New Destruxins from the Entomopathogenic Fungus *Aschersonia* sp.

Stuart B. Krasnoff* and Donna M. Gibson

USDA-ARS Plant Protection Research Unit, Tower Road, Ithaca, New York 14853

Gilbert N. Belofsky, Katherine B. Gloer, and James B. Gloer

Department of Chemistry, University of Iowa, Iowa City, Iowa 52245

Received December 14, 1995[⊗]

Two new insecticidal cyclic depsipeptides, destruxins A4 (**1**) and A5 (**2**), were isolated by bioassay-guided fractionation of organic extracts from an undescribed species of entomopathogenic fungus from the genus *Aschersonia*. Another previously identified but related compound, homodestruxin B (**3**), was also isolated as a product of this fungus. The chemical structures of **1**−**3** were determined based on analysis of NMR and mass spectral data. In common with homodestruxin B (**3**), the novel compounds destruxins A4 (**1**) and A5 (**2**) have a MeIle moiety in place of the MeVal residue more typically found in destruxins. In addition, destruxin A5 has a β-MePro unit in place of the Pro residue found in most destruxins. LC_{50} values for **1** and **2** in an insecticidal assay against *Drosophila melanogaster* were estimated at 41 and 52 ppm, respectively. Homodestruxin B (**3**) showed no activity at 400 ppm in the same assay. Destruxins A4 (**1**) and A5 (**2**) are the first biologically active secondary metabolites reported from the fungal genus *Aschersonia*, which represents a new source of destruxins.

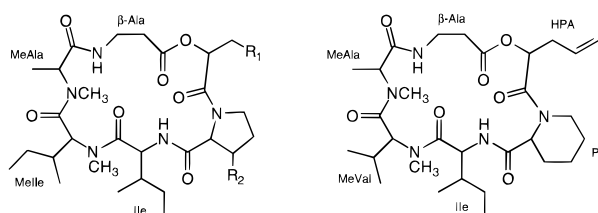
Fungi of the genus *Aschersonia* (Coelomomycetes, Deuteromycotina) are pathogens of whiteflies (Homoptera, Aleyrodidae). These fungi form leathery stromata of thickly matted hyphae and produce spindle-shaped conidia in slimy masses.[1] Because their hosts include a number of important pest species such as the citrus whitefly, *Dialeurodes citri*, and the sweet potato whitefly, *Bemisia tabaci*,[2,3] *Aschersonia* fungi have been under investigation as potential biological control agents.[2] As a consequence, their toxigenicity is of great interest.

Most *Aschersonia* species are known only in their conidial form, but a number of species have been associated with perfect stages disposed to the genus *Hypocrella*. A group of photodynamic perylenequinones, the hypocrellins, are produced by *Hypocrella bambusae*.[4] The evidence for production of biologically active substances by conidial forms of *Aschersonia* is limited, however, to a report of the production of hopanoids by *Aschersonia aleyrodis*, but with no biological activity attributed to the isolated compounds,[5] and a report that *A. aleyrodis* culture broth extracts are weakly toxic to the greenhouse whitefly adults.[6]

While screening entomopathogenic fungi in the USDA-Agricultural Research Service Collection of Entomopathogenic Fungi (ARSEF) for the production of insecticidal metabolites, we detected insecticidal activity against fruit flies (*Rhagoletis pomonella* and *Drosophila melanogaster*) in CH_2Cl_2 extracts of culture broth from a strain of an undescribed *Aschersonia* species. Herein we report the bioassay-guided isolation and characterization of three destruxins from culture filtrate extracts of this fungus: two novel insecticidal compounds, destruxins A4 (**1**) and A5 (**2**), and the previously identified homodestruxin B (**3**).

