SHORT COMMUNICATIONS

Department of Pharmaceutical Chemistry, Pharmaceutical Faculty, Comenius University, Bratislava, Slovak Republic

Study of local anaesthetics

Part 152: Some piperidinomethyl esters of alkoxy substituted phenylcarbamic acids

D. SLÁDKOVÁ and J. ČIŽMÁRIK

In the frame of study of the relations between chemical structure, physico-chemical properties and local anaesthetic activity, a series of compounds belonging to the group of piperidinomethyl esters of 2-, 3- and 4-alkoxy substituted phenylcarbamic acids were synthesized and evaluated. These compounds are local anaesthetics of the carbamate type with a short connecting chain between the lipophilic and the hydrophylic part of the molecule.

Results of their local anaesthetic activity evaluation point to the fact that these compounds are more efficient than standards, i.e. cocaine and procaine, and they are more effective as surface anaesthetics than as infiltration anaesthetics. These derivatives are expectivelly less effective than their piperdinoethyl substituted analogues [1, 2].

In general, it can be stated that the introduction of a methyl substituent, which represents the new type of connecting chain between the lipophilic and the hydrophylic part of the structure of the given compounds, leads to a decrease in local anaesthetic activity. This can be explained with lower values of partition coefficients, which is due to a shortening of the connecting chain by one methylene group and with the shorter distance between the lipophilic and the hydrophylic part of the molecule (necessary for the interaction of this local anaesthetics type with membrane receptors) [3, 4].

Table 2: Indexes of relative local anaesthetic activity for surface and infiltration anaesthesia, LD_{50} values in mice after s.c. application of compound BM4.

Comp.	Local anaesthetic activity		LD_{50}
	SLAA	ILAA	($mg \cdot kg^{-1}$)
BM4	46.5	23.5	400–500
Cocaine	1	3.6	125
Procaine	0.36	1	630

Experimental

1. Synthesis

The appropriately substitued 2-, 3- and 4-alkoxyphenylisocyanate (0.1 mol), synthesized according to Čižmárik et al. [2] in dry toluene (100 ml), were refluxed with 1-piperidinomethanol (0.1 mol) for 6 h. After refluxing, the reaction mixture was cooled and washed with H_2O (250 ml, 3 times). The solvent was distilled off. The resulting base was purified by CC (using CH_2Cl_2) and converted into its chloride. The salts were purified by crystallization from acetone or butanone.

The final derivatives were characterised by IR and UV spectra. Compound purity was verified by TLC and by determination of m.p.'s.

2. Physico-chemical properties

Dissociation constants were determined potentiometrically, surface tension of aqueous solutions was determined stalagmometrically and experimental partition coefficients were measured spectrophotometrically in a system octan-1-ol/aqueous phosphate buffer pH 7. Chromatographic parameters R_M were obtained from adsorption TLC.

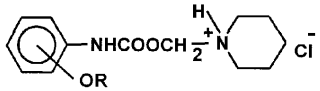
3. Pharmacology

3.1. Estimation of local anaesthetic activity

From the evaluated group the compound [(3-butoxyphenylcarbamoyloxy)-methyl]piperidiniumchloride (BM4) was selected for the estimation of local anaesthetic activity and acute toxicity. The basis for this decision was the knowledge about local anaesthetic activity of phenylcarbamic acid basic esters with shorter alkoxy substituents.

The surface local anaesthetic activity was estimated on the rabbit cornea and the infiltration local anaesthetic activity on the dorsal skin of guinea pigs after intradermal administration of drug solutions. The efficiency of compound BM4 was expressed as the index of local (surface and infiltra-

Table 1: Studied substances, R_F, R_M values from TLC, partition coefficient (log P′), surface tension γ and pK_a.



Compd. R	Formula Mol. wt.	M.p. (°C)	R_F	R_M	λ_{max}/A ε ($mol^{-1} \cdot m^2$)	λ_{max}/A ε ($mol^{-1} \cdot m^2$)	λ_{max}/A ε ($mol^{-1} \cdot m^2$)	log P′	γ ($N \cdot m^{-1}$)	pK_a
BO3 2-C_3H_7	$C_{16}H_{25}O_3N_2Cl$ 328.8	80–81[b]	0.81	−0.62	206/0.9458 18916	242/0.2482 4965	282/0.1090 2181	2.33	0.0710	6.55
BM3 3-C_3H_7	$C_{16}H_{25}O_3N_2Cl$ 328.8	96–97[b]	0.76	−0.50	216/0.7716 3858	244/0.3426 1713	280/0.0986 399	2.60	0.0700	6.10
BP3 4-C_3H_7	$C_{16}H_{25}O_3N_2Cl$ 328.8	144–145[b]	0.73	−0.43	204/0.8832 3533	244/0.6376 2550	284/0.0841 336	2.42	0.0715	6.98
BO4 2-C_4H_9	$C_{17}H_{27}O_3N_2Cl$ 342.9	51–52[a]	0.85	−0.75	208/0.7532 1673	242/0.3117 692	280/0.0924 205	2.39	0.0713	6.05
BM4 3-C_4H_9	$C_{17}H_{27}O_3N_2Cl$ 342.9	95–96[a]	0.79	−0.57	214/1.0351 5175	244/0.4296 2148	280/0.1053 526	2.97	0.0695	6.18
BP4 4-C_4H_9	$C_{17}H_{27}O_3N_2Cl$ 342.9	123–124[a]	0.75	−0.47	206/1.0039 4015	244/0.7314 2925	280/0.1033 413	3.04	0.0715	6.37

