[Chem. Pharm. Bull. 19(6)1089—1094(1971)]

UDC 547.233.04 : 541.486

Syntheses of Related Compounds of Selenocysteamine and Their Complex Formation with Metal Ions

Akira Yokoyama, Hiromu Sakurai, and Hisashi Tanaka

Faculty of Pharmaceutical Sciences, Kyoto University[1]

(Received July 25, 1970)

In order to discuss the property of selenium as a ligand atom and the effect of the substituent to complex formation in the related compounds of selenocysteamine, γ-aminopropylselenol, α-dimethyl-β-aminoethy selenol, α-methyl-β-aminoethylselenol, N-ethylaminoethylselenol and N,N'-dimethylaminoethylselenol were synthesized. Acid dissociation constants were determined by potentiometric titration and the values were compared with those of sulfur analogues. Stability constants of their metal complexes were calculated.

In the previous reports,[2,3] the acid dissociation phenomena and the reactions with various metal ions of selenocysteamine (2-aminoethaneselenol) were discussed and the results were compared with those of cysteamine which has been well known as one of the potential radiation protectors. Some marked differences were found between these two compounds. In this study, several related compounds of selenocysteamine were synthesized by the procedures shown in Chart 1 to investigate the influence of the substituents and at the same time to find general property of selenium as a ligand atom in these complexing agents. The acid dissociation constants of these compounds were calculated. The stability constants of their metal complexes were also calculated. The results were compared with those of corresponding sulfur and oxygen compounds in some cases.

Experimental

Syntheses of the Related Compounds of Selenocysteamine (Chart 1)——γ-Aminopropylselenol Hydrochloride [I]: Diaminopropyl diselenide dihydrochloride (3.5 g, 0.01 mole) synthesized according to the method of Coblentz,[4] was dissolved in 20 ml of 10% aqueous NaOH. The orange–yellow solution was then extracted with three 30 ml portions of $CHCl_3$ and the combined extracts were dried with anhydrous $MgSO_4$. The solvent was removed under reduced pressure to give a viscous, orange–yellow oil as a residue. To a solution of 0.68 g (0.0025 mole) of oily diaminopropyl diselenide in 5 ml of H_2O, 0.06 g (0.0016 mole) of sodium borohydride in 5 ml of H_2O was added with the slow introduction of nitrogen. The orange–yellow color disappeared on standing for about 15 min. After standing the reaction mixture for 30 min, H_2O was removed under reduced pressure at 50° to give white residue. The residue was acidified with 30 ml of ice–cooled saturated ethanolic HCl solution. The mixture was agitated for 30 min under nitrogen atmosphere and filtered. The filtrate was evaporated to dryness under reduced pressure to obtain 0.25 g of I. The melting point of this compound could not be measured because of its hygroscopic character. The purity of the compound was confirmed to be 99% by the iodometric determination of the selenohydryl group.

α-Dimethyl-β-aminoethylselenol Hydrochloride [II]: Dimethylethylenimine was synthesized from 2-amino-2-methyl-1-propanol according to Cairns's method.[5] Potassium selenosulfite solution was prepared from 15.8 g of fresh potassium sulfite (0.1 mole), 8.7 g of powdered selenium (0.12 mole), and 10 mg of sodium lauryl sulfate. To this solution, 7.1 g of dimethylethylenimine (0.1 mole) was added and 200 ml of 1 N sulfuric acid was added dropwise in a similar manner as in the synthesis of 2-aminoethaneselenosulfate[6] and

1) Location: *Yoshida Shimoadachi-cho, Sakyo-ku, Kyoto.*
2) H. Tanaka, H. Sakurai, and A. Yokoyama, *Chem. Pharm. Bull.* (Tokyo), **18**, 1015 (1970).
3) A. Yokoyama, H. Sakurai, and H. Tanaka, *Chem. Pharm. Bull.* (Tokyo), **18**, 1021 (1970).
4) V. Coblentz, *Ber.*, **24**, 4109 (1891).
5) T.L. Cairns, *J. Am. Chem. Soc.*, **63**, 871 (1941).
6) D.L. Klayman, *J. Org. Chem.*, **30**, 2454 (1965).

