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220. Shuji Takahashi and Eiji Ohki[*1]: Chemical Studies on Azalomycins. III.[*2] Alkaline Degradation of Azalomycin-B.

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Alkaline degradation of azalomycin-B, a macrolide antibiotic produced by *Streptomyces hygroscopicus* var. *azalomyceticus*, afforded oct-4-en-3-one (II), an unsaturated keto-alcohol (X), and 2,4-dimethylphenylacetic acid (VIII), accompanied with lower-graded degradation products. Based on elucidated structures of these products, a partial structure of (XIV) was proposed for this antibtotic.

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In previous papers, it was reported that the molecular formula of azalomycin-B, a macrolide antibiotic produced by *Streptomyces hygroscopicus* var. *azalomyceticus*, was tentatively established as $C_{52}H_{92}O_{19}$[1] and its sugar component was designated as 2-deoxy-L-fucose.[*2]

Azalomycin-B (I) is very sensitive to acids; even if a weak acid like acetic acid or phosphoric acid was used, hydrolysis of I proceeded without any control to give 2-deoxy-L-fucose and a complex mixture of aglycons shown by many spots revealed on thin-layer chromatogram. Attempted separation of this hydrolysate failed to give any simple material. In addition, I is also labile to bases; even under mild conditions such as trimethylamine in ethanol, azalomycin-B (I) liberated 2-deoxy-L-fucose and gave a rather a simple, but still inseparable mixture of products. Therefore, we carried out an alkaline degradation of I under more drastic conditions. Based on the structures of lower-graded degradation products thus obtained, we attempted to presume the partial structure of I which forms the subjects of this paper.

Degradation of I was conducted by heating it in 10% aqueous sodium hydroxide, followed by separation of a neutral fraction, which is volatile with steam, and an acid fraction, as described in the experimental section.

As for the neutral fraction, the water-soluble part was found to contain acetaldehyde, propionaldehyde, and methyl ethyl ketone. Each of them was separated and characterized as their 2,4-dinitrophenylhydrazones, whose melting point, infrared spectra, and gas chromatograms were identical with those of the authentic samples.

The water-insoluble part of the neutral fraction, whose gas chromatogram is shown in Fig. 1, was fractionally distilled to give a colorless oil (II) of b.p$_{40}$ 100~110°. The infrared absorption at 1700 cm^{-1} and the ultraviolet absorption at 225 mμ suggested the presence of monosubstituted α,β-unsaturated ketone group in II. Hydrogenation of II on palladium-charcoal gave a saturated ketone (III), b.p. 160°.

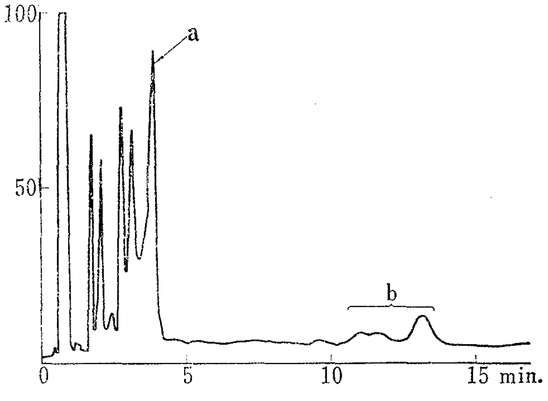

Fig. 1. Gas Chromatogram of the Neutral Fraction (Fraction 2) obtained by Alkaline Degradation of Azalomycin-B

(PEG 6000, 2.25 m., at 180°, 60 ml./min.).
a : Oct-4-en-3-one (II).
b : A high-boiling oil of b.p$_{2}$ 120~140°.

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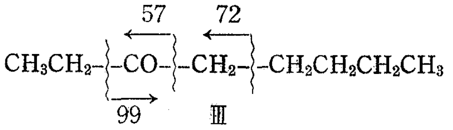
[*2] Part II: This Bulletin, **15**, 1651 (1967).
1) Part I: *Ibid.*, **15**, 1657 (1967).