Results and Discussion

Bioassay-guided fractionation of the organic extracts from liquid cultures of *Aschersonia* sp. by Si gel column chromatography and reversed-phase HPLC afforded metabolites **1**−**3**. Compound **1** was assigned the molecular formula $C_{30}H_{49}N_5O_7$, based on HRFABMS ([M + H]$^+$ at *m/z* 592.3701), ^{13}C-NMR, and DEPT data. The ^{13}C-NMR spectrum contained signals for a vinyl group (119.5 and 131.4 ppm), as well as six amide or ester carbonyl resonances (168−174 ppm). Based on these data and the required number of unsaturations for the molecular formula, compound **1** must contain two rings. The ^{1}H-NMR spectrum revealed two N−H doublets, two N−Me singlets, three vinylic proton signals, and five downfield-shifted methine signals consistent with peptide α protons. The MS and NMR data, together with a literature search, suggested that **1** is a hexadepsipeptide of the destruxin class.[7,8] Detailed comparison of the NMR data for compound **1** (Tables 1 and 2) with data for an authentic sample of the known compound destruxin Al[8] (**4**, $C_{30}H_{49}N_5O_7$) supported this conclusion. HMBC and HMQC experiments conducted on **1** and **4** permitted the corresponding proton and carbon NMR assignments to be made, and also revealed several significant differences. The MeVal residue in **4** is replaced by a MeIle residue in **1**, as evidenced by the appearance of an additonal methylene carbon at 25.8 ppm and by the ^{1}H-NMR multiplicities, ^{13}C-NMR shift values, and HMBC correlations of the β and δ-CH_3 groups. Similar NMR considerations also indicated that the pipecolic acid moiety in **4** is replaced by a proline residue in **1**. The NMR data for the remaining amino acid residues in **1** were nearly identical with those for



1 R_1= CH=CH$_2$, R_2=H
2 R_1= CH=CH$_2$, R_2=CH$_3$
3 R_1= CHMe$_2$, R_2=H
4

* To whom correspondence should be addressed. Phone: (607) 255-7744. FAX: (607) 255-2459. E-mail: sbk1@cornell.edu.

⊗ Abstract published in *Advance ACS Abstracts,* May 1, 1996.

S0163-3864(96)00121-8 CCC: $12.00 © 1996 American Chemical Society and American Society of Pharmacognosy

Table 1. ^{1}H-NMR Spectral Data for Compounds **1**−**4** [δ_H (mult., J_{HH}); $CDCl_3$]

