

Synthesis of (2S,3S,5S)-3-Hydroxy-5-methyl-2-pyrrolidinecarboxylic Acid,  
a Component of Actinomycin Z<sub>1</sub>

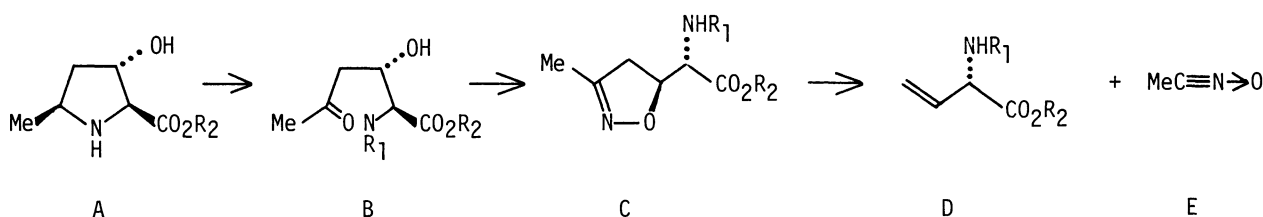
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(2S,3S,5S)-3-Hydroxy-5-methyl-2-pyrrolidinecarboxylic acid and the enantiomer, the relative configurations of which are the same as that of the natural compound found in actinomycin Z<sub>1</sub> were synthesized and the optical properties were determined.

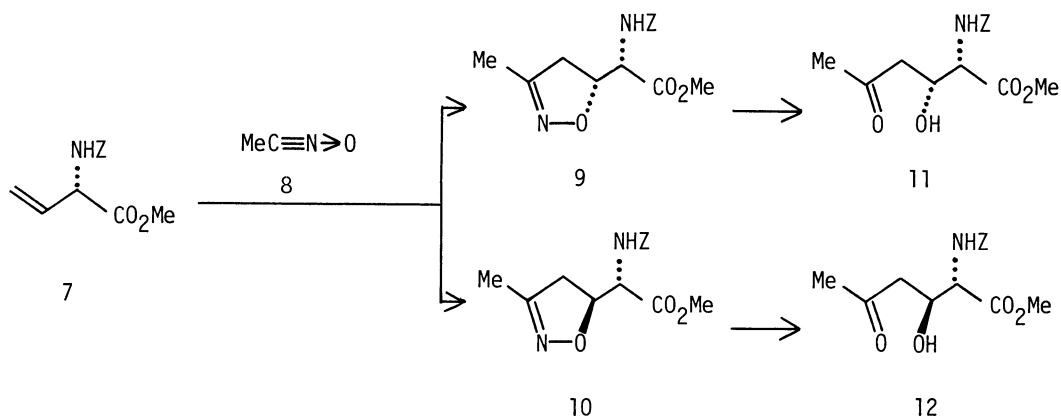
3-Hydroxy-5-methyl-2-pyrrolidinecarboxylic acid (3-hydroxy-5-methylproline) is an unusual imino acid identified as a component of a peptide antibiotic actinomycin Z<sub>1</sub>.<sup>1)</sup> The four diastereomers of the imino acid were synthesized by Mauger et al. and their relative stereochemistries were determined by NMR data and epimerization studies.<sup>2)</sup> It has been demonstrated that 3-hydroxy-5-methylproline obtained from the hydrolyzate of actinomycin Z<sub>1</sub> was identical with one of the synthetic isomers which has 2,3-trans and 2,5-cis relative stereochemistry, however, the absolute configuration (2S,3S,5S or 2R,3R,5R) remains unknown.

As a part of our efforts toward the synthesis of biologically active β-hydroxy-α-amino acid derivatives,<sup>3)</sup> we planned to synthesize (2S,3S,5S)-3-hydroxy-5-methylproline (**1**) in order to reveal the optical properties based on the retrograde pathway **A** → **B** → **C** → **D** + **E**.

