H, H-11 and H-11'), 6.55–7.65 (m, 8 H); ^{13}C NMR δ 144.6 (C-1a), 114.6 (C-1), 129.5 (C-2), 117.5 (C-3), 131.2 (C-4), 122.5 (C-4a), 53.0 (C-4b), 48.5 (C-6), 135.8 (C-6a), 126.0 (C-7), 126.3 (C-8), 126.5 (C-9), 128.4 (C-10), 136.1 (C-10a), 44.7 (C-11), 36.7 (C-10b).

Anal. Calcd for $C_{16}H_{16}N_2$: C, 81.35; H, 6.78; N, 11.86. Found: C, 81.31; H, 6.80; N, 11.84.

Acknowledgment. We thank Mrs. T. Zardin and Mr. H. R. Loosli for helpful discussion concerning the NMR data and Miss C. Fritz for the supply of large quantities of intermediates.

Toxic Trichothecenes from *Fusarium sporotrichioides* (MC-72083)

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The trichothecene mycotoxins are responsible for alimentary toxic aleukia (ATA), vomiting, weight loss, skin inflammation, and death in humans and farm animals from ingestion of *Fusarium* infected grains.[1] Recent efforts in our laboratories aimed at understanding the role secondary metabolites produced by *F. sporotrichioides* (MC-72083) play in the above noted illnesses have led to new relatively nontoxic trichothecenes.[2-4] We now report the structures and bioactivities of two new toxic metabolites: FS-2 and trichotriol, as well as 8-oxo-DAS, trichodiol, and 3-hydroxytrichothecene, a decomposition product of trichotriol.

F. sporotrichioides (MC-72083) was cultured and subjected to chromatographic workup as previously reported.[2,5]

8-Oxo-DAS (*diacetoxyscirpenol*) (**1**), 2 mg (30 ppb), $C_{19}H_{24}O_8$ (*m/z* 380.147, calcd 380.146), exhibited the following fragments in its electron-impact mass spectrum (EI-MS): *m/z* 380 (relative intensity) M^+ (10), 320 (1), 247 (5), 173 (10), 121 (20), and 43 (100). The IR (film) spectrum of **1** indicated that hydroxyl (3439, 1040, cm^{-1}), carbonyl (1739, 1734 cm^{-1}), and ether (1236 cm^{-1}) functionalities were pesent.[6] From the ^1H NMR (300 MHz, $CDCl_3$) data, four methyl groups at δ 0.82 (s, C-14), 1.84 (br s, 16-H, coupled to δ 6.62, 10-H), 2.00 (s, OAc), and 2.16 (s, OAc) were observed, together with a vinyl proton at 6.62 (dd, *J* = 2.9, 5.9 Hz, 10-H) and the characteristic splitting pattern of the 12,13-epoxide methylene protons at δ 2.82 and 3.10 (d, *J* = 3.9 Hz). Complete proton assignments were made by using COSY[7] (Table I) and are in accord with structure **1** as depicted. Comparison of the ^1H NMR data of **1** with that of a synthetic sample[8] confirmed the structure as depicted. The UV (λ_{max} 226 nm, ϵ_{max} 6100, ACN) indicated the presence of an α,β-unsaturated ketone which was suggested from the ^1H NMR and is supported by the ^{13}C NMR (75 MHz, $CDCl_3$) data (δ 136.8 d, C-10; 138.3, s, C-9). The presence of an unsaturated ketone at δ 196.6 (s, C-8) and two acetates at δ 172.6 (s) and 170.1 (s) were also indicated by ^{13}C NMR. Comparison of the NMR data of **1** with that of the closely related diacetoxyscirpenol[9] (DAS) **2** allowed ^{13}C NMR assignments to be made (Table I). This is the first report of naturally occurring 8-oxo-DAS.