^{1}H position	destruxin A4 (**1**)	destruxin A5 (**2**)	homodestruxin B (**3**)	destruxin A1 (**4**)
β-alanine				
NH	8.23 (br d, 8.3)	8.28 (br d, 9.7)	8.22 (br d, 10)	8.12 (br d, 8.7)
α-CH_2	2.68 (ddd, 19, 12, 2.0)	2.66 (ddd, 19, 12, 1.9)	2.68 (ddd, 19, 12, 2.1)	2.65 (m)
	2.57 (m)	2.52 (br dd, 19, 4.4)	2.57 (br dd, 19, 4.7)	2.57 (m)
β-CH_2	4.04 (m)	4.01 (m)	4.06 (m)	4.06 (m)
	3.07 (br t, 13)	3.05 (br t, 13)	3.08 (br t, 13)	3.01 (br t, 13)
N-Me-alanine				
NCH_3	2.72 (s)	2.72 (s)	2.73 (s)	2.72 (s)
α-CH	5.16 (q, 6.8)	5.11 (q, 6.7)	5.18 (q, 6.8)	5.18 (q, 7.0)
β-CH_3	1.31 (d, 6.8)	1.27 (d, 6.8)	1.30 (d, 6.8)	1.30 (d, 7.0)
N-Me-isoleucine[a]				*N*-Me-valine
NCH_3	3.20 (s)	3.20 (s)	3.20 (s)	3.22 (s)
α-CH	5.03 (d, 11)	5.02 (d, 11)	5.02 (d, 11)	4.87 (d, 11)
β-CH	2.05 (m)	2.06 (m)	2.08 (m)	2.32 (m)
β-CH_3	0.86 (d, 6.9)	0.84 (d, 6.6)	0.86 (d, 6.9)	
γ-CH_2	1.43 (m)	1.43 (m)	1.44 (m)	
	0.95 (m)	0.90 (m)	0.98 (m)	
γ-CH_3				0.92 (d, 6.6)
γ-CH_3				0.90 (d, 6.6)
δ-CH_3	0.92 (br t, 7.2)	0.89 (br t, 7.2)	0.93 (br t, 7.1)	
isoleucine				
NH	7.15 (d, 9.1)	7.06 (d, 9.2)	7.18 (d, 8.9)	7.07 (d, 9.0)
α-CH	4.86 (dd, 9.0, 6.6)	4.81 (dd, 9.0, 6.7)	4.86 (dd, 8.9, 6.6)	4.97 (dd, 9.1, 6.0)
β-CH	1.90 (m)	1.89 (m)	1.87 (m)	1.94 (m)
β-CH_3	0.85 (d, 6.9)	0.82 (d, 6.9)	0.86 (d, 6.9)	0.91 (d, 6.9)
γ-CH_2	1.43 (m)	1.43 (m)	1.44 (m)	1.43 (m)
	1.28 (m)	1.26 (m)	1.29 (m)	1.32 (m)
δ-CH_3	0.85 (m)[d]	0.83 (m)[d]	0.86 (m)[d]	0.84 (br t, 7.4)
proline[b]		β-Me-proline		pipecolic acid
α-CH	4.67 (d, 7.2)	4.24 (br s)	4.67 (d, 7.3)	5.26 (br d, 4.6)
β-CH		2.75 (m)		
β-CH_2	2.47 (m)		2.49 (m)	1.45 (m)
	1.94 (m)		1.94 (m)	
β-CH_3		1.06 (d, 7.0)		
γ-CH_2	2.05 (m)	2.08 (m)	2.08 (m)	1.75 (m)
	1.94 (m)	1.65 (m)	1.94 (m)	1.45 (m)
δ-CH_2				1.75 (m)
				1.43 (m)
NCH_2	3.89 (br t, 9.0)	3.83 (ddd, 9.4, 9.4, 2.8)	3.91 (br t, 8.7)	3.94 (d, 13)
	3.48 (br q, 9)	3.58 (ddd, 9.4, 9.4, 9.4)	3.43 (br q, 9)	3.19 (m)
2-hydroxy-4-pentenoic acid[c]			2-hydroxy-4-methyl-pentanoic acid	
α-CH	4.84 (br t, 7.2)	4.84 (br t, 7.3)	4.89 (dd, 10, 2.9)	5.13 (t, 7.0)
β-CH_2	2.65 (m)	2.63 (m)	1.96 (ddd, 14, 10, 4.4)	2.65 (m)
	2.54 (m)	2.59 (m)	1.38 (ddd, 14, 9.5, 3.0)	2.57 (m)
γ-CH	5.80 (dddd, 17, 10, 7.2, 7.1)	5.76 (dddd, 17, 10, 7.2, 7.2)	1.96 (m)	5.78 (dddd, 17, 10, 7.2, 7.2)
δ-CH_2	5.21 (dd, 17, 1.3)	5.21 (dd, 17, 1.3)		5.23 (dd, 17, 1.4)
	5.17 (br d, 11)	5.16 (br d, 10)		5.19 (br d, 10)
δ-CH_3			0.99 (d, 6.7)	
δ-CH_3			0.95 (d, 6.7)	

[a] This residue is *N*-Me-valine in destruxin A1 (**4**). [b] This residue is β-Me-proline in destruxin A5 (**2**) and pipecolic acid in destruxin A1 (**4**). [c] This residue is 2-hydroxy-4-methylpentanoic acid in homodestruxin B (**3**). [d] Coupling constants could not be measured due to signal overlap.

4, and the peptide was therefore determined to contain one equivalent each of β-Ala, MeAla, MeIle, Ile, Pro, and 2-hydroxy-4-pentenoic acid. HMQC and HMBC correlations were fully consistent with the presence of these six residues.

The peptide sequence of **1** was assigned on the basis of HMBC correlations from the α, β, N−H, and N−Me proton signals to the resonances for the neighboring carbonyl carbons. For example, the β-CH_3 proton signal of MeAla (1.31 ppm) and the β-CH_2 proton signal of β-Ala at 3.07 ppm both show correlations to the carbonyl carbon at 169.7 ppm, indicating that β-Ala is acylated by MeAla. Sequence-relevant correlations of this type were observed for each carbonyl signal in the molecule, thereby permitting independent confirmation of the structure of destruxin A4 (**1**) as shown.

