Analogs of Parasympathetic Neuroeffectors. I. Acetylselenocholine, Selenocholine, and Related Compounds[1]

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Acetylselenocholine, benzoylselenocholine, butyrylselenocholine, selenocholine diselenide, and some of their dimethyl and monomethyl analogs have been prepared. The use of the Bunte salts of selenocysteamines for the preparation of diselenides is described.

Acetylcholine is one of the rather small number of compounds the biological actions of which can be considered in terms of molecular interactions.[2] On the other hand, the pharmacology[3,4] of its sulfur analog, acetylthiocholine, a compound widely used as a histological stain,[5] remains rather confusing.

Recent work has indicated that in thioacyl and selenoacyl analogs, which are essentially isosteric, electron distribution and electron mobility are rather different, an effect responsible for the observation that selenoacyl compounds undergo aminolysis much more readily than isologous thioacyl compounds[6-8] and for the differences in the spectra of thioacyl and selenoacyl analogs.

Since electron distribution would be expected to differ in acetylcholine, acetylthiocholine, and acetylselenocholine, while these compounds presumably have a rather similar ability to fit receptor sites, a systematic study of the comparative pharmacology of this group of analogs, and of their reactions with true acetylcholinesterase, pseudocholinesterase, and with nucleophilic reagents, was initiated in the hope that it would provide additional information about the receptor sites to which these compounds attach themselves.

The selenium of 2-aminoethylselenium compounds can be introduced by the reaction of the corresponding 2-aminoethyl halides with potassium selenocyanate[9, 10] after protection of the amino group. It has now been found that 2-aminoethylselenols and the corresponding diselenides can be synthesized most conveniently by way of the intermediate formation of 2-aminoethyl selenosulfates, the Bunte salts of 2-aminoethylselenols.

Potassium selenosulfate, obtained by dissolving finely powdered selenium in an aqueous solution of potassium sulfite,[11] reacted smoothly with 2-amino- and 2-alkylaminoethyl halide hydrohalides, to yield the inner salts of 2-aminoethyl selenosulfates (I–IV, Table I). Hydrolysis with *N* hydrochloric acid at 50–60° of the selenosulfates yielded the corresponding diselenides. These were yellow oils from which the crystalline hydrochlorides (V–VII, Table I) could be prepared. Bis-(2-trimethylammoniumethyl) diselenide (VIII) could not be obtained in pure form by this route, due to contamination with inorganic salts.

Diselenides could be reduced to the corresponding selenols by sodium borohydride in water or in organic solvents in the presence of methanol, as noted previously.[12] In all cases it was found that slightly more than 2 molar equivalents of sodium borohydride discharged the yellow color of the diselenides, yielding the sodium salts of the selenols, which were used for further reaction without isolation. 2-Aminoethyl selenobenzoates (XI–XIII, Table I) were formed in good yield by treatment of the reduction products with benzoyl chloride in the presence of sodium bicarbonate buffer[6]; N-methyl-N,Se-dibenzoyl selenocysteamine (XII) crystallized from the reaction mixture; and 2-dimethylaminoethyl selenobenzoate (XIII) was isolated as the hydrochloride. Treatment of the free base of XIII with methyl iodide in an ether–acetone mixture gave benzoylselenocholine iodide (XVI) in excellent yield. Se-Benzoylselenocholine bromide (XVII) and Se-butyrylselenocholine iodide (XVIII) were prepared in a similar fashion. Acetylselenocholine (XIV, XV) could not be prepared *via* the Schotten–Baumann acylation since the intermediate 2-dimethylaminoethyl selenoacetate appeared to be unstable under the reaction conditions. 2-Dimethylaminoethyl diselenide (VII) was, therefore, reduced in methanol solution, a large excess of acetic anhydride was added, and the mixture was evaporated to dryness under reduced pressure. The oily residue was extracted with a mixture of acetone and ether, filtered, and permitted to react with methyl iodide to yield acetylselenocholine iodide (XIV) as colorless prisms. Acetylselenocholine bromide was prepared in an analogous fashion. Quaternization of VII yielded the diselenide form of selenocholine as its diiodide (VIII). Methylation of the borohydride reduction product of VII in an ether–acetone mixture gave 2-trimethylammoniumethyl methyl selenide iodide (X). An attempt to suppress the methylation of the selenol group by addition of glacial acetic acid, as in the analogous synthesis of thiocholine,[13] resulted in the preferred methylation of the selenol group, giving 2-dimethylaminoethyl methyl selenide hydriodide (IX) instead of the expected isomeric