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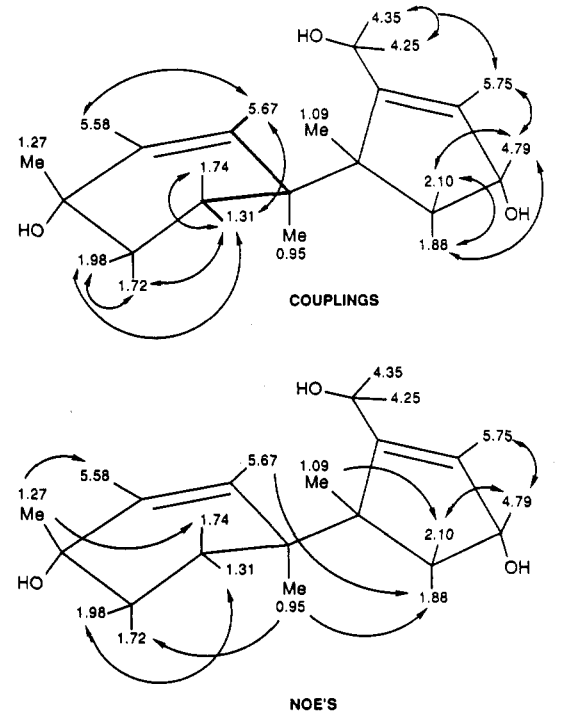

Figure 1. Couplings obtained from COSY and NOE's obtained from DNOES are shown for FS-2 (**3**). The W coupling between 11-H (δ 5.67) and 7-H (1.31) is depicted in bold lines.

FS-2 (**3**), 2 mg (30 ppb), $C_{15}H_{24}O_3$ (*m/z* 252.173, calcd 252.173), exhibited peaks at *m/z* 253 ($M^+ + 1$; 2), 235 ($M^+ - OH$; 50), 127 (85), 125 (90), 109 (127 – H_2O; 100), 107 (125 – H_2O, 85) in the CIMS. The large peaks at *m/z* 125 and 127 correspond to cleavage of the C-5/C-6 bond in **3** and gives rise to resonance stabilized fragments.

The UV (λ_{max} 194.5, ϵ_{max} 12 000, ACN) data was appropriate for two ene $\pi \rightarrow \pi^*$ transitions.[10] From inspection of the ^1H NMR spectrum (300 MHz, $CDCl_3$) three methyl singlets (δ 0.95, 1.09, 1.27), a methylene next to oxygen (δ 4.23, 4.35, J_{AB} = 14.5 Hz), two vinyl protons on an isolated *cis*-1,2-disubstituted ene (δ 5.58, d, *J* = 10.2 Hz; 5.67, dd, *J* = 1.7, 10.2 Hz), and a vinyl proton (δ 5.75, dd, *J* = 1.5, 3.3 Hz) coupled to a methine next to oxygen (δ 4.79, m) were observed. COSY[7] and DNOES (difference nuclear Overhauser effect spectroscopy) allowed completion of the structure and facilitated proton assignments (Table I). Significant couplings (COSY) and NOE's (DNOES) are shown in Figure 1, confirming the relative stereochemistry of **3** as depicted. The absolute stereochemistry is assumed

(1) (a) Ueno, Y. *Trichothecenes-Chemical, Biological and Toxicological Aspects, Developments in Food Science-4*; Elsevier: New York, 1983; Chapters 1, 5, 6. (b) Joffe, A. Z. *Fusarium Species: Their Biology and Toxicology*; Wiley-Interscience: New York, 1986.
(2) Corley, D. G.; Rottinghaus, G. E.; Tempesta, M. S. *Tetrahedron Lett.* **1986**, *27*, 427.
(3) Oltz, E. M.; Nakanishi, K.; Yagen, B.; Corley, D. G.; Rottinghaus, G. E.; Tempesta, M. S. *Tetrahedron* **1986**, *42*, 2615.
(4) Corley, D. G.; Rottinghaus, G. E.; Tracy, J. K.; Tempesta, M. S. *Tetrahedron Lett.* **1986**, *27*, 4133.
(5) Burmeister, H. R. *J. Appl. Microbiol.* **1971**, *21*, 739.
(6) Nakanishi, K.; Solomon, P. H. *Infrared Absorption Spectroscopy*, 2nd ed.; Holden-Day, Inc.: San Francisco, 1977.
(7) Bax, A.; Freeman, R.; Morris, G. *J. Magn. Reson.* **1981**, *42*, 164.
(8) Kaneko, T.; Schmitz, H.; Essery, J. M.; Rose, W.; Howele, H. G.; O'Herron, F. A.; Nachfolger, S.; Huftalen, J.; Bradner, W. T.; Partyka, R. A.; Doyle, T. W.; Davies, J.; Cundliffe, E. *J. Med. Chem.* **1982**, *25*, 579–589.
(9) Cole, R. J.; Cox, R. H. *Handbook of Toxic Fungal Metabolites*; Academic Press: New York, 1981; pp 171–174.
(10) Rao, C. N. R. *Ultra-violet and Visible Spectroscopy*; Butterworth: London, 1961; p 16.

