H), 0.56 (s, 3 H), 1.07 (d, 3 H, J = 6.3 Hz), 0.9-2.7 (m, 10 H), 1.99 (s, 6 H), 3.44 (q, 1 H, J = 6.3 Hz), 4.4-5.0 (m, 2 H), 5.1-5.8 (m, 1 H), 6.6-7.3 (m, 4 H).

Preparation of 6β-Methylestra-1,3,5(10)-trien-17-one (52d). Compound **50d** (663 mg, 1.72 mmol) (diastereoisomeric mixture) was quaternized with methyl iodide to produce ammonium iodide **51d**: ^{1}H NMR ($CDCl_3$, Me_4Si as an external reference) δ 0.15 and 0.28 (two s, 9 H), 0.82 and 0.90 (two s, 3 H), 1.2-3.2 (m, 13 H), 3.42 (br s, 9 H), 4.8-6.3 (m, 4 H), 6.9-7.6 (m, 4 H).

Thus obtained ammonium salt **51d** was treated with cesium fluoride (540 mg, 3.55 mmol) as in the preparation of 4-methoxyestra-1,3,5-(10)-trien-17-one (**52c**). The crude reaction product was subjected to preparative TLC on silica gel with a 1:1 chloroform-benzene solvent to give 440 mg (95%) of **52d** (R_f 0.46) as a pale yellow solid, which contained 7-8% of the C(9) epimer: mp 89.5-91.5 °C (recrystallized from ethyl acetate); IR (KBr disk) 1739, 768 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 0.84 (s, 3 H), 1.25 (d, 3 H, J = 6.6 Hz), 1.1-3.2 (m, 14 H), 6.9-7.3 (m, 4 H) (a small singlet at δ 0.89 may be assigned to 18-methyl protons of the C(9) epimer); ^{13}C NMR ($CDCl_3$) δ 13.60, 21.32, 24.29, 25.15, 31.35, 31.71, 33.06, 35.48, 44.29, 47.75, 50.13, 124.62, 125.65 (2C), 128.44, 139.27, 141.52, 220.05, and trace signals (7-8%) from the C(9) epimer at δ 13.38, 18.36, 21.59, 27.84, 30.04, 34.36, 34.81, 47.26, 49.96, 123.18, 138.91, 141.74, 143.68, 220.32. Anal. Calcd for $C_{19}H_{24}O$: C, 85.03; H, 9.01. Found: C, 84.77; H, 9.20.

Synthetic Studies in the Indole Series. Preparation of the Unique Antibiotic Alkaloid Chuangxinmycin by a Nitro Group Displacement Reaction

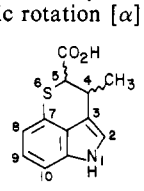
Alan P. Kozikowski,*† Michael N. Greco,† and James P. Springer‡

Contribution from the Department of Chemistry, University of Pittsburgh, Pittsburgh, Pennsylvania 15260, and the Merck Sharp & Dohme Research Laboratories, Rahway, New Jersey 07065. *Received May 17, 1982*

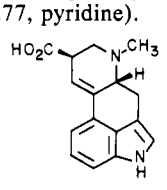
Abstract: The total synthesis of the unique sulfur-containing antibiotic indole alkaloid chuangxinmycin is described. This compound, first isolated by Chinese chemists at the Institute of Materia Medica, was assembled from 2,6-dinitrotoluene by a scheme that combines the nitro group displacement reaction with the Leimgruber indole synthesis to produce a 4-sulfur-substituted indole. Further transformations involving acetylation of the indole 3-position, an intramolecular Knoevenagel condensation to dehydrochuangxinmycin methyl ester, and a stereospecific hydrogenation reaction furnish chuangxinmycin methyl ester. The synthesis scheme does establish the cis relationship between the carboxylic acid and methyl group in the natural product.

A new species of microorganism *Actinoplanes tsinanensis* n. sp. was obtained by the Mainland Chinese from a soil sample collected in Tsinan, Shantung Province. In culture filtrates of this organism, an antibiotic was secreted that could be isolated through a process that began with adjustment of the pH to 3 and extraction with butyl acetate. The organic layer was then extracted with sodium hydroxide, the pH again brought to 3, and the butyl acetate extraction repeated. A secondary sodium hydroxide extraction and pH adjustment then led to deposition of the crystalline product. This material, named chuangxinmycin (a new kind of mycin), was found to be active in vitro against a number of Gram-positive and Gram-negative bacteria. In vivo, the product was found to be active in mice against *Escherichia coli* and *Shigella dysenteria* infections. Preliminary clinical results (140 cases) have shown that chuangxinmycin is effective in the treatment of septicaemia, urinary, and biliary infections caused by *E. coli*.[1]

The structure of the antibiotic was assigned through examination of its UV, IR, NMR, and mass spectral characteristics. The gross structure **1** reveals a unique heterocyclic skeleton bearing two centers of asymmetry. The product is optically active with a specific rotation $[\alpha]^{18}_D$ −249° (*c* 0.77, pyridine).



Chuangxinmycin (1)



Lysergic acid (2)

Chuangxinmycin is related in a structural sense to alkaloids of the ergot family (e.g., lysergic acid (**2**)). These pharmacologically important substances are also comprised of an indole unit that is specifically substituted at its 3- and 4-positions.[2] In the ergot alkaloids, however, the C-4 appendage is linked through a carbon atom rather than a sulfur atom. As a consequence of the gross structural similarity, tactics and/or methods that might thus be developed in the construction of chuangxinmycin in the laboratory could well be carried over to the development of synthetic routes to the ergot alkaloids.[3]

In selecting a synthetic route to chuangxinmycin, we took cognizance of the fact that the early work of the Chinese had failed to discern whether the carboxyl group and methyl group of the product were related in a cis or trans manner.[1] It thus seemed prudent to select a reaction scheme wherein one of these compounds, either the cis or the trans material, could be produced stereoselectively, thus allowing an unambiguous assignment of structure to be made.

