corresponding acetic acid (VI) from which the key intermediate (XIII) was prepared by several different routes. The structure of the final product (XV) was proved by converting the penultimate compound (XIV) into a well defined crystalline *rac*-c-bisnorrubremetinium bromide (XVI).

(Received July 7, 1958)

UDC 547.94.02 : 582.675.1

10. **Toshihiko Okamoto**: The Aconite Alkaloids. XIX.[1] The Structure of Kobusine. (1).

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Kobusine was first isolated from *Aconitum sachalinense* Fr. Schmidt by Suginome and F. Shimanouchi[2] and $C_{20}H_{27}O_2N$ was given as its molecular formula by the above workers. One oxygen was proved to be present as a hydroxyl group and the other was considered to be an ether oxygen.[3] Kobusine absorbed one mole of hydrogen on catalytic reduction and these workers concluded that this double bond may be attached to nitrogen.[2,3]

Previously, Ochiai and others reported the isolation of an alkaloid from the aconite roots, gathered in Shimokita Peninsula, Aomori Prefecture, which showed quite similar physical properties with kobusine.[4] Later, this sample was identified as kobusine by admixture.[5]

This paper describes some reduction and oxidation reactions of kobusine. Kobusine (I) crystallized from acetone as prisms, m.p. 267~267.5°, $[\alpha]_D^{12}$+104.4° (MeOH), and gave a picrate, m.p. 282~284°(decomp.), perchlorate, m.p. 185~187°(decomp.), and also a methiodide, m.p. 286~287°(decomp.).[4,6]

Kobusine (I) showed the bands at 1648 and 888 cm^{-1} in its infrared spectrum and consumed one mole of hydrogen on hydrogenation over platinium oxide in acetic acid, so the existence of a terminal methylene group could be considered. As was previously reported, shimoburo base-I[7] and hypognavine[1] have an allyl alcohol group in their structure and gave rearranged ketones by hydrogenation over palladium-carbon. In the relation to these alkaloids, kobusine was hydrogenated by the use of palladized carbon as the catalyst in methanol, and three products were isolated by alumina chromatography. The first fraction showed m.p. 230~232° (needles from acetone), $C_{20}H_{27}O_2N$ (II), the second showed m.p. 240~241° (needles from chloroform), $C_{20}H_{27}O_2N$ (III), and the third fraction melted at 229~231° (needles from acetone), $C_{20}H_{29}O_2N$ (IV). In the infrared spectra, the first two products (II and III) showed a strong absorption band at 1705 cm^{-1} and were considered to be ketones of a six-membered ring, which are the rearranged products in the allyl alcohol part of kobusine. The third product of m.p. 229~231° (IV) showed no absorption band of ketone in the infrared spectrum and this should be the dihydro derivative. The