III formed a 2,4-dinitrophenylhydrazone of m.p. 74∼77°, whose analytical values suggested a hydrazone of octanone, $C_8H_{16}O$. Nuclear magnetic resonance (NMR) of III ruled out the presence of a methyl ketone function and a mass spectrometric analysis of III suggested the structure of octan-3-one; the intense fragmentation peaks at m/e 99 and 57 were indicative of $[CH_3(CH_2)_4CO]^+$ and $[CH_3CH_2CO]^+$ (α-cleavage), and that at m/e 72 was indicative of $[CH_2{=}C(OH)\text{-}CH_2CH_3]^+$ (β-cleavage).



$$CH_3CH_2\text{-}\underset{\xrightarrow{99}}{\overset{\xleftarrow{57}}{CO}}\text{-}\overset{\xleftarrow{72}}{CH_2}\text{-}CH_2CH_2CH_2CH_3$$

III

$CH_3CH_2COCH{=}CHCH_2CH_2CH_3$

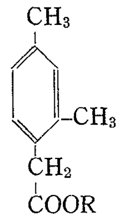
II

The structure of III was also confirmed by comparison with the authentic sample and the hydrazone derivative. NMR absorption of II, accompanied with decoupling study, suggested the presence of an ethyl ketone group; CH_3 signal centered at 1.08 p.p.m. coupled with $\text{-}CH_2CO\text{-}$ signal centering at 2.50 p.p.m. (J=7 c.p.s.). Based on these data, II was designated as oct-4-en-3-one. Furthermore, II was identified by gas-chromatography and NMR spectra with the synthetic sample prepared by treatment of α-hexenoyl chloride with ethyl zinc bromide.

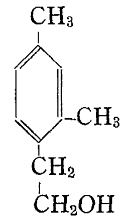
Distillation of the neutral fraction afforded a higher-boiling oil, b.p$_2$ 120∼140°, whose ultraviolet absorption at 279 mμ suggested that this oil contained a kind of $\alpha,\beta:\gamma,\delta$-conjugated dienone compounds. Gas chromatographic test, however, showed that this oil was a complex mixture and an attempt to prepare any crystalline derivative was unsuccessful.

In another run, the neutral fraction was oxidized directly with silver oxide, followed by separation of neutral and acid parts. Although pure product was not isolated from the acid part, the gas chromatography indicated the presence of tiglic acid, accompanied with acetic acid and propionic acid. Methylation of the acid mixture with diazomethane gave a mixture of methyl esters, which was found to contain methyl tiglate by gas chromatography. This methyl ester mixture was hydrogenated on palladium-charcoal to a saturated ester mixture, whose gas chromatogram also indicated the presence of methyl α-methylbutyrate by comparison with an authentic sample. This sequence of experiments showed that the neutral fraction of the alkaline degradation contained tiglic aldehyde, although it should further be ascertained.

As for the acid fraction obtained by the alkaline degradation, gas chromatographic detection indicated the presence of a considerable amount of acetic acid, less amount of propionic acid, and a trace of butyric acid as lower fatty acids. The part of higher fatty acids was directly esterified with diazomethane, followed by fractional distillation, and gave a colorless oil (IV) of b.p$_{10}$ 110°, $C_{11}H_{14}O_2$. The ultraviolet absorption of IV at 267∼268 mμ suggested the presence of an aromatic ring, and its NMR spectrum exhibited absorptions at 2.24 p.p.m. corresponding to two benzenoid methyl groups and at 3.45 p.p.m. of a methylene group located between the aromatic ring and the methoxycarbonyl group. Base on these data, IV was designated as methyl dimethylphenylacetate. Treatment of IV with lithium aluminum hydride afforded dimethylphenethyl alcohol (V) of b.p$_{20}$ 180°, $C_{10}H_{14}O$, which was characterized as its 3,5-dinitrobenzoate, m.p. 143∼145°. The infrared absorption pattern of V in the region of 5∼6 μ corresponded to that of 1,2,4-substitution of a benzene ring; therefore, V was assumed to be either 2,4-dimethyl-(V), 3,4-dimethyl-(VI), or 2,5-dimethyl-phenethyl alcohol (VII). Gas chromatograhic comparison with each of the authentic sample ruled out the structure of VII (5% neopentylglycol succinate 2.25 m. at 150°, 60 ml./min.). Whether the dimethylphenethyl alcohol was V or VI was determined by comparison of the melting point or mixed melting



Ⅳ : R=CH_3, Ⅷ : R=H



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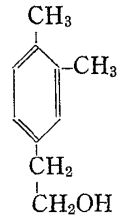

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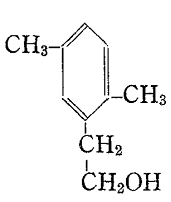

Ⅶ

point of its 3,5-dinitrobenzoate with that of the authentic sample; thus, the alcohol was designated as 2,4-dimethylphenethyl alcohol, and consequently, Ⅳ as methylphenylacetate. Therefore, one component of the acid products was found to be 2,4-dimethylphenylacetic acid (Ⅷ).