α-dimethyl-β-aminoethylselenosulfuric acid was obtained as yellowish white crystals, (yield 75%). This selenosulfuric acid (11.6 g, 0.05 mole) was dissolved in 50 ml of 10% aqueous NaOH and the reaction mixture was allowed to stand at room temperature for 2 hr. The yellow solution was extracted with three 30 ml portions of $CHCl_3$ and the combined extracts were dried with anhydrous $MgSO_4$. The solvent was removed under reduced pressure to give a moderately viscous, yellow–orange oil as a residue, (yield 50%). This oil gave only one spot on thin–layer chromatography and was used for the next step without further purification. To a solution of 0.38 g (0.0025 mole) of oily di-α,α-dimethyl-β-aminoethyl diselenide in 5 ml of H_2O, 0.052 g (0.0014 mole) of sodium borohydride in 5 ml of H_2O was added with the slow introduction of nitrogen. The yellow color of diselenide disappeared on standing for about 15 min. After standing the reaction mixture for 30 min, H_2O was removed under reduced pressure at 50° to give a white residue. The residue was acidified with 30 ml of ice–cooled saturated ethanolic HCl solution. The mixture was agitated for 30 min under nitrogen atmosphere and filtered. The filtrate was evaporated to dryness under reduced pressure to obtain II as white and slightly hygroscopic crystals. Yield was about 30% from dimethylethylenimine. mp 140° (decomp.). The purity of the compound was confirmed to be 96.8% by the iodometric determination of the selenohydryl group. *Anal.* Calcd. for $C_4H_{12}NSeCl$: C, 25.48; H, 6.43. Found: C, 25.99; H, 6.90.

α-Methyl-β-aminoethylselenol Hydrochloride [III]: 1-Amino-2-propanol (0.5 mole) dissolved in anhydrous $CHCl_3$ (100 mole) was added slowly to a stirred mixture of $SOCl_2$ (1 mole) and $CHCl_3$ (500 mole) at −5°. After standing for 1 hr, most of the $CHCl_3$ was removed on a water bath, and the white residue was dissolved in EtOH, filtered, and the filtrate was evaporated under reduced pressure. Recrystallization of the product from EtOH–ether gave stout needles of α-methyl-β-aminoethylchloride hydrochloride with about 90% yield. To the solution of 0.1 mole of potassium selenosulfite, α-methyl-β-aminoethylchloride hydrochloride in 200 ml of EtOH was added dropwise with vigorous stirring. A little amount of elemental selenium deposited by each addition of potassium selenosulfite and it dissolved again. The rate of the addition of potassium selenosulfite was controled to dissolve elemental selenium before next addition of potassium selenosulfite. The addifon was completed within 60 min. The precipitated salt was filtered off and the filtrate was chilled overnight. The crystalline deposit was collected and extracted with three 100 ml portions of 80% EtOH. The extracts were then combined and evaporated to dryness under reduced pressure to obtain crude selenosulfate which contains inorganic salts. The yield was 60%. The crude selenosulfate could be used for further reaction without purification. Selenosulfate (0.05 mole) was dissolved in 50 ml of 10% aqueous NaOH and the resulting solution was allowed to stand at room temperature for 2 hr to obtain the solution of diselenide. The yellow solution was then extracted with three 30 ml portions of $CHCl_3$ and combined extracts were dried with anhydrous $MgSO_4$. The extracts gave only one spot on thin–layer chro-