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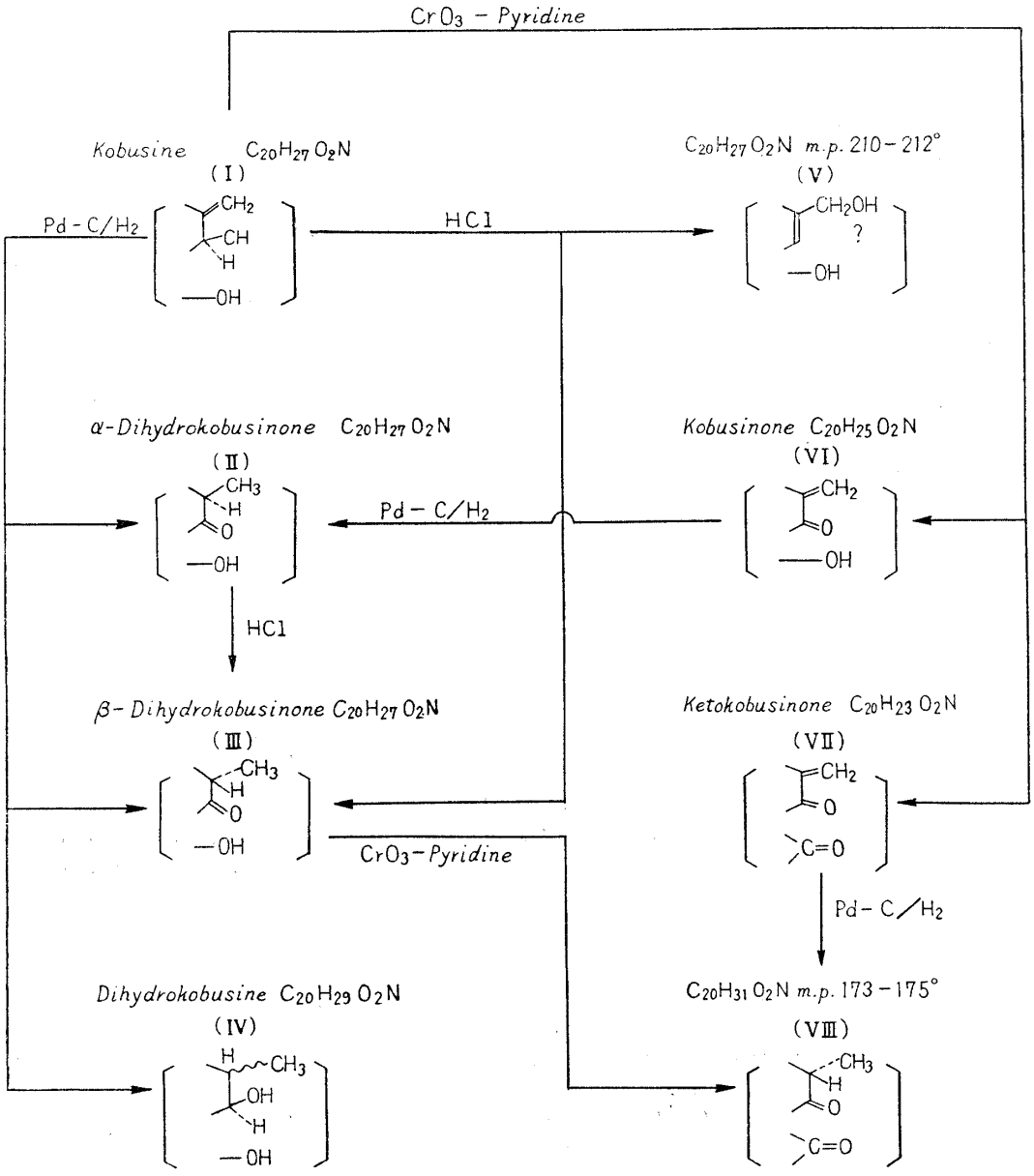
1) Part XVIII: S. Sakai: This Bulletin, **5**, 1(1957).
2) H. Suginome, F. Shimanouchi: Ann., **545**, 220(1940).
3) H. Suginome, F. Shimanouchi: J. Fac. Sci., Hokkaido Univ., IV (1942).
4) E. Ochiai, *et al.*: Yakugaku Zasshi, **75**, 639 (1955).
5) The author is grateful to Dr. H. Suginome and Dr. T. Amiya, Department of Chemistry, University of Hokkaido, for their kind help for the identification of the sample.
6) H. Suginome reported the following physical properties for kobusine in reference of Footnote(2): m.p. 268°, $[\alpha]_D^{22}$ +83.61° ($CHCl_3$); picrate, m.p. 273°(decomp.): perchlorate, m.p. 220°; methiodide, m.p. 286~287°(decomp.).
7) T. Sugasawa: This Bulletin, **4**, 6(1956).

ketone of m.p. 230~232° (II) was designated as α-dihydrokobusinone and the second one of m.p. 240~241° (III), as β-dihydrokobusinone. α-Dihydrokobusinone (II) isomerized to the β-isomer (III) on heating with 5% hydrochloric acid on a steam bath. From these facts, the presence of the α-methylenehexanol portion in kobusine was confirmed.

β-Dihydrokobusinone (III) was also obtained directly from kobusine by heating it with 5% hydrochloric acid,[8] but in this case another product of m.p. 210~212°, $C_{20}H_{27}O_2N$ (V), was isolated as the second fraction on alumina chromatography. The compound (V) showed the absence of the band for carbonyl or terminal methylene group in its infrared spectrum, and therefore it might have the structure of an isomeric allyl alcohol of the type of C=C—CH_2OH, which is the anionotropic rearrangement product[9] of kobusine. The compound (V) did not undergo rearrangement to β-dihydrokobusinone (III) by further prolonged heating with hydrochloric acid.[10]



8) cf. C. Djerassi: J. Am. Chem. Soc., **77**, 4801(1955).
9) C. K. Ingold: "Structure and Mechanism in Organic Chemistry," **1953**, 586. Cornell University Press.
10) cf. A. S. Dreiding, J. Hartman: J. Am. Chem. Soc., **78**, 1216(1956).

