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Determination of the C-7,9,12,13,17 and 18 Stereochemistries of Tautomycetin. Synthesis of the Tautomycetin Degradation Product

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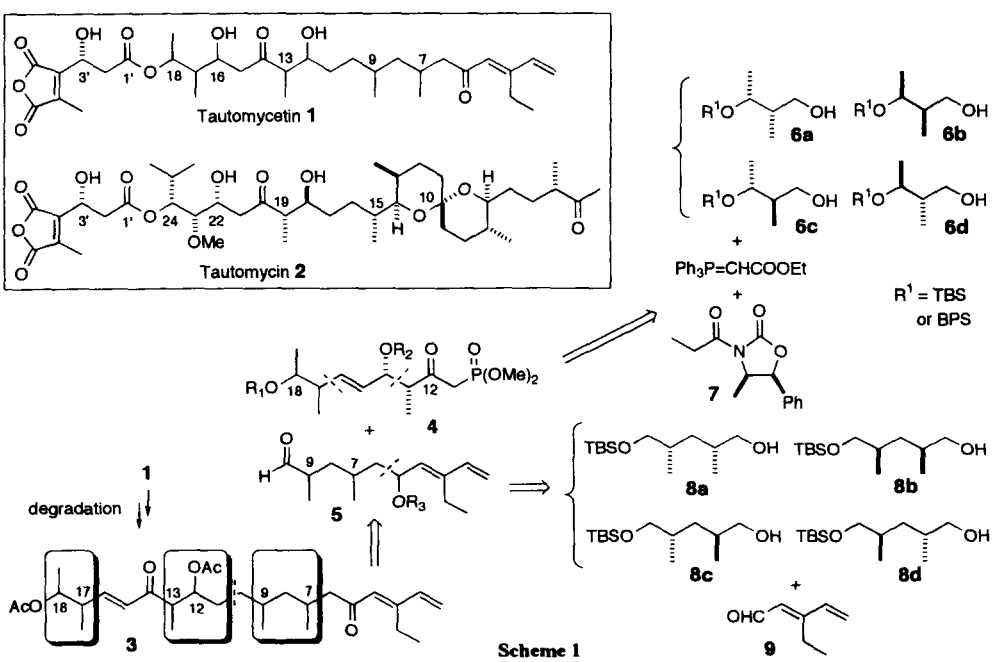
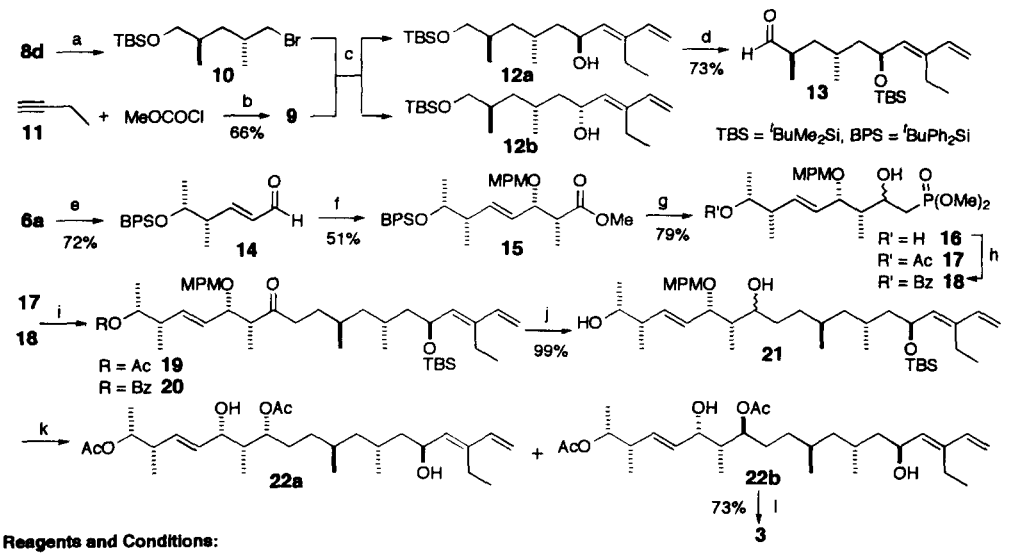
Abstract : Syntheses of the degradation product **3** of tautomycetin (**1**) and its diastereoisomers have been achieved. Comparison of the spectral data of these diastereoisomers with those of the degradation product of natural **1** indicates that tautomycetin has the 7*R*,9*S*,12*S*,13*S*,17*S*,18*R* stereochemistry.

