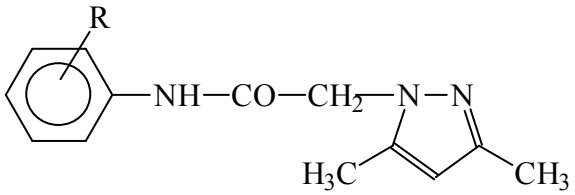
NEW SUBSTITUTED 2-(3,5-DIMETHYL-PYRAZOL-1-YL)-ACETANILIDES WITH PHARMACOLOGICAL ACTIVITY

Christina Zălaru*, M. Iovu, F. Dumitraşcu, Elena Cristea and Isabela Târcomnicu

abstract: New substituted 2-(3,5-dimethyl-pyrazol-1-yl)-acetanilides were obtained. The compounds have been characterized by UV-VIS, IR, ^{1}H-NMR, ^{13}C-NMR. spectra, mass spectrometry and pharmacology research.

Introduction

Our previous papers [1-4] have reported the synthesis and characterization of some 2-(pyrazol-1-yl)-acetanilides. It was shown that the nature and the number of substituents from the pyrazole influence the physical, chemical and pharmacological behaviour of the compounds. It seemed desirable to investigate whether or not the nature, number and position of R-substituent from the benzene ring cause the same effects in physical, chemical and pharmacological behaviour. For this, the present work examines compounds of the following type (R-substituent described in Table 1):



Experimental

The melting points were determined with a Boetius apparatus and are not corrected.

Thin layer chromatography (TLC) was made on silicagel Merck plates, in one dimensional technique; for the development solution of 7.5:1:2:1 = petroleum ether: ethyl ether: methylene chloride: ethyl acetate were used. The visualization was made with an UV lamp, λ = 254 nm.

* Dept. of Organic Chemistry, Faculty of Chemistry, University of Bucharest, 90-92 Road Panduri, Bucharest, Romania

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Molecular weight was obtained with a GC-MS 8000 MD 800 Fissions spectrometer at 70 eV, carrier gas He at 2 ml/min.

Electronic spectra within 200-800 nm range were obtained with Unicam UV-VIS spectrometer in ethanol solution 10^{-5}M.

IR spectra were recorded within 4000-400 cm^{-1} range by BIO-RAD FTS-135 spectrometer, in KBr pellets.

NMR spectra were recorded on a Varian Gemini 300 spectrometer operating at 300 MHz (^{1}H-NMR), 75MHz (^{13}C-NMR) respectively in $CDCl_3$ or DMSO-d_6 in 5 mm NOREL-57 PP grade sample tubes. The chemical shifts were referred to TMS as internal standard.

The chemical shifts of representative compounds are:

^{1}H-NMR ($CDCl_3$, δ (ppm) for compound number **1**: 5.92 s (H-4); 2.16s (Me-3); 2.27s (Me-5); 4.78s (CH_2); 8.28bs (NH); for compound number **5**: 5.93s (H-4); 2.24s (Me-3); 2.32s (Me-5); 4.84s (CH_2); 7.50bs (NH); for compound number **6**: 5.84s (H-4); 2.12s (Me-3); 2.17s (Me-5); 4.73s (CH_2); 10.16bs (NH).

^{13}C-NMR ($CDCl_3$, δ (ppm) for compound number **1**: 149.9 (C-3); 106.4 (C-4); 140.9 (C-5); 13.4 (Me-3); 10.9 (Me-5); 165.6 (CO); 52.5 (CH_2); for compound number **5**: 149.9 (C-3); 106.4 (C-4); 140.7 (C-5); 13.4 (Me-3); 11.0 (Me-5); 166.5 (CO); 52.3 (CH_2); for compound number **6**: 150.4 (C-3); 106.8 (C-4); 140.2 (C-5); 13.4 (Me-3); 10.9 (Me-5); 167.1 (CO); 52.9 (CH_2).

Acute toxicity (LD_{50}). White mice weighing 20±2 g each were used in batches. The new compounds were administered in doses ranging between 250 and 750 mg/kg of body weight. The graphic method [5,6] was used. The strain of the experimental animals has been maintained at a minimum all through the testing.

Infiltration local anaesthetic action was determined by Bianchi's method [7,8]**.** Lidocaine hydrochloride was used for comparison. The results were interpreted using the "t" student test [5,6].

Antiarrhythmic action. The latent time of the appearance of heart fibrillation was measured using Hackenberger's technique [9] for the new compounds comparatively with lidocaine hydrochloride and quinidine sulphate.