Because their infrared spectra of α- and β-dihydrokobusinone have the bands of hydroxyl group, kobusine should have two hydroxyl groups. To confirm this point, the oxidation reaction of kobusine was examined. Kobusine was oxidized with chromium trioxide in pyridine at room temperature and two products were isolated by chromatography on alumina. The first eluate was recrystallized from acetone to needles ($C_{20}H_{23}O_2N$), m.p. 189~191° (at ca. 200° it resolidified and again melted at 280~282°). In its infrared spectra, the band at 1721 cm^{-1} corresponds to a six-membered cyclic ketone, the bands at 1708 and 1628 cm^{-1} are considered as those of α,β-unsaturated ketone in a six-membered ring, and the spectrum showed the absence of a band for hydroxyl group. This ketone will be termed ketokobusinone (VII). The second eluate was crystallized from acetone as needles, m.p. 273~275°, $C_{20}H_{25}O_2N$. Its infrared spectrum showed the bands for hydroxyl group at 3100 cm^{-1}, and for α,β-unsaturated ketone at 1692 and 1632 cm^{-1}. This ketone will be called kobusinone (VI).

Ketokobusinone (VII) was hydrogenated over palladium-carbon catalyst, purified by alumina chromatography, and only one saturated diketone (dihydroketokobusinone) (VIII) was obtained. This recrystallized from acetone as prisms, m.p. 173~175°, $C_{20}H_{25}O_2N_2$, and its infrared spectrum showed a band at 1707 cm^{-1} for a six-membered ring ketone.[11] The compound (VIII) was identified as the oxidation product of β-dihydrokobusinone (III) by admixture of the two products and also by comparing their infrared spectra.

Kobusinone (VI) was hydrogenated in the same way as for ketokobusinone (VIII), purified by chromatography on alumina, and gave one product of m.p. 230~232°. This product was identified as α-dihydrokobusinone (II) by admixture with the sample obtained from kobusine by the rearrangement reaction, and this identity was also confirmed by comparing the infrared spectra of the two samples. It is rather striking that ketokobusinone gave β-type diketone and kobusinone gave α-type ketone on hydrogenation.

Kobusine was acetylated with acetic anhydride in pyridine to a diacetyl compound of m.p. 139~141°,[12] $C_{24}H_{31}O_4N$, and also it gave a dibenzoyl compound by reaction with benzoyl chloride in pyridine. Although the free benzoyl compound did not crystallize, it gave a hydrochloride, m.p. 290~292° (decomp.), $C_{34}H_{35}O_4N \cdot HCl \cdot H_2O$, and a picrate, m.p. 225~227° (decomp.), $C_{34}H_{35}O_4N \cdot C_6H_3O_7N_3$.

From the above-described rearrangement and oxidation reactions, obviously one of hydroxyl groups in kobusine is present as the allyl alcohol of the type $HO-\overset{|}{C}-\overset{|}{C}=CH_2$. Considering the oxidation and acetylation reactions, another hydroxyl group should be a secondary alcohol. From these data, the formula $C_{20}H_{29}N$ couldbe given for the basic skeleton of kobusine and it should have seven rings in the skeleton. This conclusion suggests that kobusine might have a structure quite similar to anhydroignavinol[13] and hypognavinol.[14]

The author expresses his gratitude to Dr. E. Ochiai for his constant guidance in the course of this study. The microanalyses were carried out by the members of the Central Analysis Room of this Faculty and by Messrs. T. Hattori and Y. Sato of the Institute of Applied Microbiology in this University, and the infrared spectra were determined by Mr. S. Sakai in this Faculty, to all of whom the author is also grateful.

Experimental

Determination of Double Bond in Kobusine—Determined by Tsuda-Sakamoto method.[15] Sample,

11) Possibly overlapping with two carbonyl bands.
12) H. Suginome (*loc. cit.*) also reported the acetate of m.p. 139~141° as the product from reaction with acetyl chloride.
13) E. Ochiai, *et al.*: Yakugaku Zasshi; **72**, 816(1952); This Bulletin, **1**, 60(1953); **2**, 388(1954).
14) E. Ochiai, *et al.*: This Bulletin, **1**, 152(1953); S. Sakai: Yakugaku Zasshi, **76**, 1054(1956); This Bulletin, **5**, 1(1957).
15) Yakugaku Zasshi, **57**, 1037(1937).