The ^{1}H-NMR (Table 1), ^{13}C-NMR (Table 2), HMQC, and HMBC data for destruxin A5 (**2**, $C_{31}H_{51}N_5O_7$) were nearly identical to those of **1**, except for signals that correspond to the cyclic amino acid residue. In the spectra for **2**, a new Me group (1.06/18.9 ppm) and a methine carbon at 36.1 ppm replaced the Pro methylene carbon observed at 29.1 ppm in **1**. An HMBC correlation was observed between the new Me group and the downfield-shifted α-carbon (now at 67.2 ppm), indicating that this Me group is located at the β-position of the pyrrolic ring. Additional HMBC and HMQC data confirmed the presence of a β-MePro residue. The ^{1}H- and ^{13}C-NMR signals for this residue are consistent with those observed for roseotoxin B, the only other member of the destruxin class reported to contain a β-MePro subunit.[9,10]

Although HMBC correlations of the α-, β-, N-H, and N−CH_3 proton signals with the adjacent carbonyl carbon resonances were useful in confirming the MeAla to β-Ala and 2-hydroxy-4-pentenoic acid to β-MePro segments of destruxin A5 (**2**), overlap of four key ^{13}C-

Table 2. ^{13}C-NMR Spectral Data for Compounds **1**−**4** (δ_C, $CDCl_3$)

^{13}C position	destruxin A4 (**1**)	destruxin A5 (**2**)	homodestruxin B (**3**)	destruxin A1 (**4**)
β-alanine				
C=O	173.6	173.6	173.8	173.7
α-CH_2	34.5	34.5	34.4	34.2
β-CH_2	33.2	33.2	33.2	33.2
N-Me-alanine				
NCH_3	28.1	28.1	28.1	28.1
C=O	169.7	169.7	169.7	169.6
α-CH	55.5	55.5	55.5	55.4
β-CH_3	15.2	15.2	15.3	15.3
N-Me-isoleucine[a]				*N*-Me-valine
NCH_3	31.0	30.9	31.0	30.9
C=O	171.1	171.1	171.1	171.0
α-CH	56.7	56.7	56.8	58.3
β-CH	33.6	33.5	33.6	27.3
β-CH_3	16.2	16.3	16.3	
γ-CH_2	25.8	25.7	25.8	
γ-CH_3				19.8
γ-CH_3				20.1
δ-CH_3	11.1	11.0	11.1	
isoleucine				
C=O	173.5	173.6	173.5	173.5
α-CH	53.6	53.4	53.7	53.8
β-CH	37.5	37.5	37.5	37.6
β-CH_3	15.4	15.3	15.4	15.9
γ-CH_2	24.4	24.6	24.5	24.4
δ-CH_3	11.4	11.4	11.4	11.5
proline[b]		*β*-Me-proline		pipecolic acid
C=O	171.0	171.1	171.0	169.6
α-CH	60.8	67.2	60.7	53.2
β-CH		36.1		
β-CH_2	29.1		28.9	25.2
β-CH_3		18.9		
γ-CH_2	24.1	30.7	24.1	20.8
δ-CH_2				25.6
NCH_2	46.7	45.1	46.5	44.3
2-hydroxy-4-pentenoic acid[c]			2-hydroxy-4-methylpentanoic acid	
C=O	168.8	169.1	175.1	169.8
α-CH	72.8	72.6	72.0	70.3
β-CH_2	34.9	35.2	39.0	35.6
γ-CH	131.4	130.9	24.4	131.4
δ-CH_2	119.5	119.9		119.5
δ-CH_3			23.4	
δ-CH_3			21.5	

[a] This residue is *N*-Me-valine in destruxin A1 (**4**). [b] This residue is *β*-Me-proline in destruxin A5 (**2**) and pipecolic acid in destruxin A1 (**4**). [c] This residue is 2-hydroxy-4-methylpentanoic acid in homodestruxin B (**3**).

NMR signals (two at 171.1 and two at 173.6 ppm) prevented independent differentiation of two possible peptide sequences: the sequence shown in **2**, or an alternative sequence wherein an MeIle-Ile unit acylates 2-hydroxy-4-pentenoic acid, while *β*-MePro acylates MeAla. Ultimately, structure **2** was proposed based on the very close spectral similarities of **2** with **1**[e.g., $\Delta\delta_C \leq 0.5$ ppm for all ^{13}C-NMR assignments except those where the amino acid unit differs (MePro)] and other destruxins.

