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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

2-Substituted-8-methyl-3,6-dihydroimidazo[4,5-*c*]pyrazolo[3,4-*e*] pyridazine as an anti-inflammatory agent

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Abstract A series of 8-methyl-2-substituted-3,6-dihydroimidazo[4,5-*c*]pyrazolo[3,4-*e*]pyridazine compounds have been synthesized in the present investigation utilizing Philips condensation (Philips, J Chem Soc 2393–2399, 1928). The anti-inflammatory activity of the synthesized compounds was evaluated using a carrageenin rat model.

Keywords 8-Methyl-2-substituted-3,6-dihydroimidazo [4,5-*c*]pyrazolo[3,4-*e*]pyridazine · Anti-inflammatory · Philips condensation

Introduction

Heterocyclic systems containing pyrazole and imidazole rings are characterized by several biological properties (Avasthi *et al.*, 2002, 2003) including a medicinal character which can be utilized in the management of inflammation (Deeb *et al.*, 1990, 1991; Deeb and Said, 1990; Tisler and Stanounic, 1973; Seada *et al.*, 1989). Various synthetic routes have been followed for synthesizing the fused ring pyrazolo-pyridazine system and their derivatives (Druey, 1958; Caronna *et al.*, 1986; Kobayashi *et al.*, 1971; Kasuga *et al.*, 1974; Shawali, 1977; Kurihara *et al.*, 1980). The synthesis of pyridazines from 1,4-diketones and hydrazine hydrate is one of the best known procedures (Hendrickson *et al.*, 1964). However, its major limitation is the availability of the required 1,4-dicarbonyl precursors (Huisgen *et al.*, 1968; Potts and Roy, 1968; Potts and Elliott, 1973). This, in part, is due to the difficulty in the cycloaddition procedure (Vogel, 1978), which involves ring closure of vicinal dicarbonyl compounds, to synthesize the fused pyridazine derivatives.

The present paper reports the synthesis and biological activity of a series of five tricyclic compounds: 8-methyl-2-phenyldihydroimidazo[4,5-*c*]pyrazolo[3,4-*e*]pyridazine (**6a**), 8-methyl-2-[2-phenylethany]-3,6-dihydroimidazo[4,5-*c*] pyrazolo[3,4-*e*]pyridazine (**6b**), 2-(8-methyl-3,6-dihydroimidazo[4,5-*c*]pyrazolo[3,4-*e*]pyridazin-2-yl)benzoicacid (**6c**), 3-(8-methyl-3,6-dihydroimidazo[4,5-*c*]pyrazolo[3,4-*e*]pyridazin-2-yl)propionic acid (**6d**), and 5-(8-methyl-3,6-dihydroimidazo[4,5-*c*]pyrazolo[3,4-*e*]pyridazin-2-yl)pentanoic acid (**6e**). The objective was achieved by using a unique procedure of synthesizing 4,5-disubstituted-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazin, staring from 5-methyl-2,4-dihydro-3*H*-pyrazo-3-one, already synthesized by known method (Winter *et al.*, 1962).

In Scheme 1, the synthesized 4-amino-3-methyl-1*H*-pyrazolo [3,4-*c*]pyridazin-5-yl amine (**5**), was further cyclized with different carboxylic acids (Scheme 2) to obtain the final products.

Biological activity

All the reported compounds were tested for their anti-inflammatory activities against carrageenin induced rats paw edema.

