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Pyrazoles and Pyrazolo[3,4-*d*]pyrimidines as BiologicallyActive Agents, II[1)]

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Pyrazoles with bio-active substituents at different positions are prepared and cyclised to the corresponding pyrazolo[3,4-*d*]pyrimidines in order to establish structure-activity relationships. The structure of the compounds is established by micro-analyses and spectroscopic studies.

Pyrazole und Pyrazolo[3,4-d]pyrimidine als biologisch aktive Verbindungen, 2. Mitt.

Es werden Pyrazole mit bioaktiven Substituenten in verschiedenen Positionen hergestellt und zu den entsprechenden Pyrazolo[3,4-d]pyrimidinen zyklisiert, um Struktur-Wirkungsbeziehungen aufzustellen. Die Struktur der Verbindungen wird durch Mikroanalysen und spektroskopische Daten gesichert.

The tranquilizing, muscle relaxant, psychoanaleptic[2)], hypnotic[3)], anticonvulsant[4,5)], antipyretic[6)] and anticancer[7,8)], activities of pyrazoles prompted the synthesis of 1,3,4,5-tetrasubstituted pyrazoles. Recently *Metcalf*[9)] has discussed cholinesterase inhibiting property of such compounds which show systemic insecticidal activity and related the presence of alkylthiosubstituents in these compounds and

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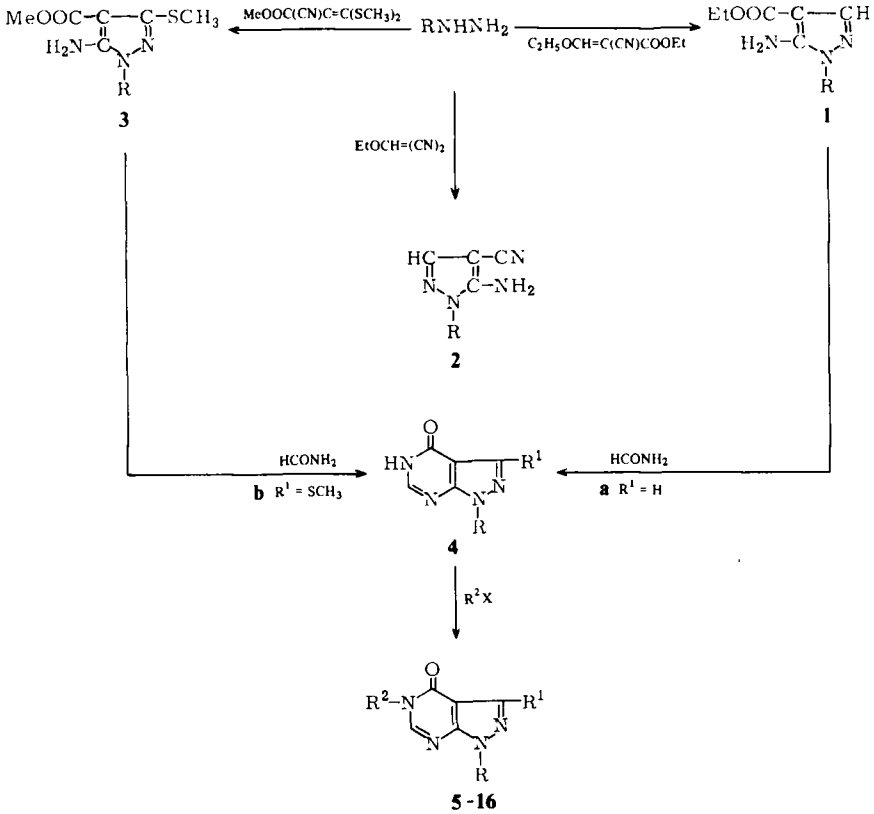
their ability to undergo oxidation in vivo to the corresponding sulphoxides and sulphones to 'delay factors' in their endometatoxic systemic properties.

Certain 3-substituted pyrazolopyrimidines have been of recent interest especially with methylmercapto substituent due to their ability to inhibit the enzyme 3',5'-cyclic AMP phosphodiesterase and their interesting cardiotropic properties[10,11]. However antipyretic, anti-inflammatory and anticancer activity has been reported for many pyrazolo[3,4-d]pyrimidine derivatives during the last decade[12,13].

Thus high therapeutic indexes of pyrazoles and their cyclised products led to the synthesis of 3-methylmercapto-pyrazoles and corresponding pyrazolo[3,4-d]pyrimidines in the anticipation that the new compounds will exhibit better bio-responses in comparision to the already reported ones. The substituted pyrazoles were prepared by the reaction of alkyl/aryl hydrazine with ethoxymethylenemalononitrile, ethoxymethylene-ethylcyanoacetate and dimethylmercaptomethylenemethylcyanoacetate separately. The resulting pyrazoles **1** and **3** were cyclised to the pyrazolo[3,4-d]pyrimidines **4** by refluxing in formamide and were converted into the N-alkyl derivatives **5–16** by the reaction of alkyl halide in aqueous sodium hydroxide (1 %).