Tautomycetin (**1**) was isolated from a culture of *Streptomyces griseochromogenes* and was reported to exhibit antifungal activity and to induce a morphological change in human leukemia cells K562.[1] **1** has also been found to affect the production of secondary metabolites in *Penicillium urticae*.[2] Tautomycin (**2**), a strong specific serine/threonine phosphatase inhibitor,[3] has been found to exhibit similar effects at a three-fold higher concentration than **1**. This data suggests a role for **1** as a powerful regulator of intracellular signal transduction. Tautomycetin has eight chiral carbon centers, but the stereochemistry of these centers has only been determined at one position (C3'). Because of its instability and limited supply determination of the stereochemistry of these chiral centers from the natural product alone will be difficult. As a step toward the total synthesis of **1**, we have determined the stereochemistry of these centers through the synthesis of the reported degradation product **3**.[1b]



Scheme 1

Our basic strategy for the stereocontrolled synthesis of **3** and its various diastereoisomers is shown in Scheme 1. The six chiral centers in **3** have been grouped into three pairs, C7,9, C12,13, and C17,18. We planned to introduce the first and third pairs of chiral centers using the stereochemically known fragments **8a-d** and **6a-d**. These fragments can be readily prepared from methyl (*R*)- or (*S*)-3-hydroxy-2-methylpropionate and ethyl (*R*)- or (*S*)-hydroxybutyrate respectively.[4,5] Using Evans chemistry the C13 and C14 stereocenters can then be set and used to direct the C12 stereochemistry.[6] Using this strategy we have synthesized fourteen diastereoisomers (some as a mixture of C12 epimers),[7] and after analysis of spectral data, we have concluded that **3** has the 7*R*,9*S*,12*S*,13*S*,17*S*,18*R* stereochemistry.



Reagents and Conditions:

a) Ph_3P, CBr_4, Et_3N, CH_2Cl_2, rt, (quant). b) i. BuLi, THF, -78 °C; ii. CuI, THF, H_2C=CHLi, -75 °C, 12 h; iii. DIBAL, CH_2Cl_2, -78 °C; iv. MnO_2, pentane, rt. c) ^{t}BuLi, THF, -100 °C, **12a** 17%, **12b** 20%. d) i. TBSCl, imidazole, DMF; ii. TBAF (1.0 M in THF), AcOH (1.0 *eq* to TBAF); iii. Dess-Martin periodinane, CH_2Cl_2, rt. e) i. Dess-Martin periodinane, CH_2Cl_2, rt; ii. Ph_3P=$CHCO_2Et$, CH_2Cl_2, rt, 48 h; iii. DIBAL, CH_2Cl_2, -78 °C; iv. MnO_2, pentane, rt. f) i. **7**, Bu_2BOTf, Et_3N, CH_2Cl_2, -78 °C; ii. MPMOC(=NH)CCl_3, 0.03 mol % TfOH, rt, Et_2O; iii. LiOH·H_2O, THF:H_2O = 3:1, rt, 12 h; iv. CH_2N_2, Et_2O. g) i. DIBAL, CH_2Cl_2, -78 °C; ii. 3,4-Dihydro-2*H*-pyran, 10 mol % PPTS, CH_2Cl_2, rt; iii. TBAF, THF; iv. Ac_2O, pyridine; v. 10 mol % PPTS, EtOH, 50 °C, 20 h; vi. Dess-Martin periodinane, CH_2Cl_2, rt; vii. $(MeO)_2P(O)Me$, BuLi, -78 °C, **16**:**17** = 58:42. h) BzCl, pyridine, CH_2Cl_2, rt, 4 h, 91%. i) i. Dess-Martin periodinane, CH_2Cl_2, rt; ii. DIPEA, LiCl, **13**, CH_3CN, rt, 50 h; iii. NaHTe, EtOH, rt, 6.0 h; **19** 49%, **20** 77% in 3 steps. j) DIBAL, CH_2Cl_2, -78 °C. k) i. Ac_2O, pyridine, CH_2Cl_2; ii. DDQ, rt, CH_2Cl_2:H_2O = 20:1; iii. TBAF (1.0 M in THF), AcOH (0.5 *eq* to TBAF), **22a** 30%, **22b** 22% in 3 steps. l) Dess-Martin periodinane, CH_2Cl_2, rt.

