Table I. Deuterium Isotope Effects[a,b] on ^{13}C Chemical Shifts of Ortho Carbons and C_{ipso}–C_{ortho} π-Bond Orders[c] in Compounds 1-5

compd	C	isotope shift	bond	π-bond order
1	C-4	−16	C-3/C-4	0.432
2	C-3	−13	C-2/C-3	0.496
3	C-3	−6	C-2/C-3	0.530
4	C-2	+1.6[d]	C-1/C-2	0.667
3	C-1	+15	C-1/C-2	0.782
2	C-1	+18	C-1/C-2	0.806
1	C-2	+20	C-2/C-3	0.859
5	C-2	−6	C-1/C-2	0.782
5	C-8a	+28	C-1/C-8a	0.509

[a] Values given in ppb (0.001 ppm); positive sign denotes deshielding in the deuterated compound. [b] Measured at 100.6 MHz of mixtures of the protium and deuterium analogues; digital resolution better than 0.5 ppb; error estimate ±2 ppb. [c] From INDO molecular orbital calculations (QCPE program No. 141) of benzene, naphthalene, anthracene and furan; experimental geometries used as input. [d] From ref 8.

the para carbons, respectively. These effects and related ones in α-deuterated carbonyl compounds[9] and carbocations[10,11] have been ascribed, at least in part, to diminished hyperconjugative electron release by the C–D relative to the C–H bonds.[5,8-12] Isotope shifts of the ortho carbons in (α-deuteroalkyl)benzenes are explained[8] as resulting from a superposition of the normal ("inductive"[10]) and the hyperconjugative effects.

Isotope effects on NMR chemical shifts would be even more useful if a better understanding could be achieved of their sizes as functions of molecular parameters. We therefore investigated the influence of benzylic deuterons on ortho carbon chemical shifts in compounds **1-4**, in which there is a large variation of double-bond character between C_{ipso} and C_{ortho} but a very similar steric environment of the trideuteromethyl groups. If (hyper)conjugation to some degree influences the sizes of the isotope shifts over three bonds, $^3\Delta$, then one would expect them to depend on the π-bond order, P_π, of the C_{ipso}–C_{ortho} bonds in the same way as regular ortho substituent chemical shifts (SCS) vary with P_π.[13] Our results (Table I) show that for compounds **1-4** the $^3\Delta$ values correlate very strongly with the π-bond order. A least-squares treatment (Figure 1) yields a good linear dependence (correlation coefficient 0.993; root mean square error 1.94 ppb) of $^3\Delta$ on $P_\pi(C_{ipso}–C_{ortho})$:

$$^3\Delta \text{ (ppb)} = 88.4P_\pi - 54.9 \quad (1)$$

According to eq 1, low double-bond character involves shielding and high double-bond character involves deshielding isotope shifts over three bonds. This finding fully supports the idea[8] of a superposition of "normal" (negative) and hyperconjugative (positive) contributions to this type of isotope shift. For π bond orders near $^2/_3$ (benzene derivatives) the opposing contributions tend to cancel each other.

Figure 1 also demonstrates that the values of the vicinal isotope shifts in 1-(methyl-d_3)naphthalene (**5**) do not fit the above correlation at all. Since in **5** the CD_3 group is in relatively close contact with the C-8–H-8 bond, this suggests that steric interactions cause substantial alterations of the magnitude of isotope shifts. We have shown previously[5] that nonbonded interactions between deuterons and carbon atoms cause appreciable "through space" isotope shifts. Possible complications by steric factors have thus to be taken into account when trying to predict isotope shifts by means of eq 1.

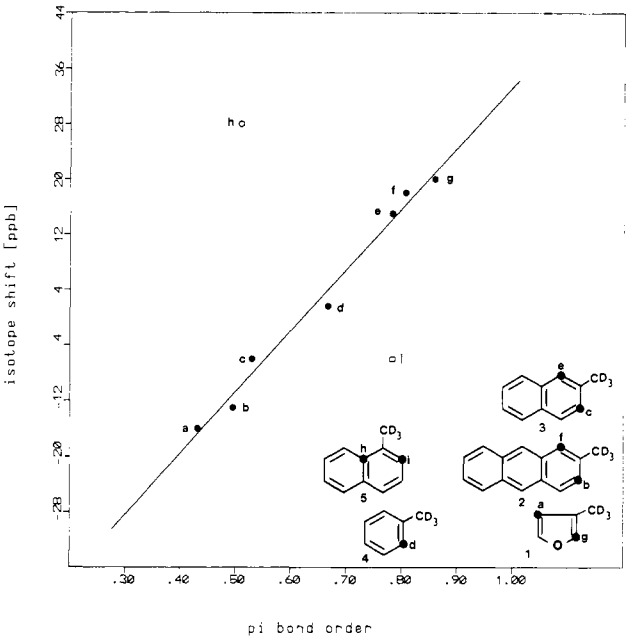

Figure 1. Correlation between the deuterium isotope effects on vicinal carbon chemical shifts and the C_{ipso}–C_{ortho} π bond order in compounds **1-5**. Only points a to g (filled circles) were considered in the calculation of the least-squares line.