The fractional distillation of the ester mixture of higher fatty acids afforded a small amount of a high boiling oil and a considerable amount of residue. Separation of simple materials from each of them was not successful. The degradation products are summarized in Table I.

As mentioned before, degradation of azalomycin-B (I) and its derivatives under other alkaline conditions gave complex mixtures, which showed at least four spots on thin-layer chromatograms and which could not be separated into any simple material.

A successful result was obtained by the reduction of perhydroazalomycin-B (Ⅸ), prepared by hydrogenation of the acetate of I,[1] with lithium aluminum hydride. Treatment of Ⅸ with this reagent in tetrahydrofuran afforded an unsaturated keto-alcohol (X), b.p$_{0.001}$ 100° and a polyhydroxyl compound*[3] (Ⅺ), m.p. 105°.

The elemental analysis of X was satisfactory with a molecular formula of $C_{10}H_{18}O_2$. The ultraviolet absorption of X at 225 mμ and the infrared absorptions at 1675 and 1635 cm^{-1} indicated the presence of an α,β-unsaturated ketone group. The presence of an additional hydroxyl group was detected by the infrared absorption at 3450 cm^{-1} and by acetylation to give a monoacetate (Ⅻ) of b.p$_{0.001}$ 80°, $C_{12}H_{20}O_3$. NMR analysis of Ⅻ, as shown in Fig. 2, was indicative of the structure of the δ-acetoxy-α,β-unsaturated ketone. The doublet, centering at 6.10 p.p.m. coupled with the quartet at 6.63 p.p.m. (J=16 c.p.s.), supported the presence of an α,β-unsaturated ketone system. Irradiation at 2.17, corresponding to an allylic methine group showed that the quartet at 6.63 p.p.m. was further coupled with the signals at 2.17 p.p.m. of the allylic methine proton, indicating the presence of $-COCH{=}CH-\overset{|}{C}H-$ system. Since this irradiation study also indicated that the ABX_3-type octet at 5.05 p.p.m., characteristic of the proton on a carbon atom bearing the acetoxyl group, was also coupled with the allylic methine proton. The system of $=CH-\underset{|}{C}H-\underset{\substack{|\\ CH_3}}{C}H-OCOCH_3$ was presumed to be present in the molecule. On the other hand, two proton absorptions at 2.63 p.p.m. (quartet) was coupled with that of one of the terminal methyl groups (J=6.5 c.p.s.), suggesting the presence of CH_3CH_2CO- group. Based on these data the acetate (Ⅻ) would be formulated as below and consequently, the keto-alcohol as X.

It was found that the keto-alcohol (X) was also obtained by the treatment of azalomycin-B (I) with potassium hydroxide in methanol at room temperature. In this case, the keto-alcohol (X) itself could not be separated due to contamination of accompanying complex products, but X was detected and identified by thin-layer and gas-liquid

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*[3] Polyhydroxyl compound (Ⅺ), whose analytical data corresponded to the formula of $C_{32}H_{64}O_{11}$, was stable to bases, but still unstable to acids and liberated 2-deoxy-L-fucose. Structural study of Ⅺ is now in progress.