Scheme 2

Synthesis of 7*R*,9*S*,12*S*,13*S*,17*S*,18*R*-**3** is shown in Scheme 2 as a representative example. Other diastereoisomers were synthesized in a similar manner. Dienal **9** was synthesized stereoselectively using the cuprate method,[8] then coupled with the dimethylpentane **10** to give dienol **12**.[4a] To simplify NMR analysis, the C5-epimers were separated and **12a**[9] was used for further reactions. The stereochemistry at C13 was set by coupling of Evans' chiral oxazolidone **7** with enal **14**.[6] After conversion of **16** and **17** to ketones **19** and **20**,[10] the carbonyl at C12 was reduced with DIBAL to give an epimeric mixture of alcohols **21**. These epimers could be separated after conversion to the diols **22a** and **22b**.[11] Final oxidation of **22b** using the Dess-Martin reagent afforded the desired tautomycetin degradation product **3**.

As described below, ^{1}H-NMR of the various synthetic diastereoisomers of **3** clearly shows the general characteristics of the relative stereochemistry of each pair of stereocenters. The relative stereochemistry at C12, 13 positions had the noticeable effect on the ^{1}H-NMR spectra. The chemical shifts of the olefinic proton at C16 and the methine proton at C12 of the 12, 13-*anti*-isomers appear at 6.87-6.92 and 5.07-5.10 ppm respectively, whereas those of 12, 13-*syn*-isomers appear at 6.75-6.81 and 5.16-5.17 ppm. These facts strongly suggest that natural tautomycetin has the 12, 13-*anti* configuration as is found in **2**. The C6-methylene protons of the 7,9-*syn* isomer are found at *ca.* 2.18 and 2.43 ppm, whereas those of the 7,9-*anti* isomers are found at *ca.* 2.28 and 2.38 ppm, suggesting that natural **1** has the 7,9-*anti* configuration. The methine protons of C17 and C18 in the 17,18-*syn* isomer appear at *ca.* 2.53 and 4.93 ppm, while those of the 17,18-*anti* isomer appear at *ca.* 2.58 and 4.90 ppm respectively. These observations suggest that the natural tautomycetin has the 7,9-*anti*, 12,13-*anti*, 17,18-*syn* configuration. Fortunately, it has not been necessary to synthesize all possible 32 diastereomers. The four possible 7,9-*anti*-12,13-*anti*-17,18-*syn*-diastereoisomers were synthesized in diastereomerically pure form, and their spectral data was carefully compared with those of **3** derived from natural **1**. The ^{1}H- and ^{13}C-NMR, and IR spectra of the 7*R*,9*S*,12*S*,13*S*,17*S*,18*R*-isomer were found to be completely superimposable on those of the natural product derivative **3**. ^{1}H-NMR spectra of both synthetic and natural product-derived **3** are shown in Figure 1. The sign of the $[\alpha]_D^{26}$ of synthetic **3** [+ 36.6° (*c* 0.24, $CHCl_3$)] was also same as that reported [+ 46.2° (*c* 2.0, $CHCl_3$)][1b] indicating that the absolute stereochemistry of the synthetic **3** is same as the natural product.[12]