While a variety of possible strategies for the assembly of this material could be envisioned on the basis of a single-stage retrosynthetic analysis, we felt that pathway B appeared to be the most promising from a stereochemical standpoint. If we consider X = O in **4**, then C ring closure by an internal Knoevenagel condensation should afford dehydrochuangxinmycin.[4] This intermediate might then be converted by a stereospecific cis hydrogenation reaction to that chuangxinmycin isomer with the methyl and carboxyl groups cis related.[5] If this cis material failed

† University of Pittsburgh.
‡ Merck Sharp & Dohme Research Laboratories.

(1) Liang, H.-T.; Hsu, H.-D.; Chang, C.-P.; Ku, H.-E.; Wang, W.-S. *Hua Hsueh Hsueh Pao* **1976**, *34*, 129; *Chem. Abstr.* **1977**, *87*, 165948z; *Sci. Sin. (Engl. Ed.)* **1977**, *20*, 106; *Chem. Abstr.* **1977**, *87*, 98565g.
(2) Berde, B.; Schild, H. O., Eds. "Ergot Alkaloids and Related Compounds"; Springer-Verlag: Berlin, 1978.
(3) Kozikowski, A. P. *Heterocycles* **1981**, *16*, 267. For a preliminary account of the chuangxinmycin work, see: Kozikowski, A. P.; Greco, M. N. *J. Am. Chem. Soc.* **1980**, *102*, 1165.
(4) Johnson, J. R. *Org. React. (N.Y.)* **1942**, *1*, 210. Jones, G. *Ibid.* **1967**, *15*, 204.

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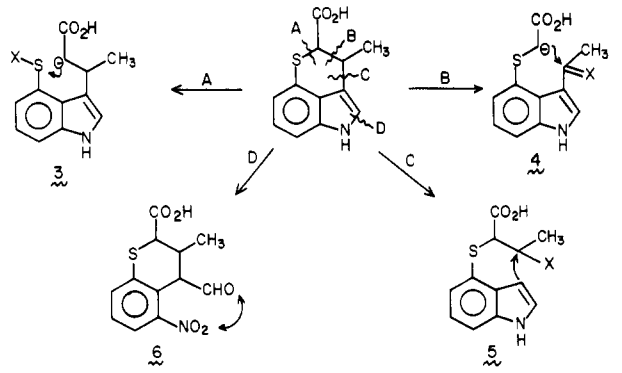
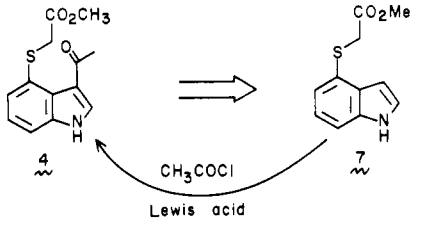


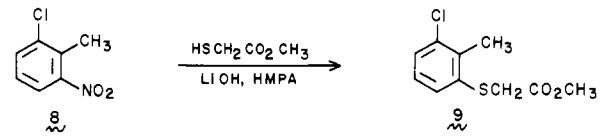
to match the natural product, then an epimerization experiment would be in order. Preparation of intermediate **4** (X = O) became the initial objective of our studies.

Intermediate **4** could, of course, be prepared by a simple Friedel–Crafts acetylation of the corresponding 4-substituted indole **7**.[6] In deciding on an approach to **7**, a search of the literature



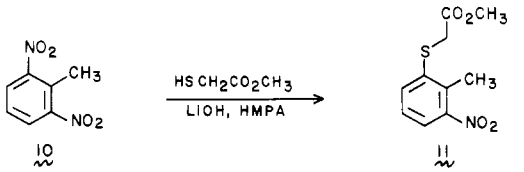
revealed that a 4-sulfur-substituted indole had in fact been prepared more than 20 years ago. E. Piers and colleagues reported a preparation of 4-mercaptoindole by the Reissert condensation of 6-(benzylthio)-2-nitrotoluene.[7] The 6-(benzylthio)-2-nitrotoluene was prepared in 26% yield by displacement of the halogen in 2-bromo-6-nitrotoluene with potassium benzylmercaptide. The nitrotoluene was then treated with potassium ethoxide and diethyl oxalate to furnish the potassium enolate of ethyl (3-(benzylthio)-2-nitrophenyl)pyruvate. A hot solution of the salt in dilute ammonium hydroxide was added to a boiling suspension of ferrous hydroxide to effect reduction with cyclization to the indole-2-carboxylic acid. Decarboxylation[8] and cleavage of the benzyl group (Na, NH_3) led to 4-mercaptoindole.

We sought to prepare **7** in an analogous fashion. An attempt was made to displace the chloro group of 2-chloro-6-nitrotoluene (2,6-CNT) with the anion of methyl thioglycolate. The chloro compound rather than the bromide was used since the former was available commercially. The reaction was conducted by treatment



of a solution of the chloronitrotoluene and methyl thioglycolate in hexamethylphophoric triamide portionwise with powdered lithium hydroxide. Much to our initial dismay, while a displacement reaction did occur readily, as evidenced by TLC analysis, it was the nitro group and not the chlorine atom that was displaced, as ascertained from mass spectral analysis of the product.