[a] substance crystallized from acetone
[b] substance crystallized from butanone

tion) anaesthetic activity in comparison to cocaine or procaine, according to the method of Vrba et al. [5].

3.2. Acute toxicity estimation

The letal effects of compound BM4 were determined after s.c. administration to mice. LD_{50} values were expressed as a range of the doses after which the animals stayed alive and those when the mortality in the group was 100% [6]. The results were recorded 24 h after administration of the compound.

References

1 Čižmárik, J.; Borovanský, A.: Chem. Zvesti **29**, 119 (1975)
2 Čižmárik, J.; Borovanský, A.; Švec, P.: Acta Fac. Pharm. Univ. Comenianae **29**, 53 (1976)
3 Pešák, M.; Kopecký, F., Čižmárik, J.; Borovanský, A.: Pharmazie **35**, 150 (1980)
4 Remko, M.; Čižmárik, J.: Česk. Farm. **35**, 199 (1986)
5 Vrba, Č.; Šekera, A.: Arch. Int. Pharmacodyn. Ther. **118**, 155 (1959)
6 Roth, Z.: Čsl. fysiol. **10**, 408 (1961)

Received December 23, 1999
Accepted March 22, 2000

PharmDr. Dagmar Sládková
Department of Pharmaceutical Chemistry
Pharmaceutical Faculty
Comenius University
Odbojárov 10
832 32 Bratislava
Slovak Republic

Department of Pharmaceutical Technology[1], Department of Pharmacognosy and Botany[2], Faculty of Pharmacy, Medical University, Sofia, Bulgaria

HPTLC densitometric determination of ruscogenins in dry extract of *Ruscus aculeatus* L.

B. Peneva[1], I. Krasteva[2], S. Nikolov[2] and E. Minkov[1]

A constant increase in the application of *Ruscus aculeatus* L. dry extract as a biologically active substance has been observed due to the possibility to include it into different drug formulations. Drugs based on *Ruscus aculeatus* L. dry extract exhibit good antiinflammatory, vasotonic, antihemorroidal and antiulcer effect [1, 2].

The dry extract used in our studies was prepared via the hot extraction method with successive drying on a Nitro Atomizer dispersion drier [4]. The product is a light brown powder, possessing specific slight odor. It is soluble in water and ethanol. Since the extract is very hygroscopic Aerosil 200 was included into its samples at concentrations 13, 24 and 32% with respect to the extract amount [5].

Ruscus aculeatus L. dry extract with and without Aerosil 200 was standardized according to the percentage of residual moisture (Table) and the quantitative content of the biologically active substances. Our previous studies have shown densitometric HPTLC to be the most appropriate method for determining the ruscogenins in *Ruscus aculeatus* L. dry extract [4, 5]. The quantitative determination was preceded by studies on the optimization of the time and conditions of the acid hydrolysis: 3–3.5 h; 1 N HCl in the presence of *n*-butanol. The results for ruscogenin content in the dry extract before and after optimization of the method run with and without Aerosil are presented in the Table as mean values obtained from five experiments. The results were estimated and the quantity of ruscogenins was calculated from the peak area in the plot and from the concentration of a standard ruscogenin solution.

Table: Characteristics of samples, following European Pharmacopoeia

Sample	Residual water (%)	Ruscogenins (mg)
Extract*	1.97	0.4300
Extract	1.97	0.4881
Extract + 13% Aerosil	1.42	0.4836
Extract + 24% Aerosil	1.36	0.4920
Extract + 32% Aerosil	1.29	0.4995

* Sample hydrolysed with 5% H_2SO_4 without *n*-butanol

As seen from the Table the increase of Aerosil concentration leads to a smooth decrease in the percentage of residual moisture. This is an evidence that Aerosil could be used as a support for the dry *Ruscus* extract. Meanwhile the amount of ruscogenins remains almost unchanged in all samples. The only exception is the sample subjected to densitometry. It contains a smaller amount of ruscogenins. This is probably due to the fact that the acid hydrolysis of this extract sample was run with 5% sulphuric acid without *n*-butanol. On the other hand, the higher ruscogenins amounts in the other samples could be explained by the good solubility of aglycons in non polar organic solvents.