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Cycloechinulin, a Trp-Ala-derived alkaloid from the fungus *Aspergillus ochraceus*

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The structure of the Trp–Ala-derived, non-basic alkaloid cycloechinulin from the mycelial mat of the fungus *Aspergillus ochraceus* is described; systematic name: (3*S*)-2,3,8,9-tetrahydro-11-methoxy-3,8,8-trimethylpyrazino[1′,2′:1,2]azocino[5,4-*b*]indole-1,4-dione–methanol (4/1), $C_{20}H_{21}N_3O_3\cdot0.25CH_4O$. There are four independent molecules plus one methanol molecule in the asymmetric unit. The conformations of molecules 1–3 are almost identical, while molecule 4 shows a different conformation in the fused 8–6 ring moiety.

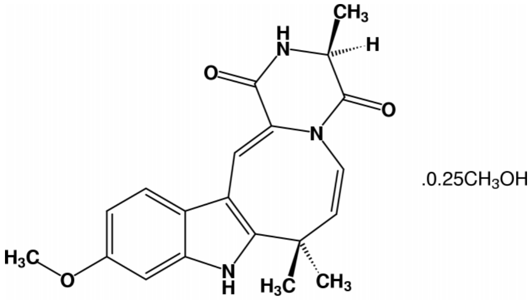
Key indicators

Single-crystal X-ray study
T = 298 K
Mean σ(C–C) = 0.011 Å
R factor = 0.064
wR factor = 0.209
Data-to-parameter ratio = 13.4

For details of how these key indicators were automatically derived from the article, see http://journals.iucr.org/e.

Comment

Aspergillus ochraceus is known to produce a number of alkaloids constructed from various amino acids, such as Phe, Ala, Trp, and Pro (Merwe *et al.*, 1965; Rahbaek & Breinholt, 1999; Rahbaek *et al.*, 1999). Cycloechinulin (I) was first isolated in 1992 in minute quantities, 1.1 mg, from the sclerotia of *A. ochraceus* by De Guzman *et al.* (1992). The Trp–Ala-derived structure was proposed on the basis of spectroscopic properties, but has not been established by X-ray diffraction. Cycloechinulin (I) is structurally related to the pentacyclic Trp–Pro-derived alkaloid (II) which was isolated from *A. ustus* by Steyn in 1973, and, more remotely, to those of the echinulin series of compounds, *e.g.* neoechinulin from *A. amstelodami* (Barbetta *et al.*, 1969; Dossena *et al.*, 1974; Quilico & Panizzi, 1943; Steyn, 1971; Yamamoto & Arai, 1986). Cycloechinulin (I) has been shown to exert moderate insecticidal activity against the lepidopteran crop pest *Helicoverpa zea* (De Guzman *et al.*, 1992). Substantial amounts of cycloechinulin (I) have now been isolated from the mycelial mat of *A. ochraceus* and its structure has been confirmed by X-ray diffraction analysis.



(I)

The title compound crystallizes in the monoclinic non-centrosymmetric space group $P2_1$ with four molecules in the asymmetric unit together with one methanol molecule. The four independent molecules are shown in Figs. 1–4. The conformations of molecules 1–3 are almost identical, while

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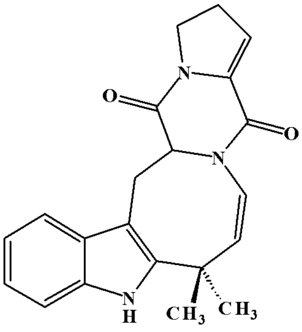
molecule 4 is found in a different conformation in the eight-membered dihydroazocine moiety and six-membered 2,5-diketopiperazine part of the molecule (see Table 1 and Figs. 5–8). The indole part of all four molecules is planar and the atoms CX23, OX01, CX10 and CX19 (X = 1–4), are also in the same plane [largest deviation −0.138 (10) Å for atom C419]. A selection of bond lengths shows that these are all within the normal range of such bonds (Allen *et al.*, 1987). The mean value for the C═O bonds is 1.232 (15) Å, while the CX02═CX03 bonds have an average bond length of 1.38 (2) Å. The mean value of the CX10═CX11 and CX18═CX17 bonds is 1.32 (3) Å. The molecules are packed in the crystal through a series of intermolecular hydrogen bonds which also involve the methanol molecule (see Table 2).