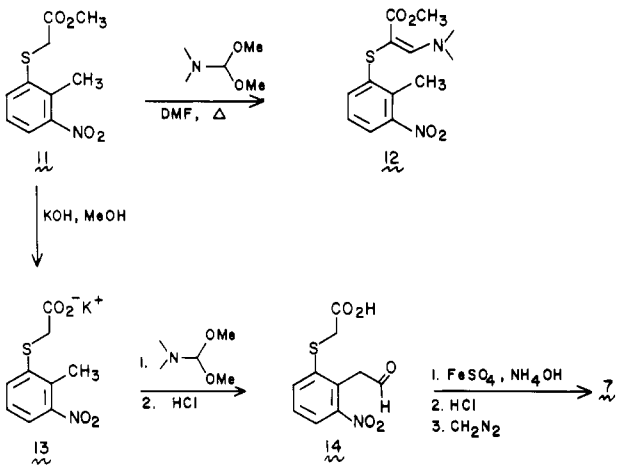
This result in itself suggested that one might possibly employ 2,6-dinitrotoluene (2,6-DNT) in the initial displacement reaction. Thus, the experiment described above was repeated with 2,6-DNT substituted for the 2,6-CNT.



Powdered lithium hydroxide was added over a 1-h period to a solution of methyl thioglycolate and the dinitrotoluene in HMPA at 0 °C. After 1 day at room temperature, the reaction mixture was poured into water and the crude isolated product chromatographed to furnish a new product in 70% yield, which proved to be the desired thioether **11** by spectral analysis.

Subsequent to our findings, an entire review article authored by Beck on the nucleophilic displacement of aromatic nitro groups appeared, which put our results into proper perspective.[9] Direct displacement of aromatic nitro groups proved to be a fairly well-studied phenomenon and a method of broad utility for heterocycle synthesis.[10] With benzenethiol anion as the nucleophile, Bartoli and Todesco had, in fact, shown previously that the relative rate of nitro displacement in 1-X-2,4-dinitrobenzenes was 2000 times that of chlorine and 40 times that of fluorine. The transformation of **8** to **9** was thus in accord with this rate data.[11]

To transform **11** to the 4-substituted-indole **7**, we chose to use the Leimgruber modification of the Reissert indole synthesis.[12] Accordingly, the ester **11** was heated with *N,N*-dimethylformamide dimethyl acetal[13] at reflux with removal of methanol. NMR



analysis of the crude enamine revealed that the condensation had occurred not at the methyl group as desired but rather at the methylene group between the acidifying ester and phenylthio substituents. To properly direct the regiochemical course of the condensation reaction, it was necessary to deactivate the methylene site by transforming the ester to its potassium carboxylate **13** (KOH, MeOH).

DMF acetal condensation in DMF (120 °C, 20 h) now occurred with the proper regioselectivity, as evidenced by ^1H NMR analysis of the aldehyde acid obtained when the crude enamine was stirred

(5) Alder, K.; Roth, W. *Chem. Ber.* **1954**, *87*, 161. Siegel, S.; Smith, G. V. *J. Am. Chem. Soc.* **1960**, *82*, 6082, 6087. Brown, C. A. *Ibid.* **1969**, *91*, 5901.

(6) Sundberg, R. J. "The Chemistry of Indoles"; Academic Press: New York, 1970.

(7) Piers, E.; Haarstad, V. B.; Cushley, R. J.; Brown, R. K. *Can. J. Chem.* **1962**, *40*, 511.

(8) Piers, E.; Brown, R. K. *Can. J. Chem.* **1962**, *40*, 559.

(9) Beck, J. R. *Tetrahedron* **1978**, *34*, 2057.

(10) The displacement of the nitro group in *m*-dinitrobenzene by several nucleophiles had also been described previously: Kornblum, N.; Cheng, L.; Kerber, R. C.; Kestner, M. M.; Newton, B. N.; Pinnick, H. W.; Smith, R. G.; Wade, P. A. *J. Org. Chem.* **1976**, *41*, 1560.

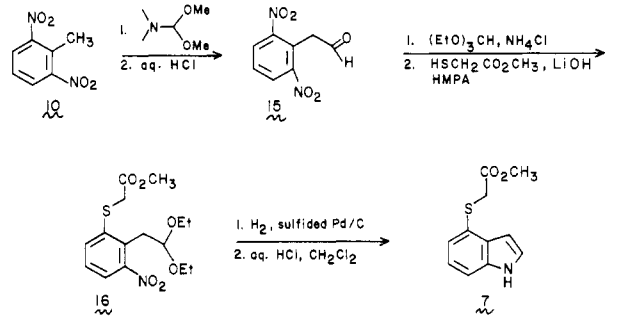
(11) Bartoli, G.; Todesco, P. E. *Acc. Chem. Res.* **1977**, *10*, 125.

(12) Leimgruber, W.; Batcho, A. D. Third International Congress of Heterocyclic Chemistry, Japan, August 23–27, 1971; U.S. Patent 3 976 639, 1971. Maehr, H.; Smallheer, J. M. *J. Org. Chem.* **1981**, *46*, 1752. Garcia, E. E.; Fryer, R. I. *J. Heterocycl. Chem.* **1974**, *11*, 219. Bennington, F.; Morin, R. D.; Bradley, R. J. *Ibid.* **1976**, *13*, 749. Gassman, P. G.; Schenk, W. N. *J. Org. Chem.* **1977**, *42*, 3240.