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Table I. ^1H (300 MHz) and ^{13}C (75.5 MHz) NMR Data on 8-Oxo-DAS (1), DAS (2), FS-2 (3), Trichodiol (4), Trichotriol (5), and 3-Hydroxytrichothecene (6)

	1		2		3		4		5		6	
atom	$^1H^a$	$^{13}C^a$	$^1H^a$	$^{13}C^a$	$^1H^a$	$^{13}C^b$	$^1H^a$	$^{13}C^a$	$^1H^a$	$^{13}C^b$	$^1H^a$	$^{13}C^a$
2	3.79 d (5.0)	78.5 d	3.64 d (4.9)	77.4 d	5.75 dd (1.5, 3.3)	133.0 d	4.49 m (br)	68.6	3.75 m (br)	78.1 d	3.51 d (4.4)	80.0
3	4.23 dd (2.9, 5.0)	78.9 d	4.13 dt (4.9, 3.0)	78.6 d	4.79 m	73.4 d	2.06 dd (5, 14) 1.85 m	46.2	4.32 m (br)	69.3 d	4.46 m	69.4
4	5.07 d (2.9)	83.9 d	5.13 d (3.0)	83.8 d	2.10 m	47.4 t	1.45–1.85	45.8	2.15 dd (7, 14) 1.80 dd (6, 14)	42.3 t	2.08 m	41.6
5		48.7 s		48.4 s		54.6 s		n.o.[c]		44.0 s		n.o.
6		47.4 s		43.5 s		40.0 s		n.o.		39.8 s		n.o.
7	2.94 d (15.8)	38.2 t	1.7–2.1	20.7 t	1.75 m	27.8 t	1.46–1.85	27.7	1.73 m	27.8 t	1.42 m	24.6
	2.45 dd (1.5, 15.8)				1.31 m				1.32 m		1.97 m	
8		196.6 s		27.5 t	1.98 m 1.73 m	35.0 t		35.3	1.62 m 1.72 m	34.8 t	1.92 m 2.15 m	28.3
9		138.3 s		140.0 s		65.7 s		n.o.		65.8 s		139.7
10	6.62 dd (2.9, 5.9)	136.8 d	5.49 br d (5.3)	118.1 d	5.58 d (10.2)	135.6 d	5.53 d (10)	133.8	5.57 d (10)	136.0 d	5.49 dd (1.4, 5.6)	119.2
11	4.54 d (5.9)	68.3 d	4.06 br d (5.3)	67.5 d	5.67 dd (1.7, 10.2)	133.2 d	5.47 dd (1.5, 10)	133.5	5.76 br d (10)	132.3 d	4.09 br d (5.6)	72.1
12		64.4s		64.0 s		154.0 s		n.o.		69.9 s		n.o.
13	3.10 d (3.9)	46.7 t	3.01 d (4.0)	46.7 t	4.35 br AB q (14.5)	60.7 t	3.25 d (4)	50.7	3.61 d (4)	50.0 t	3.08 d (4.1)	48.6
	2.82 d (3.9)		2.74 d (4.0)		4.23		2.76 d (4)		2.99 d (4)		2.85 d (4.1)	
14	0.82 s	6.3 q	0.77 s	6.7 q	1.09 s	21.3 q	1.04 s	19.6	1.10 s	20.5 q	0.75 s	11.3
15	4.17 AB q (12.4)	64.5 t	4.12 AB q (12.5)	63.1 t	0.95 s	22.0 q	0.98 s	22.0	0.95 s	21.8 q	0.82 s	16.1
	4.12		3.94									
16	1.84 br s	15.2 q	1.68 br s	22.8 q	1.27 s	30.9 q	1.28 s	29.6	1.30 s	31.3 q	1.73 bd	23.3
$COCH_3$		172.6 s		171.6 s								
		170.1 s		170.2 s								
$COCH_3$	2.16	21.0 q	2.16	20.6 q								
	2.00	20.6 q	2.00									

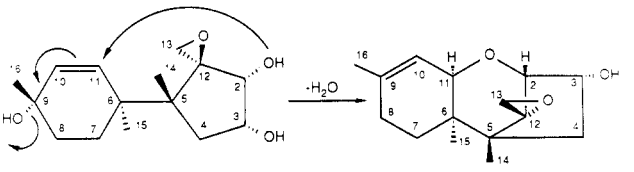
[a] $CDCl_3$. [b] Acetone-d_6. [c] n.o. = signal not observed.

to be the same[11] as all previously isolated trichothecenes. ^{13}C NMR data are in accord with **3** (Table I).