(II)

Experimental

Aspergillus ochraceus, an Australian isolate D2306 (Connole *et al.*, 1981), was grown on Sabourauds agar plates (pH 5.9, 298 K, 10–14 d). The mycelial mat was washed with distilled water (4 × 5 ml). The extracts were combined, the volume reduced to *ca* 50%, filtered, centrifuged, and extracted with $CHCl_3$. Preparative TLC (silica gel) furnished *ca* 100 mg cycloechinulin per m^2 mycelial mat. Cycloechinulin (I) was spectroscopically characterized before recrystallization. Single crystals of (I) were obtained on slow evaporation of MeOH at room temperature.

Crystal data

$C_{20}H_{21}N_3O_3\cdot0.25CH_4O$
M_r = 359.41
Monoclinic, $P2_1$
a = 15.222 (2) Å
b = 16.848 (2) Å
c = 15.930 (2) Å
β = 114.27 (1)°
V = 3724.3 (8) Å^3
Z = 8
D_x = 1.282 Mg m^{-3}
Mo $K\alpha$ radiation
Cell parameters from 25 reflections
θ = 12–18°
μ = 0.09 mm^{-1}
T = 298 (2) K
Block, colourless
0.6 × 0.3 × 0.3 mm

Data collection

Enraf–Nonius CAD-4 diffractometer
ω–2θ scans
14 273 measured reflections
13028 independent reflections
4745 reflections with $I > 2\sigma(I)$
$R_{\rm int}$ = 0.028
$\theta_{\rm max}$ = 25.0°
$h = -18 \rightarrow 18$
$k = -19 \rightarrow 19$
$l = -18 \rightarrow 18$
3 standard reflections
frequency: 120 min
intensity decay: 12%

Refinement

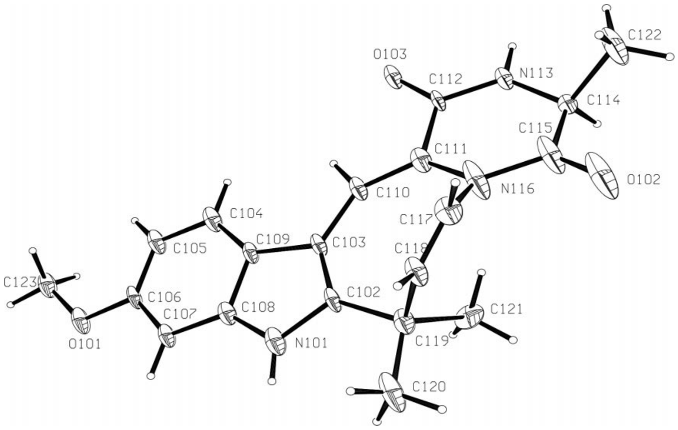


Figure 1
A view of molecule 1 of (I) with the atomic numbering scheme. Displacement ellipsoids are drawn at the 10% probability level.

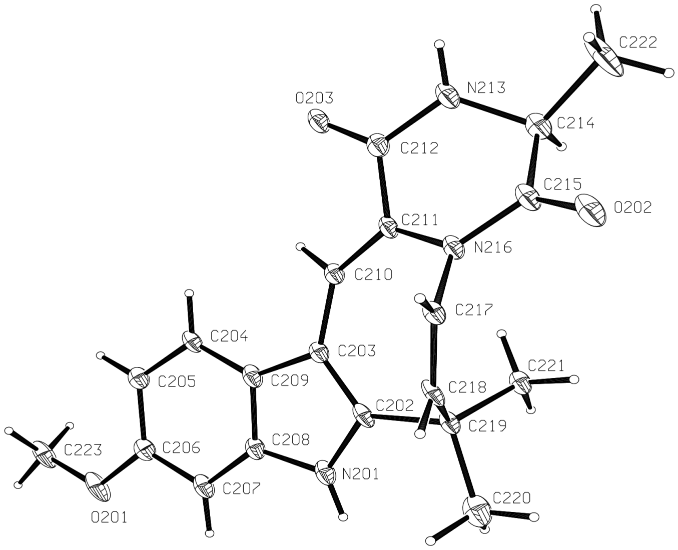


Figure 2
A view of molecule 2 of (I) with the atomic numbering scheme. Displacement ellipsoids are drawn at the 10% probability level.

