Notes

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Michiya Kimura, Masahiko Tohma, and Itsuo Yoshizawa : Constituents of Convallaria. Ⅵ.*1 Isolation of Convallatoxol.

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A steroidal glycoside, $C_{29}H_{44}O_{10}$, m.p. 168～169°, was isolated from the flowers of Japanese lily of the valley, *Convallaria keisukei* Miq. and was identified as convallatoxol (I). A nitrogen compound, $C_{16}H_{24}O_8N_4$, m.p. 189.5～190.5°(Ⅱ), was also isolated.

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Rabauld and Kraus[1] reported that the aldehyde group at C_{10} in strophanthidin was reduced to the alcohol giving strophanthidol through the Meerwein-Pondorf reaction employing aluminum amalgam or aluminum isopropylate and that the reduction of strophanthoside heptaacetate occurred also in a similar way. Hunger and Reichstein[2] found that strophanthidin-rhamnoside, convallatoxin, was not reduced by these reagents and that sodium borohydride instead could give the corresponding strophanthidol-glycoside which was then named as convallatoxol (I). Later, Tschesche and Seehofer[3] succeeded in isolating I from the leaves of lily of the valley, *Convallaria majalis* L.

In the previous papers[4~8] of this series, the authors reported that convallatoxin, desglucocheirotoxin, chelidonic acid and new steroidal saponins, convallasaponin-A, -B, and -C, were isolated from the blossoms of Japanese lily of the valley, *Convallaria keisukei* Miq. As described in the experimental part, I, $C_{29}H_{44}O_{10}$, m.p. 168～169°, was isolated from the aqueous layer[7] on the course of extraction of these new saponins.

Table I. Alumina Chromatography of Aqueous Layer

Fraction No.	Solvent	Vol. (L.)	Weight (g.)
I	MeOH-$CHCl_3$ (10:90)	10	6.78 (oil)
Ⅱ	〃 (15:85)	15	21.28
Ⅲ	〃 (20:80)	12	3.55
Ⅳ	〃 (30:70)	6	2.38
Ⅴ	〃 (40:60)	11	2.70
Ⅵ	〃 (60:40)	16	5.08
Ⅶ	MeOH	29	11.20
Ⅷ	H_2O	30	24.24

A colorless nitrogen compound, $C_{16}H_{24}O_8N_4$, m.p. 189.5～190.5°, (Ⅱ), was also isolated from the aforementioned aqueous layer and was purified chromatographically on Celite 545-Florisil (1:2) as shown in Table Ⅱ. Details on the chemical structure of this compound will be presented in the forthcoming paper.

*1 Part Ⅴ : This Bulletin, **14**, 55 (1966).

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2) A. Hunger, T. Reichstein : Chem. Ber., **85**, 635 (1952).
3) R. Tschesche, F. Seehofer : *Ibid.*, **87**, 1108 (1954).
4) M. Ishidate, M. Kimura, M. Kawada, S. Tamikado : Yakugaku Zasshi, **77**, 679 (1957).
5) M. Kimura, M. Tohma, T. Kikuchi : *Ibid.*, **82**, 1320 (1962).
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Experimental

Spectrophotometry——The ultra violet (UV) spectra were measured in EtOH using a self-recording ultraviolet spectrophotometer (Hitachi Model EPS 2U) and the IR spectra in KBr disks using an infrared spectrophotometer (Nihon Bunko Model Koken DS 101).

Chromatography on Alumina (Table I)——The aq. layer shown on the flow-sheet in the previous paper[7] was evaporated to give yellow powder (90.8 g.) under reduced pressure. Ninety grams of the powder dissolved in 300 ml. of 10% MeOH–$CHCl_3$ and was submitted to chromatography on alumina (1.8 kg.) giving the results as shown in Table I. Fraction II gave yellow powder (21.28 g.).

Thin-layer chromatography of fraction II : Five spots showing positive Kedde reaction[9] were found on the thin-layer chromatography using Wakogel B-5 and 30% MeOH–$CHCl_3$ as adsorbent and solvent respectively. The Rf values and color changes of these spots by 5% H_2SO_4 at 120° were as follows : Rf 0.47 (yellow → green; convallatoxin), 0.41 (red → violet; convallatoxol), 0.38 (green; unidentified), 0.27 (grey → violet; unidentified), 0.23 (green; unidentified). Four other spots showing negative Kedde reaction were also detected by 5% H_2SO_4 at 120° showing Rf values as follows : 0.95, 0.78, 0.65, 0.45 (nitrogen compound).

Chromatography on Celite-Florisil (Table II)——The powder (13.4 g.) from fraction II (Table I) was dissolved in 1% MeOH–$CHCl_3$ (100 ml.) and was submitted again to chromatography on Celite 545–Florisil (1:2; 390 g.) as shown in Table II. Fraction 3 gave colorless powder (392 mg.) which was recrystallized from MeOH–H_2O to afford needles, m.p. 234~238°. Liebermann-Burchard reaction : red-green; positive Kedde and Legal reactions. IR spectrum, mixed melting point and Rf value (0.47) on thin-layer chromatography described above were found to be identical with those of authentic convallatoxin.