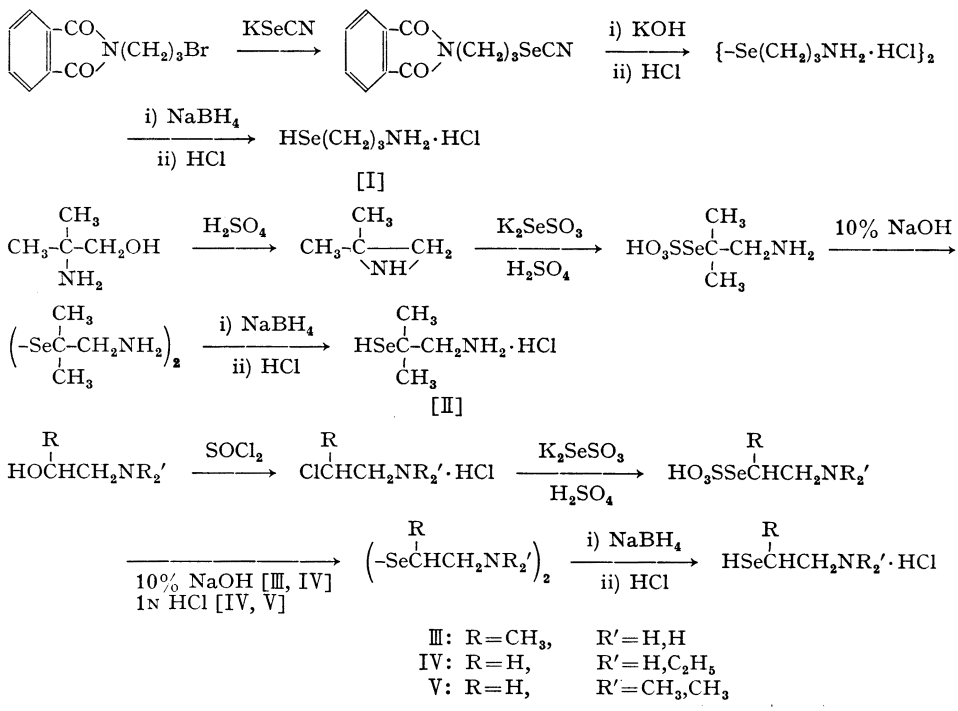

Chart 1

matography. The solvent was removed under reduced pressure to obtain viscous orange oil. Diselenide was reduced by sodium borohydride in a same manner as II. The yield was 50%. III was slightly yellowish white hygroscopic crystal. mp 130—135° (decomp.). The purity of this compound was confirmed to be 98% by the iodometric titration of the selenohydryl group. *Anal.* Calcd. for $C_3H_{10}NSeCl$: C, 20.64; H, 5.78, Found: C, 21.53; H, 6.02.

N-Ethylaminoethylselenol Hydrochloride [IV]: IV was prepared from 25 g (0.28 mole) of N-ethylaminoethanol by the similar procedure to that of III. However, in this case, the conversion of selenosulfate into diselenide was also achieved by the use of 50 ml of 1 N HCl (yield 50%). The purity of selenohydryl group of IV was confirmed to be 93.3% by the iodometric titration of the selenohydryl group. *Anal.* Calcd. for $C_4H_{12}NSeCl$: C, 25.48; H, 6.43. Found: C, 25.94; H, 6.76.

N,N'-Dimethylaminoethylselenol Hydrochloride [V]: V was prepared from 25 g (0.3 mole) of N,N'-dimethylaminoethanol by almost similar procedure to that of III. However, in this case, the conversion of selenosulfate to diselenide was achieved only by the use of 50 ml of 1 N HCl instead of 50 ml of 10% NaOH (yield 50%). The purity of this compound was confirmed to be 96.0% by the iodometric titration of the selenohydryl group. *Anal.* Calcd, for $C_4H_{12}NSeCl$: C, 25.48; H, 6.43. Found: C, 25.37; H, 6.51.

α-Dimethyl-β-aminoethylthiol Hydrochloride: From 10 g of dimethylethylenimine[5] and H_2S in absolute EtOH, 16.2 g of α-dimethyl-β-aminoethylthiol hydrochloride was obtained according to Mills and Bogert's method,[7] (yield 85%), mp 202°—203°. The purity of the compound was confirmed to be 98% by the iodometric determination of the sulfhydryl group.

pH Titration and Acid Dissociation Constants——Titrations and the calculations of the acid dissociation constants were carried out in a same manner as in the previous report.[2]

Stability Constants of Metal Complexes——Stability constants of nickel (II), cobalt (II), zinc (II), lead (II), cadmium (II) and copper (II) complexes were determined in a manner as in the previous report.[3]

Measurement of NMR Spectra——The NMR spectra were recorded at 60 Mc in D_2O solution using tetramethylsilane as internal standard.