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TABLE I

$R^1R^2R^3N^+CH_2CH_2SeR^4 \cdot X^-$

No.	R^1	R^2	R^3	R^4	X	% yield	M.p., °C.[d]	Recrystn. solvent
I	H	H	H	SO_3^-	...	80	196[c]	80% EtOH
II	CH_3	H	H	SO_3^-	...	60	153[b]	MeOH
III	CH_3	CH_3	H	SO_3^-	...	60	152[b]	MeOH
IV	CH_3	CH_3	CH_3	SO_3^-	..	42	260[b]	Aq. MeOH
V	H	H	H	*a*	2Cl	55	177–179[b]	C_6H_6–EtOH
VI	CH_3	H	H	*a*	2Cl	50	195–198[b]	EtOH
VII	CH_3	CH_3	H	*a*	2Cl	60	210–212[b]	EtOH
VIII	CH_3	CH_3	CH_3	*a*	2I	73	245[b]	MeOH
IX	CH_3	CH_3	H	CH_3	I	34	120[b]	MeOH–ether
X	CH_3	CH_3	CH_3	CH_3	I	100	205[b]	EtOH
XI	C_6H_5CO	H	...	C_6H_5CO	...	90	99–100	Aq. MeOH
XII	C_6H_5CO	CH_3	...	C_6H_5CO	...	90	91–92	Petr. ether
XIII	CH_3	CH_3	H	C_6H_5CO	Cl	99	167–170	EtOH–ether
XIV	CH_3	CH_3	CH_3	$COCH_3$	I	69	177–178[b]	EtOH
XV	CH_3	CH_3	CH_3	$COCH_3$	Br	65	202–203[b]	EtOH
XVI	CH_3	CH_3	CH_3	COC_6H_5	I	65	228–230[b]	EtOH
XVII	CH_3	CH_3	CH_3	COC_6H_5	Br	73	186–188[b]	EtOH
XVIII	CH_3	CH_3	CH_3	CO-*n*-C_3H_7	I	58	167[b]	MeOH–EtOH

[a] The symmetrical diselenide. [b] M.p. dec. [c] Compound resolidifies, then decomposes at 270°. [d] Melting points were determined using an electrically heated Scientific Glass Co. copper block apparatus and were corrected. [e] Microanalyses were performed at the

reduced selenocholine iodide. That IX was a tertiary amine rather than a quaternary ammonium salt was established by the pK_a of this compound being 8.7, a value similar to the pK_a 8.4 of VII. The selenol form of selenocholine has, so far, not been obtained in pure form. Various attempts to reduce its diselenide or to hydrolyze its acetyl or benzoyl esters under alkaline conditions resulted largely in a decomposed product with elimination of trimethylamine. Attempts to precipitate the selenol with various heavy metal salts yielded impure precipitates or resulted in removal of selenium from the organic moiety and precipitation of the inorganic metal selenides.