Table II. Celite 545–Florisil Chromatography of Fraction II

Fraction No.	Solvent	Vol. (L.)	Weight (mg.)	Rf on T.L.C.
1	$CHCl_3$	10	100	0.95, 0.78, 0.65
2	MeOH–$CHCl_3$ (1:99)	13	90	0.78, 0.65
3	〃 (2:98)	14	392	0.47
4	〃 (2:98)	12	803	0.47, 0.45
5	〃 (3:97)	12	407	0.47, 0.45
6	〃 (4:96)	12	630	0.45
7	〃 (5:95)	14	1720	0.45, 0.41
8	〃 (6:94)	16	2350	0.45, 0.41
9	〃 (6:94)	4	560	0.41
10	〃 (6:94)	4	470	0.41, 0.38
11	〃 (10:90)	12	1440	0.41, 0.38, 0.27
12	〃 (20:80)	8	176	0.38, 0.27, 0.23
13	〃 (40:60)	2	123	0.27, 0.23

Isolation of Convallatoxol (I)——Fraction 9 gave colorless powder (560 mg.) which was recrystallized from MeOH–H_2O to afford colorless needles, m.p. 168~169°, undepressed on admixture with authentic convallatoxol prepared from convallatoxin under the usual method.[2] Liebermann-Burchard reaction : pink → green; positive Kedde and Legal reactions. Rf value (0.41) on thin-layer chromatography described above and IR spectrum were also found to be the same as those of authentic specimen. UV λ_{max}^{EtOH} mμ (log ε) : 217 (4.31). *Anal.* Calcd. for $C_{29}H_{44}O_{10}$: C, 63.02; H, 8.03. Found : C, 63.08; H, 7.63.

Hydrolysis of I——i) by Mannich-Siewert Method[10] : I (70 mg.) was dissolved in acetone (10 ml.) containing conc. HCl (0.1 ml.) and was allowed to stand for a week at room temperature. Water (10 ml.) was then added to the solution and acetone was evaporated after heating for 30 min. on a water-bath. The $CHCl_3$-extracts of the residue (30 mg.) were recrystallized from MeOH–$CHCl_3$ to give colorless needles, m.p. 135~138°, undepressed on admixture with the authentic strophanthidol.[1,2] Thin-layer chromatography using AcOEt–*iso*PrOH–H_2O (65:25:15) as a solvent gave also the same Rf value (0.68) with that of authentic specimen. *Anal.* Calcd. for $C_{23}H_{34}O_6$: C, 67.95; H, 8.43. Found : C, 67.99; H, 8.39.

7) M. Kimura, M. Tohma, I. Yoshizawa : This Bulletin, **14**, 50 (1966).
8) *Idem* : *Ibid.*, **14**, 55 (1966).
9) E. Schenker, A. Hunger, T. Reichstein : Helv. Chim. Acta, **37**, 680 (1954).
10) C. Mannich, G. Siewert : Ber., **75**, 737 (1942).

ii) by the Kiliani Mixture[11]: I (10 mg.) was dissolved in the Kiliani mixture (AcOH–H_2O–conc. H_2SO_4 =35:55:10; 1 ml.) and was heated on a water-bath for 1 hr. Water (1 ml.) was then added and the filtrate was neutralized with Amberlite IR-4B. Evaporation of the aq. solution gave yellow syrup which was revealed to contain L-rhamnose (Rf 0.58) on the paper (Toyo Roshi No. 51) chromatography using AcOEt–Pyridine–H_2O (2:1:2) and aniline hydrogen phthalate as a solvent and developer respectively.

Isolation of Nitrogen Compound (II)——Fraction 6 shown in Table II afforded colorless needles, m.p. 189.5~190.5°, recrystallized from MeOH–$CHCl_3$. Molecular weight determined by the Rast method was shown as revealed 400±20. *Anal.* Calcd. for $C_{16}H_{24}O_8N_4$ (M. W. =400.38): C, 47.99; H, 6.04; N, 13.99. Found: C, 47.82; H, 5.93; N, 14.07. UV λ_{max}^{EtOH} mμ (log ε): 209 (4.17), 268 (4.17). IR ν_{max}^{KBr} cm^{-1}: 3310, 3020, 2830, 1710, 1660, 1479, 1435, 1315, 1289, 1273, 1252, 1222, 1120, 1098, 1065, 1010, 906, 869, 851.

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Takeo Ueda and Kumi Ishizaki: Syntheses of 3-Aminopropylguanidine Derivatives.

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The fourteen compounds of 3-(substituted amino)propylguanidine were synthesized by reacting S-methylisothiourea sulfate with 3-(substituted amino)propylamine: 3-(substituted amino) propylamine having an aliphatic substituent group at 3-position were synthesized by reacting the aliphatic amine with acrylonitrile and by reducting with lithium aluminum hydride, while 3-(substituted amino)propylamine having an aromatic substituent group at 3-position, synthesized by the Ing-Manske modification of the Gabriel synthesis of primary amines.

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Recently, we have found that guanidine salts[1] and their related compounds[2] exerted inhibitory effect on several pathogenic viruses in tissue culture. In those studies, many compounds having guanidino moiety were synthesized and examined as to their antiviral activity. At the same time, attempts were also made to find pharmacologically active agents among those compounds. Especially, it was noted that some compounds of aminoalkylguanidine were found to have hypertensive and analgesic activities.

This finding prompted the authors to synthesize compounds of substituted amino-alkylguanidine.

This report is concerned with the syntheses of 3-(substituted amino)propylguanidine.

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2) M. Furukawa, S. Toyoshima, T. Ueda: This Bulletin, **11**, 1247 (1963).