3 **1** **2** **4** **5-16**

IR spectra of **2a** showed four peaks in the region 3410, 3350, 3240 and 3100 cm^{-1} due to NH- stretching vibrations. The former two peaks are attributed to free NH_2 group while latter are assumed due to sulphonamido-group because these disappeared in **3b**. A strong and sharp peak at 2210 cm^{-1} is assigned for C – N stretching vibrations. Carbonyl group frequency in **3a–d** appeared at ~1675 cm^{-1} which is a bit lower than the normal ester carbonyl function. This lowering is expected due to intramolecular hydrogen bonding. The carbonyl frequency in the cyclised products **4** and **5–16** appeared at ~1675 cm^{-1} due to cyclic amide group.

NMR spectrum of **3a** showed singlets at δ (ppm) = 2.44, 3,72 due to SCH_3 and $COOCH_3$ protons. The latter peak resonates in low field due to greater desheilding effect of carbomethoxy group. Similar phenomena is observed in the case of NH_2 and SO_2NH_2 groups which resonate as singlet at 6.64 and 7.48 ppm resp. The signal due to the aromatic protons appeared as multiplet at 7.72–8.04 ppm.

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Experimental

All the m. ps. are uncorrected.

1-(p)-sulphonamidophenyl-4-carbethoxy-5-aminopyrazole **(1a)**

It was prepared by refluxing 1 : 1 molar quantity of ethoxymethylenecyanoethylacetate and p-sulphonamidophenylhydrazine hydrochloride in the presence of fused sodium acetate (in the molar quantity) in alcohol for 3 h. The solution was cooled. The precipitate was washed with ethanol and finally crystallised from ethanol. Yield: 70 %, m. p. 230°C. $C_{12}H_{14}N_4O_4S$ Calcd.: C 46.5 H 4.51 N 18.1 Found: C 46.6 H 4.7 N 18.2. M^+ 310; 265 ($-OC_2H_5$); 264 ($-C_2H_5OH$); 184 ($-SO_2NH_2$); 156 (-CO) NMR (DMSO d^6) δ (ppm) = 1.28 (t, CH_3); 4.04 (m, CH_2); 6.52 (s, NH_2); 7.49 (s, SO_2NH_2); 7.8–7.96 (m, C_6H_4); 8.04 (CH, s).

1-(p)-Chlorophenyl-4-carbethoxy-5-aminopyrazole **(1d)**

It was prepared following the procedure described in the preceding experiment from 0.74 g ethoxymethylenecyanoethylacetate and 0.62 g p-chlorophenylhydrazine. The crude product was crystallised from ethanol-water mixture. Yield: 0.46 g (50 %), m. p. 141°, M^+ 265.5. $C_{12}H_{12}ClN_3O_2$ Calcd: C 54.2 H 4.51 N 15.8; Found: C 54.4 H 4.6 N 16.1.

1-(p)-sulphonamidophenyl-4-cyano-5-aminopyrazole **(2a)**

A mixture of 1.83 g ethoxymethylenemalononitrile, 3.3 g p-sulphonamidophenylhydrazine hydrochloride and 1.21 g fused sodium acetate in 10 ml ethanol was refluxed for 3 h and cooled. The precipitate was washed with a little ethanol and crystallised from ethanol. Yield: 0.67 g (35 %), m. p. 222°. $C_{10}H_9N_5O_2S$ Calcd.: C 45.6 H 3.42 N 26.2; Found: C 45.8 H 3.6 N 26.4. M^+ 263; 247 ($-NH_2$); 199 (-SO); 183 ($-SO_2NH_2$); 156 (-HCN); 129 (-HCN).

General procedure for the preparation of 1-substituted-3-methylmercapto-4-carbomethoxy-5-aminopyrazoles **3**

An equimolar mixture of dimethylmercaptomethylenemethylcyanoacetate and appropriate hydrazine in alcohol was refluxed for 3–4 h. On cooling, a pricipitate obtained which was washed with little ethanol and then crystallised from a suitable solvent. Compounds thus prepared are listed in Table 1.

Table 1: *1-Substituted-3-methylmercapto-4-carbmethoxy-5-aminopyrazoles* **3**

3	R	M.P. °C	Yield %	Molecular formula	M^+	Analysis Calcd. C	H	N	Found C	H	N
a	p-Sulphonamidophenyl-	229[a]	76	$C_{12}H_{14}N_4O_4S_2$	342	42.1	4.09	16.4	42.3	4.12	16.6
b	Isopropyl-	70[a]	50	$C_9H_{15}N_3O_2S$	229	47.1	6.5	18.3	47.2	6.4	18.4
c	p-Tolyl-	147[a]	75	$C_{13}H_{15}N_3O_2S$	277	56.3	5.4	15.2	56.4	5.6	15.3
d	p-Chlorophenyl-	187[a]	84	$C_{12}H_{12}ClN_3O_2S$	297.5	48.4	4.0	14.1	48.6	4.2	14.3
e	1,2,4-Triazino-[5,6-b]indole-3-yl-	273[d]	83	$C_{15}H_{13}N_7O_2S$	355	50.7	3.6	27.6	50.8	3.5	27.5

Solvent for crystallisation: a = ethanol; d = dimethylformamide.

