, 1570, 1500. ^{1}H-NMR: δ 1.6 [m, $-CH_2(CH_2)_2CH_2-$], 2.2 (s, CH_3), 2.3 (t, $-CH_2CH_2OH$), 2.4 and 2.6 [2 t, $-CH_2(CH_2)_2CH_2-$], 3.5 (t, $-CH_2CH_2OH$), 4.6 (bs, OH). Anal. $C_{14}H_{18}N_4O_3$ (C, H, N).

4.1.6. 1-(5-Ethoxycarbonyl-4-oxo-3H-pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one ***10***

A solution of **3** (1.84 g, 10 mmol) and diethyl ethoxmethylenemalonate (2.0 mL, 10 mmol) in absolute ethanol (40 mL) was refluxed for 1 h in the presence of anhydrous potassium carbonate (0.7 g, 5 mmol). The separated sodium salt of the title compound was filtered and worked up as described for **8**; yield: 3.0 g (97.3%); m.p. 180–182 °C (EtOH). IR ν cm^{-1}: 3500–7400, 1740, 1710, 1650, 1600, 1500. ^{1}H-NMR: δ 1.3 (t, CH_3CH_2-), 2.2 (s, CH_3), 2.4 (t, $-CH_2CH_2OH$), 3.5 (t, $-CH_2CH_2OH$), 4.3 (q, CH_3CH_2), 8.5 (s, H at C-6 of pyrimidine). Anal. $C_{13}H_{16}N_4O_5$ (C, H, N).

4.1.7. 1-(5-Cyano-4-oxo-3H-pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one ***11***

As described for **10**, from **3** (1.84 g, 10 mmol), ethyl (ethoxymethylene)cyanoacetate (1.7 g, 10 mmol) and anhydrous potassium carbonate (0.7 g, 5 mmol); yield: 1.6 g (61.3%); m.p. 220–222 °C (DMF). IR ν cm^{-1}: 3500, 3300–2400, 2220, 1700, 1660, 1590, 1550. ^{1}H-NMR: δ 2.2 (s, CH_3), 2.35 (t, $-CH_2CH_2OH$), 3.5 (t, $-CH_2CH_2OH$), 4.6 (bs, OH), 8.6 (s, H at C-6 of pyrimidine). Anal. $C_{11}H_{11}N_5O_3$ (C, H, N).

4.1.8. 1-(5-Carboxy-4-oxo-3H-pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one ***12***

It was prepared by stirring **10** (1.45 g, 5 mmol) with 1 N sodium hydroxide (25 mL) at 80 °C for 3 h. The product was obtained after cooling and neutralizing the reaction mixture with hydrochloric acid; yield 0.9 g (64.2%), m.p. 219–220 °C (DMF). IR ν cm^{-1}: 3500–2500, 1720, 1650, 1610, 1530, 1490. ^{1}H-NMR: δ 2.2 (s, CH_3), 2.4 (t, $-CH_2CH_2OH$), 3.5 (t, $-CH_2CH_2OH$), 8.6 (s, H at C-6 of pyrimidine). Anal. $C_{11}H_{12}N_4O_5$ (C, H, N).

*4.1.9. 1-(6-Methyl or 6-phenyl-4-oxo-3H-pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one (**13**, **14**; tables V, VI)*

A solution of **3** (1.84 g, 10 mmol) and the appropriate β-keto ester (10 mmol) in absolute ethanol (40 mL) was refluxed for 15 h in the presence of anhydrous potassium carbonate (0.7 g, 5 mmol). After cooling, the separated sodium salt of the respective title compound was filtered, worked up as described for **8** and crystallized from dimethylformamide. IR ν cm^{-1} (**13**, **14**): 3500–2400, 1670, 1640, 1600–1590, 1570–1560.