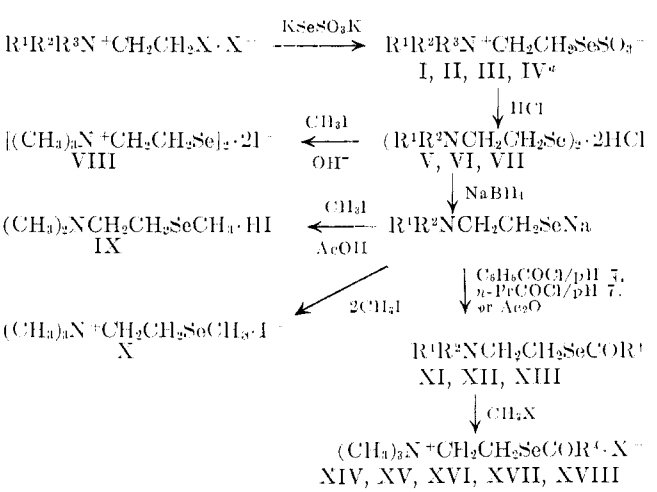
An investigation of the comparative pharmacological actions of acetylcholine, acetylthiocholine, and acetylselenocholine has been undertaken. It was found that while the hydrolysis product of acetylcholine, choline, is relatively inert in the guinea pig ileum and frog rectus abdominis preparations, the hydrolysis products cholinethiol and cholineselenol exhibit muscarinic activities exceeding those of the esters from which they are derived.[14, 15] Thus, while physostigmine, an acetylcholinesterase inhibitor, will greatly enhance the muscarinic activity of acetylcholine by preventing its destruction, it reduces the activities of the analogous thiolester and selenolester by preventing the formation of the more active hydrolysis products. Oxidation of cholinethiol and cholineselenol to the disulfide and diselenide, respectively, reduces muscarinic activity. In view of the pharmacologically important differences between "single armed" and "doubled armed" onium compounds, it should be noted that simple oxidation of cholinethiol or cholineselenol represents a convenient method of converting "single armed" into "doubled armed" compounds.

Relative potencies of the above compounds in the frog rectus abdominis preparation are summarized in Table II. Experimental details will be published elsewhere.[15]

TABLE II

Compd.	Equipotent molar ratios, frog rectus abdominis
Acetylcholine	1
Acetylthiocholine	40
Acetylselenocholine	60
Choline	5000
Cholinethiol	30
Cholineselenol	*a*
Choline diselenide	560

[a] Due to rapid oxidation to the weakly active diselenide, it is difficult to obtain reproducible quantitative data with this compound. The activity of cholineselenol definitely exceeds that of acetylselenocholine.



[a] Numbering refers to compounds in Table I.

Experimental

2-Aminoethyl Selenosulfates (I, II, III, IV).—A stirred suspension of 7.9 g. of finely powdered selenium in a solution of 16 g. of potassium sulfite in 50 ml. of water was heated for 30–60 min. at 70–80° until most of the selenium had dissolved. The solution was then filtered into a 500-ml. flask fitted with a mechanical stirrer, reflux condenser, and dropping funnel. To the vigorously

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Formula	% C[e] Calcd.	% C[e] Found	% H[e] Calcd.	% H[e] Found	% N[e] Calcd.	% N[e] Found	% Se[e] Calcd.	% Se[e] Found	Calcd.	Found
$C_2H_7NO_3SSe$	11.77	12.01	3.46	3.50	6.86	6.86	...	...	...	...
									Sulfur	
$C_3H_9NO_3SSe$	16.51	16.50	4.16	4.11	6.42	6.65	36.19	35.81	14.70	14.50
$C_4H_{11}NO_3SSe$	20.69	20.98	4.78	4.95	6.03	6.05	34.01	34.09	13.81	14.00
$C_5H_{12}NO_3SSe$	24.39	24.59	5.32	5.39	5.69	5.79	32.07	32.17	13.02	12.82
$C_4H_{14}N_2Se_2Cl_2$	*f*									
$C_6H_{18}N_2Se_2Cl_2$	20.76	20.41	5.23	5.27	8.07	8.26	45.5	45.15		
$C_8H_{22}N_2Se_2Cl_2$	25.61	25.40	5.91	5.83	7.47	7.78	42.1	41.9	Iodine	
$C_{10}H_{26}N_2O_2Se_2I_2$	20.49	19.86	4.47	4.12	4.78	4.97	26.95	26.71	43.31	43.58
$C_5H_{14}INSe$	20.42	20.49	4.80	4.63	4.76	5.09	26.85	26.52	43.16	43.59
$C_6H_{16}NSeI$	23.39	23.75	5.24	5.61	4.55	4.40	25.63	25.4	...	...
$C_{16}H_{15}NO_2Se$	*g*									
$C_{17}H_{14}NO_2Se$	58.96	59.26	4.95	4.34	4.05	4.10	22.80	22.78		
$C_{11}H_{16}ClNOSe$	45.14	44.92	5.50	5.50	4.79	4.88	26.98	27.22		
$C_7H_{16}NOSeI$	25.02	24.93	4.80	4.89	4.17	4.24	23.49	23.03	37.76	37.73
$C_7H_{16}NOSeBr$	29.08	29.00	5.58	5.51	...	...	...	...	...	...
$C_{12}H_{18}NOSeI$	36.20	36.38	4.56	4.73	3.52	3.29	19.83	19.85	31.87	32.17
$C_{12}H_{18}NOSeBr$	41.40	41.25	5.16	5.12	...	...	...	...	...	...
$C_9H_{20}NOSeI$	29.68	29.89	5.53	5.68	3.85	4.11	21.68	21.36	...	...