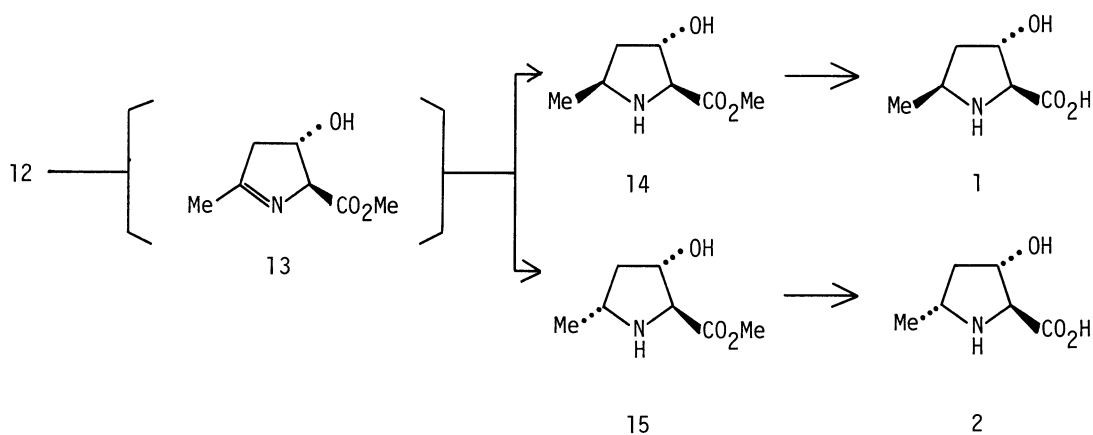


The main features are (i) [3+2] dipolar cycloaddition<sup>4)</sup> of **D** and **E**, (ii) reductive cleavage of an isoxazole ring to a β-hydroxyketone<sup>5a)</sup> (**C** → **B**), and (iii) reductive cyclization (**B** → **A**). The present communication describes an efficient synthesis of (2S,3S,5S)-3-hydroxy-5-methylproline (**1**) and the stereoisomers (**2**-**6**) along these lines.<sup>6)</sup>

Reaction of L-vinylglycine **7**<sup>7)</sup> (Z: benzyloxycarbonyl) and nitrile oxide **8** generated *in situ* (nitroethane, p-chlorophenylisocyanate,<sup>8)</sup> Et<sub>3</sub>N, benzene, 25 °C, 19 h, then reflux, 6.8 h) afforded a 2.3:1 mixture of *threo*- and *erythro*-isoxazolines,<sup>9)</sup> **9**, [α]<sub>D</sub> -0.69° (c 0.57, CHCl<sub>3</sub>) and **10**, [α]<sub>D</sub> +71° (c 0.51, CHCl<sub>3</sub>), respectively in 53% yield. These diastereomers were separated by column

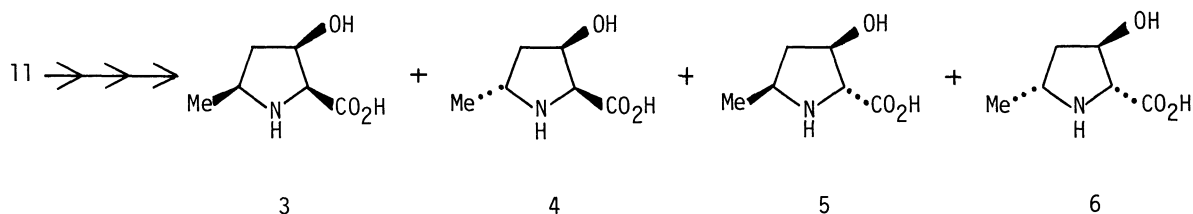


chromatography on silica gel (hexane/ethyl acetate, 2:1) and, the stereochemistry of the isolated compounds was determined by converting them to the corresponding proline derivatives shown below. Hydrogenolytic cleavage of isoxazoline **10** according to the Curran's method<sup>5b)</sup> (Raney-Ni,  $\text{B}(\text{OH})_3$ ,  $\text{MeOH}/\text{H}_2\text{O}$ ,  $\text{H}_2$ ) afforded a  $\beta$ -hydroxyketone **12**,  $[\alpha]_{\text{D}} -6.5^\circ$  (c 0.71,  $\text{MeOH}$ ) in 88% yield. After removal of the amino protecting group of **12** ( $\text{H}_2/10\% \text{Pd-C}$ ,  $\text{EtOH}$ ), the resulting cyclic imine **13** was reduced ( $\text{NaBH}_3\text{CN}$ ,  $\text{EtOH}$ , pH 5)<sup>10)</sup> to a diastereomeric mixture of 3-hydroxy-5-methylproline esters (**14** and **15**). The mixture was hydrolyzed (0.15 mol  $\text{Ba}(\text{OH})_2$ ,  $20^\circ\text{C}$ , 4 h) without separation and the hydrolyzate was subjected to a Dowex 50W x 4 column chromatography eluting with ammonia-formate buffer (pH 2.70) to afford (2*S*,3*S*,5*S*)-3-hydroxy-5-methylproline **1**, mp  $247-249^\circ\text{C}$  (dec.),  $[\alpha]_{\text{D}} -17^\circ$  (c 0.50,  $\text{H}_2\text{O}$ ) and (2*S*,3*S*,5*R*)-isomer **2**, mp  $252-255^\circ\text{C}$  (dec.),  $[\alpha]_{\text{D}} -10^\circ$  (c 0.48,  $\text{H}_2\text{O}$ ) in 44% and 26% yields (from **12**), respectively. The  $^1\text{H-NMR}$  data of **1** and **2** were in accord with those reported about racemic **1** and **2**, respectively.<sup>2)</sup>