Trichodiol (**4**), 2 mg (30 ppb), was isolated and characterized (except stereochemistry) previously as the acetate from *Trichothecium roseum*.[12] ^1H and ^{13}C NMR data are reported here for the first time (Table I) on the parent compound. ^1H NMR assignments are based on COSY. The quaternary carbons were not observed in ^{13}C NMR due to very small sample size.

Trichotriol (**5**), 5 mg (75 ppb), $C_{15}H_{24}O_4$ ($M^+ - H_2O$, m/z 250.157, calcd 250.157, CIMS), exhibited the following fragments in the fast atom bombardment mass spectrum (FABMS), m/z 269 ($M^+ + H$), 251 ($M^+ - H_2O$, 15), 185 (15), 109 (30), and 43 (100) and also exhibited the following fragments in the EIMS, m/z 125 (20), 107 (125 – H_2O; 40), 43 (100). The IR spectrum[13] (film) indicated that hydroxyls (3367, vs; 1066, 1030 cm^{-1}) were present. The UV (λ_{max} 194.5, ϵ_{max} 4900, ACN) data are consistent with a single ene. The ^1H NMR data indicated that three methyl singlets (δ 0.95, 1.10, 1.30), a characteristic 12,13-epoxide methylene (δ 2.99, 3.61, both d, J = 4 Hz), an isolated *cis*-1,2-disubstituted ene (δ 5.57, d, J = 10 Hz, 5.76, br d, J = 10 Hz), and two methines bearing oxygen (δ 3.75, 4.32, both m) were present. COSY allowed proton assignments to be made (Table I), giving the gross structure of **5** as indicated. Complete stereostructural assignments were made in two ways. The W coupling observed in **3** between 11-H and 7-H appears present in both **4** and **5** (both 11-H's in **4** and **5** are broadened or split). This coupling *and* the C-9 methyl chemical shift similarities in **3** (δ 1.27), **4** (δ 1.28), and **5** (δ 1.30) indicates that the stereochemistry at C-9 is most likely the same in all three compounds. Evidence for the other chiral centers in **5** was provided by the fortuitous decomposition of **5** to **6** in an NMR tube containing $CDCl_3$ (with a trace of H_2O present) as shown in Scheme I. From the spectral data (see below), 3α-hydroxytrichothecene **6** was characterized as shown, allowing assignment of all the remaining chiral centers in **5**. By biosynthetic analogy with **5**, **4** is also expected to have the stereochemistry shown.

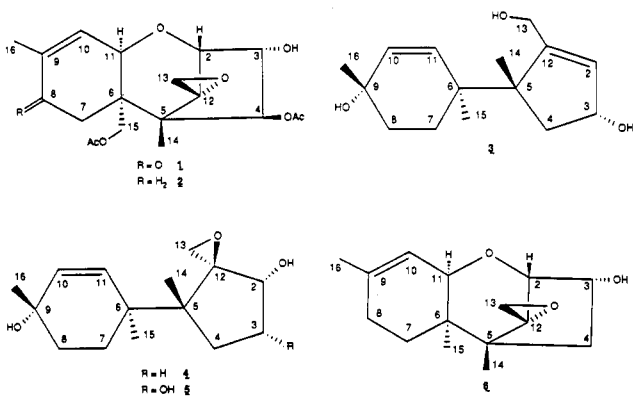
Scheme I



3-Hydroxytrichothecene (**6**), $C_{15}H_{22}O_3$ (m/z 250.157, calcd 250.157), had the following peaks in the EIMS: m/z 250 (M^+, 5), 235 ($M^+ - CH_3$, 5), 125 (20), 107 (100), 91 (40), and 43 (55). The IR spectrum (film) shows hydroxyl (3387, 1100 cm^{-1}) and ether (1158 cm^{-1}) functionalities present. The UV (λ_{max} 196.7, ϵ_{max} 10 000, ACN) spectrum indicates an ene present. The ^1H and ^{13}C NMR are consistent with that of the normal trichothecene nucleus. This is based on chemical shift similarities in ^1H and ^{13}C NMR of **6** with a number of closely related compounds[13] (Table I) *and* key J values (2-H/3-H, J = 4.4 Hz; 10-H/11-H, J = 5.6 Hz) in ^1H NMR.