*4.1.10. 1-(6-Hydroxy-5-substituted-4-oxo-3H-pyrimidin-2-yl)-4-(2-hydroxyethyl)-3-methyl-3-pyrazolin-5-one (**15–18**; tables V, VI)*

A mixture of **3** (1.84 g, 10 mmol) and the appropriate substituted diethyl malonate (10 mmol) was refluxed with methanolic sodium methoxide solution (20 mmol), prepared from (0.4 g) of sodium in absolute methanol (40 mL), for 15 h. The solvent was removed under vacuum, the residue was dissolved in water and extracted with chloroform from any unreacted esters. The aqueous solution was acidified to pH 3–4 with hydrochloric acid to give a white product of the respective title compounds. A gummy product may be formed during acidification, in this case a few mL of ethanol were added with vigorous stirring until the formation of an amorphous product. It was filtered and recrystallized from aqueous ethanol. IR ν cm^{-1} (**15–18**): 3600–2200, 1670–1660, 1640–1620, 1600–1580, 1500.

4.1.11. 2-(5-Ethoxycarbonyl-4-oxo-3H-pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one ***19***

It was prepared as described for **10**, from **7** (1.8 g, 10 mmol), diethyl ethoxymethylenemalonate (2.0 mL, 10 mmol) and anhydrous potassium carbonate (0.7 g, 5 mmol); yield: 2.85 g

Table V. Experimental data of 1-(pyrimidin-2-yl)-3-pyrazolin-5-ones **13–18** and 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **22–27**.

Compound	R	M.p. (°C)	Yield (%)	Formula[a]
13	CH_3	228–229	64	$C_{11}H_{14}N_4O_3$
14	C_6H_5	253–254	45	$C_{16}H_{16}N_4O_3$
15	CH_3	240–241	42	$C_{11}H_{14}N_4O_4$
16	C_2H_5	218–220	29	$C_{12}H_{16}N_4O_4$
17	C_4H_9	250–253	62	$C_{14}H_{20}N_4O_4$
18	C_6H_5	202–203	31	$C_{16}H_{16}N_4O_4$
22	CH_3	220–222	61	$C_{12}H_{14}N_4O_2$
23	C_6H_5	291–293	59	$C_{17}H_{16}N_4O_2$
24	CH_3	282–284	57	$C_{12}H_{14}N_4O_3$
25	C_2H_5	294–296	75	$C_{13}H_{16}N_4O_3$
26	C_4H_9	285–287	69	$C_{15}H_{20}N_4O_3$
27	C_6H_5	305–307	67	$C_{17}H_{16}N_4O_3$

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

Table VI. ^{1}H-NMR spectral data of the 4-(2-hydroxyethyl)-3-methyl-1-(pyrimidin-2-yl)-3-pyrazolin-5-ones **13–18**.

Compound	CH_3 (s)	CH_2 (t)	CH_2O (t)	–OH (bs)	H at C-5 (s)	R
13	2.2	2.35	3.5	4.60	6.0	2.25 s (CH_3)
14	2.3	2.40	3.5	4.65	6.8	7.5–8.2 m (C_6H_5)
15[a]	2.2	2.40	3.5	4.50	–	1.8 s (CH_3)
16	2.2	2.40	3.5	4.60	–	1.0 t, 2.45 q (C_2H_5)
17	2.2	2.20	3.5	4.60	–	0.90 t, 1.4 m, 2.3 t (C_4H_9)
18[b]	2.2	2.40	3.5	4.50	–	7.2–7.6 m (C_6H_5)

[a]NH (s, 13.0); [b]NH (s, 13.5).

(93.7%); m.p. 260–262 °C (DMF); IR ν cm^{-1}: 3300–2500, 1740, 1650, 1560. ^{1}H-NMR: δ 1.3 (t, CH_3CH_2–), 1.7 [m, –$CH_2(CH_2)_2CH_2$–], 2.2 and 2.6 [2 t, –$CH_2(CH_2)_2CH_2$–], 4.3 (q, CH_3CH_2–), 8.5 (s, H at C-6 of pyrimidine). Anal. $C_{14}H_{16}N_4O_4$ (C, H, N).

4.1.12. 2-(5-Cyano-4-oxo-3H-pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one ***20***

It was prepared as described for **10**, from **7** (1.8 g, 10 mmol), ethyl (ethoxymethylene)cyanoacetate (1.7 g, 10 mmol) and anhydrous potassium carbonate (0.7 g, 5 mmol); yield: 1.75 g (68.0%); m.p. 290–293 °C (DMF); IR ν cm^{-1}: 3450–2500, 2240, 1680, 1650, 1600. ^{1}H-NMR: δ 1.7 [m, –$CH_2(CH_2)_2CH_2$–], 2.2 and 2.6 [2 t, –$CH_2(CH_2)_2CH_2$–], 8.6 (s, H at C-6 of pyrimidine). Anal. $C_{12}H_{11}N_5O_2$ (C, H, N).