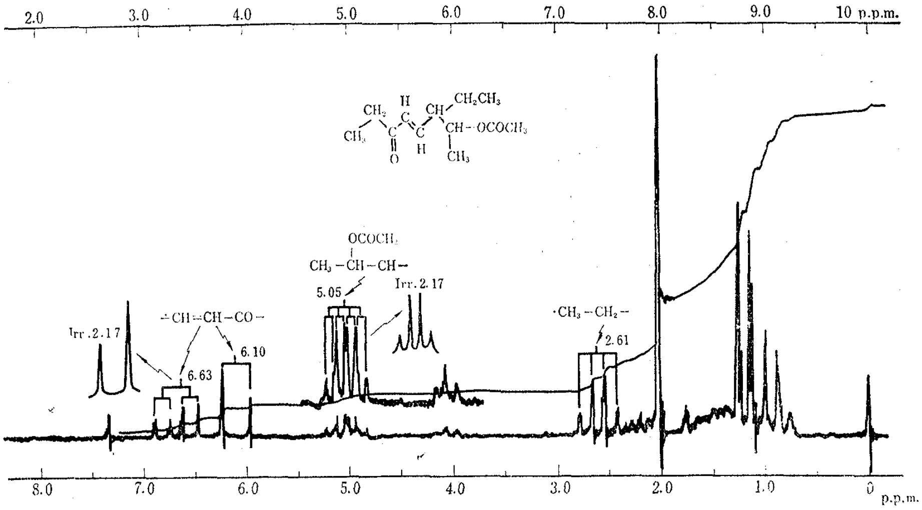
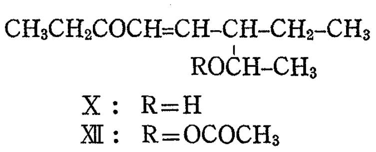


Fig. 2. NMR Spectrum and Irradiation Study of Unsaturated Acetoxy Ketone (XII) in $CDCl_3$

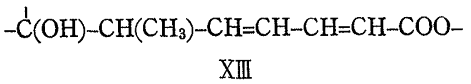
chromatography. Hence, X was assumed to be one of the alkaline degradation products, which was not produced primarily by reduction with lithium aluminum hydride, but by the basic conditions at the time of decomposition of the lithium aluminum hydride reaction complex.



$$CH_3CH_2COCH{=}CH\text{-}\underset{\displaystyle ROCH\text{-}CH_3}{CH}\text{-}CH_2\text{-}CH_3$$

X : R=H

XII : $R=OCOCH_3$

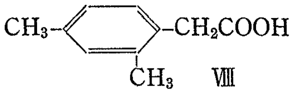


$$\text{-}\overset{|}{C}(OH)\text{-}CH(CH_3)\text{-}CH{=}CH\text{-}CH{=}CH\text{-}COO\text{-}$$

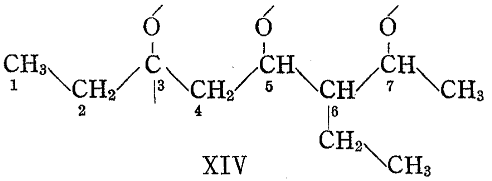
XIII

In a previous paper,[1] it was suggested that azalomycin-B (I) has an unsaturated lactone function (XIII) as a chromophor. The presence of any other ketonic function must be ruled out, because perhydro-acetate (IX) was not catalytically hydrogenated with platinum oxide and IX also exhibited no remarkable exaltation on the optical rotatory dispersion curve.

Table I. Components obtained by Alkaline Degradation of Azalomycin-B

CH_3CHO	CH_3COOH
CH_3CH_2CHO	CH_3CH_2COOH
$CH_3CH{=}CCH_3\text{-}CHO$	$CH_3CH_2CH_2COOH$
$CH_3CH_2COCH{=}CHCH_2CH_2CH_3$ II	 VIII
$CH_3CH_2COCH{=}CH\text{-}\underset{\displaystyle CH(OH)CH_3}{CH}\text{-}CH_2CH_3$ X	

Although how a ketone function was generated by the treatment of azalomycin-B with a base is still obscure, presumption of a presence of a carbon-like XIV suggests one of plausible passways which illustrate these retro-aldol degradation products; initial generation of a ketone function at the position of carbon-3, followed by removal of the O-substituent at the 5-position, and hydrolysis of the O-substituent at 7-position to give the unsaturated keto-alcohol (X). Further retro-aldol splitting at the bond between the carbon-6 and -7 would afford the octenone (II) and acetaldehyde. Butyraldehyde propionaldehyde, and methyl ethyl ketone are also explainable by the same consideration of the reteo-aldol degradation process of the presumed formula (XIV).



XIV

Formation of 2,4-dimethylphenylacetic acid (VIII) will be discussed in a future paper.

Experimental

Melting points are not corrected. Ultraviolet spectra were determined in 95% EtOH on a Beckman Model DK-2 and infrared spectra on a Perkin-Elmer Model 21. Proton magnetic resonance spectra were taken on a Varian A-60 or HA-101 spectrometer with Me_4Si as an internal standard. Analyses of gas-liquid chormatography were conducted with a Shimadzu Model GC-IB programmed vapor-phase chromatograph. Molecular weight determinations were carried out on a Vapor Pressure Osmometer Model 301A "Mechrolab." Plates for thin-layer chromatography were prepared with Silica-Gel G (E. Merck AG). Visualisation of spots was effected by spraying conc. H_2SO_4, followed by heating.