Acknowledgment. Support of part of this work by the Fonds der Chemischen Industrie is gratefully acknowledged. We thank Norbert Krause and Hartwig Drögemüller for preparing some of the deuterated compounds.

Registry No. **1**, 930-27-8; **2**, 613-12-7; **3**, 91-57-6; **4**, 108-88-3; **5**, 90-12-0.

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(9) Maciel, G. E.; Ellis, P. D.; Hofer, D. C. *J. Chem. Phys.* **1967**, *71*, 2160-2164.
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(14) Other isotope shifts [ppb]: **1**, −141 (C-3); **2**, −102 (C-2), +13 (C-4a); **3**, −104 (C-2), +12 (C-4a), −5 (C-4), −6 (C-7); **5**, −91 (C-1), −7 (C-4a), −6 (C-5), −7 (C-7), +5 (C-8); one-bond isotope shifts were not determined. Isotope shifts not mentioned were not resolved.

Electron Transfer at Crystallographically Known Long Distances (25 Å) in [Zn^{II},Fe^{III}] Hybrid Hemoglobin

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In this report we describe the first example of long-range electron transfer[1-3] between chromophores that are rigidly held at a fixed *and* crystallographically known distance and orientation. We employ mixed-metal, [M,Fe], hybrid hemoglobins that have both chains of one type, either α or β, substituted with a closed shell zinc(II) protoporphyrin (ZnP), whereas the two chains of the other type contain a ferriheme (Fe^{III}P): [Zn^{II},Fe^{III}].[4] In the

(1) For general reviews of long-range electron transfer, see ref 2. Elegant experiments with goals paralleling those here have been reported more recently.[3]
(2) (a) DeVault, D. *Q. Rev. Biphys.* **1980**, *13*, 387-564. (b) Chance, B. et al., Eds. "Tunneling in Biological Systems"; New York, 1979. (c) See also: Sutin, N. *Acc. Chem. Res.* **1982**, *15*, 275-283.
(3) (a) Winkler, J. R.; Nocera, D. G.; Yocam, K. M.; Bordignon, E.; Gray, H. B. *J. Am. Chem. Soc.* **1982**, *104*, 5798-5800. (b) Isied, S. S.; Worosila, G.; Atherton, S. J. *Ibid.* **1982**, *104*, 7659-7661. (c) Calcaterra, L. T.; Closs, G. L.; Miller, J. R. *Ibid.* **1983**, *105*, 670-671.

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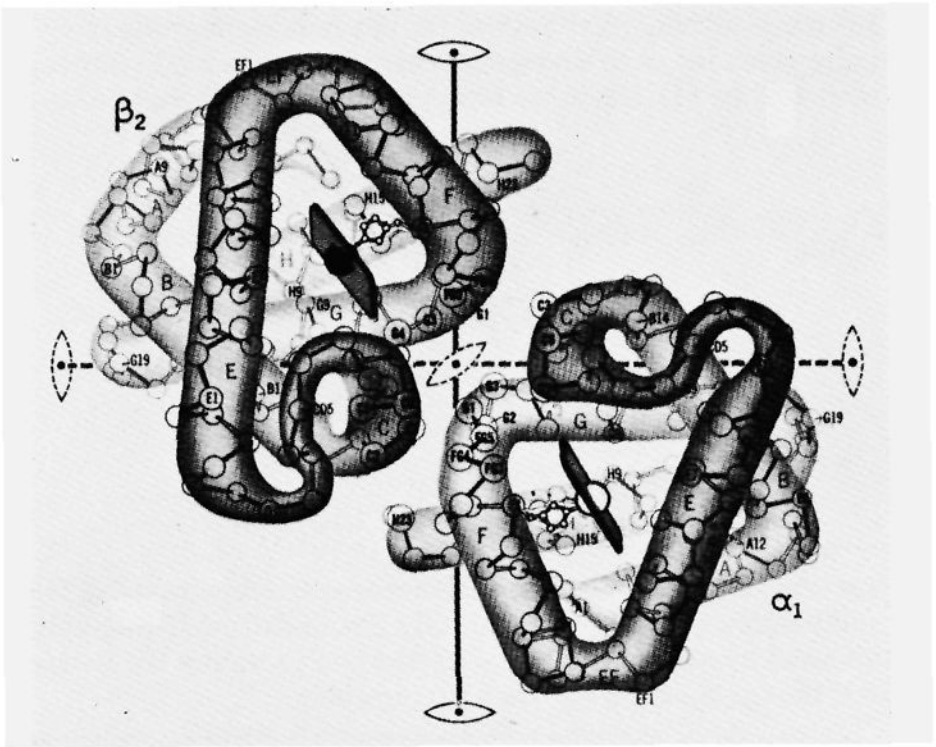