Result and Discussion

The decomposition reaction of selenosulfate to diselenide was achieved by the use of 10% sodium hydroxide in the case of monoalkylated amino compounds[IV]as in the case of the preparation of selenocystamine, and hydrochloric acid could also be used for this reaction.

Table I. Signals of Methylene Protons in Nuclear Magnetic Resonace Spectra of Selenocysteamine Hydrochloride and Related Compounds in D_2O Solution

Compounds	τ		
	[a] X-CH_2-C	C-CH_2-N	
$HSeCH_2CH_2NH_2 \cdot HCl$	6.68	7.13	
$HSeCH(CH_3)CH_2NH_2 \cdot HCl$	6.41	7.13	8.62 (CH_3)
$HSeC(CH_3)_2CH_2NH_2 \cdot HCl$		7.12	8.58 (CH_3)
$HSCH_2CH_2NH_2 \cdot HCl$	6.75	7.12	
$HSC(CH_3)_2CH_2NH_2 \cdot HCl$		7.18	8.61 (CH_3)
$HOCH_2CH_2NH_2 \cdot HCl$	6.17	6.85	
$HSeCH_2CH_2CH_2NH_2 \cdot HCl$	6.85	7.31	7.96 (C-CH_2-C)
$CH_3SeCH_2CH_2NH_2 \cdot HCl$	6.70	7.18	7.94 (CH_3)

a) X=Se, S or O

7) E.J. Mills and M.T. Bogert, *J. Am. Chem. Soc.*, **62**, 1173 (1940).

On the contrary, in the case of dialkylated amino compound[V], sodium hydroxide caused the complete decomposition of selenosulfate to elemental selenium. In this case the use of hydrochloric acid in place of sodium hydroxide was effective to obtain diselenide. All compounds synthesized here are hygroscopic yellowish white crystals, and very unstable to air oxidation similarly to selenocysteamine. A little decomposition to elemental selenium seems to occur gradually even when they are stored in vacuum desicator on phosphorous pentoxide.

The chemical shifts in τ-values in D_2O solution of these compounds and related sulfur and oxygen analogues are shown in Table I.

In sulfur and selenium compounds, chemical shifts of the protons bonded with the carbon atom which is adjacent to the nitrogen atom are scarcely different from each other but those of the protons bonded with the carbon atom which is adjacent to the sulfur and selenium atom differ from each other significantly. The signals of the protons adjacent to the amino group were always observed at higher magnetic field in both sulfur and selenium compounds than those of protons adjacent to sulfur or selenium atom. When a solution of about equimolar of NaOD was added into the selenocysteamine hydrochloride in D_2O solution, the chemical shift corresponding to 6.68 τ of selenocysteamine hydrochloride disappeared as seen in Fig. 1 (c). Selenocysteamine dihydrochloride, whereas gave a complicated spectrum (b). The disappearance of the signal of 6.68 τ may be explained by the influence of the negative charge on the selenium atom to the adjacent carbon atom in the zwitter-ionic form, $Na^+Se^- -CH_2-CH_2- {}^+NH_3Cl^-$. This observation in nuclear magnetic resonance spectra corresponds to the conclusion in the previous reports[2] that selenocysteamine exist almost completely as the zwitter–ionic form in neutral pH region.

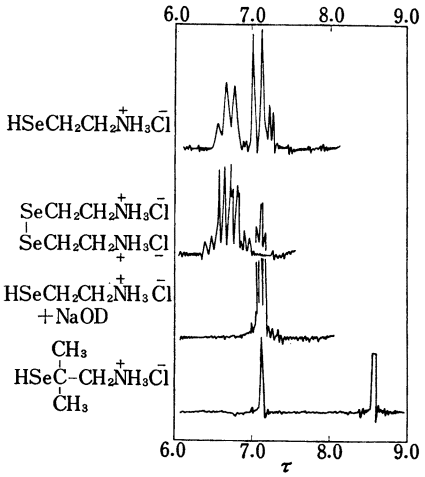

Fig. 1. Nuclear Magnetic Resonance Spectra

The acid dissociation constants of the related compounds of selenocysteamine are shown in Table II.