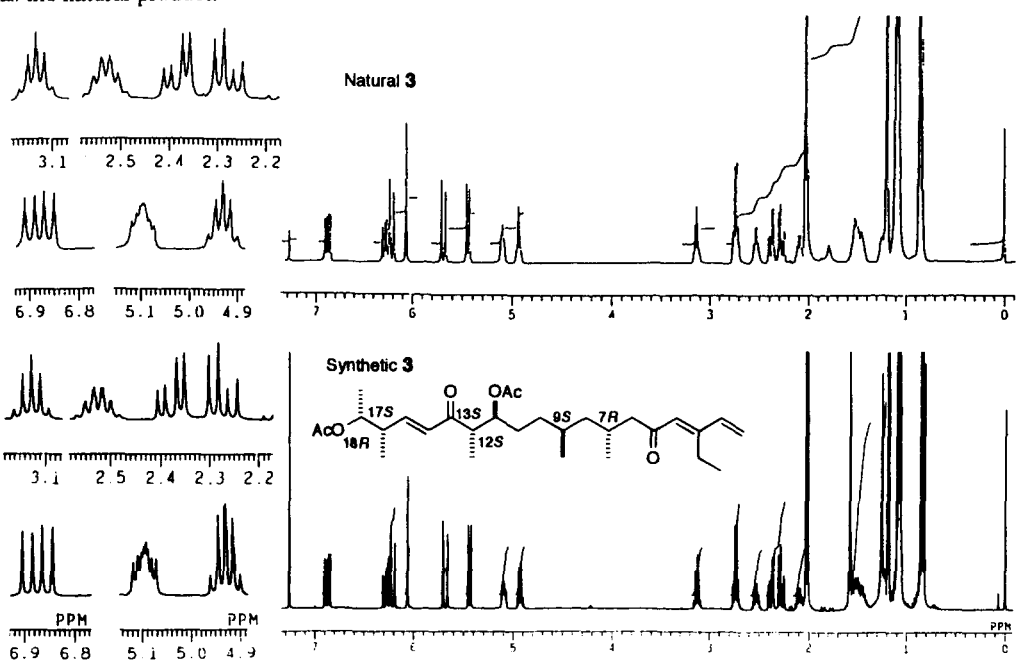


Figure 1

In conclusion, we have assigned the stereochemistries of tautomycetin (**1**) to be 7*R*,9*S*,12*S*,13*S*,17*S*,18*R* through the synthesis of **3**. The structural information described here will be

crucial for the total synthesis of **1**, and the configuration of C16 may be readily determined on completion of this synthesis. The total synthesis of tautomycetin **1** will pave the way for further biological studies, and work along this line is currently underway.

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References and Notes

(1) (a) Chen, X.-C.; Kihara, T.; Ying, X.; Uramoto, M.; Osada, H.; Kusakabe, H.; Wang, B.-N.; Kobayashi, Y.; Ko, K.; Yamaguchi, I.; Shen, Y.-C.; Isono, K. *J. Antibiotics.* **1989**, *42*, 141. (b) Chen, X.-C.; Ubukata, M.; Isono, K. *J. Antibiotics.* **1990**, *43*, 890.

(2) Sakuda, S.; Miki, K.; Kitaoka, S.; Reugjitchachawaly, M.; Yamada, Y. *Biosci. Biotech. Biochem.* **1995**, *59*, 133.

(3) (a) Chen, X.-C.; Kihara, T.; Kusakabe, H.; Magae, J.; Kobayashi, Y.; Fang, R.-P.; Ni, Z.-F.; Shen, Y.-C.; Ko, K.; Yamaguchi, I.; Isono, K. *J. Antibiotics.* **1987**, *40*, 907. (b) Chen, X.-C.; Ubukata, M.; Isono, K. *J. Antibiotics.* **1990**, *43*, 809. For other biological and synthetic studies of tautomycin, see: (c) Oikawa, M.; Ueno, T.; Oikawa, H.; Ichikawa, A. *J. Org. Chem.* **1995**, *60*, 5048, (d) Ichikawa, Y.; Tsuboi, K.; Jiang, Y.; Naganawa, A.; Isobe, M. *Tetrahedron Lett.* **1995**, *36*, 7101. (e) Nakamura, S.; Shibasaki, M. *Tetrahedron Lett.* **1994**, *35*, 4145, and references cited therein.

(4) For preparation of **8a-d**, see: (a) Romo, D.; Johnson, D. D.; Plamondon, L.; Miwa, T.; Schreiber, S. L. *J. Org. Chem.* **1992**, *57*, 5060. (b) Evans, D. A.; Morrissey, M. M.; Dow, R. L. *Tetrahedron Lett.* **1985**, *26*, 6005. (c) Johnson, M. R.; Kishi, Y. *Tetrahedron Lett.* **1979**, *20*, 4343.

(5) For preparation of **6a,b**: i) LDA, MeI; ii) BPSCl (**6a**) or TBSCl (**6b**), imidazole, DMF, 65% (**6a**), 85% (**6b**) in 2 steps; iii) DIBAL, CH_2Cl_2, -78 °C, 82-88%. For stereoselective methylation, see: (a) Brown, J. M. *Angew. Chem. Int. Ed. Engl.* **1987**, *26*, 190. The compound **6c,d** was prepared from **6a** and **6b**: i) MPMOC(=NH)CCl_3, PPTS, CH_2Cl_2, 90%; ii) TBAF, THF, 86%; iii) MsCl, NEt_3, CH_2Cl_2, iv) CsOAc, 18-crown-6, benzene, 60 °C, 90% in 2 steps; v) NaOH, MeOH-H_2O; TBSCl, imidazole, DMF, 96% in 2 steps; vi) DDQ, CH_2Cl_2-H_2O (20:1), 83%. For inversion of the β-hydroxy group, see (b) Torisawa, Y.; Okabe, H.; Ikegami, S. *Chem. Lett.* **1984**, 1555. (c) Huffman, J. W.; Desai, R. C. *Synth. Commun.* **1983**, *13*, 553. (d) Kruizinga, W. H.; Strijtveen, B.; Kellog, R. M. *J. Org. Chem.* **1981**, *46*, 4321.