Figure 1. Representation of the (α_1^{Fe},β_2^{Zn}) subunit pair, electron-transfer complex, within [α^{Fe},β^{Zn}] hybrid hemoglobin in the deoxy-Hb (T state) quaternary structure. The distance between Fe (○) and Zn (●) atoms is 24.7 Å.[5] Adapted from ref 5b.

deoxy (T) hemoglobin structure,[5] normally adopted by these hybrids,[4d] the prosthetic groups of closest approach are α_1^{Fe}–β_2^{Zn} (or the reverse substitution; Figure 1). The ZnP and FeP planes are roughly parallel, as in a hypothetical model for the electron-transfer complex between cytochrome *c* and cytochrome *c* peroxidase.[6] The chromophores are separated by two heme pocket walls, one from each chain; the metal–metal distance is 25 Å, and the shortest distance between atoms of the rings is ca. 20 Å. Since the α_1^{Fe}–β_1^{Zn} distances are much longer (35 Å), for present purposes we can treat the molecule as two independent electron-transfer complexes, each composed of an α_1–β_2 subunit pair.

Electron transfer within the α_1–β_2 complex[7] is initiated by flash photoexcitation, which forms the long-lived (τ_T > 10 ms) ZnP triplet state (^{3}ZnP) with good quantum yield.[8] The ^{3}ZnP is a good reductant ($E_0' \sim -1.1$ V)[9] and can reduce the ferriheme ($E_0' \sim +0.15$ V)[10] by the long-range electron-transfer process

$$^3ZnP + Fe^{III}P \xrightarrow{k_t} (ZnP)^+ + Fe^{II}P \qquad \Delta E_0' \sim +1.25 \text{ V} \quad (1)$$

Figure 2 presents the progress curve for the decay of the ^{3}ZnP produced by flash excitation[11] of hybrids in which electron transfer is blocked because the heme is initially in the high-spin Fe^{II} state,