Alkaline Degradation of Azalomycin-B——Twenty grams of azalomycin-B was added to 700 ml. of 10% NaOH solution and the mixture was heated by direct introduction of steam to boiling under stirring. The steam distillate was collected in a cooled mixture of ether and H_2O. This distillation process was continued for about 20 hr. until practically no more precipitate formed by adding aqueous 2,4-dinitrophenylhydrazine hydrochloride to the distillate. Separation of the reaction products was carried out as shown in Chart 1.

After washing three times with ether, the aqueous layer of the distillate was treated with sufficient amount of aqueous 2,4-dinitrophenylhydrazine hydrochloride to give 1.72 g. of a hydrazone mixture (Fraction 1). The ether washings and ether layer of the distillate were combined and dried over anhyd. Na_2SO_4 (Fraction 2).

The distillation residue was washed twice with ether. After drying, the ether was evaporated to give 0.2 g. of a yellow oil which was not further investigated. The aqueous layer was acidified with 20% HCl under cooling and extracted with ether. Removal of the solvent from the ether extract afforded 7 g. of an acid fraction (Fraction 3). The remaining aqueous solution was steam-distilled again. The distillate was made basic with conc. NH_4OH and evaporated *in vacuo* to give 1 g. of an ammonium-salt mixture of lower fatty acids (Fraction 4).

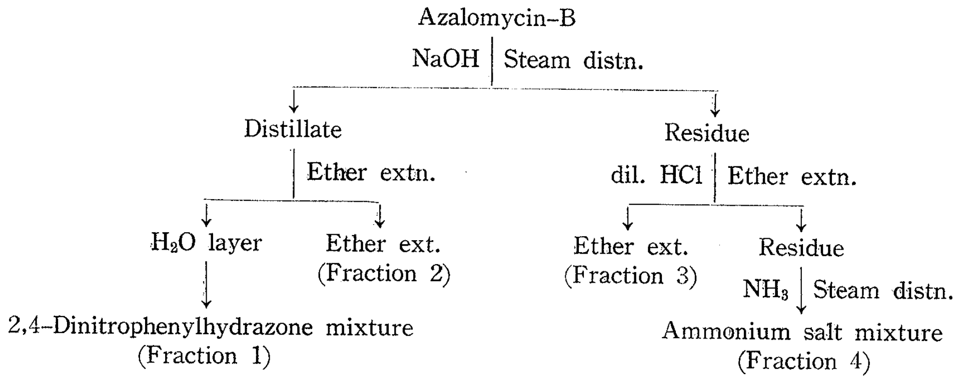


Chart 1.

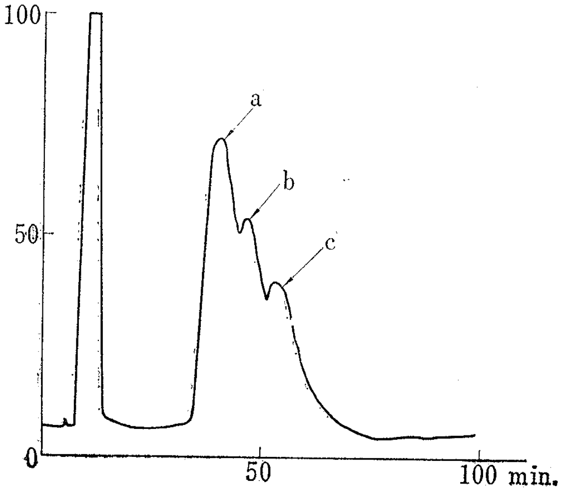


Fig. 3. Gas Chromatogram of Fraction 1

(1.5% SE-30 on Chromosorb W, 2.25 m., at 220°, 60 ml./min.)
a : 2,4-Dinitrophenylhydrazones of acetaldehyde
b : propio aldehyde c : methyl ethyl ketone

i) **Fraction 1**——Thin-layer and gas-liquid chromatographies (cf Fig. 3) indicated that this fraction contained three components. Following the method of Gordon, *et al.*,[2] this fraction was chromatographed on a silicic acid-Celite (2:1) mixture, and the column was eluted with petr. ether containing a small amount of ether. The hydrazones thus separated by chromatography, followed by repeated recrystallization, were identified with authentic samples by mixed m.p., infrared and NMR spectrometry, thin-layer and gas-liquid chromatography. 2,4-Dinitrophenylhydrazones of acetaldehyde, m.p. 147°, that of propionaldehyde, m.p. 156°, and of methyl ethyl ketone, m.p. 115°, were separated and identified.