The *threo*-isoxazoline **9** was independently converted by the same series of reactions to 3-hydroxy-5-methylprolines via  $\beta$ -hydroxyketone **11**,<sup>11)</sup>  $[\alpha]_{\text{D}} +2.8^\circ$  (c 0.71,  $\text{MeOH}$ ) (**9**  $\rightarrow$  **11**, 83% yield). A mixture of (2*S*,3*R*,4*S*)- and (2*S*,3*R*,4*R*)-isomers (**3/4**, 2:1) was obtained in 23% yield (from **11**) together with (2*R*,3*R*,4*S*) isomer **5** and (2*R*,3*R*,4*R*)-isomer **6** (7% and 5% yields, respectively).<sup>12)</sup> These results

suggest that epimerization attributable to a tautomerism of the cyclic imine intermediate occurred at the C-2 position. The (2R,3R,5R)-isomer 6 and (2R,3R,5S)-isomer 5 exhibited the same spectral properties as those of 1 and 2, respectively except the opposite optical rotations, 6,  $[\alpha]_D +18^\circ$  (c 0.32, H<sub>2</sub>O), 5,  $[\alpha]_D +10^\circ$  (c 0.20, H<sub>2</sub>O).



In conclusion, we demonstrated an efficient synthesis of optically active 3-hydroxy-5-methylprolines (1-6) and clarified their optical properties, which will serve to elucidate the absolute configuration of the natural compound from actinomycin Z<sub>1</sub>.<sup>13)</sup>

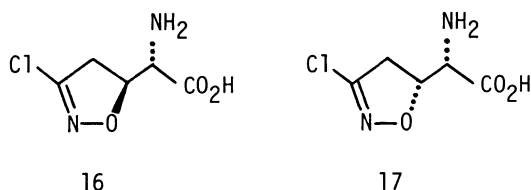
The present methodology seems to be very useful for preparations of  $\beta$ -hydroxy- $\alpha$ -amino and imino acids especially considering the wide variation of molecules containing nitryl oxide groups.

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#### References

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- 9) The ratio of the diastereomers was determined by <sup>1</sup>H-NMR analysis of the purified mixtures. The coupling constants between C<sub>( $\alpha$ )</sub>-H and C<sub>( $\beta$ )</sub>-H in the <sup>1</sup>H-NMR spectra of *threo*-isomer 9 and *erythro*-isomer 10 are 2.0 and 4.0 Hz respectively. These values do not correspond to those of AT-125 (*erythro*, 3.0 Hz) (16) and the C<sub>(3)</sub>-epimer (*threo*, 7.0 Hz) (17). J. E. Baldwin, L. I. Kruse, and J. K. Cha, *J. Am. Chem. Soc.*, **103**, 942 (1981); Low stereo-selectivity was observed in nitrile oxide cycloadditions to chiral terminal