25.600 mg. in AcOH 4 cc., K=0.246, h=8.95, F=1.03. Sample, 24.102 mg. in AcOH 4 cc., K=0.246, h=7.13, F=1.02.

Catalytic Hydrogenation of Kobusine—Kobusine (150 mg.) was reduced in MeOH (20 cc.) over Pd-C (prepared from 50 mg. of carbon and 5 cc. of 1% $PdCl_2$ solution), and 7.60 cc. of hydrogen was consumed at 11° (calcd. for 1 mole, 11.3 cc.) in 2 hrs. After filtration of the catalyst, the solvent was distilled off and 142 mg. of resinous product was obtained. The crude products were separated by alumina chromatography, using $CH_2Cl_2 \rightarrow CH_2Cl_2$(MeOH, 0.5~1%) as the solvent.

Chromatography 1.

Fraction No.	m.p. (°C)	Yield (mg.)
1	229~233	10
2	235~237	74
3	224~228	34
4	220~224	18

The second fraction (74 mg.) was submitted again to chromatographic separation.

Chromatography 2.

Fraction No.	m.p. (°C)	Yield (mg.)
1	224~227	18
2	237~240	51
3	230~237	2

The first fractions in chromatography 1 and 2 were combined (28 mg.), again purified by chromatography, and 17 mg. of crystals were obtained as the less-adsorbed fraction, which recrystallized from Me_2CO as needles (II), m.p. 230~232°. *Anal.* Calad. for $C_{20}H_{27}O_2N$: C, 76.64, H, 8.68. Found: C, 76.81, H, 8.02. IR ν_{max}^{Nujol} 3115 cm^{-1} (OH), 1705 cm^{-1} (six-membered ring ketone).

The second fraction in chromatography 2 (51 mg.) was purified by chromatography and recrystallized from $CHCl_3$ to needles (III), m.p. 240~241°. $[\alpha]_D^{10}$ +36.6 (c=2.03, MeOH). Yield, 43 mg. *Anal.* Calcd. for $C_{20}H_{27}O_2N$: C, 76.64, H, 8.68. Found: C, 76.59, H, 8.26. IR ν_{max}^{Nujol} 3115 cm^{-1} (OH), 1705 cm^{-1} (six-membered ring ketone).

The third fraction in chromatography 1 was chromatographed again.

Chromatography 3.

Fraction No.	m.p. (°C)	Yield (mg.)
1	220~240	3
2	226~228	20
3	210~220	5

The fraction 4 in chromatography 1 was chromatographed again.

Chromatography 4.

Fraction No.	m.p. (°C)	Yield (mg.)
1	220~225	12
2	210~215	2

Fraction 2 in chromatography 3 and the fraction 1 in chromatography 4 were combined (32 mg.) and chromatographed again.

Chromatography 5.

Fraction No.	m.p. (°C)	Yield (mg.)
1	226~228	11
2	210~220	7

The first fraction (11 mg.) was recrystallized from acetone as needles (IV), m.p. 229~231; $[\alpha]_D^{11}$ +78.8° (c=1.05, MeOH). *Anal.* Calcd. for $C_{20}H_{29}O_2N$: C, 76.15, H, 9.27. Found: C, 76.68, H, 8.79. IR ν_{max}^{Nujol} 3465, 3125 cm^{-1} (OH), no absorption band of ketone.

Isomerisation of α-Dihydrokobusinone (II) to β-Dihydrokobusinone (III)—A solution of 8 mg. of (II) was heated with 5% HCl (5 cc.) on a steam bath for 5 hrs., basified with NH_4OH, and extracted with $CHCl_3$. After removal of the solvent, the residue (7 mg.) was recrystallized from $CHCl_3$ as needles, m.p. 240~241°. It showed no depression by admixture with β-dihydrokobusinone (III) and the identity was also confirmed by comparing their infrared spectra.