A third destruxin analogue (**3**, $C_{31}H_{53}N_5O_7$) was also isolated from *Aschersonia* sp. cultures. The ^{1}H-NMR data for **3** (Table 1) were similar to those for destruxin A4 (**1**), but contained additional aliphatic signals in place of the olefinic signals corresponding to the 2-hydroxy-4-pentenoic acid moiety. This compound was identified as the known compound homodestruxin B by analysis of FABMS, ^{1}H-NMR (Table 1), ^{13}C-NMR (Table 2), and HMBC data and by comparison with available literature data.[11] Homodestruxin B differs from **1** only in the substitution of a 2-hydroxy-4-methyl-pentanoic acid residue for the 2-hydroxy-4-pentenoic acid unit in **1**. Although compound **3** and destruxin A1 (**4**) have been previously reported,[8,11] neither ^{13}C-NMR nor complete ^{1}H-NMR assignments have been reported for either compound. Therefore, these data are provided here in Tables 1 and 2. The absolute configurations for the amino acid residues in compounds **1**−**4** were not determined.

Using the probit model,[12] LC_{50} values in the *Drosophila* assay were estimated at 41 ppm (95% confidence interval: 32−50 ppm) for destruxin A4 (**1**) and 52 ppm (95% confidence interval: 44−63 ppm) for destruxin A5 (**2**) (Figure 1). Homodestruxin B (**3**) produced no mortality in the assay after 48 h at a dosage of 400 ppm, indicating less than one-eighth the insecticidal potency of the destruxin A analogues. This result is consistent with prior structure/activity studies[13] reporting weaker insecticidal activity of destruxin B analogues relative to destruxin A analogues and supports the view that the olefinic side chain in the α-hydroxy acid moiety of the destruxin A subfamily of compounds is responsible for their greater insecticidal activity.

Destruxin A4 (**1**) and A5 (**2**) are the 28th and 29th destruxins to be characterized and the first biologically active metabolites to be reported from the genus *Aschersonia*, which represents yet another source of destrux-

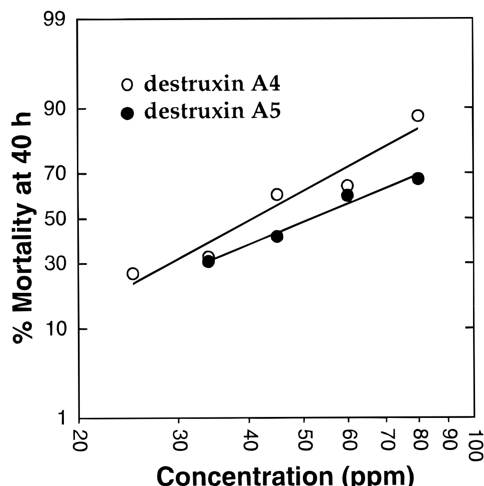


Figure 1. Insecticidal effect of destruxins A4 and A5 against *Drosophila melanogaster*. Percentage of mortality is plotted on a probability scale and concentration of formulated destruxins on a $\log_{10}$ scale. Each point represents a sample of 40–65 insects. Control mortality (response to 4.75% EtOH/10% sucrose solution) was zero in this experiment. The probit regression lines shown (Probit mortality = 3.51 ($\log_{10}$ dosage) − 0.66; probit mortality = 2.73 ($\log_{10}$ dosage) + 0.31) were used to estimate effective doses for destruxins A4 and A5, respectively.

ins. In all, 23 destruxins have been isolated as products of the entomopathogenic fungus *Metarhizium anisopliae* Sorokin (Deuteromycota).[7,8,14–20] Homodestruxin B (**3**) was originally isolated from the plant pathogenic fungus *Alternaria brassicae* (Berkeley) Saccardo,[11] and roseotoxin B is known only from *Trichotecium roseum.*[10,21,22] Two additional compounds, bursaphelocides A and B, which possess a unique α-hydroxy acid (2-hydroxy-3-methylpentanoic acid) and a 4-MePro in place of the usual Pro, have been isolated and characterized from an unidentified imperfect fungus.[23]

There is evidence from other fungi that there may be species-specific tendencies to incorporate particular amino acid precursors at certain variable positions in microheterogeneous families of peptides.[24,25] Given the large number of destruxins now characterized, it would seem that directed-biosynthesis experiments would be useful for determining whether this phenomenon occurs with destruxin-producing fungi.