ii) **Fraction 2**——Removal of the solvent from ether extract afforded 2.5 g. of a pale yellow oil. The removed solvent was treated with aqueous 2,4-dinitrophenylhydrazine hydrochloride to give 0.1 g. of a hydrazone mixture which was also found to contain 2,4-dinitrophenylhydrazones of acetaldehyde, propionaldehyde, and methyl ethyl ketone by means of thin-layer and gas-liquid chromatography. The thin-layer

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2) B. E. Gordon, F. Wapot, Jr., H. D. Burnham, L. C. Jones, Jr. : Anal. Chem., 23, 1754 (1951).

chromatograpy of this hydrazone mixture indicated the presence of other higher homologs. Attempted separation of the mixture afforded a substance of m.p. 164～170° (*Anal.* Calcd. for $C_{12}H_{14}O_4N_4$: C, 51.79; H, 5.07; N, 20.14. Found : C, 53.51; H, 5.27; N, 19.26). The structure of $CH_3CH_2CH{=}CCH_3{-}CHO$ or $CH_3CH_2C(CHO){=}CHCH_3$ was suggested by NMR analysis but could not be further investigated for the lack of the sample.

The pale yellow oil (2.5 g.) obtained above, whose gas chromatogram is shown in Fig. 1, was fractionally distilled twice to give 0.7 g. of oct-4-en-3-one (II) of b.p$_{40}$ 100～110° as a main product. UV λ_{max} 225 mμ (ε 9800). IR $\nu_{max}^{liq.}$ 1700 cm^{-1} (CO). *Anal.* Calcd. for $C_8H_{14}O$: C, 76.19; H, 11.11. Found : C, 76.88; H, 11.51.

The NMR spectrum of II showed absorptions at 1.08 (3H, triplet with J=7 c.p.s. : $C\underline{H}_3-$), 2.50 (2H, doublet with J=7 c.p.s. : $-C\underline{H}_2CO-$), 6.10 (1H, doublet with J=15 c.p.s. : $-CO-C\underline{H}=$), and 6.29 p.p.m. (1H, doublet triplet with J=15 c.p.s. : $-CO-CH{=}C\underline{H}-$). Hydrogenation of II on 5% Pd-C in ether afforded octan-3-one (III) of b.p. 160°, $C_8H_{16}O$, which formed 2,4-dinitrophenylhydrazone of m.p. 74～77° (from hexane). *Anal.* Calcd. for $C_{14}H_{20}O_4N_4$: C, 54.53; H, 6.54; N, 18.49. Found : C, 54.45; H, 6.59; N, 18.49. III was identical with the authentic sample by infrared, NMR, and mass spectrometry, gas chromatography (PEG 6000, 150°, 2.25 m., 60 ml./min.), and mixed m.p. of its 2,4-dinitrophenylhydrazone. Oct-4-en-3-one (II) could not be derived to any crystalline derivative, but was identified with the sample prepared by the method described below by means of infrared and NMR spectrometry, and by gas chromatography.

Fractional distillation of the residue gave a small amount of high-boiling liquid of b.p$_2$ 120～140°, which was found to be still impure by gas chromatography (diethylene glycol succinate, 2.25 m., 180°, 60 ml./min.). UV spectrum of the fraction exhibited an absorption at 279 mμ, suggesting the presence of a conjugated linear dienone compound. Attempted preparation of any crystalline derivatives was not successful.