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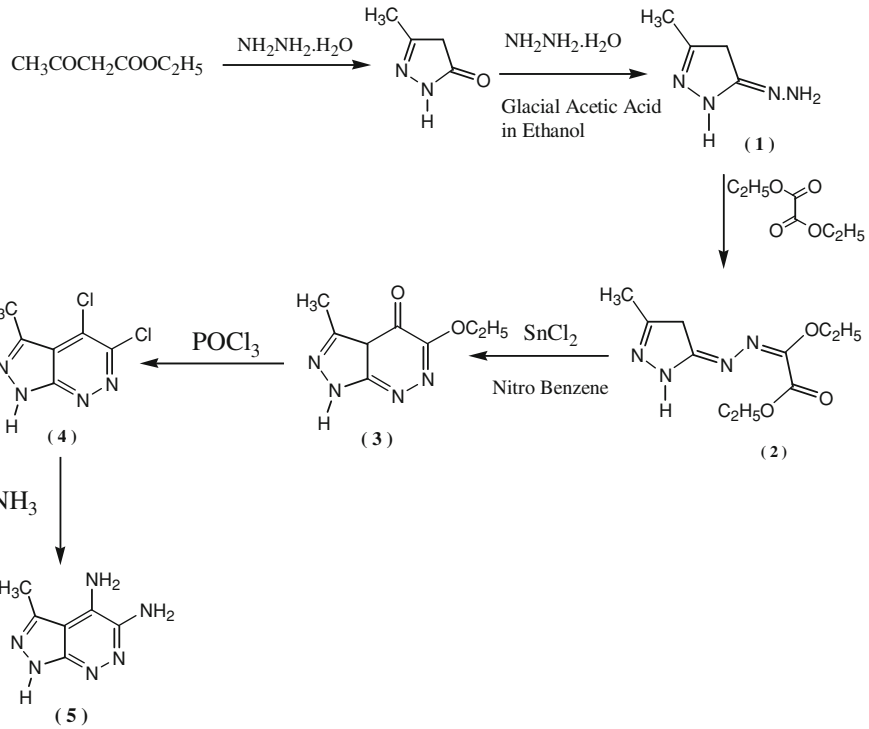
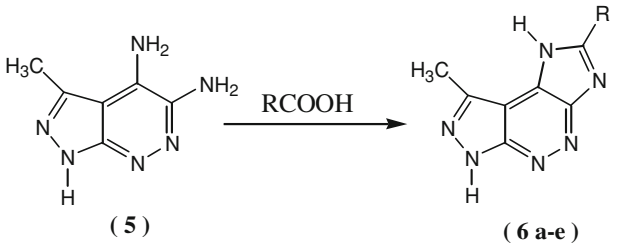
Scheme 1 Synthesis of 4-amino-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazin-5-yl amine





Scheme 2 Synthesis of 2-substituted-8-methyl-3,6-dihydroimidazo [4,5-*c*]pyrazolo[3,4-*e*]pyridazine

Anti-inflammatory activity against carrageenin induced rats paw edema

The anti-inflammatory activity of the synthesized compound was evaluated against carrageenin induced paw edema in albino rats. The weight of the rats ranged between 80 and 180 g each. Food and water was given to the experimental animals prior to the experiments. 0.05 ml of a freshly prepared suspension of carrageenin (1.0%) in 0.9% of saline was injected beneath the *planter aponeurosis* of the right paw of the rats and saline was injected in the opposite paw of the rat using the method of Winter (Segura *et al.*, 1998). Three sets of five rats each were maintained. One set was kept as control and the other two sets were pretreated with the test drug and standard drug, respectively, at a dose (Olayemi and Ajaiyeoba, 2007) of 100 mg kg^{-1}, orally 1 h before the carrageenin treatment given (Table 1). Rat paw was measured in the interval of 30 min after carrageenin treatment by the micropipette method as described by Buttle *et al.* (1957). The increase in the volume of the paw in each group was measured and percent anti-inflammatory activity was calculated by following formula:

$$\text{Percent anti-inflammatory activity} = [1 - V_t/V_c] \times 100$$

where V_t and V_c are the volume of the paw edema in drug treated and control group, respectively, where V_c is volume of the paw edema of control − volume of the paw edema of saline treated rats. Results in parentheses indicate percentage change from respective control group (Table 2).

Experimental section

Melting points were taken by an electrically heated instrument and are uncorrected. Compounds were routinely checked for their purity on silica gel G TLC plates and the spots were visualized by iodine vapors. Column chromatography was carried out by packing the column with 60–120 mesh silica gel G, used for purifying the compound. IR spectra were recorded on Varian 3100 FT-IR spectrometer. ^{1}H and ^{13}C-NMR spectra were recorded on a JEOL AL 300 MHz FT NMR instrument. NMR data represented as follows: chemical shifts δ (in ppm relative to $\delta_{TMS} = 0$), multiplicity, coupling constant J (quoted in hertz, Hz) integration, and assignment.