(6) (a) Evans, D. A.; Bartroli, J.; Shih, T. L. *J. Am. Chem. Soc.* **1981**, *103*, 2127. (b) Evans, D. A.; Dow R. L. *Tetrahedron Lett.* **1986**, *27*, 1007. (c) Evans, D. A.; Polniaszek, R. P. *Tetrahedron Lett.* **1986**, *27*, 5683. (d) Hayashi, K.; Hamada, Y.; Shioiri, T. *Tetrahedron Lett.* **1991**, *32*, 7287.

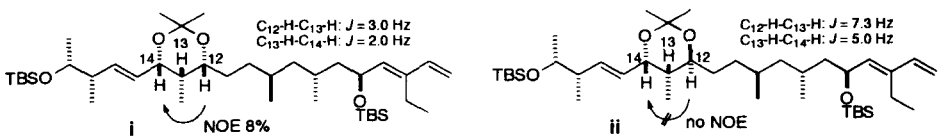
(7) Synthesized diastereomers: *7R,9S,12S,13S,17S,18R*; *7R,9S,12R,13S,17S,18R*; *7R,9S,12S,13S,17R,18S*; *7R,9S,12R,13S,17R,18S*; *7S,9R,12S,13S,17S,18R*; *7S,9R,12R,13S,17S,18R*; *7S,9R,12S,13S,17R,18S*; *7S,9R,12R,13S,17R,18S*; *7R,9R,12S/R,13S,17S,18R*; *7S,9R,12S/R,13S,17S,18S*; *7S,9R,12S/R,13S,17R,18R*.

(8) (a) Corey, E. J.; Katzenellenbogen, J. A. *J. Am. Chem. Soc.* **1969**, *91*, 1851. (b) Siddall, J. B.; Biskup, M.; Fried, J. H. *J. Am. Chem. Soc.* **1969**, *91*, 1853. (c) Bowlus, S. B.; Katzenellenbogen, J. A. *J. Org. Chem.* **1973**, *38*, 2733. (d) House, H. O.; Chu, C.-Y.; Wilkins, J. M.; Umen, M. J. *J. Org. Chem.* **1975**, *40*, 1460. (e) Piers, E.; Chong, J. M.; Morton, H. E. *Tetrahedron* **1989**, *45*, 363.

(9) The stereochemistry at the C5 position of **12a** was tentatively assigned as *S* because reduction of the corresponding C5-ketone with (*S*)-BINOL-$LiAlH_4$ reagent gave **12a** as a major diastereoisomer. See: Noyori, R.; Tomino, I.; Yamada, M.; Nishizawa, M. *J. Am. Chem. Soc.* **1984**, *106*, 6717.

(10) (a) Blanchette, M. A.; Choy, W.; Davis, J. T. *Tetrahedron Lett.* **1984**, *25*, 2183. (b) Yamashita, M.; Kato, Y.; Suemitsu, R. *Chem. Lett.* **1980**, 847.

(11) The stereochemistry at the C12 position was determined by NMR analysis including NOE experiments of the acetonide **i** and **ii** derived from **22a** and **22b** respectively (i. K_2CO_3, MeOH-H_2O, rt, 72%; ii. $(CH_3)_2C(OCH_3)_2$, cat. PPTS, CH_2Cl_2, rt; iii. TBSCl, imidazole, DMF, rt, 40%, 2 steps).



(12) Purification of the final product **3** was further performed by using preparative chiral phase HPLC (Daicel Chiralcel OD, hexane:2-PrOH = 60:1) to remove a trace amount of impurities. The homogeneity of this sample was confirmed by analytical HPLC. Optical rotations of other stereoisomers are as follows: *7R,9S,12S,13S,17R,18S*: $[\alpha]_D^{24}$ + 19.8° (*c* 0.065, $CHCl_3$); *7S,9R,12S,13S,17S,18R*: $[\alpha]_D^{24}$ + 16.5° (*c* 0.164, $CHCl_3$); *7S,9R,12S,13S,17R,18S*: $[\alpha]_D^{29}$ + 10.0° (*c* 0.059, $CHCl_3$).

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