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- 10) R. F. Borch, M. D. Bernstein, and H. D. Durst, *J. Am. Chem. Soc.*, **93**, 2897 (1971).
- 11) In this case, Kozikowski's procedure using  $\text{AlCl}_3$  in the place of  $\text{B(OH)}_3$  was used. See Ref. 5a.
- 12) The mixture of 3 and 4 could not be separated under the chromatographic condition (Dowex 50w x 4, pH 2.70 buffer) however, the structures and the ratio were confirmed by comparison of  $^1\text{H-NMR}$  spectrum of the mixture with those of racemic 3 and 4.
- 13) The natural 3-hydroxy-5-methylproline from actinomycin  $\text{Z}_1$  was not obtained in sufficient quantity for optical rotation. Private communication from Dr. A. B. Mauger.
- 14)  $^1\text{H-NMR}$  data: 9 (100 MHz,  $\text{CDCl}_3$ )  $\delta$  1.80 (3H, br s), 2.69-3.06 (2H, m), 3.68 (3H, s), 4.39 (1H, dd,  $J=2.0, 10.0$  Hz,  $\text{H}_{(\alpha)}$ ), 4.79-5.59 (2H, m,  $\text{H}_{(\beta)}$  and NH), 5.03 (2H, s), 7.24 (5H, s). 10 (100 MHz,  $\text{CDCl}_3$ )  $\delta$  1.94 (3H, br s), 3.07 (2H, br d,  $J=8.0$  Hz), 3.69 (3H, s), 4.40 (1H, dd,  $J=4.0, 8.2$  Hz,  $\text{H}_{(\alpha)}$ ), 4.78 (1H, dt,  $J=4.0, 8.0$  Hz,  $\text{H}_{(\beta)}$ ), 5.09 (2H, s), 5.63 (1H, br d,  $J=8.2$  Hz), 7.30 (5H, s). 11 (100 MHz,  $\text{CDCl}_3$ )  $\delta$  2.13 (3H, s), 2.68 (2H, br d,  $J=6.2$  Hz), 3.35 (1H, br d,  $J=3.2$  Hz, OH), 3.73 (3H, s), 4.34 (1H, br d,  $J=9.2$  Hz,  $\text{H}_{(\alpha)}$ ), 4.47-4.83 (1H, m,  $\text{H}_{(\beta)}$ ), 5.11 (2H, s), 5.65 (1H, br d,  $J=9.2$  Hz, NH), 7.32 (5H, s). 12 (100 MHz,  $\text{CDCl}_3$ )  $\delta$  2.14 (3H, s), 2.74 (2H, br d,  $J=5.6$  Hz), 3.55 (1H, br d,  $J=4.2$  Hz, OH), 3.71 (3H, s), 4.08-4.45 (2H, m,  $\text{H}_{(\alpha)}$ ,  $\text{H}_{(\beta)}$ ), 5.06 (2H, s), 5.70 (1H, br d,  $J=8.0$  Hz, NH), 7.27 (5H, s). 1 (500 MHz,  $\text{D}_2\text{O}$ )  $\delta$  1.51 (3H, d,  $J=6.7$  Hz, Me), 1.76 (1H, ddd,  $J=4.3, 11.5, 14.1$  Hz,  $\text{H}_{(4a)}$ ), 2.20 (1H, ddd,  $J=1.0, 6.0, 14.1$  Hz,  $\text{H}_{(4b)}$ ), 4.07 (1H, br s,  $\text{H}_{(2)}$ ), 4.07 (1H, ddd,  $J=6.0, 6.7, 11.5$  Hz,  $\text{H}_{(5)}$ ), 4.68 (1H, br dd,  $J=1.0, 4.3$  Hz,  $\text{H}_{(3)}$ ). 2 (500 MHz,  $\text{D}_2\text{O}$ )  $\delta$  1.48 (3H, d,  $J=6.8$  Hz, Me), 1.74 (1H, dddd,  $J=1.0, 4.6, 7.4, 14.0$  Hz,  $\text{H}_{(4b)}$ ), 2.44 (1H, ddd,  $J=5.9, 8.0, 14.0$  Hz,  $\text{H}_{(4a)}$ ), 3.93 (1H, ddd,  $J=6.8, 7.4, 8.0$  Hz,  $\text{H}_{(5)}$ ), 4.03 (1H, dd,  $J=1.0, 3.1$  Hz,  $\text{H}_{(2)}$ ), 4.62 (1H, ddd,  $J=3.1, 4.6, 5.9$  Hz,  $\text{H}_{(3)}$ ).
- 15)  $^{13}\text{C-NMR}$  data: 1 (25 MHz,  $\text{D}_2\text{O}$ )  $\delta$  17.4 (q), 39.7 (t), 56.0 (d), 69.9 (d), 74.6 (d), 172.0 (s). 2 (25 MHz,  $\text{D}_2\text{O}$ )  $\delta$  17.9 (q), 38.9 (t), 55.0 (d), 68.1 (d), 73.8 (d), 171.9 (s).

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