143. G. Regnier, R. Canevari, and M. Laubie, Experientia, 28, 814 (1972).
144. M. Laubie and F. Diot, J. Pharmacol. (Paris), 3, 363-374 (1972).
145. M. Gastaud, C. Dolisi, J. P. Camous, et al., C. R. Soc. Biol., 171, 169-175 (1977).
146. S. Chiba, Y. Nagawa, and T. Yui, Nippon Yakurigaku Zasshi, 66, 76-82 (1970).

QUATERNARY AMMONIUM SALTS WITH A LABILE $\overset{+}{N}$—C BOND AS PRECURSORS OF MEDICINAL COMPOUNDS.

II. COMPOUNDS WITH A LABILE $\overset{+}{N}$—CH_2—N GROUPING

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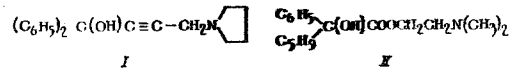
UDC 615.014.47:546.39

We have already reported on the reversible chemical modification of several pharmacologically active compounds, containing a tertiary amino group, by conversion into quaternary ammonium salts (QS).

We showed that because of the presence of this group, salts containing a $-CH_2OCOR$ grouping attached to a nitrogen atom can undergo a hydrolytic cleavage at the $\overset{+}{N}$—C bond with regeneration of a tertiary amine [1]. It was also found [2] that medicinal compounds modified in the form of their acyloxymethylates have a more prolonged action and are less toxic.

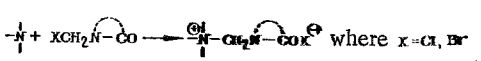
It could be assumed that, in analogy with the compounds already studied, quaternary salts with $\overset{+}{N}$—CH_2NHCOR or N—$CH_2\overset{+}{N}$—CO groupings can also be converted into the initial tertiary amines as the result of hydrolysis. But they should differ from their oxygen-containing analogs in the rate of hydrolysis, lipophilicity, and possibly in other properties, determining their pharmacokinetics. The present investigation deals with the development of methods of synthesis and study of certain properties of QS containing an acylamido(imido)methyl grouping.

For the investigation we used known pharmacologically active compounds containing a tertiary amino group: butynoline (azulone) (I) and cyclosil (II) [3].

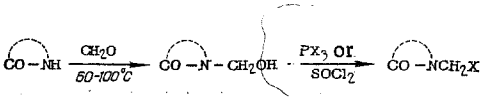


I II

To obtain the QS, we alkylated the free bases of these compounds by methyl halide derivatives of amides and imides.



The alkylating agents were synthesized by a two-stage method [4, 5].



By this method we obtained the halomethyl derivatives of benzamide, succinimide, phthalimide, benzoxazolone, benzimidazolone, hydantoin, andbarbituric acid. Because of the formation of a polymer at the last stage of the synthesis, we were unable to obtain bis-N-halomethylurea.

The amines were alkylated in acetone or in chloroform for 1-6 days at a temperature not above 55°C, since at a higher temperatre the halomethyl derivatives undergo a partial disproportionation [6].

All the synthesized QS (Table 1) are colorless crystalline compounds, readily soluble in alcohol, DMFA and DMSO, and somewhat less soluble in water, chloroform, acetone, and methylene

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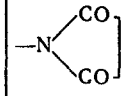
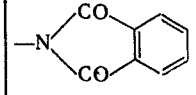
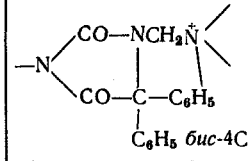
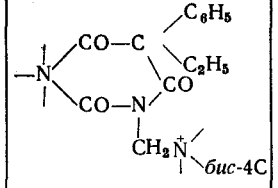
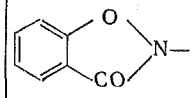
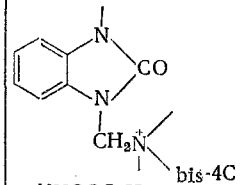
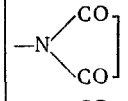
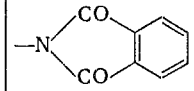
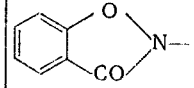
TABLE 1. Properties of QS with General Formula $\left[\diagup\!\!\diagdown N-CH_2R\right]^{\oplus}X^{\ominus}$