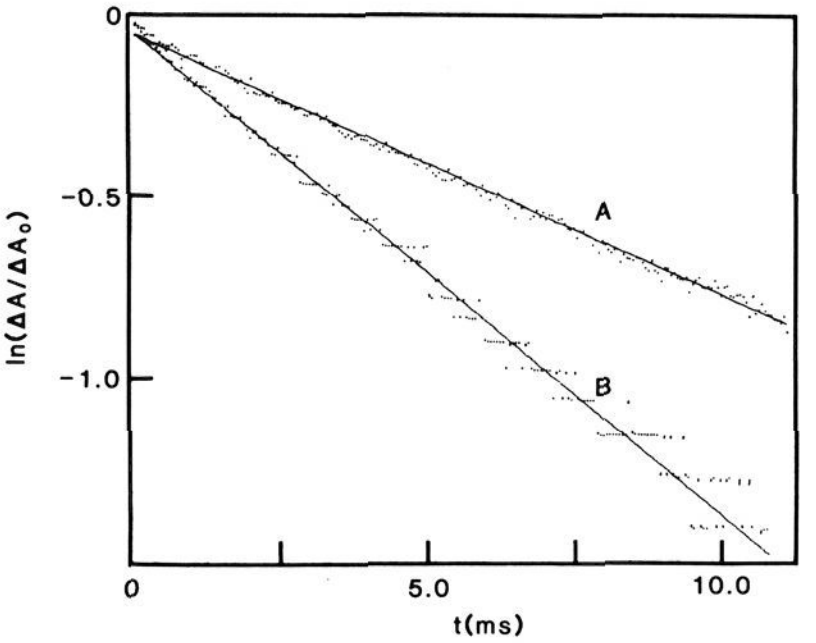

Figure 2. Progress curve of the [^{3}ZnP–ZnP] difference spectrum subsequent to flash photolytic excitation of [α^{Fe},β^{Zn}] hybrid hemoglobins, as monitored at 415 nm, the isosbestic of the [Fe^{III}P–Fe^{II}P] difference spectrum. Conditions: 0.01 M KP_i, pH 7; [inositol hexaphosphate] = 30 μM; room temperature; concentration of tetramer ~3 μM. (A) [$\alpha^{Fe^{II}}$,β^{Zn}]; decay rate, 80 ± 5 s^{-1}. (B) [$\alpha^{Fe^{III}}$,β^{Zn}]; decay rate, 140 ± 20 s^{-1}.

[$\alpha^{Fe^{II}}$,β^{Zn}]. The triplet decay rate, k_D = 80 ± 5 s^{-1}, is comparable to that for ^{3}ZnP in an individual subunit of fully substituted ZnHb,[8] and thus long-range paramagnetic quenching of ^{3}ZnP is negligible. However, when the Fe-containing partner subunit is initially in the aquoferriheme, Fe^{III}, state (Figure 2), the triplet decay rate is almost doubled: k_{obsd} = 140 ± 20 s^{-1}. The triplet decay enhancement is found to be independent of protein concentration and thus, as confirmed by observations of ferriheme reduction (see below), represents the contribution from intramolecular electron transfer within the α_1–β_2 complex. In this case, the observed rate constant is the sum of the intrinsic decay (k_D) and electron-transfer rates: $k_{obsd} = k_D + k_t$; thus, the rate constant for electron transfer at room temperature between a ^{3}ZnP and aquoferriheme rigidly held 25 Å apart within the α_1–β_2 electron-transfer complex is quite substantial: k_t = 60 ± 25 s^{-1}.

The $(ZnP)^+$ π-cation radical product of electron transfer is strongly oxidizing ($E_0' \sim +0.74$ V[9b]), and it rapidly oxidizes its ferroporphyrin partner (rate constant, k_e) in a reaction that mimics, say, the oxidation of a ferroprotein by a peroxidase compounds I.[6,12]

$$(ZnP)^+ + Fe^{II}P \xrightarrow{k_e} ZnP + Fe^{III}P \qquad \Delta E_0' \sim +0.6 \text{ V} \quad (2)$$

When following the Fe^{II}P concentration subsequent to a flash excitation, one observes a small net heme reduction corresponding to ca. 15% of the ^{3}ZnP formed, with the appearance of Fe^{II}P following the observed ^{3}ZnP decay. Reaction 2 thus is not quantitative; $(ZnP)^+$ in the heme pocket of Hb at room temperature in part also undergoes a competing spontaneous reduction (rate constant, k_m) without accompanying ferroheme oxidation, by as yet unidentified (but possibly tyrosyl[13]) protein residues.[14] Analysis of these observations with kinetic equation for the full reaction scheme indicates that $k_m + k_e > k_t$ and $k_e/k_m \sim 2$.

In order to obtain k_e and k_m individually, the measurements were repeated in the presence of high CO concentrations. In this case electron transfer according to eq 2 must compete with CO binding to the ferroheme:

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(5) (a) Fermi, G.; Perutz, M. F. "Haemoglobin and Myoglobin: Atlas of Molecular Structures in Biology"; Phillips, D. C., Richard, F. M., Eds.; Oxford University Press: 1981; Vol. 2. (b) See: Dickerson, R. E.; Geis, I. "The Structure and Action of Proteins"; Harper and Row: New York, 1969. (c) Model of Hb assembled by John Mack for Prof. I. M. Klotz.

(6) Poulos, T. L.; Kraut, J. *J. Biol. Chem.* **1980**, *255*, 10322, 10330.

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(9) (a) $E_0[(ZnP)^+/^3ZnP] = E_0'[(ZnP)^+/ZnP] - E_T$ = +0.74 V (ref 9b) − 1.84 (ref 9c). (b) Kaneko, Y.; Tamura, M.; Yamazaki, I. *Biochemistry* **1980**, *19*, 5795–5799. (c) Stanford, M. A.; Hoffman, B. M. *J. Am. Chem. Soc.* **1981**, *103*, 4104–4114.

(10) For example: Bull, C.; Hoffman, B. M. *Proc. Natl. Acad. Sci. U.S.A.* **1975**, *72*, 3382.