A mixture of 0.5 g. of Fraction 2 and 1 g. of Ag_2O in dil. NaOH was stirred for 5 hr. at room temperature. After filtration of the mixture, the filtrate was washed three times with ether. The aqueous layer was acidified with conc. H_2SO_4 and extracted with 50 ml. of ether by using a continuous extractor. After drying over Na_2SO_4, the ether extract was evaporated to leave 0.3 g. of an oil. Acetic acid, propionic acid, and tiglic acid, constituents of the oil, were identified with the authentic samples by gas-liquid chromatography (DEGS-H_3PO_4, 2.25 m, 130°, 100 ml./min.). The oil was methylated with CH_2N_2-Et_2O. Gas-liquid chromatography (PEG 6000, 2.25 m., 130°, 60 ml./min.) of the mixture of methyl esters showed the presence of methyl tiglate. The methylated oil was hydrogenated in ether using 100 mg. of 5% Pd-C to give an oil, whose constituent, methyl α-methylbutyrate, was identified with the authentic sample by gas-liquid chromatography (PGE 6000, 2.25 m., 175°, 60 ml./min.).

iii) Fraction 3——To a solution of Fraction 3 in 50 ml. of ether sufficient amount of a CH_2N_2-Et_2O solution was added and the mixture was allowed to stand for 1 hr. Removal of the solvent gave 8.5 g. of a methyl ester mixture whose gas chromatogram is shown in Fig. 4. Fractional distillation of this mixture gave 1.5 g. of a colorless oil of b.p$_{20}$ 90～150° which was further fractionated to afford 0.45 g. of methyl 2,4-dimethylphenylacetate, (IV), b.p$_{10}$ 110°. UV λ_{max} mμ (ε) : 267～268 (500). IR $\nu_{max}^{liq.}$ cm^{-1}: 1610 (aromatic). *Anal.* Calcd. for $C_{11}H_{14}O_2$: C, 74.13; H, 9.92. Found : C, 72.41; H, 8.02.[*4]

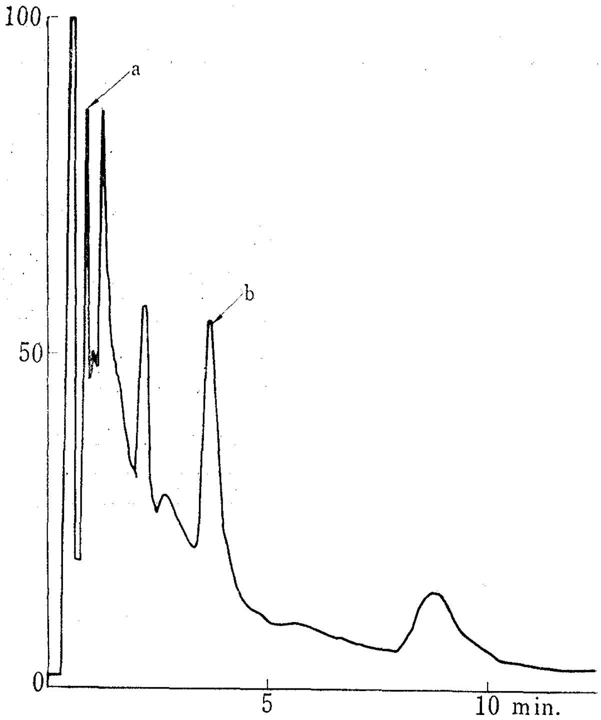

Fig. 4. Gas Chromatogram of the Methyl Ester Mixture obtained from Fraction 3

(1.5% SE-30 on Chromosorb W, 2.25 m., at 210°, 60 ml./min.)
a : Methyl 2,4-dimethylphenyl acetate.
b : The high-boiling acid ester of b.p$_{0.005}$ 150～180°.

IV thus obtained was treated with $LiAlH_4$ in ether in the usual manner and gave 2,4-dimethylphenethyl alcohol (V) of b.p$_{20}$ 180°. UV λ_{max} mμ (ε) : 267～268 (510). IR $\nu_{max}^{liq.}$ cm^{-1} : 1600, 820, 870 (aromatic). *Anal.* Calcd. for $C_{11}H_{14}O$: C, 79.95; H, 9.39. Found : C, 79.41; H, 9.20.