4.1.13. 2-(5-Carboxy-4-oxo-3H-pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one ***21***

As described for **12**, from **19** (1.52 g, 5 mmol) and (1 N) sodium hydroxide (25 mL); yield: 1.2 g (87.2%); m.p. 270–272 °C (DMF); IR ν cm^{-1}: 3240, 3100–2800, 1720, 1680, 1650, 1610. ^{1}H-NMR: δ 1.7 [m, –$CH_2(CH_2)_2CH_2$–], 2.2 and 2.5 [2 t, –$CH_2(CH_2)_2CH_2$–], 8.6 (s, H at C-6 of pyrimidine). Anal. $C_{12}H_{12}N_4O_4$ (C, H, N).

*4.1.14. 2-(6-Methyl- or -6-phenyl-4-oxo-3H-pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one (**22**, **23**; tables V, VII)*

As described for (**13**, **14**), from **7** (1.8 g, 10 mmol), the appropriate β-keto ester (10 mmol) and anhydrous potassium carbonate (0.7 g, 5 mmol). The products were recrystallized from dimethylformamide. IR ν cm^{-1} (**22**, **23**): 3300–2700, 1660, 1610–1600, 1580.

*4.1.15. 2-(6-Hydroxy-5-substituted-4-oxo-3H-pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3H-indazol-3-one (**24–27**; tables V, VII)*

As described for **15–18**, from **7** (1.8 g, 10 mmol), the appropriate substituted diethyl malonate (10 mmol) and sodium methoxide (20 mmol). The products were recrystallized from aqueous dimethylformamide. IR ν cm^{-1} (**24–27**): 3400–2300, 1650–1640, 1600–1570.

Table VII. ^{1}H-NMR spectral data of the 2-(pyrimidin-2-yl)-1,2,4,5,6,7-hexahydro-3*H*-indazol-3-ones **22–27**.

Compound	–$CH_2(CH_2)_2CH_2$–			H at C-5	2 NH	OH	R
	(m)	+	(2 t)	(s)	(bs)	(bs)	
22	1.7		2.2, 2.5	6.0	12.5	–	2.3 s (CH_3)
23	1.7		2.2, 2.6	6.7	–	–	7.5–8.3 m (C_6H_5)
24	1.7		2.2, 2.5	–	11.5	12.0	1.8 s (CH_3)
25	1.7		2.2, 2.5	–	11.4	11.9	1.0 t, 2.3 q (C_2H_5)
26	1.7		2.2, 2.5	–	11.3	11.8	0.9 t, 1.3 m, 2.3 t (C_4H_9)
27	1.7		2.2, 2.6	–	12.0	12.0	7.2–7.6 m (C_6H_5)

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 I, II)

Dexatran-induced paw edema in rats (table I): 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 compounds (at a dose of 50 mg/kg). The paw volume was then measured and remeasured again 1, 2 and 3 h after administration of dextran. One group of 5 rats was kept as control and one group recieved the standard drug indomethacin (at a dose of 5 mg/kg) [21]. The percentage antiinflammatory activity was calculated by the formula % antiinflammatory activity = $(1 - dt / dc) \times 100$, where dt = difference in paw volume in drug treated groups; dc = difference in paw volume in control treated group.

Determination of serum transaminases in arthritic rats (table II): Formaldehyde (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 seven days and serum was obtained on the 8th day [21]. The levels of both serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were measured according to the method described by Reitman and Frankel [22].

4.2.2. Ulcerogenic activity [23]

Groups of 5 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.

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 g – 4.0 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 III)

The analgesic activity was determined using the hot-plate method [24]. 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 determinations ±S.E. and the percentage increase of the reaction time was calculated in comparison with the basal values.

4.2.5. Effect on body temperature

Hypothermic and antipyretic activities (table IV): The body temperature was measured at the beginning as a control for each group and then remeasured after 1/2, 1, 2, 4 and 6 h after administration of the test compounds (50 mg/kg, po). Indomethacin was used as a reference drug (5 mg/kg). Furthermore, 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 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).

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