Results and discussion

The new compounds were synthesized by N-alkylation reaction of 3,5-dimethylpyrazole with several 2- iodoacetanilides as described in reference [1-4].

Physical properties are given in Table 1. The purity of new compounds was checked through TLC.

The electronic spectra of the compounds recorded in ethanolic solution show the λ_{max} values which exist in the characteristic ranges (222-236 nm and 254-324 nm) of the chromophores present in the molecule (>C=O, >C=C<, >C=N-). These bands are assigned to the π-π* transitions.

It was noticed that in the electronic spectra of the new compounds there occur the bands λ_1,λ_2 assigned to the π-π* transitions characteristic of disubstituted benzene ring, but they are shifted either hypsochrome or batochrome depending in the nature of the substituent on the benzene ring.

In the case of compound number **6** (R = *o'*-NO_2) we noticed the occurrence of a wide and strong band with three peaks (234, 258,324 nm) whose λ_{max} is much shifted batochrome.

Table 1. Analytical physical data

No	R	Formula	Molecular mass		Base peak m/e 100%	M.P. (°C)	Yield (%)	R_F
			Calc.	Exp. (MS)				
1	2'-Me	$C_{14}H_{17}N_3O$	243.31	243	109	151-152	36.2	0.17
2	3'-Me	$C_{14}H_{17}N_3O$	243.31	243	109	136-138	79.5	0.13
3	4'-Me	$C_{14}H_{17}N_3O$	243.31	243	109	145-147	52.5	0.20
4	2',4'-diMe	$C_{15}H_{19}N_3O$	257.34	257	109	158-159	26.4	0.17
5	2',6'-diEt	$C_{17}H_{23}N_3O$	285.39	285	109	149-150	79.9	0.16
6	2'-NO_2	$C_{13}H_{14}N_4O_3$	274.28	274	109	130-131	40.5	0.19

IR spectra recorded in the 4000-400 cm^{-1} range in KBr pellets reflected the molecular structure of the new compounds and showed the bands characteristic of the secondary amides [13]. The strong band due to the stretching frequency, ν_{NH} appears within the 3242-3278 cm^{-1} range. The very strong amide band **I**, ν_{CO} appears within the 1660-1690 cm^{-1} range; the strong amide band **II**, due to the δ_{NH} + ν_{CN} coupling is present within the 1531-1587 cm^{-1} range. The bands due to the stretching of the pyrazole ring can be found within the 1375-1490 cm^{-1} range.

^{1}H-NMR and ^{13}C-NMR spectra recorded at 300 MHz in the $CDCl_3$ and DMSO-d_6 solution support the structure formulas assigned to these compounds, deduced from the equation of the synthesized reaction [1-4]. The positions of the protons and of the substituents on the ring can be found out using HETCOR and COSY.

Table 1 gives the m/e values % of the base peak in the mass spectra of the new compounds. Fragmentation processes can support the structure formulas assigned to the compounds [2,3]. The fragmentation process specific to the aromatic amides occurring by the cleavage of the bond between the N and C atoms of the carbonyl group followed by CO elimination [2,3]. The base peak thus appears [10]. As expected, the base peak is the same with all compounds.

Pharmacological results. The acute toxicity (LD_{50}) of the compounds ranges within 438-650 mg/kg of body weight. The compounds evince acute toxicity on mice which is low versus lidocaine hydrochloride. The least toxic compound is number **4** and the most toxic is compound number **6**.

The anaesthetic and antiarrhythmic actions were tested using 1/10 of the LD_{50} value of the compounds as a working dose.

Local anaesthetic action. The anaesthetic action was tested using Bianchi's [7,8] experiment on mice using lidocaine hydrochloride as reference substance. All compounds have moderate infiltration anaesthetic action versus lidocaine hydrochloride (41.92%-58.03%).

Antiarrhythmic action. The influence of the studied compounds was determined on the experimental fibrillation induced in mouse by an atmosphere of chloroform [9]. The reference substances were quinidine sulphate and lidocaine hydrochloride.

All tested compounds delayed the appearance of the toxic fibrillating effect of chloroform (32.22%-88.08%) and (20.61-56.35%). Compound number 6 has high antiarrhythmic activity versus lidocaine hydrochloride (88.08%) and quinidine sulphate (56.35%) respectively. Structurally, this most active compound was characterized by the presence of the nitro group in the o-position on the benzene ring, but it is also the most toxic.

Conclusions

The new compounds were characterized by UV-VIS, IR, ^{1}H-NMR, ^{13}C-NMR, SM and pharmacological tests.

The studied compounds have infiltration local anaesthetic actions and antiarrhythmic action, but their potency is lower than that of lidocaine and quinidine respectively.

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