The trichothecenes **1**, **3**, and **5** were all bioassayed for toxicity by using the chicken egg yolk sac inoculation method.[14,15] Preliminary results indicate that all three

(11) Abrahamsson, S.; Nilsson, B. *Acta Chem. Scand.* **1966**, *20*, 1044.
(12) Nozoe, S.; Machida, Y. *Tetrahedron* **1972**, *28*, 5105.
(13) Cole, R. J.; Cox, R. H. *Handbook of Toxic Fungal Metabolites*; Academic Press: New York, 1981; pp 152–224.



compounds have similar embryotoxicity to that of T-2 toxin, which has an LD_{50} value of 55 ng/egg.[15] Trichotriol is the first highly toxic modified trichothecene reported. This suggests that in the structure/activity relationship of the trichothecene skeleton, the tetrahydropyran ring may play a minor role in contributing to toxicity. These results warrant more detailed bioevaluation, which is underway.

Trichotriol is also interesting biosynthetically. It is the first example of a modified trichothecene with C-3 oxidation before ring closure as shown in the scheme to the trichothecene nucleus.

Experimental Section

Physical Analyses. All ultraviolet spectra were obtained on a Perkin-Elmer 576 ST spectrophotometer. Infrared spectra were obtained on a Fourier transform Nicolet 20 DXB. Samples were cast as a film on a NaCl plate.

Mass spectra of the samples were obtained on a Kratos MS-25 mass spectrometer equipped with a DS-55 data system. The instrument was operated in several different modes: low resolution (1/600) electron impact (EI) at 70 eV and 1 s/decade scanning speed; medium resolution (1/6500) at 70 eV and 3 s/decade for measurement of exact masses.

All NMR experiments were performed on a Nicolet NT-300 WB spectrometer equipped with 5-mm ^1H and ^{13}C NMR probes operating at 300.06 and 75.45 MHz, respectively. All ^1H NMR chemical shifts were referenced to internal tetramethylsilane (TMS) (0.0 ppm) and all ^{13}C NMR chemical shifts were referenced against the deuteriated solvent used ($CDCl_3$ = 77.0 ppm). The two-dimensional *J*-correlated experiment COSY (9), used a delay of 3 s, a 90-deg pulse of 8 μs, a 512 block size, 128 increments, quadrature detection in both dimensions, zero-filling once in t_1, and an acquisitions multiple of 4. The distortionless enhancement by polarization transfer experiment (DEPT) used the pulse sequence by Doddrell.[16]

Fungal Materials. The fungus *Fusarium sporotrichioides* MC-72083 was obtained from Professor John Tuite, Department of Botany and Plant Pathology, Purdue University. The fungus was isolated in 1972 from a wheat sample in Miskolc, Hungary. The wheat had caused mycotoxicoses in hen layers and was shown to be very toxic to rats, chickens, and swine.[17] It was originally classified as *F. sporotrichiella* var. *sporotrichioides* by Bilay's system and latter reclassified as *F. sporotrichioides* by Booth and Nelson.[18]

Culture Conditions. *F. sporotrichioides* was grown[5] on yeast malt agar plates for 14 days at 25 °C. Sterilized H_2O was added to the agar plates and the conidia were scraped into a larger volume of sterilized distilled H_2O (100 mL/agar plate). One-quart Ball canning jars (100–200) containing 100 g of Quaker Oats white corn grits were autoclaved for 30 min. Aliquots (2 mL) of the mycelium–H_2O mixture were pipetted into each jar of corn grits and 33 mL of sterile H_2O was added. The jars were shaken and the lids loosened to allow for respiration. After 24 h of incubation at 10 °C in darkness, the jars were shaken again to insure complete dispersal of the mycelium. The jars were incubated for a total of 21 days at 10 °C in darkness.