Table 1 Percentage edema growth relative to control at different time intervals (mean ± SEM)

Group	0 (min)	30 (min)	90 (min)	180 (min)
Control	100 ± 0	130.1 ± 6.54(30.1)	138.7 ± 4.47(38.7)	122.3 ± 5.17(22.3)
6a	100 ± 0	114.1 ± 2.88(14.1)	123.4 ± 3.37(23.4)	114.9 ± 4.25(14.9)
6b	100 ± 0	103.9 ± 2.59(3.9)	106.2 ± 3.98(6.2)	99.6 ± 2.54(0.4)
6c	100 ± 0	115.1 ± 10.02(15.1)	123.3 ± 3.83(23.3)	120.6 ± 8.34(20.6)
6d	100 ± 0	106.5 ± 3.67(6.5)	118.4 ± 3.86(18.4)	121.4 ± 4.55(21.4)
6e	100 ± 0	133.9 ± 3.56(33.9)	146.5 ± 6.54(46.5)	133.6 ± 7.58(33.6)
Ibuprofen	100 ± 0	101.7 ± 2.11	108 ± 3.27	11.6 ± 4.19

Table 2 Anti-inflammatory activity screening data of the compounds

Compound no.	Mean difference	Percent activity (100 mg/kg)
6a	28.76	30
6b	30.17	35
6c	22.31	41
6d	29.08	33
6e	28.73	31
Ibuprofen	18.76	49

5-Methyl-2,4-dihydro-3*H*-pyrazol-3-one hydrazone (1)

To a mixture of 60 ml of hydrazine hydrate, and 60 ml of glacial acetic acid, about 30 ml of water and 5-methyl-2,4-dihydro-3*H*-pyrazol-3-one (0.13 mol) was added in fractions for about 30 min. The mixture was stirred continuously on a magnetic stirrer during the addition. After complete addition, the mixture was warmed on a water bath for 1 h. On cooling and scratching the sides of round-bottom flask **1** was precipitated. Compound was recrystallized from ethanol. yield: 54% m.p 241°C ^{1}H NMR (DMSO): δ 0.91 (1H, s, C$\underline{H}_3$), δ 1.34 (3H, s, C$\underline{H}_2$), δ 7.10 (1H, s, N$\underline{H}$) δ 7.11 (2H, s, N$\underline{H}_2$); ^{13}C NMR (DMSO): δ 15.4 (C$\underline{H}_3$), δ 36.1 (C$\underline{H}_2$), δ 155.6 (C$\underline{C}$H$_3$), δ 155 (C$\underline{=}$N); IR (KBr): 3513 (sym), 3316 (assy) (N–H), 1387 (C–N); Elemental analysis for $C_4H_8N_4$: calcd.: C; 42.85%, H; 7.14%, N; 50% Found: C; 43.27%, H; 7.87%, N; 48.98%; MS: m/z 112.

Ethyl-2-[2-(5-methyl-2,4-dihydro-3*H*-pyrazol-3-ylidene)hydrazine]-2-oxoacetate (2)

Ethyl-2-[2-(5-methyl-2,4-dihydro-3*H*-pyrazol-3-ylidene) hydrazine]-2-oxoacetate (**2**) was synthesized by adding 5-methyl-2,4-dihydro-3*H*-pyrazol-3-one hydrazone (**1**) (0.18 mol) in fractions to a well stirred mixture of diethyl oxalate (0.18 mol) and 8 g of anhydrous sodium acetate in 30 ml of ethanol and 20 ml of water. Hydrazone derivatives were added to the stirring mixture for about 1 h. Then, temperature of stirrer was increased up to 20°C and the whole mixture was stirred for 5 h. The solid material was filtered to yield title compound. Yield: 48% m.p. 286°C; ^{1}H NMR ($CDCl_3$): δ 0.3 (t, 3H, C$\underline{H}_3$, $J = 6$), δ 0.9 (s, 3H, C$\underline{H}_3$), δ 1.11 (t, 3H, C$\underline{H}_3$, $J = 6$), δ 1.4 (s, 2H, C$\underline{H}_2$), δ 3.7 (q, 2H, C$\underline{H}_2$, $J = 6.0$), δ 4.2 (q, 2H, C$\underline{H}_2$, $J = 6$), δ 7.0 (s, 1H, N$\underline{H}$); ^{13}C NMR ($CDCl_3$); δ 13.3 (C$\underline{H}_3$), δ 14.5 (C$\underline{H}_3$), δ 15.4 (C$\underline{H}_3$), δ 30.0 (C$\underline{H}_2$), δ 56.3 (C$\underline{H}_2$), δ 59.6 (C$\underline{H}_2$), δ 155.6 (C$\underline{C}$H$_3$), δ 161 (CO), δ 163 (CO), δ 164 (C$\underline{=}$N); IR (KBr): 3575 (O–H), 1718 (C=O), 1413 (C–N), 3413 (N–H); Elemental analysis for $C_{10}H_{16}N_4O_3$: calcd: C; 50.00%, H; 6.66%, N; 23.33% Found: C; 50.1%, H; 6.65%, N; 23.29%; M S: m/e 184.