(13) For an excellent review on the chemistry of formamide acetals, see: Abdulla, R. F.; Brinkmeyer, R. S. *Tetrahedron* **1979**, *35*, 1675.

with cold 2 N HCl. This crude aldehyde acid was subjected directly to nitro group reduction with ferrous sulfate/ammonium hydroxide.[14] The crude indole acid was then treated with diazomethane in ether to furnish, after silica gel chromatography, the indole ester **7** (43% overall from **11**).

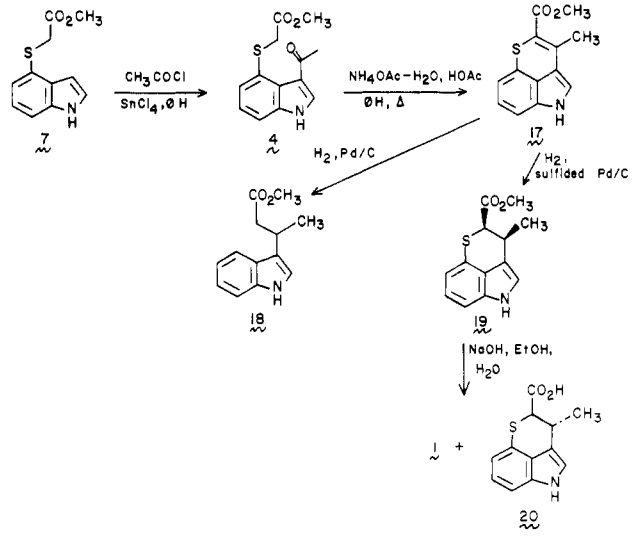
As an alternate route to **7**, we examined a scheme in which the methyl group of 2,6-DNT was functionalized first and then the nitro group displacement carried out. Accordingly, **10** was treated with DMF acetal in DMF (125 °C, 24 h) and the condensation product hydrolyzed to (2,6-dinitrophenyl)acetaldehyde (mp 117–118 °C). The aldehyde group was protected as its diethyl



acetal (triethyl orthoformate, ammonium chloride),[15] and the nitro group displacement reaction with methyl thioglycolate was carried out in a manner similar to the experiment described above. The remaining nitro group of the thioether diethyl acetal **16** was reduced to an amine with a sulfided palladium catalyst at 40 psi of hydrogen. The crude reduction product was refluxed in a two-phase mixture consisting of dichloromethane and aqueous 6 N HCl. Deprotection of the aldehyde group took place with concurrent cyclization to furnish the indole ester **7**.

The overall yield for this scheme is approximately 9–10%, and it is hence less efficient than the first route we had developed.

To complete the chuangxinmycin synthesis, it was now necessary to acetylate the C-3 position of the sulfur-substituted indole. Acylation was effected in a straightforward fashion by treatment of a cold benzene solution of **7** with acetyl chloride and anhydrous stannic chloride and then stirring the mixture at 0° for 20 min and at room temperature for 2 h.[16] In this fashion, the 3,4-di-



substituted-indole **4** was obtained in 92% yield. After **4** was refluxed in benzene in the presence of ammonium acetate monohydrate and acetic acid for 15 h, a 94% yield of the brilliant yellow product dehydrochuangxinmycin methyl ester was obtained.[4]

While the chemistry up to this point proceeded fairly rapidly, the last stage of the synthesis proved rather difficult. Reduction of a heavily substituted double bond that was moreover a vinyl sulfide was required.[17] As anticipated, attempts to effect saturation by hydrogenation over catalysts such as palladium on charcoal led to rapid desulfurization with formation of the 3-substituted-indole **18**. A system consisting of $NaBH_4$–$NiCl_2$ was equally effective in promoting this transformation.[18] A host of other reducing agents including stannous chloride/hydrochloric acid, zinc/acetic acid, borane, and diimide were also examined and found to give none of the desired reduction product. *Only* when the poisoned catalyst sulfided palladium on charcoal[19] was employed did reduction take place at 70 psi of hydrogen pressure to produce a single new product in good yield. The ^1H NMR of this synthetic ester was identical with the ^1H NMR of chuangxinmycin methyl ester prepared by diazomethane treatment of natural chuangxinmycin. Since double-bond isomerization during this hydrogenation reaction did not appear likely,[20] we reasoned that the hydrogenation reaction would most likely deliver the product with carbomethoxy and methyl in a cis stereochemical relationship. Thus, the natural acid must also possess the cis relationship between these functional groups.

The stereochemical assignment derived further support from the small coupling constant (4 Hz, δ_{H_5} = 4.08) present for the vicinal protons of the C ring. Additionally, when the methyl ester of chuangxinmycin was converted to the acid by aqueous sodium hydroxide/ethanol treatment, a second new product was isolated, which was assigned the trans stereochemistry (δ_{H_5} = 3.76, J_{vic} = 6 Hz).[21] While epimerization does take place during the basic hydrolysis, the pure *cis*-**1** is readily separated from the trans-product **20** by fractional crystallization. An X-ray analysis of the methyl ester **19** has confirmed independently the cis relationship of the vicinal substituents.[22,23]

The total synthesis of chuangxinmycin has been completed through a scheme that establishes the relative stereochemical relationship of the C-ring vicinal substituents. The nitro group displacement reaction when combined with the Leimgruber indole synthesis does appear to provide a fairly general strategy for the construction of 4-substituted indoles.

In closing, we note that the Chinese research group at the Institute of Materia Medica has also succeeded in the total synthesis of chuangxinmycin.[24] Their route is somewhat similar to our own in that it too passes through a dehydrochuangxinmycin intermediate. In brief, they prepared 4-bromoindole from *o*-nitrotoluene by a sequence of reactions involving bromination, isomer separation, diethyl oxalate condensation, reduction, and decarboxylation (a typical Reissert indole synthesis). The 4-bromoindole was thence converted to its *N*-magnesium iodide, and the organometallic reacted with acetyl chloride to furnish the

(14) Blaikie, K. G.; Perkin, W. H. *J. Chem. Soc.* **1924**, 296. Rydon, H. N.; Tweedle, J. C. *Ibid.* **1955**, 3499.