Isolation. A large-scale workup of the culture filtrate used a modified method of Burmeister.[15] Approximately 800 jars were harvested in batches of 100–200 jars over a period of 1 year. The corn grits were extracted with 85:15 $CHCl_3$/acetone (400 mL/jar) by blending at high speed until homogenized. The fungal–solvent mixture was allowed to stand overnight and suction-filtered. The solid residue was reextracted with acetone, suction-filtered, autoclaved, and discarded. The acetone extract was combined with the 85:15 $CHCl_3$/acetone extract and concentrated under vacuum. The dark-red oil (ca. 0.5 L/200 jars) was subjected to a hexane drip to remove the nonpolar constituents. This was achieved by dripping the oil into a stirring solution of 85:15 hexane/acetone (ca. 50 mL oil/2 L solution) and then allowing the solution to stand for 24 h. The solvent was decanted and concentrated under vacuum and the remaining solid residue from the drip was discarded. The orange-red oil was subjected to Florisil column chromatography. The oil was dissolved in 2:1 C_6H_6/hexane to obtain a nonviscous solution (ca. 1:4 oil/solvent). The 100 mL of dissolved oil was applied to the column and washed with 300 mL of 2:1 C_6H_6/hexane and 300 mL of CH_2Cl_2. The trichothecenes were then eluted with 400 mL of 95:5 $CHCl_3$/MeOH, followed by 400 mL of acetone. Flash chromatography[19] (toluene/acetone, 3:1) followed by preparative HPLC (toluene/acetone) and preparative RPTLC (MeOH/H_2O, 7:3) resulted in pure compounds.

All solvents used for extraction and Florisil chromatography were ACS grade purchased from Fisher. All solvents used for flash chromatography, HPLC, and TLC were glass-distilled solvents purchased from Burdick & Jackson.

The Florisil was Fisher 60–100 mesh and packed by pouring the Florisil into a 5 × 20 cm gravity column (C_6H_6/hexane, 2:1) to a depth of 8 cm. A 2-cm layer of Fisher anhydrous Na_2SO_4 was added on top of the Florisil. Flash chromatography used EM reagent 40–63 μm Kieselgel 60 silica gel.

Normal-phase TLC plates were silica gel HLF uniplates, 250-μm thick, purchased from Analtech. Preparative TLC entailed applying 5–10 mg of material to a 10 × 20 cm silica gel Analtech HLF uniplate, 250-μm thick. After development, compounds were identified by their quenching behavior at 254 nm or by spray visualization using a chromogenic reagent. To avoid destruction of compounds, a thin slice in the middle of the TLC plate was removed and used for spray development. The bands of interest were scraped and sonicated with 10 mL of acetone for 1 min and allowed to stand for 2 h with occasional stirring. The sample was filtered through a Rainin 0.45-μM nylon-66 filter, concentrated under vacuum, and prepared for NMR analyses by using standard procedures. Preparative RPTLC plates were Whatman KC18F with 200-μm thickness and used the same procedure as described above for preparative TLC. The spray reagent for TLC analysis was prepared with MeOH/HOAc/H_2SO_4/anisaldehyde (Eastman Organic Chemicals) (85:15:5:0.5 v/v). The blue spray reagent[20] is administered in two parts. Spray no. 1 (Aldrich 1% 4-(*p*-nitrobenzyl)pyridine in $CCl_4/CHCl_3$ 3:2 v/v) is sprayed liberally on the air-dried plate. The plate is oven-heated at 150 °C for one-half hour, cooled, and sprayed with spray no. 2 (Aldrich 10% tetraethylenepentamine in $CCl_4/CHCl_3$ 3:2 v/v). The epoxide-containing trichothecenes give a sky-blue color.

Preparative HPLC was conducted on a Perkin-Elmer series 3B chromatograph using a linear gradient solvent system solvent system. The column was silica gel 3.5 × 28 cm Perkin-Elmer 0258-3002. Quantitative HPLC was conducted on a Perkin-Elmer series 2 chromatograph using a Waters 3.9 mm × 30 cm μBondapak C-18 column and a Perkin-Elmer LC-85 spectrophotometric detector operating at 195 nm.

(14) McLaughlin, J., Jr.; Marbiac, J. P.; Verret, M. J.; Mytchler, M. K.; Fitzhugh, O. G. *Toxicol. Appl. Pharmacol.* **1963**, *5*, 760.
(15) Lafont, J.; Lafont, P. *Bull. Acad. Vet. Fr.* **1979**, *52*, 119.
(16) Doddrell, D. M.; Pegg, D. T.; Bendall, M. R. *J. Magn. Reson.* **1982**, *48*, 323.
(17) Szigeti, G. C. Sc. and Ph.D. Dissertation, Miskolc, Hungary, 1976.
(18) Tuite, J. Indiana University, personal communication, 1987.
(19) Still, W. C.; Kahn, M.; Mitra, A. *J. Org. Chem.* **1978**, *43*, 2923.
(20) Takitani, S.; Asabe, Y.; Kato, T.; Suzuki, M.; Ueno, Y. *J. Chromatogr.* **1979**, *116*, 335.

