thiosulfate, or potassium permanganate at room temperature, but not with hydrogen peroxide. It was also found that the phenol fraction shows two polarographic reduction waves in phosphate buffer of pH 6.8 at $E_{1/2}=-0.41$ and $E_{1/2}=-0.61$.

The two active components in the the phenol fraction were separated from each other by paper chromatography, using several different solvent systems successively. Both components were positive to potassium cyanide-nitroprusside. Details of these experiments will be published elsewhere.

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Osaka Factory,
Takeda Chemical Industries, Ltd.,
Juso-nishinocho, Higashiyodogawa-ku, Osaka.

Fumihiko Tanaka (田中文彦)
Shigeru Shintani (新谷 茂)
Momoyoshi Nakamura (中村桃吉)
Iwao Nakayama (中山 巌)

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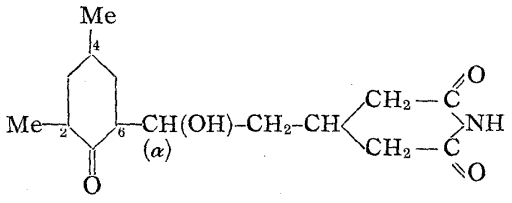
Absolute Configuration of Cycloheximide*1

Previous studies on the stereochemistry of cycloheximides made it possible to assume most of the configurations and conformations of the four isomers as illustrated in Table I and depicted in Chart 1.[1] However, it still remained to determine which of the configurations (I) or (II) is to be assigned to cycloheximide (Naramycin-A).

Table I. Configuration and Conformation of Cycloheximides

Isomers	Octant projection	Configuration	Conformation	[φ] Value at extrema of acetates
Cycloheximide (Naramycin-A)	(I) or (II)	4S:6S:αS	4-ax:6-eq	− 449° (at 312.5 mμ)
Naramycin-B	(VI)	2S:4S:6R:αS	2-ax:4-eq:6-eq	+2,923° (at 312.5 mμ)
Isocycloheximide	(V)	2R:4S:6R:αS	2-eq:4-eq:6-eq	+ 575°[b] (at 307.5 mμ)
α-Epiisocycloheximide[a]	(III)	2R:4S:6R:αR	2-eq:4-eq:6-eq	+ 449° (at 315 mμ)

a) A compound referred to as A_{II} in J. Antibiotics, **14A**, 158 (1961).
b) [φ] Value of synthesized acetyl-isocycloheximide (m.p. 166~167°).



Plane Structure of Cycloheximides

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*1 "Studies on Streptomyces Antibiotic, Cycloheximide. XVII.[2]"

1) Part IV. T. Okuda: This Bulletin, **7**, 659 (1959); Part VI. *Ibid.*, **7**, 671 (1959); Part VII. *Ibid.*, **8**, 335 (1960); Part XIV. Yakugaku Kenkyu, **33**, 532 (1961).

2) Parts XV and XVI. Presented before the 81st Annual Meeting of the Pharmaceutical Society of Japan (July, 1961). Preliminary Note. T. Okuda, M. Suzuki, Y. Egawa: J. Antibiotics, **14A**, 158 (1961).

When a molecule possesses more than one asymmetric center, rotation of the molecule is assumed to be the summation of the rotations of each individual asymmetric center. Therefore, adaptation of the "rule of optical superposition" and "octant rule" to the four acetyl cycloheximides, whose molecular rotations at the extreme ([ϕ] values) are shown in Table I, makes it possible to determine the absolute configuration of cycloheximide, because all of these acetates have a substituent at 6-position, taking an equatorial orientation in a chair-formed cyclohexanone ring and they are released from the interaction between carbonyl and hydroxyl groups.

Geometric considerations will now be made to see how much [ϕ] values can be expected of the configurations (I) and (II).