[e] Schwarzkopf Laboratories, Woodside, N. Y. [f] For carbon and other analyses, see ref. 9 and 10. [g] For carbon and other analyses, see ref. 6.

stirred solution was added dropwise, with continued heating, a solution of 0.1 mole of 2-aminoethylhalide hydrohalide in 200 ml. of ethanol at such a rate that the deposit of red selenium redissolved before the next drop was added and that the reaction mixture maintained a pale green color. The addition was completed within 60 min.; the solution was filtered from precipitated salts and chilled overnight. The crystalline deposit was collected and extracted with 100-ml. portions of hot 80% ethanol until a sample extract did not turn yellow on addition of an equal volume of concentrated hydrochloric acid. The filtrate and the extracts were then combined and evaporated to dryness under reduced pressure. Recrystallization of the residue from methanol or dilute ethanol gave the selenosulfates as the inner salts in 60–80% yield. The crude selenosulfates contained inorganic salts. They were suitable for further reactions without any purification; repeated crystallizations were necessary for the analytical samples.

Bis(2-alkylaminoethyl) Diselenides (V, VI, VII).—A solution of 2-alkylaminoethyl selenosulfate (0.1 mole) in *N* hydrochloric acid (150 ml.) was slowly evaporated to dryness under reduced pressure at a bath temperature of 50–60°. The procedure was repeated and the bright yellow residue was then extracted with boiling absolute ethanol until the extracts were colorless. The combined ethanol solutions were evaporated to a smaller volume and the products were allowed to crystallize in ice. The diselenides were obtained as the hydrochlorides in 50–60% yield; a white residue which could not be extracted with absolute ethanol contained potassium chloride and unidentified material.

N-Methyl-N,Se-dibenzoylselenocysteamine (XII).—A solution of bis(2-methylaminoethyl) diselenide dihydrochloride (VII) (5.5 g.) in water (75 ml.) was reduced in a nitrogen atmosphere by gradual addition of sodium borohydride (1.6 g.) until the solution was colorless. Sodium hydrogen carbonate (15 g.) was added, followed by benzoyl chloride (12 g.) and the mixture was stirred vigorously with cooling in an ice bath until the smell of benzoyl chloride was no longer noticeable. The colorless solid which precipitated was collected and recrystallized from dilute methanol or from petroleum ether to yield colorless needles of the dibenzoyl compound (XII).

2-Dimethylaminoethyl Selenobenzoate Hydrochloride (XIII).—A solution of bis(2-dimethylaminoethyl) diselenide dihydrochloride (VII, 3.75 g.) in water (25 ml.) was reduced under nitrogen with sodium borohydride (0.8 g.) until the solution was colorless. Sodium hydrogen carbonate (5 g.) was then added, followed by benzoyl chloride (2.5 ml.). The mixture was stirred vigorously in an ice bath until the smell of benzoyl chloride was no longer noticeable. The product was then extracted quickly with several portions of ether; the ether solution washed with saturated sodium sulfate solution and dried over anhydrous sodium sulfate. One half of this solution was evaporated to dryness, the residue taken up in absolute ethanol, and the calculated amount of concentrated hydrochloric acid in absolute ethanol added. On addition of ether and cooling in ice, shiny plates (2.9 g.) of XIII separated.