Biological activities demonstrated for various destruxins include phytotoxicity,[26] antitumor activity,[27] antiviral activity,[28] and toxicity to insects.[29] Destruxin-challenged insects exhibit a tetanic paralysis attributed to an activating effect on calcium channels in muscle.[30] Also, evidence suggests that destruxins serve a role in pathogenesis by interfering with the immune response of host insects,[31,32] possibly by inducing degranulation of hemocytes and thus depleting a critical defensive resource before it can be deployed against the proliferating fungus.[33] In view of the economic importance of the homopteran hosts of *Aschersonia* fungi, the potential role of destruxins as determinants of virulence should be investigated to determine whether identifying highly toxigenic isolates would facilitate the discovery of isolates with greater potential for exploitation as biological control agents.

Experimental Section

General Experimental Procedures. ^{1}H-NMR data were obtained at 300 or 600 MHz on Bruker AC-300 or AMX-600 spectrometers, respectively. ^{13}C-NMR data were recorded at 75.5 MHz. HMBC and HMQC data were obtained at 600 MHz (^{1}H-dimension), and experiments were optimized for $^nJ_{CH}$ = 8.3 Hz and $^1J_{CH}$ = 150.2 Hz, respectively. All NMR spectra were recorded in $CDCl_3$, and chemical shifts were referenced using the corresponding solvent signals (7.24/77.0 ppm). FABMS experiments employed a DTT/DTE matrix and were performed on a VG ZAB-HF spectrometer.

Fungal Material and Culture Conditions. Cryogenically preserved mycelial material of *Aschersonia* sp. (strain ARSEF no. 2356) obtained from the ARSEF collection was used to inoculate stock cultures maintained on slants of Sabouraud's dextrose agar plus yeast (1.0%). Three-week-old solid cultures were used to inoculate liquid cultures grown in 250-mL Erlenmeyer flasks containing 100 mL each of Sabouraud's dextrose broth plus yeast (1.0%). After 16 days, 25-mL aliquots of these cultures were used to inoculate each of four 2-L Fernbach flasks containing 1 L of Czapek-Dox broth (Difco) supplemented with 0.5% bactopeptone (Difco). Cultures were grown for 21 days on a rotary shaker (160 rpm, 20 ± 1 °C), were harvested by vacuum filtration (Whatman no. 1), and extracted three times with CH_2Cl_2.

Isolation. A portion (4 mg) of the crude extract (74 mg) was removed for testing and produced 80% mortality in the *R. pomonella* assay (see below) at 48 h at a concentration of 2 mg/mL. The remainder of the extract (70 mg) was dissolved in CH_2Cl_2, applied to a Si gel column (20 g Baker 40-μm) and flash-chromatographed by elution with 100% CH_2Cl_2 (4 × 100 mL) and then 3 × 100 mL each of 0.1%, 1%, 2%, 5%, 10%, 20%, and 50% MeOH in CH_2Cl_2. Fractions 11 (20.4 mg) and 12 (13.4 mg), eluting with 2% MeOH, displayed activity accounting for that observed for the crude extract in the *D. melanogaster* assay (see below).

These two fractions were separated by semipreparative reversed-phase HPLC (5-μm C18 column; 250 × 10 mm; 50:50 MeCN-H_2O at 3 mL/min with UV detection at 230 nm) to give three major components. The first and second major components to elute (**1** and **2**) showed activity accounting for the effects of the parent Si gel fraction (*D. melanogaster* assay--100% mortaility at 200 μg/mL). The third major component (**3**) showed no activity at 400 μg/mL.