General procedure for the cyclisation of **1** *and* **3** *into* **4** *1-(p)-sulphonamidophenyl-4-oxo-pyrazolo[3,4-d]pyrimidine* **(4a)**

A solution of 1.0 g **1a** in 4 ml formamide was refluxed for 3 h. When fumes of ammonia ceased the content was cooled. The product was crystallised from formamide-water mixture, yield: 0.8 g, m. p. > 300°. $C_{11}H_9N_5O_3S$ Calcd.: C 45.4 H 3.1 N 24.1; Found: C 45.6 H 3.2 N 24.6. M^+ 291; 275 ($-NH_2$); 211 ($-SO_2NH_2$); 184 (-HCN); 156 (-CO); 129 (-HCN).

1-Isopropyl-3-methylmercapto-4-oxo-pyrazolo[3,4-d]pyrimidine **(4b)**

It was prepared by the cyclisation of 1.0 g **3b** in 4 ml formamide as described in preceding experiment and was crystallised from ethanol, yield: 0.7 g, m. p. 189°. $C_9H_{12}N_4OS$ Calcd.: C 48.2 H 3.35 N 25.0; Found: C 48.4 H 3.5 N 25.1. NMR (DMSO d^6): δ (ppm), 1.44 (t, CH_3); 2.52 (s, S-CH_3); 4.88 (m, CH); 8.04 (s, CH). M^+ 224; 209 ($-CH_3$); 194 ($-CH_3$); 182 (209-HCN); 181 (194-CH); 149 (-S).
Other compounds were prepared similarly and are listed in Table 2.

General procedure for the preparation of 1-substituted-3-methylmercapto-4-oxo-5-N-alkyl/-aralkyl-pyrazolo[3,4-d]pyrimidines **9–16**

Procedure A: – To a solution of 0.01 mole 1-substituted-3-methylmercapto-4-oxo-pyrazolo[3,4-d]-pyrimidine in 5 proc. aqueous sodium hydroxide, 0.02 mole alkyl/aralkyl halide were added. To make the solution clear, a little ethanol was added and the mixture was stirred for 3 h at room temp. The precipitate obtained was washed with water and crystallised from a suitable solvent. The yield was

found quantitative in each case and thus compounds. **9–12** (Table 2) were prepared by this procedure.
Procedure B: – 1-substituted-3-methylmercapto-4-oxo-pyrazolo[3,4-d]pyrimidine and potassium carbonate in equimolar quantities were dissolved in dimethylformamide and then alkyl/aralkyl halide was added. The solution was stirred for 3–4 h at room temp. The precipitate was washed with water and crystallised from a suitable solvent. Compounds **13–16** (Table 2) were prepared by this procedure.

Table 2: *1-Substituted-3H-methylmercapto-4-oxo-5-substituted pyrazolo[3,4-d]pyrimidines* **5–16**

Nr.	R	R^2	M.P. °C	Yield %	Molecular formula	M^+
5	p-Chlorophenyl	H	300[c]	86	$C_{11}H_7ClN_4O$	246.5
6	p-Sulphonamidophenyl	H	290[b]	53	$C_{12}H_{11}N_5O_3S_2$	337
7	p-Tolyl	H	280[c]	86	$C_{13}H_{12}N_4OS$	272
8	p-Chlorophenyl	H	270[c]	59	$C_{12}H_9ClN_4OS$	292.5
9	p-Tolyl	Methyl	168[a]	80	$C_{14}H_{14}N_4OS$	286
10	p-Tolyl	Ethyl	182[a]	40	$C_{15}H_{16}N_4OS$	300
11	p-Tolyl	Allyl	150[a]	30	$C_{16}H_{16}N_4OS$	312
12	p-Tolyl	Benzyl	185[c]	32	$C_{20}H_{18}N_4OS$	362
13	p-Chlorophenyl	Benzyl	165[c]	55	$C_{19}H_{15}ClN_4OS$	382.5
14	p-Chlorophenyl	Allyl	270[c]	60	$C_{15}H_{13}ClN_4OS$	332.5
15	p-Chlorophenyl	Methyl	231[a]	60	$C_{13}H_{11}ClN$	306.5
16	p-Chlorophenyl	p-Bromobenzyl	140[c]	55	$C_{19}H_{14}ClBrN_4OS$	382.5

All the compounds were analysed for C, H, N satisfactorily and crystallised from a = ethanol-water; b = formamide-water; c = dimethyl-formamide-water mixture. R^1 = H for **5** and for **6–16** $-SCH_3$.

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