Rearrangement of Kobusine to β-Dihydrokobusinone (III) with Hydrochloric Acid—A solution of 60 mg. of kobusine was heated with 5% HCl (10 cc.) on a steam bath for 2 hrs. The reaction solution was made basic with NH_4OH, extracted with $CHCl_3$, and evaporated to dryness (55 mg.). The crude product was separated by alumina chromatography, using $CHCl_3$ (1% MeOH) as the solvent.

Chromatography 1.

Fraction No.	m.p. (°C)	Yield (mg.)
1	230~234	3
2	200~220	45
3	208~210	6

Fraction 2 (45 mg.) was chromatographed again.

Chromatography 2.

Fraction No.	m.p. (°C)	Yield (mg.)
1	238~241	11
2	170~200	8
3	208~210	17

Fractions 1 in chromatography 1 and 2 were combined and chromatographed again and 11 mg. of the compound of m.p. 240~241° was obtained, which recrystallized from $CHCl_3$ as needles. This showed no depression on admixture with β-dihydrokobusinone (III) and their infrared spectra were completely the same. Fractions 3 in chromatography 1 and 2 were combined (23 mg.) and separated again by alumina chromatography. 17 mg. of the compound (V), m.p. 210~212°, was obtained and recrystallized from Me_2CO as needles. *Anal.* Calcd. for $C_{20}H_{27}O_2N$: C, 76.64; H, 8.68. Found: C, 76.25; H, 7.79. IR ν_{max}^{Nujol} 3105 cm^{-1} (OH). No band for ketone group.

The compound (V) (10 mg.) was heated with 5% HCl (5 cc.) on a steam bath for 5 hrs. The reaction solution was made basic with NH_4OH, extracted with $CHCl_3$, and 8 mg. of starting material was recovered.

Oxidation of Kobusine—To a suspension of CrO_3 (100 mg.) in dry pyridine (2 cc.), 60 mg. of kobusine was added and the mixture was allowed to stand over night at room temperature. The reaction solution was diluted with water, a small amount of conc. NH_4OH was added, and extracted with $CHCl_3$ (1% MeOH). The extract was dried over anhyd. Na_2SO_4 and evaporated to dryness. The crude product was separated by chromatography on alumina, $CHCl_3 \rightarrow CHCl_3$ (1% MeOH) was used as the solvent. The first eluate (13 mg.) (ketokobusinone) (VII) was recrystallized from petr. ether to needles, m.p. 189~191° (it resolidified at ca. 200° and melted again at 280~282°). *Anal.* Calcd. for $C_{20}H_{23}O_2N$: C, 77.64, H, 7.49. Found: C, 77.13, H, 7.39. IR ν_{max}^{Nujol} cm^{-1}; 1718 (six-membered ring ketone), 1705, 1625 cm^{-1} (α,β-unsaturated ketone in a six-membered ring).

The second eluate (12 mg.) (kobusinone) (VI) was recrystallized from Me_2CO-petr. ether mixture to needles, m.p. 273~275°. *Anal.* Calcd. for $C_{20}H_{25}O_2N$: C, 77.13, H, 8.09. Found: C, 76.02, H, 7.56. IR ν_{max}^{Nujol} cm^{-1}: 3120 (OH), 1692, 1632 (α,β-unsaturated ketone in a six-membered ring).

Oxidation of β-Dihydrokobusinone (III)—To a suspension of CrO_3 (20 mg.) in pyridine (1 cc.), β-dihydrokobusinone (III) (10 mg.) was added and the mixture was allowed to stand over night at room temperature. The reaction solution was diluted with water and extracted with CH_2Cl_2. After removal of the solvent, the residue (8 mg.) was purified by alumina chromatography, using CH_2Cl_2 (0.5% MeOH) as a solvent. The product (7 mg.) was recrystallized from Me_2CO as prisms, m.p. 173~175°. *Anal.* Calcd. for $C_{20}H_{25}O_2N$: C, 77.13, H, 8.09. Found: C, 76.43, H, 7.54. IR ν_{max}^{Nujol} 1707 cm^{-1} (six-membered ring ketone).

Catalytic Reduction of Ketokobusinone—10 mg. of ketokobusinone (VII) was reduced in MeOH (10 cc.), usiug Pd-C (prepared from ca. 10 mg. of carbon and 2 cc. of 1% $PdCl_2$ solution) as the catalyst, (the volume of absorbed H_2 was not determined). After filtration of the catalyst, the solvent was evaporated and the residue (7 mg.) was purified by alumina chromatography, using CH_2Cl_2 (0.5% MeOH) as the solvent. Only one product was obtained. Recrystallization from acetone gave prisms, m.p. 173~175°, undepressed when mixed with the oxidation product of β-dihydrokobusinone (III). This identity was also confirmed by comparing the infrared spectra.