(15) Claisen, L. *Chem. Ber.* **1893**, *26*, 2729; *Liebigs Ann. Chem.* **1897**, *297*, 76.

(16) Degraw, J. I.; Kennedy, J. G.; Skinner, W. A. *J. Heterocycl. Chem.* **1966**, *3*, 9.

(17) Vinyl sulfides have been reduced successfully with high catalyst loadings. See, for example: Markgraf, J. H.; Hess, B. A.; Nichols, C. W.; King, R. W. *J. Org. Chem.* **1964**, *29*, 1499. Schneider, H. J.; Bagnell, J. J.; Murdock, G. C. *Ibid.* **1961**, *26*, 1987. Parham, W. E.; Groen, S. H. *Ibid.* **1964**, *29*, 2214.

(18) Satoh, D.; Hashimoto, T. *Chem. Pharm. Bull.* **1976**, *24*, 1950.

(19) The catalyst is available commercially from Engelhard Industries.

(20) Many cases have been documented where the net result of catalytic hydrogenation is not the expected cis addition of two hydrogen atoms. See, for example: Sauvage, J. F.; Baker, R. H.; Hussey, A. S. *J. Am. Chem. Soc.* **1961**, *83*, 3874.

(21) The use of *n*-PrSLi in HMPA (Bartlett, P. A.; Johnson, W. S. *Tetrahedron Lett.* **1970**, 4459) for ester cleavage does also result in some epimerization.

(22) The X-ray structure, atomic coordinates, temperature parameters, bond distances, and bond angles are available as supplementary material.

(23) An X-ray structure determination has also been carried out by the Chinese: Hsu, H.-C.; Shao, M.-C.; Chang, C.-Y.; Li, K.-P.; Chou, K.-C.; Tang, Y.-C. *K'o Hsueh T'ung Pao* **1980**, *25*, 350; *Chem. Abstr.* **1980**, *93*, 2678w.

(24) Chang, C.-P.; Hsu, H.-D.; Huang, L.-C.; Lin, Y.-C.; Li, H.-S.; Yu, C.-L.; Chao, C.-L. *Acta Chim. Sin* **1976**, *34*, 133; *Chem. Abstr.* **1978**, *88*, 62309h.

4-bromo-1,3-diacetylindole. Exposure of this compound to the cuprous salt of ethyl mercaptoacetate in a mixture of quinoline and pyridine at 170–180 °C produced the ethyl ester of 1-acetyldehydrochuangxinmycin. The ester was reduced in a nonstereospecific fashion with stannous chloride in a hot mixture of hydrochloric acid and acetic acid and the ester group hydrolyzed with sodium hydroxide to produce racemic chuangximycin on fractional crystallization. The racemic material showed 50% of the antibacterial activity associated with the natural product.

Since the fermentation route to chuangxinmycin can be difficult to control, the development of a good industrial-scale synthesis of the natural product does appear valuable.[25] In both of the present syntheses, the reduction step can be problematic and alternative stereoselective strategies for completing the C ring should be sought.

Experimental Section

Analytical thin-layer chromatography was performed on Brinkman MN polygram 0.25-mm silica gel plastic plates with G/UV_{254} inorganic phosphar fluorescent indicator or E. Merck 0.25-mm silica gel glass plates with PF_{254} indicator. Gravity column chromatography was performed on E. Merck 0.063–0.200-mm silica gel with distilled reagent-grade solvents. High-pressure liquid chromatography was carried out on a Waters Associates instrument with a μ-porasil column.

When required, solvents and reagents were dried prior to use. Dimethylformamide, hexamethylphsophoric triamide, and benzene were distilled from calcium hydride.

Methyl ((2-Methyl-3-nitrophenyl)thio)acetate (11). To a solution of 12 g (67 mmol) of 2,6-dinitrotoluene and 8.53 g (80 mmol) of freshly distilled methyl thioglycolate in 15 mL of HMPA was added 1.9 g (80 mmol) of anhydrous lithium hydroxide via a solid addition tube over 1 h at 0 °C. The dark red mixture was stirred at room temperature for 1 day. The reaction mixture was poured into water and extracted three times with diethyl ether. The organic extracts were dried ($MgSO_4$) and concentrated by rotary evaporation. The crude product was chromatographed on silica gel with 10% ethyl acetate/hexanes as eluent to yield 11.3 g (70%) of **11**: mp 46–47 °C (methylene chloride/petroleum ether); IR ($CHCl_3$) 1736 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.53 (s, 3 H), 3.65 (s, 2 H) 3.70 (s, 3 H), 7.07–7.75 (m, 3 H).

Anal. Calcd for $C_{10}H_{11}NO_4S$: C, 49.78; H, 4.61; N, 5.80. Found: C, 49.50; H, 4.71; N, 5.58.