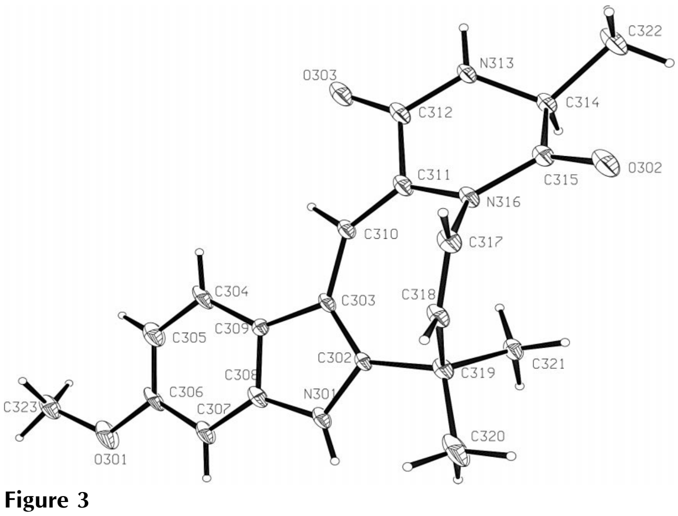


Figure 3
A view of molecule 3 of (I) with the atomic numbering scheme. Displacement ellipsoids are drawn at the 10% probability level.

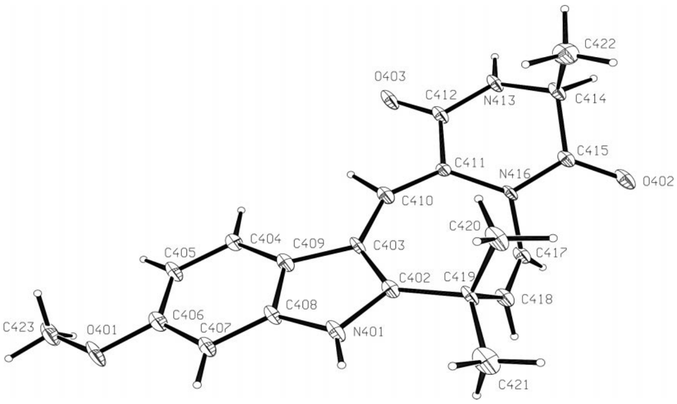


Figure 4
A view of molecule 4 of (I) with the atomic numbering scheme. Displacement ellipsoids are drawn at the 10% probability level.

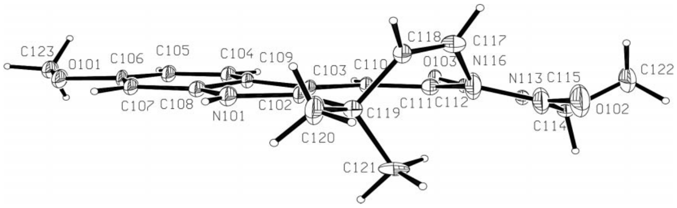


Figure 5
A side view of molecule 1 of (I) with displacement ellipsoids drawn at the 10% probability level.

Refinement on F^2
$R[F^2 > 2\sigma(F^2)] = 0.064$
$wR(F^2) = 0.209$
$S = 0.83$
13028 reflections
973 parameters
H-atom parameters constrained

$w = 1/[\sigma^2(F_o^2) + (0.1209P)^2]$
where $P = (F_o^2 + 2F_c^2)/3$
$(\Delta/\sigma)_{\max} = 0.121$
$\Delta\rho_{\max} = 0.52$ e Å^{-3}
$\Delta\rho_{\min} = -0.31$ e Å^{-3}
Absolute structure: (Flack, 1983), 6232 Friedel pairs
Flack parameter = 2.7 (19)

Table 1
Selected torsion angles (°).

C102—C119—C118—C117	−84.1 (11)	C302—C319—C318—C317	−80.2 (10)
C111—N116—C117—C118	73.0 (13)	C311—N316—C317—C318	76.8 (10)
C202—C219—C218—C217	−84.3 (8)	C402—C419—C418—C417	82.3 (9)
C211—N216—C217—C218	74.5 (9)	C411—N416—C417—C418	−71.4 (10)

Table 2
Hydrogen-bonding geometry (Å, °).

