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**Synthesis and Determination of Stereochemistry of Four
Diastereoisomers at the C-24 and C-25 Positions
of $3\alpha, 7\alpha, 12\alpha, 24$ -Tetrahydroxy- 5β -
cholestan-26-oic Acid***

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In the major pathway for the biosynthesis of cholic acid from cholesterol, the latter compound is first converted into $3\alpha, 7\alpha, 12\alpha$ -trihydroxy- 5β -cholestane and then ω -oxidation takes place at the side chain terminal leading to $3\alpha, 7\alpha, 12\alpha$ -trihydroxy- 5β -cholestan-26-oic acid. Side chain cleavage of this acid is thought to proceed in a similar manner to the β -oxidation of long chain fatty acids. Thus, $3\alpha, 7\alpha, 12\alpha, 24$ -tetrahydroxy- 5β -cholestan-26-oic acid (varanic acid) is considered to be one of the intermediates in this process, and some evidence for the putative role of the tetrahydroxy acid intermediate has been reported. However, it is not known which of the four stereoisomers at the C-24 and C-25 positions is the true intermediate.

In this paper, four diastereoisomers at the C-24 and C-25 positions of varanic acid were synthesized in a stereochemically defined manner which contains the two key reactions: aldol condensation using a chiral imide enolate developed by Evans *et al.*, and Mitsunobu inversion reaction and, also by a non-stereoselective route, followed by chromatographic separation. Their stereochemistry at the C-24 and C-25 positions was established on the basis of the known stereochemical course of the reactions employed for the synthesis, and ^1H - and ^{13}C -nuclear magnetic resonance spectroscopic data of these isomers. On the basis of the evidence presented here it is concluded that the previous assignment for (24R, 25S) and (24R, 25R) isomers made by Hosita and co-workers must be revised.

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