4,5-Dihydroxy-3-methyl-1*H*–pyrazolo[3,4-*c*]pyridazine (3)

4,5-Dihydroxy-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazine (**3**) has been synthesized by refluxing ethyl-2-[2-(5-methyl-2,4-dihydro-3*H*-pyrazol-3-ylidene) hydrazino]-2-oxoacetate (**3**) (0.05 mol) with nitrobenzene (30 ml) and $SnCl_2$ (10 g) for 5 h. The reaction was continued until a black jelly was settled on the bottom of round-bottomed flask. On the completion of cyclization nitrobenzene was distilled out. The solid black material was shaken vigorously with water in which insoluble material was filtered through suction. Recrystallization from alcohol yielded the desired product (**4**) Yield: 80%, m.p. 324°C; ^{1}H NMR (DMSO d_6): δ 0.9 (s, 3H, C$\underline{H}_3$), δ 1.11 (t, 3H, C$\underline{H}_3$, $J = 6$), δ 2.2 (s, 3H, C$\underline{H}$), δ 3.57 (q, 2H, C$\underline{H}_2$, $J = 6$), δ 5.2 (s, 1H, N$\underline{H}$); ^{13}C NMR (DMSO d_6): δ 12.7 (C$\underline{H}_3$), δ 14.5 (C$\underline{H}_3$), δ 44.5 (C$\underline{C}$C), δ 56.4 (C$\underline{H}_2$), δ 155.6 (C$\underline{C}$H$_3$), δ 163 (NCO), δ 164 (C$\underline{=}$N) δ 200 (CO); IR (KBr): 3575 (O–H), 1718 (C=O), 1413 (C–N), 3413 (N–H); Elemental analysis for $C_8H_9N_4O_2$: calcd: C; 49.74%, H; 4.66%, N; 29.01% Found: C; 49.71%, H; 4.62%, N; 29.15%; MS: m/e (%) 166 (M^+, 100.0), 137 (84.9), 108 (19.3), 81 (33.8) and 56 (13.4).

4,5-Dichloro-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazine (4)

4,5-Dichloro-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazine (**4**) has been synthesized by refluxing 4,5-dihydroxy-3-methyl-1*H*-pyrazolo [3,4-c]pyridazine (**3**) (0.05 mol) with phosphorous oxychloride (200 ml) for 50 h. Excess $POCl_3$ was removed by vacuum distillation. The residual mixture was poured into crushed ice to give **4** yields: 95.2%, m.p. 298°C; ^{1}H NMR (DMSO d_6): δ 2.32 (s, 3H, C$\underline{H}_3$), δ 12.67 (S, 1H, N$\underline{H}$); ^{13}C NMR (DMSO d_6): δ 12.43 (C$\underline{H}_3$), δ104.6 (C$\underline{C}$C), δ 134.56 (C$\underline{C}$l), δ 135 (C$\underline{=}$N), δ 145 (C$\underline{=}$N), δ 154.21 (N$\underline{C}$Cl); Elemental analysis for $C_6H_4N_4Cl_2$: calcd.: C; 35.64%, H; 1.98%, N; 27.72% Found: C; 35.63%, H; 1.94%, N; 27.70%; MS: m/z 202.