Se-Benzoylselenocholine Iodide (XVI).—The second half of the dried ether solution from the above experiment was mixed with a solution of methyl iodide (2 ml.) in acetone (25 ml.). The colorless precipitate was collected after 1 hr. at room temperature; it crystallized from ethanol in colorless needles. The corresponding quaternary ammonium bromide (XVII) was made in an analogous fashion.

Se-Butyrylselenocholine iodide (XVIII) was prepared in a procedure analogous to that used for the benzoyl ester XVI.

Se-Acetylselenocholine Iodide (XIV).—Bis(2-dimethylaminoethyl) diselenide (from 3.7 g. of the dihydrochloride VII) in methanol (20 ml.) was reduced with sodium borohydride (0.8 g.) and then mixed with acetic anhydride (80 ml.). After 15 min. at room temperature the solution was evaporated to dryness under reduced pressure and dried on an oil pump; the residue was extracted with ether. The ether solution was filtered and methyl iodide (3 ml.) added. The colorless precipitate was collected after 2 hr. at room temperature and recrystallized from absolute ethanol. The corresponding quaternary ammonium bromide (XV) was made using methyl bromide.

Bis(2-trimethylammoniumethyl) Diselenide Diiodide (Diselenide Form of Selenocholine.) (VIII).—Bis(2-dimethylaminoethyl) diselenide dihydrochloride (3.75 g.) was mixed with *N* sodium hydroxide solution (50 ml.) and the orange-yellow oil was extracted with ether (100 ml.). The ether solution was washed with saturated sodium sulfate solution, dried over sodium sulfate, diluted with an equal volume of acetone, and methyl iodide (2 ml.) was added. The precipitate of VIII (4.3 g., 73%) was collected after 2 hr.; it recrystallized from methanol as yellow flakes.

2-Trimethylammoniumethyl Methyl Selenide Iodide (X).—A solution of bis(2-dimethylaminoethyl) diselenide (from 18.7 g. of the dihydrochloride VII) in methanol (50 ml.) was reduced by addition of sodium borohydride (3.7 g.) until the mixture was colorless. Acetone (200 ml.) was added, followed after 1 min. (to allow for the discharge of any excess borohydride) by methyl iodide (29 g.), and the stoppered flask was kept at room temperature overnight. The crystalline precipitate (25.0 g.) was collected and an equal volume of ether was added to the mother liquor. This yielded another 5.0 g. of product, making the yield quantitative.

2-Dimethylaminoethyl Methyl Selenide Hydriodide (IX).—Bis(2-dimethylaminoethyl) diselenide dihydrochloride (2.0 g.) was suspended in methyl alcohol (10 ml.) in a 40-ml. centrifuge tube. Solid sodium borohydride was added until the mixture appeared colorless; then the turbid solution was diluted with glacial acetic acid (7 ml.) and the tube filled with dry, peroxide-free ether. The tube was stoppered and centrifuged to remove salt precipitates. The supernatant was transferred quickly (to

avoid air oxidation) to another centrifuge tube containing methyl iodide (2 ml.), mixed, and centrifuged for 1 min. to remove a yellow oil. From the last supernatant, colorless needle clusters separated which were collected after 1 hr. at room temperature.

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Synthesis of A New Class of Antishock Agents[1a]