Methyl (4-Indolylthio)acetate (7). A solution of the thioether **11** (2.1 g, 9 mmol) and potassium hydroxide (0.4 g, 9 mmol) in 5 mL of methanol was stirred overnight under an argon atmosphere at room temperature. The resulting precipitate was collected by filtration and washed several times with hexanes to yield 2.4 g (100%) of the crude potassium salt. After thorough drying, the potassium salt was dissolved in a mixture of 10 mL of DMF and 3.2 g (27 mmol) of dimethylformamide dimethyl acetal contained in a two-necked, round-bottomed flask equipped with a Vigreux column and solvent take-off head. The reaction mixture was refluxed for 20 h with removal of methanol. On cooling, the dark colored mixture was poured into 20 mL of ice cold 2 N HCl, and the crude aldehyde–acid extracted with diethyl ether (3×). After drying ($MgSO_4$) and concentration, the acid was dissolved in 20 mL of a 1:1 mixture of concentrated ammonium hydroxide and water. This solution was added to 7.0 g (46 mmol) of ferrous sulfate dissolved in 4.3 mL of concentrated ammonium hydroxide and 20 mL of water. The mixture was refluxed for 50 min, then cooled, neutralized with concentrated HCl, and extracted with ethyl acetate (3×). The extracts were dried and concentrated in vacuo to furnish the crude acid, which was immediately esterified by reaction with excess diazomethane in ether. The crude product was chromatographed on silica gel with 25% ethyl acetate/hexanes as eluent to furnish 855 mg (43% overall) of the indole ester **7**: mp 83–85 °C (benzene/petroleum ether); IR ($CHCl_3$) 1730 cm^{-1}; ^1H NMR ($CDCl_3$) δ 3.67 (s, 3 H), 3.70 (s, 2 H), 6.65 (m, 1 H), 7.20 (m, 4 H), 8.33 (br s, 1 H).

Anal. Calcd for $C_{11}N_{11}NO_2S$: C, 59.89; H, 5.05; N, 6.30. Found: C, 59.71; H, 5.01; N, 6.33.

(2,6-Dinitrophenyl)acetaldehyde (15). A 100-mL, two-necked, round-bottomed flask fitted with Vigreux column and solvent take-off head was charged with 6.4 g (35 mmol) of 2,6-dinitrotoluene, 12.5 g (105 mmol) of *N,N*-dimethylformamide dimethyl acetal, and 26 mL of dry DMF. The mixture was heated under argon at 125 °C for 24 h with removal of methanol. The mixture was cooled, poured into ice cold 20% HCl, and extracted repeatedly with ether. The extracts were dried ($MgSO_4$) and concentrated, and the residue was chromatographed on silica gel with 50% ethyl acetate/hexanes to yield 3.8 g (51%) of **15**: mp 117–118 °C (ethyl acetate/hexanes); IR ($CHCl_3$) 2836, 2732, 1731 cm^{-1}; ^1H NMR ($CDCl_3$) δ 4.27 (s, 2 H), 7.17–8.18 (m, 3 H), 9.65 (s, 1 H); mass spectrum (15 eV), m/e = 165.

(2,6-Dinitrophenyl)acetaldehyde Diethyl Acetal. Aldehyde **15** (3.0 g, 14.3 mmol), triethyl orthoformate (2.54 g, 17.2 mmol), and ammonium chloride (40 mg, 0.81 mmol) in 25 mL of ethanol were refluxed for 1 day. The reaction mixture was cooled, filtered, and concentrated. The residue was chromatographed on silica gel with 25% ethyl acetate/hexanes as eluent to provide 3.7 g (93%) of the diethyl acetal of **15**: mp 75–76 °C (ethyl acetate/hexanes); IR ($CHCl_3$) 1120, 1060 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.10 (t, 6 H, J = 7 Hz), 3.37–3.63 (m, 6 H), 4.60 (t, 1 H, J = 5 Hz), 7.22–7.92 (m, 3 H).

Exact mass calcd for $C_{12}H_{16}N_2O_6 - OC_2H_5$: 239.0668. Found: 239.0669.

Methyl [(2-(2,2-Diethoxyethyl)-3-nitrophenyl)thio]acetate (16). Powdered lithium hydroxide (220 mg, 9.2 mmol) was added from a solid addition tube over a 1-h period to a solution of methyl thioglycolate (974 mg, 9.2 mmol) and the diethyl acetal of **15** (2.4 g, 8.3 mmol) in 100 mL of HMPA cooled to 0 °C. The resulting red solution was stirred at room temperature for 1 day, poured into water, and extracted repeatedly with ether. The combined extracts were dried ($MgSO_4$) and concentrated. The crude product was chromatographed on silica gel with 20% ethyl acetate/hexanes as eluent to yield 1.82 g (63%) of **16**: IR ($CHCl_3$) 1729 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.11 (t, 6 H, J = 7 Hz), 3.38–3.72 (m, 6 H), 3.65 (s, 2 H), 3.68 (s, 3 H), 4.55 (t, 1 H, J = 4.75 Hz), 7.22–7.69 (m, 3 H); mass spectrum (15 eV), m/e = 298 ($M^+ - OC_2H_5$).

Methyl (4-Indolylthio)acetate (7) from 16. A solution of the diethyl acetal **16** (50 mg, 0.15 mmol) in 2 mL of ethyl acetate was hydrogenated at 40 psi of hydrogen pressure (Parr shaker) over 318 mg of 5% Pd/C sulfided catalyst for 3 h. Filtration and concentration of the hydrogenation mixture gave an oil, which was refluxed in a two-phase mixture of 1 mL of dichloromethane and 0.1 mL of 6 N HCl for 2 h. Preparative TLC of the crude isolated product on silica gel with 25% ethyl acetate/hexanes as solvent gave 10 mg (31%) of the indole ester **7**: mp 83–84 °C; spectral data as above.