Compound	$-\overset{|}{N}-$	R	X	Yield, % (medium)	mp, °C (system of solvents)	Found, Cl or Br, %	Empirical formula	Calculated Cl or Br, %	PMR spectrum (in deuteromethanol) $\overset{+}{N}-CH_2-N$	δ ppm $-\overset{+}{N}-CH_2C\equiv C$ or $\overset{\alpha}{\diagup\!\!\diagdown}\overset{+}{N}-CH_2CH_3$
I	Butynoline		Cl	79 (chloroform)	161—2 (chloroform—acetone)	7,86	$C_{25}H_{27}ClN_2O_3$	8,09	5,03	4,54
II	"		Cl	68 (chloroform)	195—196,5 (chloroform—acetone)	6,97	$C_{29}H_{27}ClN_2O_3$	7,29	5,22	4,58
III	"		Br	80 (chloroform)	147—150 (with decomposition; chloroform—acetone)	15,38	$C_{57}H_{56}BrN_4O_4$	15,68	5,39 5,17	4,45
IV	"		Br	42 (chloroform)	200—202,5 (chloroform—methanol)	15,85	$C_{54}H_{56}Br_2N_4O_5$	15,98	5,25	4,61

V	Butynoline		Cl	65 (chloroform)	201—3 (acetone—ethanol)	7,30	$C_{28}H_{27}ClN_2O_3$	7,47	5,26	4,63
VI	"		Cl	55 (chloroform)	172—4 (with decomposition; acetone—ethanol)	8,78	$C_{49}H_{50}Cl_2N_4O_3$	8,72	5,48	4,54
VII	"	—NHCOC$_6$H$_5$	Cl	61 (chloroform)	121—3 (with decomposition; chloroform—dioxane)	7,20	$C_{28}H_{29}ClNO_2$	7,70	5,0	4,65
VIII	Cyclosil		Cl	58 (acetone)	97—9 (acetone—methanol)	7,80	$C_{22}H_{31}ClNO_5$	8,09	4,85	4,58
IX	"		Br	96 (acetone)	102—3 (acetone—methanol)	14,40	$C_{26}H_{31}BrN_2O_5$	15,01	4,86	4,52
X	"		Cl	68,5 (acetone)	191—2,5 (not recrystallized)	7,40	$C_{25}H_{31}ClN_2O_5$	7,48	4,90	4,55
XI	"	—NHCOC$_6$H$_5$	Cl	64 (absolute ether)	154—5 (acetone—ether)	7,38	$C_{25}H_{33}Cl_2O_4$	7,70	4,8	4,48

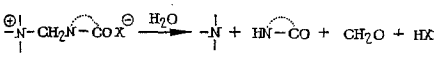
chloride. It should be noted that salts containing the $\overset{+}{N}-CH_2N$ grouping have a greater affinity towards organic weakly polar solvents than the previously obtained butynoline and cyclosil acyloxymethylates, which are practically insoluble in them.

The synthesized salts were found to be stable: on storage for 1 year in a closed vessel their physicochemical parameters did not change.

The structure of QS (I–XI) was confirmed by PMR spectra, characterized by the presence of a proton signal of the $\overset{+}{N}-CH_2N$ grouping in the 4.82-5.48 ppm region (Table 1). The chemical shifts of protons of salts (I) and (VIII) are listed in Table 2, as an example.

The most important characteristic of QS with a labile $\overset{+}{N}-C$ bond is their ability to decompose at this bond in aqueous solutions under close to physiological conditions (pH 7.4, temperature 37°C).

The study of the behavior of QS under more rigid conditions (pH 10.0-12.0) showed that most of them decompose quantitatively to form the initial tertiary amine (confirmed by GLC), a corresponding amide or imide (confirmed by TLC and GLC), a hydrogen halide (confirmed by titration) and formaldehyde (qualitatively confirmed by the method of Nesh [7]). From these results, the hydrolysis of QS containing the $\overset{+}{N}-CH_2N$ grouping can be represented by an overall scheme similar to that for the dissociation of acyloxymethylates of tertiary amines.