4-Amino-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazin-5-yl amine (5)

4-Amino-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazin-5-yl amine (**5**) was synthesized by stirring **5** (0.05 mol) with liquid ammonia for 6 h. The product was filtered and recrystallized from MeOH to **5**. Yield: 90%; ^{1}H NMR (DMSO d_6): δ 2.56 (s, 3H, C$\underline{H}_3$), δ 3.67 (S, 2H, N$\underline{H}_2$), δ 3.67 (S, 2H, N$\underline{H}_2$), δ 12.67 (S, 1H, N$\underline{H}$); ^{13}C NMR (DMSO d_6): δ 11.46 (C$\underline{H}_3$), δ 105 (C$\underline{C}$C), δ 132.56 (C$\underline{N}H_2$), δ 134.87 (C$\underline{=}$N), δ 144.31 (C$\underline{=}$N), δ 150.10 (N$\underline{C}$NH_2); IR (KBr): 3548 (Sym), 3380 (Assy), (–NH_2), 3489 (N–H), 1328 (C–N); Elemental analysis for $C_6H_8N_6$: calcd.: C; 43.90%, H; 4.87%, N; 51.21% Found: C; 43.1%, H; 4.89%, N; 51.00%; MS: m/e 164.

2-Substitued-8-methyl-3,6-dihydroimidazo[4,5-*c*]pyrazolo[3,4-*e*]pyridazine (6a–e) **general method**

2-Substitued-8-methyl-3,6-dihydroimidazo[4,5-*c*]pyrazolo [3,4-*e*]pyridazine (**6a-e**) have been synthesized by stirring 4-amino-3-methyl-1*H*-pyrazolo[3,4-*c*]pyridazin-5-yl amine (**5**) (8.20 g, 0.05 mol) and carboxylic acids (0.05 mol) in 4 N hydrochloric acid (50 ml) for 4 h at 80°C. The reaction mixture was made alkaline by adding liquor ammonia solution. Precipitate obtain was filtered and purified by column chromatography using $CHCl_3$ and MeOH.

6a: R = –C_6H_5

Yield: 74.44% ^{1}H NMR (DMSO d_6): δ 7.6 (s, 5H, Ph$\underline{H}$), δ 5.3 (s, 1H, N$\underline{H}$), δ 4.8 (s, 1H, N$\underline{H}$), δ 2.1 (s, 3H, C$\underline{H}_3$); ^{13}C NMR (DMSO d_6): δ 10.46 (C$\underline{H}_3$), δ 104.89 (C$\underline{C}$C), δ 121.56 (C$\underline{C}$N), δ 122 (N$\underline{C}$N), δ 127.1 (Ar$\underline{C}$H), δ 128.32 (Ar$\underline{C}$H), δ 129.2 (Ar$\underline{C}$H), δ 135.5 (N$\underline{C}$N), δ 135.87 (C–C), δ 136 (C$\underline{C}$N), δ 144.23 (C–CH_3); Elemental analysis for $C_{13}H_{10}N_6$: calcd.: C; 62.40%, H; 4.00%, N; 33.60% Found: C; 62.82%, H; 4.32%, N; 32.82%; MS: m/e 250.

6b: R = –CH=CH–C_6H_5

Yield: 81.6% m.p. 138°C. ^{1}H NMR (DMSO d_6): δ = 7.7 (m, 5H, Ph $\underline{H}$), δ 5.3 (s, 1H, N$\underline{H}$), δ 4.7 (s, 1H, N$\underline{H}$), δ 3.9 (d, J = 2.8, 1H, $\underline{H}1'$), δ 3.4 (d, J = 2.8, 1H, $\underline{H}2'$), δ 2.2 (s, 3 H C$\underline{H}_3$); ^{13}C NMR (DMSO d_6): δ 10.46 (C$\underline{H}_3$), δ 104.89 (C$\underline{C}$C), δ 121.56 (C$\underline{C}$N), δ 122 (N$\underline{C}$N), δ 124 (C$\underline{=}$C), δ 126.1 (Ar$\underline{C}$H), δ 127.32 (Ar$\underline{C}$H), δ 128.2 (Ar$\underline{C}$H), δ 130.2 (C$\underline{=}$C), δ 134.76 (C–C), δ 135.5 (N$\underline{C}$N), δ 135.87 (C–C), δ 144.23 (C–CH_3); Elemental analysis for $C_{15}H_{12}N_6$: calcd.: C; 65.21%, H; 4.35%, N; 30.44% Found: C; 66.78%, H; 4.82%, N; 30.12%; MS: m/e 276.