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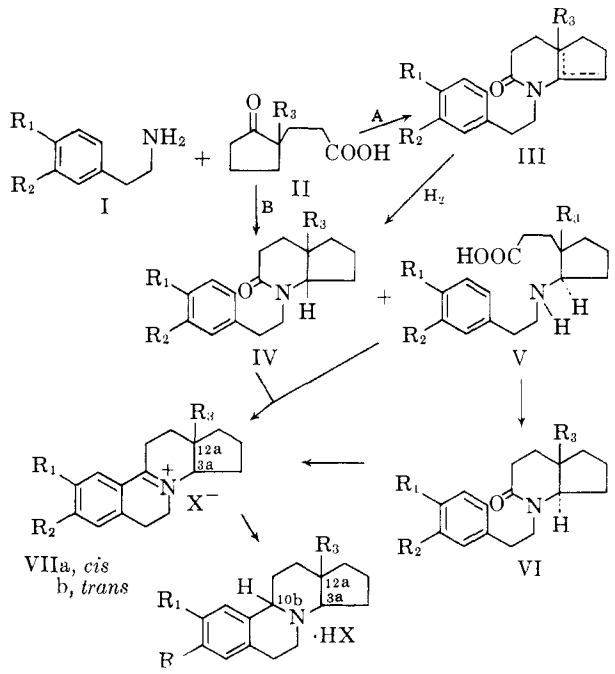
Antishock activity has been found in a series of octahydro- and decahydrobenzo[*a*]cyclopenta[*f*]quinolizines. Methods of synthesis of such compounds and the relative antishock activities are described. From the data available, no structure–activity relationship is evident.

The cardiovascular, and, in particular, the potent antishock properties of 2,3,3a,5,6,11,12,12a-octahydro-8-hydroxy-1H-benzo[*a*]cyclopenta[*f*]quinolizinium bromide (VIIa, R_1, R_3 = H; R_2 = OH) have been described by Osborne.[1b] For the same compound, Detar[2] has reported a positive inotropic effect on the cat papillary muscle preparation. In view of the interest in this and related products as representatives of a new class of antishock agents, a preliminary discussion of their synthesis is presented in this paper. Although a representative number of such products have been prepared, no structure-activity relationship has yet been uncovered. This work, however, is still in progress, and a subsequent report will be presented at a later time to delineate the scope of this activity.

CHART I



VIIIa, *syn–cis*[a]
b, *anti–cis*[a]
c, *anti–trans*[a]

[a] *syn–anti* refers to the 10b–12a relationship; *cis–trans* refers to the 12a–13a ring fusion.

Two procedures have been used for the preparation of these compounds, as indicated by routes A and B shown in Chart I.

According to route A, the starting substituted phenethylamine (I, R_1 and R_2 = H or OMe) and cyclopentanone-2-propionic acid (II, R_3 = H or Me) were condensed in refluxing xylene to afford moderate (50–70%) yields of the unsaturated lactam (III). This product underwent stereospecific reduction over palladium–carbon catalyst to give the *cis* lactam (IV). According to route B, IV was obtained directly in *ca.* 65% yield by a reductive condensation of the two starting materials in ethanol over palladium–carbon. This procedure also gave a 25% yield of the *trans* amino acid (V), which cyclized at its melting point to the *trans* lactam (VI). Ring closures on either of the lactams IV or VI or the amino acid V were carried out in high yields with phosphorus oxychloride in benzene. The resulting *cis* and *trans* quaternary salts (VIIa and b, R_1 and R_2 = H or OMe) were then demethylated directly with hydrobromic acid to afford the corresponding phenolic salts.

Tentative assignments of the *cis* and *trans* configurations to IV and V, respectively, were made initially by consideration of the relative ease of lactam formation from the initially formed amino acid mixture. Inspection of molecular models of the *cis* and *trans* forms of amino acid V supported the contention that the more easily formed lactam was the *cis*. Chemical proof of these assignments was obtained by the reaction sequence outlined in Chart II for the series where R_1 = R_3 = H and R_2 = OMe.

The isomeric lactams IV and VI were reduced with lithium aluminum hydride to the corresponding isomeric tertiary bases, IXa and b, characterized as their crystalline hydrobromide salts. There were then prepared samples of the known[3] *cis* (Xa) and *trans* (Xb) octahydro-1-pyrindines, and these two bases reacted with *m*-methoxyphenylacetyl chloride to give the oily amides (XIa, XIb). Reduction of XIa and XIb with lithium

(1) (a) This work was presented in part before the Division of Organic Chemistry, 144th National Meeting of the American Chemical Society, Los Angeles, Calif., April, 1963, Abstracts, p. 39M; (b) M. W. Osborne, M. M. Winbury, and W. M. Govier, *Federation Proc.*, **22**, 308 (1963).

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