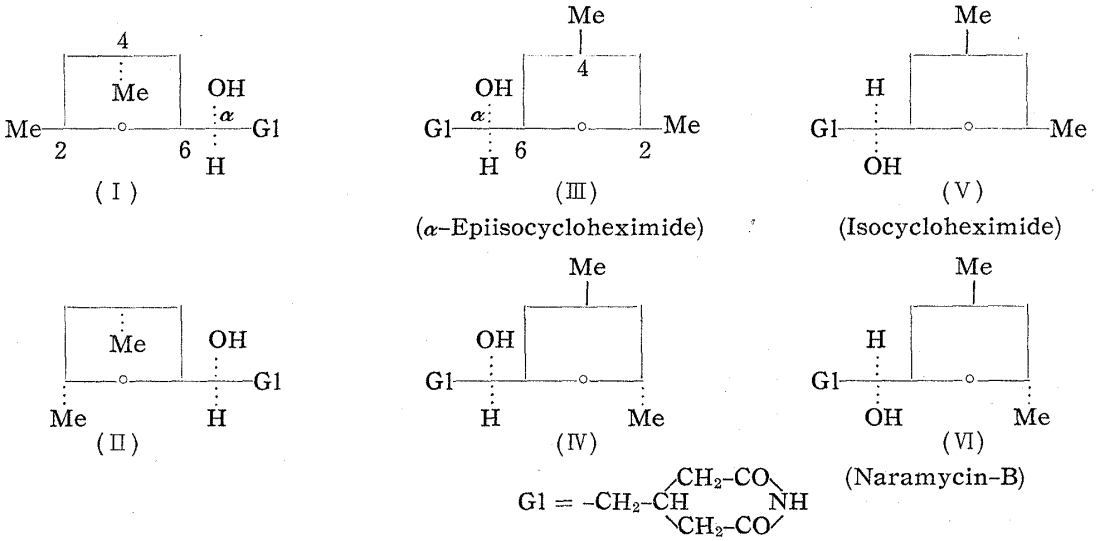

Chart 1. Octant Projection of Cycloheximides

As seen in the Chart, configurations (I) and (II) are quite antipodal to configurations (III) and (IV), respectively, in their stereochemical surroundings except C-4 configuration. According to the "octant rule," C-4 methyl group makes no substantial contribution to partial rotations in the Cotton effect and can be ignored.[3] Therefore, the [ϕ] value, which may be expected of configuration (I), will be reversed approximately to that of configuration (III) and [ϕ] value of configuration (II) will be quite opposite to that of configuration (IV). The [ϕ] value of this imaginary configuration (IV) can be approximately calculated as $[\phi]_B+([\phi]_E-[\phi]_I)$ $(=+2{,}797°)$, where $[\phi]_B$, $[\phi]_E$ and $[\phi]_I$ are respectively [ϕ] values of the acetates of Naramycin-B, α-epiisocycloheximide, and isocycloheximide, and $[\phi]_B-[\phi]_I$ corresponds to the difference which accompanies the change of C-α configuration. Thus, [ϕ] values of configurations (I) and (II) may be expected approximately as $-449°$ and $-2{,}797°$, respectively.

The fact that the $[\phi]_A$ observed of acetyl-cycloheximide is $-449°$ and identical with the value expected of configuration (I) shows that cycloheximide has the configuration depicted as (I) and belongs to (2S:4S:6S:αS)-series with C-2 methyl group equatorially oriented.*[2] The fact that observed [ϕ] value of configuration (I) was incidentally found to be identical with the one calculated gives good support to the correctness of the "octant rule" as to the contribution of C-4 center.

3) C. Djerassi: "Optical Rotatory Dispersion—Applications to Organic Chemistry," 181 (1960). McGraw-Hill Book Co., Inc., New York.

*[2] Antipodal relationship between acetyl-cycloheximide and acetyl-α-epiisocycloheximide is observed not only in [ϕ] values at extrema but also in full figures of their RD curves (cf. Parts XIV and XV).

Mathematical treatment of $[\phi]$ values of acetyl-cycloheximides also leads to the same conclusion as above. Moreover, this treatment shows that contribution of axial (2S)-2-methyl group is represented as $[\phi]_B-[\phi]_I$ (=2,348°), provided that the contribution of equatorial 2-methyl group is ignored.*3,*4

Recently, Lawes[4] found that cycloheximide undergoes pyrolysis at about 200° to afford (2S:4R)-2,4-dimethylcyclohexanone and further, from conformational considerations and known isomerization reactions,[5] he deduced the configuration of cycloheximide tentatively as depicted as (IV) or (VI). This is different from the conclusion reached in the present series of considerations, but his experimental observations are also well explicable from the structure assigned by the present authors.

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Tokyo Research Laboratory,
Tanabe Seiyaku Co., Ltd.,
Toda-machi, Kitaadachi-gun, Saitama-ken.

Tomoharu Okuda (奥田朝晴)
Makoto Suzuki (鈴木真言)

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*3 The contribution of axial (2S)-2-methyl group is correctly represented as $[\phi]_B-[\phi]_I-X_e$, in which X_e is the contribution of equatorial (2S)-2-methyl group.

*4 Effect of the bulky substituent at C-2 position of equatorial orientation was treated by Prof. C. Djerassi (J. Am. Chem. Soc., **83**, 3334 (1961)).

4) B. C. Lawes: Private Communication (cf. Abstracts of Papers, 139th Meeting of the American Chemical Society, 1961, p. 33N).

5) A. J. Lemin, J. H. Ford: J. Org. Chem., **25**, 344 (1960).

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Reaction of Dehydroacetic Acid under a Mild Condition

It was reported in previous papers[1] that dehydroacetic acid is very reactive with ammonia, primary amines, and some of the compounds possessing amino group, such as amino acids and sulfanilamides. The structure of the primary reaction product easily formed from them under a mild condition was deduced to be Schiff base-type compound derived from the combination of $>C=O$ group of 3-acyl side-chain of dehydro-acetic acid and NH_2 group in the amino compound.

Later, it became necessary to investigate the question of whether or not the reaction of dehydroacetic acid always stopped at the formation of a Schiff base in solution. Therefore, the reaction between dehydroacetic acid and amino compounds was reëxamined in view of the expectation that the primary reaction product (Schiff base) might change secondarily in solution, at least to a certain extent, even under a mild condition. For this purpose, the experiment was first carried out using ammonia and methylamine. The test solutions were prepared by adding dehydroacetic acid (1 g.) into the solution (50 cc.) of various concentrations of ammonia or methylamine, and kept in an incubator (37°). Progress of the reaction was traced periodically by paper chromatography. As was

1) S. Iguchi, *et al.*: Yakugaku Zasshi, **77**, 1258 (1957); This Bulletin, **7**, 323 (1959); *Ibid.*, 8, 1 (1960).