Bioassay. The specific-pathogen-free (SPF) fertile chicken eggs, used for the chick embryo bioassay,[14,15] were purchased from Larson Lab-Vac Eggs, Inc., Gowrie, IA 50543. The incubators used were the Imperial II produced by Lab-Line Instruments, Inc. and Model 3212-10 produced by National Appliance Company. The temperature was maintained at 37 °C and the relative humidity was kept at ca. 60%. Embryos were received within 24 h of laying, incubated for 5 days, and candled for viability. Typically a batch of 150 eggs would yield 120 acceptable eggs. Before dosing, the air cells were circled with a pencil and swabbed with 200 proof EtOH (Midwest Solvents Company of Illinois). An 18-gauge needle, sterilized in 200 proof ethanol, was used to puncture the shell above the encircled air cell. The toxins were dissolved in 200 proof EtOH, serial diluted, and 10 μL injected into the air cell with a microsyringe. The hole was then sealed with Scotch tape; 10 μL of 200 proof EtOH was injected into the control eggs. In 4 days the eggs were candled and the number of dead embryos were determined for each dose level.

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Registry No. 1, 77620-47-4; 3, 69344-87-2; 4, 40522-81-4; 5, 109890-37-1; 6, 104155-10-4.

Reaction of Cyclopropenones with Trimethylsilyl Cyanide with the Aid of Transition-Metal Complexes or Phosphines.[1] A New Synthesis of 5-Amino-2-furancarbonitriles

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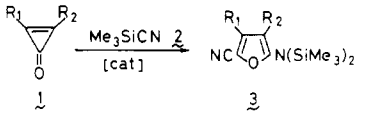
Diphenylcyclopropenone (**1a**) was first synthesized in 1959 by Breslow et al.[2] and Vol'pin et al.[3] Since that time there have been reported many reactions[4] involving cycloaddition[5] and nucleophilic addition. The interaction between cyclopropenones and transition-metal complexes has also been of interest.[6] We have now found that transition-metal complexes and phosphines are effective catalysts for the reaction of trimethylsilyl cyanide, Me_3SiCN (**2**), with cyclopropenones to give 5-amino-2-furancarbonitriles (eq 1).

Table I. Reaction of Diphenylcyclopropenone (1a) with Trimethylsilyl Cyanide (2)[a]

entry	catalyst	temp, °C	yield,[b] %
1	$Fe_2(CO)_9$	reflux[c]	41
2	$Ni(PPh_3)_2(CO)_2$	reflux	48
3	$Rh(PPh_3)_3Cl$	reflux	62
4	$Pt(PPh_3)_2Cl_2$	reflux	56
5	$Ir(PPh_3)_2(CO)Cl$	reflux	61
6	PPh_3	reflux	52
7	PPh_3	60	55
8	PPh_3	25	4
9	$P(4\text{-}MeOC_6H_4)_3$	60	55
10	$P(4\text{-}MeC_6H_4)_3$	60	46
11	PBu_3	60	14
12	$Ph_2CH_2CH_2PPh_2$	60	24

[a] Reaction conditions: diphenylcyclopropenone (0.8 mmol, 165 mg), Me_3SiCN (4.8 mmol, 0.65 mL), catalyst (0.032 mmol, 4 mol %). [b] Isolated yields based on **1a**. [c] Reflux temperature of reaction mixture; bp (Me_3SiCN) 115 °C.



(1)

1 3

a $R_1 = R_2 = Ph$
b $R_1, R_2 = -(CH_2)_5-$

The reaction of diphenylcyclopropenone (**1a**) with **2** in the presence of $Fe_2(CO)_9$ under reflux gave 5-[*N*,*N*-bis(trimethylsilyl)amino]-3,4-diphenyl-2-furancarbonitrile (**3a**)[7] in 41% yield (entry 1 in Table I). $Co_2(CO)_8$, $CpCo(CO)_2$, $Rh_6(CO)_{16}$, $Ru_3(CO)_{12}$, and $[RhCl(CO)_2]_2$ showed little or no catalytic activity. On the other hand, transition metal–phosphine complexes were effective for the present reaction (entries 2–5).