Insect Bioassay. Details of bioassay protocols have been described previously.[34] Briefly, adult fruit flies were confined in small tubular cages (5 flies per tube for *R. pomonella*, 20–35/tube for *D. melanogaster*) and allowed to feed *ad libitum* on material formulated in a 4.75% EtOH/10% sucrose solution supplied via a cotton wick. For bioassays used to guide purification, mortality was assessed at 24 and 48 h after treatment. For bioassays used to estimate effective doses of pure compounds, mortality was assessed at 40 h after treatment. In all assays, flies unable to right themselves were counted as dead.

Destruxin A4 (1): white solid; mp 200–204 °C; $[\alpha]^{25}_D$ −222° (*c* 0.31, $CHCl_3$); HPLC t_R = 10.7 min; ^{1}H- and ^{13}C-NMR data, see Tables 1 and 2; HMBC data, see the supporting information; FABMS *m/z* 746 ([M + matrix + H]$^+$; rel int 45), 592 ([M + H]$^+$, 100), 535 (1), 465 (7), 437 (7), 419 (2), 380 (8), 352 (3), 331 (9), 309 (8);

HRFABMS (thioglycerol matrix); obsd *m/z* 592.3701 (M + H)$^+$; calcd for $C_{30}H_{49}N_5O_7$ + H; 592.3710.

Destruxin A5 (2): colorless oil; HPLC t_R = 13.7 min; ^{1}H- and ^{13}C-NMR data, see Tables 1 and 2; HMBC data, see the supporting information; FABMS *m/z* 760 ([M + matrix + H]$^+$; rel int 27), 606 ([M + H]$^+$, 100), 549 (1), 479 (7), 451 (6), 433 (2), 409 (2), 366 (3), 338 (7), 309 (2).

Homodestruxin B (3): white solid; HPLC t_R = 17.3 min; ^{1}H- and ^{13}C-NMR data, see Tables 1 and 2; HMBC data, see the supporting information; FABMS *m/z* 630 ([M + Na]$^+$, rel int 23), (608 ([M + H]$^+$, 100), 551 (2), 481 (10), 453 (9), 368 (5), 340 (4), 281 (6). Other data for **3** (mp, MS, IR, partial ^{1}H-NMR data) have been previously reported.[11]

Destruxin A1 (4): white solid; HPLC t_R = 12.8 min; ^{1}H- and ^{13}C-NMR data, see Tables 1 and 2; HMBC data, see the supporting information; Other data for **4** (mp, $[\alpha]_D$, MS, partial ^{1}H-NMR data) have been previously reported.[8]

Acknowledgments. This research was supported in part by a grant from the Biotechnology Research and Development Corporation, Peoria, IL. Thanks go to Matt Chapple, Claudine Hollant, Ellen Kwan, Rahmatullah Rahmati, and Craig Umscheid for technical assistance. We are grateful to Dr. Mary Païs of Institut de Chimie des Substances Naturelles, C.N.R.S., for kindly providing an authentic sample of destruxin A1, and to Dr. Sandeep Gupta for helpful discussion.

Supporting Information Available: HMBC data for destruxins A4 (**1**) and A5 (**2**), homodestruxin B (**3**), and destruxin A1 (**4**) (2 pages). See any current masthead page for ordering information.