(11) (a) The concentration of ^{3}ZnP subsequent to flash excitation was monitored photometrically at the [Fe^{III}P–Fe^{II}P] isosbestic point, λ 415 nm, or in the presence of CO, at the [Fe^{II}P–(CO)Fe^{II}P] isosbestic, λ 424 nm; the reduction of Fe^{III}P to Fe^{II}P was monitored at the [^{3}ZnP–ZnP] difference spectrum isosbestic, λ 437 nm.[8] The apparatus has been described.[8] (b) Preliminary results for α^{Fe} and β^{Fe} hybrids are similar.

(12) (a) Hewson, W. D.; Hager, L. P. (**1979**) *Porphyrins* **1979**, *7*, 295–332. (b) Dunford, H. B., Dolphin, D., Raymond, K. N., Sieker, L., Eds. "The Biological Chemistry of Iron"; D. Reidel: Dordrecht, The Netherlands, 1982.

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(14) Careful comparison of static optical spectra of [Zn^{II},Fe^{III}] hybrid samples taken before photolysis and after reoxidation subsequent to photolysis shows that the spontaneous reduction of $(ZnP)^+$ to ZnP is not associated with ZnP degradation. The process, which is under investigation, plausibly is analogous to that described in ref 13.

$$Fe^{II}P + CO \xrightarrow{k^{CO}} Fe^{II}P(CO) \qquad (1a)$$

which process would tend to augment the degree of net heme reduction. With the fully T-state hybrid[4d] under 1 atm of CO, CO binding is monophasic, $k^{CO} = 10^2\ s^{-1}$,[5] and the presence of CO does not significantly increase the extent of heme reduction subsequent to ^3ZnP formation. This indicates that $k_e > k^{CO}$, namely, $k_e > 10^2\ s^{-1}$. Such experiments performed with partially R-state tetramer,[4d] for which $k^{CO} = 5 \times 10^4\ s^{-1}$,[15] also show no increase in reduction. Although the response of multiple protein forms make analysis difficult, the observations suggest that the lower bound to k_e may well be 10-fold higher: $k_e > 10^3\ s^{-1}$. In any event, this result for k_e strongly suggests that rapid electron transfer within protein redox complexes such as that of cytochrome *c* and cytochrome *c* peroxidase, in which the heme edge-to-edge separation is thought to be 16 Å, need not involve complicated electron relay mechanisms.[6] Reaction 2 is less exothermic than reaction 1 yet is faster. Perhaps the latter process or both are in the "anomalous" regime, in which increasing the exothermicity actually reduces transfer rates because of the difficulty in dissipating the energy released.[2]

Measurements of temperature dependences, currently in progress, will allow us to separate tunneling from thermally activated contributions to both k_t and k_e; ligation of the ferriheme can be employed to change its redox potential, spin state, and, more generally, the nature of the Franck–Condon overlap between oxidized and reduced heme states.[16] Because these hybrids provide electron-transfer partners of known geometries, it is distinctly possible that the rates will be amenable to detailed analysis with current electron-transfer theories.

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Registry No. ZnP, 15442-64-5; heme, 14875-96-8; CO, 630-08-0.

(15) For further discussions of CO binding to these hybrids, see ref 4b.
(16) We have already observed that k_t varies with ligation of the ferriheme.

Total Synthesis of the Trichothecene Mycotoxin Anguidine

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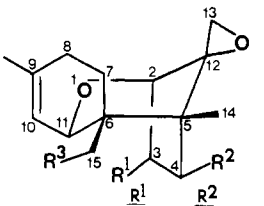
The trichothecene mycotoxins have been the focus of considerable interest due to the wide variety of biological activities they exhibit depending on the respective functional groups present on their unique tricyclic skeleton.[1] A variety of synthetic studies of trichothecenes have been reported.[2] Our interest in elucidating structure–activity relationships for this important group of sesquiterpenes led us to develop an enantioselective total synthesis of anguidine (**1**), which we report herein.[3] We chose to follow



	R^1	R^2	R^3
anguidine (1)	OH	OAc	OAc
trichodermol (2)	H	OH	H
verrucarol (3)	H	OH	OH

a scheme for assembling the trichothecene skeleton involving the addition of an A-ring unit to a fully functionalized C-ring unit, followed by an intramolecular cyclization providing the B-ring and thus completing the tricyclic skeleton.