It was observed that the introduction of the methyl group to the β-position of selenocysteamine or cysteamine increases slightly the acid dissociation constant of selenohydryl or sulfhydryl group, whereas the introduction of alkyl group to the amino group causes the decrease in those values.

In compound [I], which involves three methylene groups between selenium and nitrogen atom, the basicity of the selenohydryl group is higher than that in selenocysteamine. It was found that in this series of compounds, pK_a values of selenohydryl and sulfhydryl groups are about 5—6 and about 8 respectively. The methylations of sulfhydryl, selenohydryl and hydroxyl groups in these compounds do not show remarkable change in the dissociation constants of ammonium groups. The results of the titrations showed that the reactions of these selenium containing compounds with various metal ions are essentially identical with those of selenocysteamine. As an example, the titration curves of α-methyl-β-aminoethylselenol with various concentrations of nickel(II) ion are shown in Fig. 2, and those with various metal ions are shown in Fig. 3. In the previous paper,[3] it was reported that selenocysteamine forms cationic metal complexes of the type $^+NH_3-CH_2-CH_2-Se-M-Se-CH_2-CH_2-{}^+NH_3$ with various metal ions. In the case of the selenohydryl compounds presented here, the formation of the complexes of the similar type can be reasonably assumed from the titration

TABLE II. Dissociation Constants of Selenocysteamine and Related Compounds

Compound	pK_a	Compound	pK_a	Compound	pK_a
$HOCH_2CH_2NH_2$[a]	9.60	$HSCH_2CH_2NH_2$[a]	8.27 10.53	$HSeCH_2CH_2NH_2$	5.01 10.99
				CH_3 $HSeCHCH_2NH_2$	5.19 10.85
				CH_3 $HSeC-CH_2-NH_2$ CH_3	5.21 10.95
		CH_3 $HSC-CH_2NH_2$ CH_3	8.48 11.00		
				$HSeCH_2CH_2CH_2NH_2$	6.27 11.30
				$HSeCH_2CH_2NHC_2H_5$	5.04 10.91
		$HSCH_2CH_2N(CH_3)_2$	7.70 10.71	$HSeCH_2CH_2N(CH_3)_2$	4.74 10.81
$CH_3OCH_2CH_2NH_2$[a]	9.45	$CH_3SCH_2CH_2NH_2$[a]	9.45	$CH_3SeCH_2CH_2NH_2$	9.53
		$(-SCH_2CH_2NH_2)_2$[a]	8.82 9.58	$(-SeCH_2CH_2NH_2)_2$	8.63 9.96
$HOCH_2CHCOOH$[a] NH_2	2.26 9.12	$HSCH_2CHCOOH$[a] NH_2	1.96 8.36 10.28	$HSeCH_2CHCOOH$[b] NH_2	2.01 5.24 9.96

(22°, μ=0.1)

a) L.G. Sillen and A.E. Martel, "Stability Constants of Metal Ion Complexes," Special Publication on No. 17, The Chemical Society, Burlington House, W. 1, London, 1964

b) R.E. Huber and R.S. Criddle, *Arch. Biochem. Biophys.*, **122**, 164 (1967)

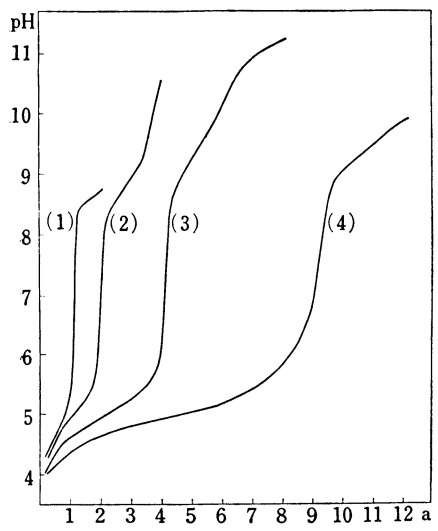

Fig. 2. Titration Curves of α-Methyl-β-aminoethylselenol Hydrochloride with Ni (II)

ligand: Ni (II), (1) 1:1 (2) 2:1 (3) 4:1 (4) 10:1
a: moles of KOH added per mole of Ni (II)
concentration of ligand: 5×10^{-3} M
μ=0.1 ($NaClO_4$), 22°