Interestingly, phosphines alone were found to catalyze the present reaction.[8] Triphenylphosphine was effective enough to produce the furan **3a** in 55% yield even at 60 °C, at which temperature transition-metal complexes, e.g., $Fe_2(CO)_9$ and $Rh(PPh_3)_3Cl$, showed little catalytic activity (entry 7). Triphenylphosphine, tris(4-methylphenyl)phosphine, and tris(4-methoxyphenyl)phosphine all were effective catalysts. Tris(2-methylphenyl)phosphine, trimethyl phosphite, triphenylarsine, diphenyl sulfide, pyridine, and triethylamine did not show catalytic activity. PPh_3-catalyzed reaction of cycloheptenocyclopropenone

(1) For the previous papers in this series, see: (a) Chatani, N.; Hanafusa, T. *J. Org. Chem.* **1986**, *51*, 4714. (b) Chatani, N.; Hanafusa, T. *Tetrahedron Lett.* **1986**, *27*, 4201. (c) Chatani, N.; Takeyasu, T.; Hanafusa, T. *Tetrahedron Lett.* **1986**, *27*, 1841. (d) Chatani, N.; Hanafusa, T. *J. Chem. Soc., Chem. Commun.* **1985**, 838.

(2) Breslow, R.; Haynie, R.; Mirra, J. *J. Am. Chem. Soc.* **1959**, *81*,247.

(3) Vol'pin, M. E.; Koreshkov, Y. D.; Kursanov, D. N. *Izv. Akad. Nauk SSSR, Otd. Khim. Nauk* **1959**, 560; *Chem. Abstr.* **1959**, *53*, 21799. Kursanov, D. N.; Vol'pin, M. E.; Koreshkov, Y. D. *Zh. Obshch. Khim.* **1960**, *30*, 2879; *Chem. Abstr.* **1960**, *54*, 16473.

(4) For reviews on cyclopropenones, see: Yoshida, Z. *Top. Curr. Chem.* **1973**, *40*, 47. Potts, K. T.; Baum, J. S. *Chem. Rev.* **1974**, *74*, 189.

(5) For reviews on cycloaddition of cyclopropenones, see: Deem, M. L. *Synthesis* **1972**, 675. Deem, M. L. *Synthesis* **1982**, 701.

(6) (a) For reactions of **1a** with transition-metal complexes, see: Bird, C. W.; Briggs, E. M.; Hudec, J. *J. Chem. Soc. C* **1967**, 1862. Fichteman, W. L.; Schmidt, P.; Orchin, M. *J. Organomet. Chem.* **1968**, *12*, 249. Wong, W.; Singer, S. J.; Pitts, W. D.; Watkins, S. F.; Baddley, W. H. *J. Chem. Soc., Chem. Commun.* **1972**, 672. Visser, J. P.; Ramakers-Blom, J. E. *J. Organomet. Chem.* **1972**, *44*, C63. Noyori, R.; Umeda, I.; Takaya, H. *Chem. Lett.* **1972**, 1189. Bird, C. W.; Briggs, E. M. *J. Organomet. Chem.* **1974**, *69*, 311. (b) For $Ni(CO)_4$-catalyzed reactions of **1a** with heterocumulenes see: Ohshiro, Y.; Nanimoto, H.; Tanaka, H.; Komatsu, M.; Agawa, T.; Yasuoka, N.; Kai, Y.; Kasai, N. *Tetrahedron Lett.* **1985**, *26*, 3015 and references cited therein.

(7) (a) To the best of our knowledge, there is no literature for the synthesis of 5-amino-2-furancarbonitriles. On the other hand, some of other regioisomers have been known. 2-Amino-3-furancarbonitriles: Nixon, W. J., Jr.; Garland, J. T.; Blanton, C. D., Jr. *Synthesis* **1980**, 56. 5-Amino-3-furancarbonitriles: Aran, V. J.; Soto, J. L. *Synthesis* **1982**, 513. (b) 5-Amino-2-furancarboxylic esters: Stibranyi, L.; Peera, M.; Kozempelova, Z.; Lesko, J.; Kovac, J. *Collect. Czech. Chem. Commun.* **1986**, *51*, 2817. 5-Amino-2-furancarbaldehydes: Ebetino, F. F. *Heterocycles* **1974**, *2*, 391.

(8) For PPh_3-catalyzed reactions of **1a**, see: Obata, N.; Takizawa, T. *Tetrahedron Lett.* **1970**, 2231. Kascheres, A.; Joussef, A. C.; Duarte, H. C. *Tetrahedron Lett.* **1983**, *24*, 1837.

0022-3263/87/1952-4408$01.50/0