This is also confirmed by time-dependent changes observed in the PMR spectra of solutions of the salts in a deutero-alkaline buffer at pH 8.3-8.7. For example, in the PMR spectra of QS (VIII), (IX) and (XI), besides a decrease in the intensity of the proton signals of the $\overset{+}{N}-CH_2N$ grouping (δ = 4.85 ppm) and proton signals of the methyl groups bound to the quaternary nitrogen atom (δ = 2.91–2.98 ppm), the appearance and increase in the intensity of the singlet of the protons of the methyl groups attached to the tertiary nitrogen atom (δ = 2.80-2.85 ppm) was also observed.

To evaluate the rate of hydrolysis of the QS studied, we developed a gas–liquid chromatographic procedure for the quantitative determination of the tertiary amine (butynoline, cyclosil) formed during dissociation of the salt. Since in the case of quaternary derivatives of cyclosil, the study of the process that interested us was complicated by partial dissociation of the molecule at the ester bond, main attention was paid to the butynoline derivatives.

As the result of our investigation (Table 3), we were able to find a dependence between the rate of hydrolysis of the QS of butynoline and the structure of the imide radical bound to the quaternary nitrogen atom through a methylene group. These data show that the rate of hydrolysis changes over wide limits.

While the QS (I), (II) are in this respect comparable with butynoline methacryloyloxymethylate, salt (V) is much more stable, and compound (VI), even at high pH values, does not generally show any tendency to hydrolyze. The dependence of the rate of hydrolysis of QS on temperature and pH of the medium is also sharply expressed. As expected, increase in these parameters accelerates the hydrolysis. In water and in slightly acid media, the salts dissociate extremely slowly.

From the results of the analysis *in vitro*, we can assume that the modification of the medicinal compounds at the tertiary nitrogen atom in the form of QS containing structurally different $\overset{+}{N}-CH_2\overset{|\cdots|}{N}-CO$ groups will make it possible to enlarge the number of precursors of pharmacologically active tertiary amines, and from these to select compounds liberating the active principle in the living organism at the required rate.

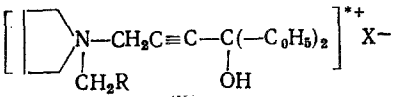
EXPERIMENTAL

The PMR spectra were run on the RYa-2310 apparatus (60 MHz) in deuteromethanol and deuterated DMSO. Gas–liquid chromatography was carried out on the "Hewlett–Packard" chromatograph (USA) with a flame ionization detector, glass column (length 1.8 m, diameter 2 mm), phase – 3% OV-17 on chromosorb W = DNCS (100-120 mesh), thermostat temperature of the column 210°C for cyclosil and 260°C for butynoline, vaporizer temperature 300°C, detector temperature 300°C. Gas carrier – nitrogen (45 ml/min). The pH of the buffer solutions and the potentiometric titration were determined on an apparatus model pH-121 (USSR). The borate and phosphate buffers were prepared by known methods [9].

TABLE 2. Data on PMR Spectra of QS (I), (VIII) (deuteromethanol)

QS	C_6H_5	C_5H_9	$+\underset{\alpha}{N}CH_2\underset{\beta}{CH_2}$	$+NCH_2C\equiv C$	$+NCH_2N$	$+N(CH_3)_2$	
I	7,34—7,55 multiplet	—	—	4,54 singlet	5,03 singlet	—	α 3,72 multiplet β 2,19 multiplet
VIII	7,32—7,65 multiplet	1,44—1,6[a] multiplet	4,59 multiplet 5,74 multiplet	—	4,82	2,93 singlet	—