Methyl ((3-Acetyl-4-indolyl)thio)acetate (4). As a suspension of the indole ester **7** (158 mg, 0.71 mmol) in 2 mL of dry benzene was cooled slowly to 0 °C, acetyl chloride (93 mg, 1.2 mmol) was added dropwise. A solution of anhydrous stannic chloride (316 mg, 1.2 mmol) in 2 mL of benzene was then added. The reaction mixture was stirred at 0 °C for 20 min and at room temperature for 2 h. The mixture was quenched with water and extracted with ethyl acetate (2×). The extracts were dried ($MgSO_4$) and concentrated, and the residue was chromatographed on silica gel with 50% ethyl acetate/hexanes as eluent to afford 172 mg (92%) of **4**: mp 140–141.5 °C (benzene); IR ($CHCl_3$) 3474, 1725, 1647 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.48 (s, 3 H), 3.63 (s, 3 H), 3.68 (s, 2 H), 7.06–7.23 (m, 4 H), 7.78 (d, 1 H, J = 3 Hz).

Exact mass calcd for $C_{13}H_{13}NO_3S$: 263.0616. Found: 263.0623.

Dehydrochuangxinmycin Methyl Ester (17). A mixture of **4** (108 mg, 0.41 mmol), $NH_4OAc \cdot H_2O$ (144 mg, 1.87 mmol), and acetic acid (227 mg, 3.8 mmol) in 8 mL of benzene was refluxed under an argon atmosphere for 15 h. The reaction mixture was cooled, poured into 10 mL of water, and extracted with ethyl acetate (3×). The extracts were dried ($MgSO_4$) and concentrated, and the residue was chromatographed on silica gel with 25% ethyl acetate/hexanes as eluent to yield 94 mg (94%) of **17** as a yellow solid: mp 167–168 °C (benzene); IR ($CHCl_3$) 1710 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.30 (s, 3 H), 3.77 (s, 3 H), 6.40–6.92 (m, 4 H), 7.92 (br s, 1 H).

Exact mass calcd for $C_{13}H_{11}NO_2S$: 245.0511. Found: 245.0510.

Chuangxinmycin Methyl Ester (19). A solution of dehydrochuangxinmycin methyl ester (25 mg, 0.1 mmol) in 3 mL of ethyl acetate was hydrogenated over 350 mg of sulfided 5% palladium on carbon at 70 psi in a Parr shaker for 72 h. The hydrogenation mixture was filtered through Celite. The filter cake was stirred with ethyl acetate for several hours and the mixture filtered. The combined filtrates were concentrated by rotary evaporation, and the residue was purified by HPLC on a μ-porasil column with 25% ethyl acetate/hexanes as eluent to yield 10 mg (40%) of analytically pure chuangxinmycin methyl ester: mp 145–146 °C (petroleum ether/methylene chloride); IR ($CHCl_3$) 1730 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.20 (d, 3 H, J = 7 Hz), 3.64 (overlapping m, 1 H and s, 3 H), 4.08 (d, 1 H , J = 4 Hz), 6.76–7.12 (m, 4 H), 7.88 (br s, 1 H).

Exact mass calcd for $C_{13}H_{13}NO_2S$: 247.0667. Found: 247.0666.

Chuangxinmycin (1). A solution of chuangxinmycin methyl ester (13 mg, 0.053 mmol), sodium hydroxide (26 mg, 0.65 mmol), 0.5 mL of water, and 0.7 mL of ethanol was stirred at room temperature for 2 h and let stand overnight. The solution was concentrated under reduced pressure and extracted with ether (2×). The aqueous layer was overlayed with ether and acidified with 10% HCl to pH 1. The ether layer was

(25) Zhang, Z.-P., personal communication, 1981.

separated, the aqueous layer was extracted with additional ether, and the combined ethereal extracts were dried ($MgSO_4$) and concentrated. The crude product was crystallized from petroleum ether/methylene chloride to yield 4 mg (40%) of chuangxinmycin: mp 145-145.5 °C; ^1H NMR ($CDCl_3$) δ 1.36 (d, 3 H J = 7 Hz), 3.80 (d of q, 1 H, J = 7, 3.5 Hz), 4.30 (d, 1 H, J = 3.5 Hz), 6.9-7.3 (m, 4 H), 8.6 (br s, 1 H).

Exact mass calcd for $C_{12}H_{11}NO_2S$: 233.0511. Found: 233.0507.

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Registry No. *cis*-(-)-**1**, 63339-68-4; **4**, 83561-15-3; **7**, 73363-65-2; **10**, 606-20-2; **11**, 73363-64-1; **13**, 83561-16-4; **15**, 78283-22-4; **15** diethyl acetal, 83561-17-5; **16**, 83561-18-6; **17**, 73363-63-0; **19**, 83602-19-1; **20**, 83602-20-4; $HSCH_2CO_2CH_3$, 2365-48-2; *N,N*-dimethylformamide dimethyl acetal, 4637-24-5; triethyl orthoformate, 122-51-0.

Supplementary Material Available: X-ray structure and listings of atomic coordinates, temperature parameters, bond distances, and bond angles (6 pages). Ordering information is given on any current masthead page.