Catalytic Reduction of Kobusinone (VI)—10 mg. of kobusinone (VI) was hydrogenated in the same way as for ketokobusinone. The product was purified by chromatographic method and one product was obtained. Recrystallization from Me_2CO gave needles, m.p. 230~232°. Yield, 6 mg. The product showed no depression on admixture with α-dihydrokobusinone (II) and their infrared spectra were the same.

Diacetylkobusine—To a solution of kobusine (50 mg.) in pyridine (2 cc.), Ac_2O (0.2 cc) was added and the mixture was allowed to stand over night at room temperature. It was diluted with water, a small amount of conc. NH_4OH was added, and extracted with CH_2Cl_2. After removal of the solvent, the residue (57 mg.) was purified by alumina chromatography, using CH_2Cl_2-benzene mixture (50:50) as the solvent. Recrystallization from petr. ether gave needles, m.p. 139~141°. *Anal.* Calcd. for $C_{24}H_{31}O_4N$: C, 72.51, H, 7.86, CH_3CO, 21.66. Found: C, 72.33, H, 8.04, CH_3CO, 21.21.

Dibenzoylkobusine—To a solution of kobusine (94 mg.) in pyridine (5 cc.), BzCl (420 mg.) was added and the mixture was allowed to stand for 2 days at room temperature. The reaction solution was diluted with water, a small amount of conc. NH_4OH was added, and extracted with $CHCl_3$. The solvent was evaporated to dryness and the residue was purified by alumina chromatography, using $CHCl_3$ as the solvent. 53 mg. of amorphous benzoyl compound was obtained. This free base was treated with 5% HCl and solidified hydrochloride was recrystallized from dil. Me_2CO to needles, m.p. 290~292°(decomp.). *Anal.* Calcd.

for $C_{34}H_{35}O_4N \cdot HCl \cdot H_2O$: C, 70.90, H, 6.60. Found: C, 71.19, H, 6.57.
Acyl determination. Sample, 9.735 mg. N/50 NaOH (F=1.004). Calcd. for 3 H^+: 2.54 cc. Found: 2.81 cc.
Picrate: Yellow needles (from MeOH), m.p. 225~227° (decomp.). *Anal.* Calcd. for $C_{34}H_{35}O_4N \cdot C_6H_3O_7N_3$: C, 64,00, H, 5.07. Found: C, 64.17, H, 4.92.

Summary

Kobusine, $C_{20}H_{27}O_2N$, m.p. 267~267.5°, was hydrogenated over palladium-carbon in methanol, and α-dihydrokobusinone, $C_{20}H_{27}O_2N$, m.p. 230~232°, β-dihydrokobusinone, $C_{20}H_{27}O_2N$, m.p. 240~241°, and dihydrokobusine, $C_{20}H_{29}O_2N$, m.p. 229~231°, were obtained as the product. α- and β-Dihydrokobusinones were proved to be a ketone in six-membered ring. The β-isomer was also obtained from α-isomer or directly from kobusine on heating with 5% hydrochloric acid. Oxidation of kobusine with chromium trioxide in pyridine gave ketokobusinone, $C_{20}H_{23}O_2N$, m.p. 189~191°, and kobusinone, $C_{20}H_{25}O_2N$, m.p. 273~275°, both products showed the bands for α,β-unsaturated ketone in their infrared spectra. On catalytic hydrogenation, the former gave a saturated diketone, $C_{20}H_{25}O_2N$, m.p. 173~175, which was identified with the oxidation product of β-dihydrokobusinone. The latter gave α-dihydrokobusinone. Diacetylkobusine, $C_{24}H_{31}O_4N$, m.p. 139~141°, and also dibenzoylkobusine (hydrochloride, $C_{34}H_{35}O_4N \cdot HCl \cdot H_2O$, m.p. 290~292°) were prepared. Allyl alcohol of the type of $HO-\overset{|}{C}-\overset{|}{C}=CH_2$ and secondary hydroxyl group were thus confirmed as the functional groups in kobusine.

(Received July 10, 1958)