TABLE 3. Data on Hydrolysis of Butynoline Derivatives with General Formula



Compound	pH	Temperature, °C	$K_{obs} \cdot 10^3$ min^{-1}*	$\tau_{1/2}$, min
I	10,12	22	—	5
	9,71	22	35	20
	9,20	22	10	65
	8,74	22	4,8	145
	8,74	37	23	30
	8,35	37	12,8	55
	7,45	37	4,8	145
II	8,74	22	6,9	100
	9,20	37	53	13
III	8,74	37	16	62
	8,35	37	7,3	95
	7,45	37	0,8	850
V	10,12	22		160
	9,20	22		250 30% hydrolysis
	8,35	22		275 16% hydrolysis
VI	10,12	22	—	Without change for 350 min
Butynoline methacryloyloxymethylate	8,7	22	13,8	50
	7,5	37	5,5	125

*K_{obs} is a pseudo-first order constant obtained by the Guggenheim method [8] from data of 2-3 parallel experiments.

The solvents used in the investigation were thoroughly dehydrated and purified by known methods [10].

N-(4,4-Diphenyl-4-hydroxy-2-butynyl)-N-phthalimidomethyl Pyrrolidinium Chloride (II). The reaction mixture containing 1.5 g (0.005 mole) of butynoline base and 1.25 g (0.006 mole) of N-chloromethylphthalimide in 20 ml of anhydrous chloroform was left to stand for 5 days at room temperature, and then boiled for 6 h. Part of the solvent was distilled, and 30 ml of ether were added. The precipitate was filtered, washed with acetone (twice with 20 ml), ether (twice with 20 ml), and dried in a vacuum exciccator. The product was recrystallized from a mixture of chloroform and acetone (4:3). Yield 1.7 g (68%), mp 195-196.5°C.

All the remaining QS, whose properties are listed in Table 1, were obtained by a similar procedure. The reaction was controlled by TLC on Silufol UV-254 plates (CSR), and the purity of the salts obtained was controlled by GLC.

Chloro-N-benzamidomethylate of N,N-Dimethylaminoethyl Ester of Phenylcyclopentylglycolic Acid (XI). A 0.32 g portion (0.0019 mole) of N-chloromethylbenzamide in 10 ml of ether was

added, with stirring, to a solution containing 0.5 g (0.0017 mole) of cyclosil base in 8 ml of absolute ether. After 2 h, the precipitate was filtered, washed with ether (three) times with 20 ml), pentane (twice with 20 ml), and dried in a vacuum exciccator. Yield, 0.47 g (64%), mp 154-155°C.

Gas-Liquid Chromatography Procedure for Quantitative Determination of Butynoline and Cyclosil in Hydrolysate on Example of QS I. An aqueous solution of QS I (0.048 g in 4 ml of double-distilled water) is poured in portions of 0.2 ml into test tubes containing 1 ml of a buffer at a given pH, thermostated at 22°C or 37°C. After a given period of time, 2 ml of toluene containing an inner standard (dipheridine — $(C_6H_5)_2COH-C{\equiv}CCH_2N$⟨⟩, are added. The mixture is shaken, the toluene layer is removed by centrifugation, and the residue chromatographed. The content of butynoline in the test probe is determined from the ratio of peak areas of amine and inner standard, according to a calibration graph.

LITERATURE CITED

1. N. D. Vinogradova, S. G. Kuznetsov, and S. M. Chigareva, Khim.-Farm. Zh., No. 9, 41 (1980).
2. S. G. Kuznetsov, N. D. Vinogradova, and S.M. Chigareva, Inventor's Certificate No. 724498 USSR).
3. M. Negwer, Organic-Chemical Drugs and Their Synonyms, Vol. 12-13, Berlin (1978).
4. H. E. Laugg and W. B. Martin, Organic Reactions, Vol. 14 (1965), p. 52.
5. S. A. Vida and W. R. Wilber, J. Med. Chem., 14, 190 (1971).
6. M. K. Hagreaves, Chem. Rev., 70, 439 (197).
7. C. Nesh, Biochem. J., 55, 416 (1953).
8. E. A. Guggenheim, Philos. Mag., 2, 538 (1926).
9. E. N. Vinogradova, Methods of Determination of Concentration of Hydrogen Ions [in Russian], 2nd edn., Moscow (1956).
10. L. Fieser and M. Fieser, Reagents for Organic Synthesis [Russian translation], Vol. 1-5 (1970-1971).