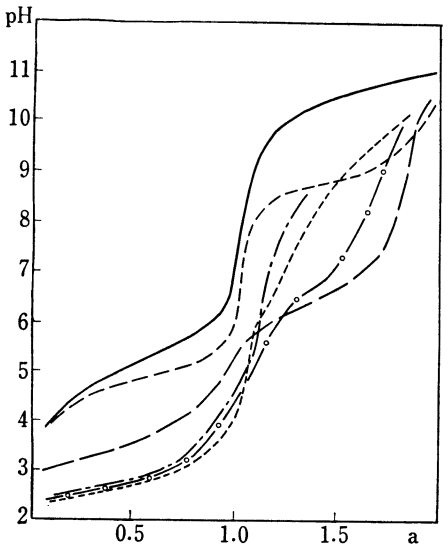

Fig. 3. Titration Curves of α-Methyl-β-aminoethylselenol Hydrochloride with Various Metal Ions

————: ligand ----------: Ni
- - - - - - -: Cu —o—o—: Cd
— — — —: Zn ..... : Pb

concentration of ligand: 5×10^{-3}M
concentration of metal ion: 2.5×10^{-3}M
μ=0.1 ($NaClO_4$). 22°
a: moles of KOH added per mole of ligand

TABLE III. Stability Constants of Metal Complexes of Selenocysteamine and Related Compounds

Ligand		Ni (II)	Co (II)	Zn (II)	Pb (II)	Cd (II)	Cu (II)
$HSeCH_2CH_2NH_2$	log K_1	2.93	2.76	3.85	>5.2	>5.4	
	log K_2	2.77	1.65	1.68	3.50	4.86	
	log β_2	5.70	4.41	5.53	> 8.7	>10.3	
CH_3	log K_1	3.48		4.45	>5.1	>5.7	
$HSeCHCH_2NH_2$	log K_2	3.22		4.05	5.25	5.46	
	log β_2	6.70		8.50	>10.4	>11.2	
CH_3	log K_1	3.10		4.32	5.35		
$HSeC-CH_2NH_2$	log K_2	3.05		3.97	4.85		
CH_3	log β_2	6.15		8.29	>10.2		
$HSeCH_2CH_2CH_2NH_2$	log K_1	3.53	3.20	5.12	>6.4	>6.6	
	log K_2	1.71	0.58	3.17	1.96	6.54	
	log β_2	5.24	3.73	8.29	8.4	>13.1	
$HSeCH_2CH_2NHC_2H_5$	log K_1			4.35			
	log K_2			4.10			
	log β_2			8.45			
$HSeCH_2CH_2N(CH_3)_2$	log K_1			>5.2		>6.0	
	log K_2			5.00		5.78	
	log β_2			>10.2		>11.8	
$CH_3SeCH_2CH_2NH_2$	log K_1	3.48		4.27	5.15	3.56	5.98
	log K_2	3.35		4.47	4.07	3.27	5.60
	log β_2	6.83		8.74	9.22	6.83	11.58

(22°, μ=0.1)

curves. Base on this assumption, the stability constants of these metal complexes shown in Table III were calculated in a same manner as those of selenocysteamine.[3]

In the reaction with cobalt(II), stability constants of the complex could not be calculated except for a few cases, because the titration curves of the ligand and those with cobalt(II) ion almost coincide, although clear green colorations were always observed in these reactions.

General relationship between the stability constants and the effects of the substituent could not be derived from these data, but in nickel(II) complexes the introduction of the methyl group at β-position in the ligand seems to increase the stability of the complex and the increase in the stability of the complex seems to correlate with the increase of pK_a value of the ligand.