Our synthesis originates with an asymmetric microbial reduction of 2-allyl-2-methyl-1,3-cyclopentanedione by actively fermenting bakers' yeast (*Saccharomyces cerevisiae*), which effects a stereoselective reduction of one of the two enantiotopic homomorphic carbonyl groups to provide the chiral starting material (2*S*,3*S*)-2-allyl-3-hydroxy-2-methylcyclopentanone, which was further elaborated to the fully functionalized C-ring precursor **4**.[4,5] With a fully functionalized C-ring precursor available, we next addressed the task of stereoselectively introducing the A-ring unit (see Scheme I). Our plan called for a Robinson annelation sequence on the hydroxymethylene **6**. We assumed that this bicyclo[3.2.1] system would add methyl vinyl ketone in a preferred exo manner similar to the model system reported by Roush.[6] The preparation of **6** was achieved by heating **4** with neat tris(dimethylamino)methane,[7] followed by mild acidic hydrolysis of the enamine **5**. The Michael reaction of **6** with methyl vinyl ketone proceeded as expected with exclusive exo addition providing the desired adduct **7**. An intramolecular aldol condensation was accomplished by treating **7** with lithium diisopropylamide at −78 °C and followed by elimination of the corresponding mesylates to give the enone **8**.[8] The A-ring unit was completed by treating **8** with methyllithium at −78 °C to provide the allylic alcohol **9**.[9]

Reduction of the A + C ring unit **9** with lithium aluminum hydride to give the tetraol **10** set the stage for an interesting study of possible intramolecular acid-catalyzed rearrangements. Inspection of molecular models indicated four possible modes of tetrahydropyran (B ring) formation by hydroxyl quenching of an intermediate allylic carbocation. Conformational analysis of these possibilities indicated that unfavorable interactions were minimized in a rearrangement that leads to the trichothecene skeleton. Experimental results of acid-catalyzed rearrangements of similar systems supported this prediction.[10] Treatment of the tetraol **10** with *p*-toluenesulfonic acid in dichloromethane gave a mixture of the desired trichothecene **11** and the unexpected [2.2.2]allyl ether **13**, resulting from cyclization of the C15 hydroxyl group in varying ratios depending on the reaction conditions.[11] Extended

(1) Reviews: (a) Tamm, C *Fortschr. Chem. Org. Naturst.* **1974**, *31*, 63. (b) Bamberg, J. R.; Strong, F. M. In "Microbial Toxins"; Kadis, S., Ed.; Academic Press: New York, 1973; Vol. 3, pp 207–292. (c) Doyle, T. W.; Bradner, W. T. In "Anticancer Agents Based on Natural Product Models"; Cassidy, J. M., Douros, J., Eds.; Academic Press: New York, 1980; Chapter 2. Isolation and structure of anguidine: (d) Dawkins, A. W. *J. Chem. Soc. C* **1966**, 116.

(2) Several total synthesis of trichothecenes have been reported: (a) Colvin, E. W.; Malchenko, S.; Raphael, R. A.; Roberts, J. S. *J. Chem. Soc., Perkin Trans. 1* **1973**, 1989. (b) Still, W. C.; Tasi, M. Y. *J. Am. Chem. Soc.* **1980**, *102*, 3654. (c) Schlessinger, R. H.; Nugent, R. A. *Ibid.* **1982**, *104*, 1116. (d) Kraus, G. A.; Roth, B.; Frazier, K.; Shimagaki, M. *Ibid.* **1982**, *104*, 1114. (e) Trost, B. M.; McDougal, P. G. *Ibid.* **1982**, *104*, 6110. (f) Roush, W. R.; D'Ambra, T. E. *Ibid.* **1983**, *105*, 1058. Other pertinent synthetic studies are referenced in the above reports.

(3) Early portions of this work were presented at the 25th Congress, International Union of Pure and Applied Chemistry, Vancouver, B. C., Canada, Aug 1981. The work described in this communication was presented at the 185th American Chemical Society National Meeting, Seattle, WA, Mar 1983.

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(5) Brooks, D. W.; Grothaus, P. G.; Palmer, J. T. *Tetrahedron Lett.* **1982**, *23*, 4187.

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(7) Martin, S. F.; Moore, D. R. *Tetrahedron Lett.* **1976**, *4459*.

(8) Attempts to effect the aldol condensation under equilibrating conditions resulted in a retro-Michael reaction to give **6**.

(9) Only one isomer seemed to be produced as indicated by ^1H NMR, but the assignment of stereochemistry is tentative.

(10) Refer to the results reported in ref 2c,f.

0002-7863/83/1505-4472$01.50/0