References and Notes

(1) Fransen, J. J. IN *Whiteflies: Their Bionomics, Pest Status and Management*; D. Gerling, Ed.; Intercept Ltd.: Andover, England, 1990; pp 187–210.
(2) Fransen, J. J.; Winkelman, K.; Van, L. J. C. *J. Invertebr. Pathol.* **1987**, *50*, 158–165.
(3) *USDA-ARS Collection of Entomopathogenic Fungal Cultures*; Humber, R. A., Ed.; USDA-ARS Plant Protection Research Unit: Ithaca, New York, 1992.
(4) Zhenjun, D.; Lown, J. W. *Photochem. Photobiol.* **1990**, *52*, 609–616.
(5) Van Eijk, G. W.; Roeijmans, H. J.; Seykens, D. *Tetrahedron Lett.* **1986**, *27*, 2533–2534.
(6) Kurbatskaya, Z. A.; Subbota, A. G. *Mikrobiol. Zh.* **1989**, *51*, 56–58.
(7) Gupta, S.; Roberts, D. W.; Renwick, J. A. A. *J. Chem. Soc., Perkin Trans. 1* **1989**, 2347–2357.
(8) Païs, M.; Das, B. C.; Ferron, P. *Phytochemistry* **1981**, *20*, 715–723.
(9) Stringer, M. A.; Timberlake, W. E. *Plant Cell* **1993**, *5*, 145–146.
(10) Engstrom, G. W.; DeLance, J. V.; Richard, J. L.; Baetz, A. L. *Agric. Biol. Chem.* **1975**, *23*, 244–253.
(11) Ayer, W. A.; Pena, R. L. M. *J. Nat. Prod.* **1987**, *50*, 400–407.
(12) Finney, D. *Probit Analysis*, 3rd ed.; Cambridge University Press: Cambridge, 1971.
(13) Dumas, C.; Robert, P.; Païs, M.; Vey, A.; Quiot, J.-M. *Comp. Biochem. Physiol.* **1994**, *108C*, 195–203.
(14) Kodaira, Y. *Agric. Biol. Chem.* **1961**, *25*, 261–262.
(15) Tamura, S.; Kuyama, Y.; Kodaira, Y.; Higshikawa, S. *Agric. Biol. Chem.* **1962**, *28*, 137–138.
(16) Suzuki, A.; Taguchi, H.; Tamura, S. *Agric. Biol. Chem.* **1970**, *34*, 813–816.
(17) Suzuki, A.; Tamura, S. *Agric. Biol. Chem.* **1972**, *36*, 896–898.
(18) Jegorov, A.; Matha, V.; Sedmera, P.; Roberts, D. W. *Phytochemistry* **1992**, *31*, 2669–2670.
(19) Wahlman, M.; Davidson, B. S. *J. Nat. Prod.* **1993**, *56*, 643–647.
(20) Chen, H. C.; Yeh, S. F.; Ong, G. T.; Wu, S. H.; Sun, C. M.; Chou, C. K. *J. Nat. Prod.* **1995**, *58*, 527–531.
(21) Engstrom, G. W. *J. Agric. Food. Chem.* **1978**, *26*, 1403–1406.
(22) Springer, J. P.; Cole, R. J.; Dorner, J. W.; Cox, R. H.; Richard, J. L.; Barnes, C. L.; van der Helm, D. *J. Am. Chem. Soc.* **1984**, *106*, 2388–2392.
(23) Kawazu, K.; Murakami, T.; Ono, Y.; Kanzaki, H.; Kobayashi, A.; Mikawa, T.; Yoshikawa, N. *Biosci. Biotech. Biochem.* **1993**, *57*, 98–101.
(24) Krasnoff, S. B.; Gupta, S. *J. Chem. Ecol.* **1991**, *17*, 1953–1962.
(25) Krasnoff, S. B.; Gupta, S. *J. Chem. Ecol.* **1992**, *18*, 1727–1741.
(26) Buchwaldt, L.; Green, H. *Plant Pathol.* **1992**, *41*, 55–63.
(27) Odier, F.; Vey, A.; Bureau, J. P. *Biol. Cell* **1992**, *74*, 267–271.
(28) Sun, C. M.; Chen, H. C.; Yeh, S. F. *Planta Med.* **1994**, *60*, 87.
(29) Poprawski, T. J.; Robert, P.-H.; Maniania, N. K. *Can. Entomol.* **1985**, *117*, 801–802.
(30) Samuels, R. I.; Reynolds, S. E.; Charnley, A. K. *Comp. Biochem. Physiol. C: Comp. Pharmacol. Toxicol.* **1988**, *90*, 403–412.
(31) Vey, A.; Quiot, J. M.; Vago, C.; Fargues, J. *C.R. Acad. Sci. (Paris), Ser D* **1985**, *300*, 647–651.
(32) Huxham, I. M.; Lackie, A. M.; McCorkindale, N. J. *J. Insect Physiol.* **1989**, *35*, 97–106.
(33) Cerenius, L.; Thornqvist, P. O.; Vey, A.; Johansson, M. W.; Soderhall, K. *J. Insect Physiol.* **1990**, *36*, 785–790.
(34) Krasnoff, S. B.; Gupta, S. *J. Chem. Ecol.* **1994**, *20*, 293–302.

NP9601216