6c: R = C_6H_4COOH

Yield: 79% ^{1}H NMR (DMSO d_6): δ 2.2 (s, 3H, C$\underline{H}_3$), δ 3.8 (s, 1H, O$\underline{H}$), δ 4.5 (s, 1H, N$\underline{H}$), δ 5.3 (s, 1H, N$\underline{H}$), δ 7.6 (m, 4H, Ph$\underline{H}$); ^{13}C NMR (DMSO d_6): δ 10.46 (C$\underline{H}_3$), δ 105.89 (C$\underline{C}$), δ 121.56 (C$\underline{C}$N), δ 122 (N$\underline{C}$N), δ 126.1 (Ar$\underline{C}$H), δ 129 (C$\underline{C}$O), δ 128.7 (Ar$\underline{C}$H), δ 130.21 (Ar$\underline{C}$H), δ 134 (Ar$\underline{C}$H), δ 136 (C$\underline{C}$), δ 139.21 (C–C), δ 172 (C$\underline{O}$), δ 144.21 (C–CH_3); IR (KBr): 1731 (C=O), 3512 (O–H), 3483 (N H), 1429 (C–N); Elemental analysis for $C_{15}H_{12}N_6$: calcd.: C; 57.14%, H; 3.40%, N; 28.57% Found: C; 58.73%, H; 2.86%, N; 29.31%; MS: m/e 294.

6d: R = –CH_2CH_2COOH

Yield: 86% m.p. 234°C ^{1}H NMR (DMSO d_6): δ 6.2 (s, 1H, O$\underline{H}$), 5.3 (s, 1H, N$\underline{H}$), 4.5 (s, 1H, N$\underline{H}$), 3.1 (t, 2H, H-2′), 2.8 (t, 2H, H-1′), 2.2 (s, 3H, C$\underline{H}_3$); ^{13}C NMR (DMSO d_6): δ 10.9 (C$\underline{H}_3$), δ 36.78 (C$\underline{H}_2$CO), δ 105.3 (C$\underline{C}$), δ 122.23 (C$\underline{C}$N), δ 122.67 (N$\underline{C}$N), δ 125.1 (C$\underline{C}H_2$), δ 134.98 (N$\underline{C}$N), δ 144.21 (C-CH_3), δ 148.67 (C$\underline{C}H_2$), δ 177.0 (C$\underline{O}$); IR (KBr): 3513 (O–H), 3418 (N_H), 1692(C–O); Elemental analysis for $C_{10}H_{10}N_6O_2$: calcd.: C; 48.78%, H; 4.06%, N; 34.14% Found: C; 49.26%, H; 3.96%, N; 33.82%; MS: m/e 248.

6e: R = –$CH_2CH_2CH_2CH_2COOH$

Yield: 83.3% m.p. 152°C ^{1}H NMR (DMSO d_6): δ 6.7 (s, 1H, OH), 5.3 (s, 1H, N$\underline{H}$), 4.5 (s, 1H, N$\underline{H}$), 3.8 (t, 2H, H-4′), 3.1 (m, 4H, H-2′, H-3′), 2.6 (t, 2H, H-1′), 2.1 (s, 3H, C$\underline{H}_3$); ^{13}C NMR (DMSO d_6): δ 10.59 (C$\underline{H}_3$), δ 25.00 (C$\underline{H}_2$), δ 29.4 (C$\underline{H}_2$), δ 30.21 (C$\underline{H}_2$), δ 31.78 (C$\underline{H}_2$), δ 35.7 (C$\underline{C}$O), δ 105.36 (C$\underline{C}$), δ 122.12 (C$\underline{C}$N), δ 122.43 (N$\underline{C}$N), δ 135.8 (N$\underline{C}$N), δ 144 (C–CH_3), δ 148.57 (C$\underline{C}H_2$), δ 176.78 (C$\underline{O}$); IR (KBr): 1711 (C=O), 3445 (O–H), 3313 (N–H), 3396 (–H). Elemental analysis for $C_{12}H_{14}N_6O_2$: calcd.: C;

52.56%, H; 5.11%, N; 30.65% Found: C; 53.12%, H; 6.12%, N; 30.04%; MS: m/e 274.