V formed 3,5-dinitrobenzoate of m.p. 143～145°. *Anal.* Calcd. for $C_{17}H_{16}O_6N_2$: C, 59.30; H, 4.68; N, 8.14. Found : C, 58.90; H, 4.43; N, 8.19. V was identical with the authentic sample by infrared spectrum and mixed melting point of its 3,5-dinitrobenzoate. The fractional distillation of the ester mixture afforded

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[*4] Analytical sample was not purified further because of the lack of the material.

a high-boiling acid ester of b.p$_{0.005}$ 150~180°. UV λ_{max} mμ: 226, 285. *Anal.* Found: C, 71.22, 71.21; H, 8.83, 8.82; mol. wt., 340 (in tetrahydrofuran), 367 (in $CHCl_3$). The highest mass number of this fraction was 302.

iv) **Fraction 4**——Gas-liquid chromatography of Fraction 4 (DC 550-stearic acid or dioctyl phthalate B, 2.25 m., at 120°, 60 ml./min.) showed that this fraction contained acetic acid, a small amount of propionic acid, and a trace of butyric acid. Paper partition chromatography ($EtOH:H_2O:NH_3=80:10:4$) did not show any spot of higher homologs other than acetic, propionic, and butyric acids.

Synthesis of Oct-4-en-3-one——A mixture of 7.6 g. (0.054 mole) of freshly-fused anhyd. $ZnCl_2$ and 20 ml. of ether was stirred and 0.054 mole of EtMgBr was added. After the initial reaction subsided, the mixture was heated to boiling for 2 hr., during which sufficient ether was distilled from the mixture to reduce its volume to about 60 ml. A solution of 2.65 g. (0.04 mole) of 1-hexenyl chloride[3] in 20 ml. of dry benzene was added with stirring and the resulting mixture was heated to boiling for 3 hr. After the mixture was cooled, 100 ml. of 2*N* HCl was added. Treatment of the reaction mixture in the usual manner afforded 2.1 g. of oct-4-en-3-one of b.p$_{40}$ 90~94°.

Reduction of Perhydro-azalomycin-B Acetate (IX) with Lithium Aluminum Hydride. i) Unsaturated Keto-alcohol (X)——To a solution of 10 g. of perhydro-azalomycin-B acetate[1] (IX) of m.p. 188~191° in 500 ml. of tetrahydrofuran, 7 g. of $LiAlH_4$ was added in small portions under cooling and stirring, and the mixture was refluxed on a steam bath for 24 hr. After excess of the reagent was decomposed with water, the precipitate was filtered, and the filtrate was concentrated to leave 2.5 g. of an oil which was chromatographed on Alumina III. The benzene eluate, which was found to be pure by gas chromatography (1.5% SE 30 on Chromosorb-W, 2.25 m., at 180°, 60 ml./min.), was distilled to give 200 mg. of an unsaturated keto-alcohol (X), as colorless liquid of b.p$_{0.001}$ 100°. UV λ_{max} mμ (ε): 225 (8000). IR $\nu_{max}^{liq.}$ cm^{-1}: 1675 (CO), 1635 (C=C), 3450 (OH). *Anal.* Calcd. for $C_{10}H_{18}O_2$: C, 70.54; H, 10.66; mol. wt., 170. Found: C, 70.15; H, 10.63; mol. wt., 198.63 (in benzene). Acetytation of X was made in the usual manner to afford an acetate (XII) of b.p$_{0.001}$ 80°. IR $\nu_{max}^{liq.}$ cm^{-1}: 1680 (C=O), 1740 (acetyl). *Anal.* Calcd. for $C_{12}H_{20}O_3$: C, 67.89; H, 9.50. Found: C, 67.54; H, 9.51. The NMR spectrum of (XII) is shown in Fig. 2.

A solution of 3 g. of azalomycin-B (I) or perhydroazalomycin-B[1] in 100 ml. of MeOH containing 0.5 g. of KOH was allowed to stand for 3 days. The chemical change that underwent was checked through thin-layer chromatography (benzene:$Me_2CO=3:1$). One spot revealed on the chromatogram corresponded to that of X. The reaction mixture was diluted with H_2O and extracted with ether. Removal of the solvent from the extract afforded 3.1 g. of an oil, whose gas-liquid and thin-layer chromatography exhibited the presence of the unsaturated keto-alcohol (X).

ii) **Polyhydroxyl Compound (XI)**——The precipitate obtained in the $LiAlH_4$ reduction was dissolved in dil. HCl under cooling and extracted with EtOAc. The extract was dried over Na_2SO_4 and removal of the solvent from the extract gave 2.5 g. of crystals which were recrystallized from EtOAc to a polyhdroxyl compound (XI) of m.p. 105°. *Anal.* Calcd. for $C_{32}H_{64}O_{11}$: C, 62.51; H, 10.49; mol. wt., 614.8. Found: C, 61.63; H, 10.57; mol. wt., 626.3 (Tetrahydrofuran).

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