Structure and Conformation of the Antiviral Nucleoside 2′-Fluoro-5-iodoarabinosylcytosine (FIAC). The Gauche Effect in Nucleosides[1]

George I. Birnbaum,*[2a] Miroslaw Cygler,[2a] Kyoichi A. Watanabe,[2b] and Jack J. Fox[2b]

Contribution from the Department of Biological Sciences, National Research Council of Canada, Ottawa, Ontario, Canada K1A 0R6, and the Sloan-Kettering Institute for Cancer Research, New York, New York 10021. *Received April 15, 1982*

Abstract: The three-dimensional structure of 2′-fluoro-5-iodoarabinosylcytosine (FIAC), an inhibitor of herpes simplex and herpes zoster viruses, was determined by X-ray crystallography. The crystals belong to the monoclinic space group $P2_1$, and the cell dimensions are a = 4.747 (2), b = 14.017 (2), and c = 18.514 (3) Å, β = 90.28°. Intensity data were measured with a diffractometer, and the structure was solved by the heavy-atom method. Least-squares refinement, which included the coordinates of the hydrogen atoms, converged at R = 4.8%. In the asymmetric unit there are two crystallographically independent molecules of FIAC that are related by a pseudo-2-fold screw axis. The conformation about the glycosidic bond is anti, χ_{CN} having the rather low value of 19.1°. Contrary to expectations based on the substituent effect, the furanose ring adopts the C(3′) endo-C(2′) exo (type N) pucker. The influence of electronegative substituents on the ring conformation is being discussed in terms of the gauche effect. The $-CH_2OH$ side chain is disordered, giving rise to equal populations of the gauche$^+$ and trans rotamers.

The antiviral activities of some arabinonucleosides as well as of some 5-substituted 2′-deoxyribonucleosides have been known for several years.[3] Recently, Watanabe et al.[4] synthesized a new series of variously 5-substituted 2′-deoxy-2′-fluoroarabinofuranosylpyrimidines, several of which exhibited marked antiherpetic activity. One of these, 1-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-5-iodocytosine (also called 2′-fluoro-5-iodoarabinosylcytosine or FIAC) was found[4,5] to be especially capable of suppressing the replication of various strains of herpes simplex virus types 1 and 2, as well as of herpes zoster and cytomegalovirus. Continuing our structural studies of chemotherapeutic nucleosides,[6] we decided to carry out an X-ray analysis of FIAC. In particular, we wished to compare the conformation in the solid state with that determined in solution[7] and with the conformations of other arabinonucleosides. The effect of electronegative ring substituents on the conformation of the furanose ring in nucleosides has been the subject of some discussion in recent literature.[8-10] Since no fluoroarabinonucleoside has ever been subjected to an X-ray analysis, the crystal structure determination of FIAC was considered to be particularly valuable.

Experimental Section

2′-Fluoro-5-iodoarabinosylcytosine (FIAC), $C_9H_{11}N_3O_4FI$, was prepared as described by Watanabe et al.[4] and crystallized from methanol. Systematic absences and the symmetry of reflection intensities on precession photographs indicated the orthorhombic space group $P2_12_12_1$. All crystals in our possession were rather mosaic (0.8-0.9°); nevertheless we collected data with molybdenum radiation on a CAD-4 diffractometer. The cell dimensions were as follows: a = 4.742 (4), b = 14.012 (5), and c = 18.504 (6) Å. The angles α, β, and γ were 90°, within experimental error (±0.2°). The data were corrected for Lorentz and polarization factors and for absorption. The structure was determined by the heavy-atom method and refined by block-diagonal least squares with anisotropic temperature parameters for non-hydrogen atoms. The refinement converged at R = 0.069 (R' = 0.077) for 1464 observed re-

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(2) (a) National Research Council. (b) Sloan-Kettering Institute for Cancer Research.
(3) For a recent review, see: De Clercq, E.; Descamps, J.; Verhelst, G.; Walker, R. T.; Jones, A. S.; Torrence, P. F.; Shugar, D. *J. Infect. Dis.* **1980**, *141*, 563-574.
(4) Watanabe, K. A.; Reichman, A.; Hirota, K.; Lopez, C.; Fox, J. J. *J. Med. Chem.* **1979**, *22*, 21-24.
(5) (a) Lopez, C.; Watanabe, K. A.; Fox, J. J. *Antimicrob. Agents Chemother.* **1980**, *17*, 803-806. (b) Fox, J. J.; Lopez, C.; Watanabe, K. A. In "Medicinal Chemistry Advances"; De las Heras, F. G.; Vega, S., Eds.; Pergamon Press: New York, 1981; pp 27-40.
(6) (a) Birnbaum, G. I.; Lin, T.-S.; Shiau, G. T.; Prusoff, W. H. *J. Am. Chem. Soc.* **1979**, *101*, 3353-3358. (b) Birnbaum, G. I.; Deslauriers, R.; Lin, T.-S.; Shiau, G. T.; Prusoff, W. H. *Ibid.* **1980**, *102*, 4236-4241. (c) Birnbaum, G. I.; Watanabe, K. A.; Fox, J. J. *Can. J. Chem.* **1980**, *58*, 1633-1638. (d) Birnbaum, G. I.; Cygler, M.; Kusmierek, J. T.; Shugar, D. *Biochem. Biophys. Res. Commun.* **1981**, *103*, 968-974.
(7) Lipnick, R. L.; Fissekis, J. D. *Biochim. Biophys. Acta* **1980**, *608*, 96-102.
(8) Uesugi, S.; Miki, H.; Ikehara, M.; Iwahashi, H.; Kyogoku, Y. *Tetrahedron Lett.* **1979**, 4073-4076.
(9) Klimke, G.; Cuno, I.; Lüdemann, H.-D.; Mengel, R.; Robins, M. J. *Z. Naturforsch., C: Biosci.* **1980**, *35C*, 853-864.
(10) (a) Guschlbauer, W.; Jankowski, K. *Nucleic Acids Res.* **1980**, *8*, 1421-1433. (b) Haertlé, T.; Wohlrab, F.; Guschlbauer, W. *Eur. J. Biochem.* **1979**, *102*, 223-230.

0002-7863/82/1504-7626$01.25/0