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References

Avasthi K, Tewari A, Rawat DS, Sharon A, Prakash R, Maulik PR (2002) A stacked pyrazolo[3,4-*d*]pyrimidine-based flexible molecule: the effect of a bulky benzyl group on intermolecular stacking in comparison with methyl and ethyl groups. Acta Crystallogr Sect C 58:494–496

Avasthi K, Farooq SM, Tewari AK, Sharon A, Maulik PR (2003) Intermolecular stacking in pyrazolo[3,4-*d*]pyrimidine-based pentamethylene-linked flexible molecules. Acta Crystallogr Sect C 59:o42–o45

Buttle GAH, D'Arcy PF, Howard EM, Kellett DN (1957) Plethysmometric measurement of swelling in the feet of small laboratory animals. Nature 179:629

Caronna S, Migliara O, Petruso S (1986) Synthesis of pyrazole(3,4-a)pyridazine derivatives. Bull Chem Farm 125:114–116

Deeb A, Said SA (1990) Studies on polyazaindenes synthesis of several new condensed pyridazine derivatives. Collect Czech Chem Commun 55:2795

Deeb A, Said SA, Homed MM Yasine F (1990) J Chim Soc (Taipei) 37 287

Deeb A, Bayoumy B, Essawy AE, Fikry R (1991) Pyridazine derivatives and related compounds part 5. Pyrazolo[3,4-c]pyridazine: synthesis and some reactions. Heterocycles 32:895–900

Druey J (1958) Angew Chem 70:5

Hendrickson JB, Rees R, Templeton JF (1964) A new general heterocycle synthesis; use of acetylenedicarboxylic esters. J Am Chem Soc 86:107–111

Huisgen R, Gotthardt H, Groshey R (1968) Chem Ber 101:1062

Kasuga K, Hirobe M, Okamoto T (1974) Reaction of N-aminopyridinium derivatives. XIII. Syntheses of pyrazolodiazines by cyclization of N-aminodiazinium salts. Chem Pharm Bull 22:1814

Kobayashi Y, Kutsuma T, Morinaga K (1971) Studies on the reactions of heterocyclic compounds. V. Syntheses of diazinium ylides and their conversion to azaindolizines. Chem Pharm Bull (Tokyo) 19:2106–2115

Kurihara T, Uno T, Sakomoto Y (1980) Reaction of ethyl 3-thoxymethylene-2,4-dioxovalerate with monosubstituted hydrazines. J Heterocycl Chem 17:231–233

Olayemi JO, Ajaiyeoba EO (2007) Short communication—anti-inflammatory studies of yam (*Dioscorea esculenta*) extract on wistar rats. Afr J Biotechnol 6:1913–1915

Philips MA (1928) The formation of 2-substituted benziminazoles. J Chem Soc 2393–2399. doi:10.1039/JR9280002393

Potts KT, Elliott AJ (1973) o-Dibenzoyl heterocycles via cycloaddition reactions. Convenient route to fused pyridazine systems. J Org Chem 38:1769

Potts KT, Roy DN (1968) 1,3-Dipolar cyclo-addition of mesoionic anhydro-2-aryl-5-hydroxy-3-methylthiazolium hydroxides. Chem Commun 536:1061–1062

Seada M, Fawzy MM, Johine H, Magid MA, Saad RR (1989) J Chin Soc (Taipei) 36:241

Segura L, Vila R, Gupta MP, Avella ME, Adzet T, Canigueral S (1998) Antiinflammatory activity of *Anthurium cerrocampanense* Croat in rats, mice. J Ethnopharm 61:243–248

Shawali AS (1977) Synthesis and tautomeric structure of some 2H-pyrazolo[3,4-d]pyridazines. J Heterocycl Chem 14:375–381

Tewari AK, Mishra A (2001) Synthesis and anti-inflammatory activities of N4, N5-isubstituted-3-methyl-1H-pyrazolo[3,4-c]pyridazines. Bioorganic Med Chem 9:715–718

Tewari AK, Mishra L, Verma HN, Mishra A (2002) Synthesis and antifungal activity of 4-substituted-3,7-dimethyl-pyrazolo[3,4-e][1,2, 4]triazine. Indian J Chem 41B:664–667

Tisler M, Stanounic B (1973) In: Castle RN (ed) The chemistry of heterocyclic compounds. Wiley, New York, p 785

Vogel AL (1978) Text-book of practical organic chemistry, 4th edn. New York: Longman, p 595

Winter CA, Risley EA, Nuss GW (1962) Carrageenin-induced edema in hind paw of the rat as an assay for antiinflammatory drugs. Proc Soc Exp Biol Med 111:544–547