D—H···A	D—H	H···A	D···A	D—H···A
N313—H313···O203$^{\rm i}$	0.86	2.01	2.847 (6)	165
N301—H301···O302$^{\rm ii}$	0.86	2.13	2.919 (6)	153
N201—H201···O202$^{\rm iii}$	0.86	2.20	2.936 (6)	144
N213—H213···O303$^{\rm ii}$	0.86	2.03	2.834 (6)	155
N113—H113···O403	0.86	2.02	2.861 (6)	167
N413—H413···O103	0.86	2.08	2.900 (6)	158
N401—H401···O402$^{\rm iv}$	0.86	2.07	2.859 (6)	152
N101—H101···O102$^{\rm v}$	0.86	2.12	2.928 (6)	156
O501—H501···O103$^{\rm i}$	0.82	1.87	2.677 (6)	167

Symmetry codes: (i) $1-x, \frac{1}{2}+y, 1-z$; (ii) $1-x, y-\frac{1}{2}, 1-z$; (iii) $2-x, \frac{1}{2}+y, -z$; (iv) $1-x, \frac{1}{2}+y, 2-z$; (v) $2-x, y-\frac{1}{2}, 1-z$.

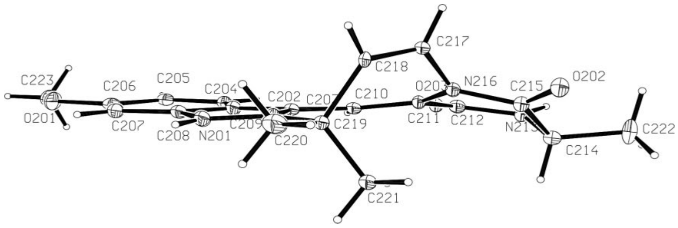


Figure 6
A side view of molecule 2 of (I) with displacement ellipsoids drawn at the 10% probability level.

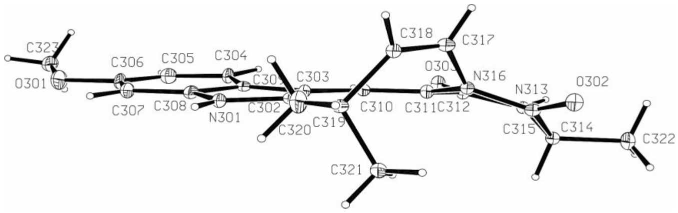


Figure 7
A side view of molecule 3 of (I) with displacement ellipsoids drawn at the 10% probability level.

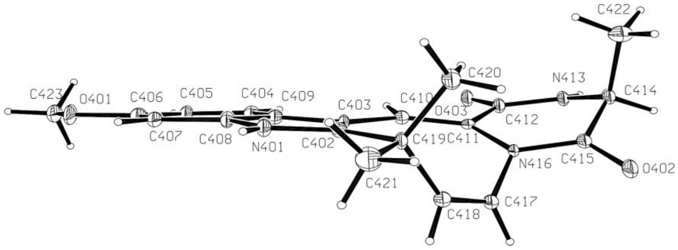


Figure 8
A side view of molecule 4 of (I) with displacement ellipsoids drawn at the 10% probability level.

The large s.u. value of the Flack (1983) parameter, 2.7 (19), indicates that the absolute structure cannot be determined with Mo radiation. The stereostructure of the asymmetric carbon, C114 (C214, C314 or C414) in the Scheme and Figs. 1–8 was just assumed tentatively. The value of $(\Delta/\sigma)_{\max}$ is 0.121 (>0.10) and the C119 has ADP max/min ratio 4.20. These results may be due to the packing of four independent molecules.

Data collection: *CAD*-4-*PC Software* (Enraf–Nonius, 1992); cell refinement: *CELLDIM* in *CAD*-4-*PC Software*; data reduction: *XCAD*4 (McArdle & Higgins, 1995); program(s) used to solve structure: *SHELXS*97 (Sheldrick, 1997); program(s) used to refine structure: *SHELXL*97 (Sheldrick, 1997); molecular graphics: *ORTEX* (McArdle, 1995); software used to prepare material for publication: *OSCAIL* (McArdle, 1993).

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