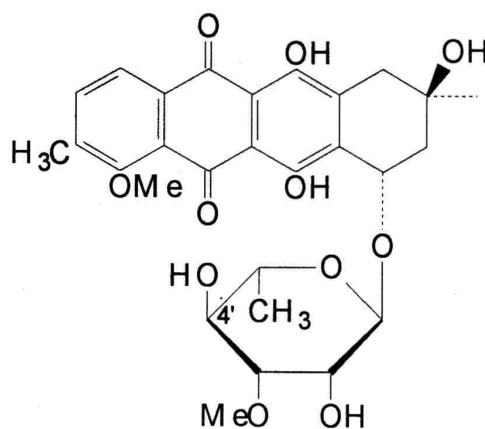
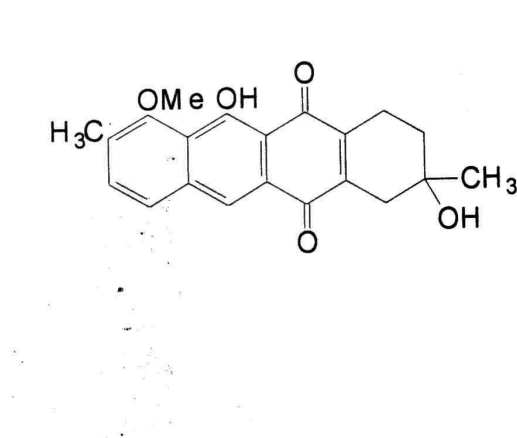


Genus	<i>Nocardia</i>
Species	<i>brasiliensis</i>
Subspecies	
Author	(Lindenberg 1909) Pinoy 1913
Reclassification	
Status	valid
Type species	ATCC 19296, DSM 43758
Hazard group	2 (German classification)



Metabolites described from *N. brasiliensis*

Antibiotic M 4 and Mutactimycin A, two antracycline antibiotics from *N. brasiliensis*. Mutactimycin shows activity against gram-positive bacteria and viruses.



Genus: *Nocardia*

FH 2197

Species: *brasiliensis*

Numbers in other collections: ATCC 19296

Morphology:

	G	R
<u>ISP 2</u>	good	yellow orange
	A	SP
	none	none
	G	R
<u>ISP 3</u>	good	dahlia yellow
	A	SP
	none	none
	G	R
<u>ISP 4</u>	good	dahlia yellow
	A	SP
	none	none
	G	R
<u>ISP 5</u>	good	dahlia yellow
	A	SP
	none	none
	G	R
<u>ISP 6</u>	good	dahlia yellow
	A	SP
	none	none
	G	R
<u>ISP 7</u>	good	orange yellow
	A	SP
	none	none

Spore chains:

Spore surface: smooth

Sporangia:

Fragmentation: +

Melanoid pigment: - - - -

NaCl resistance: 2,5 %

Lysozyme resistance:

pH: Value-

Optimum-

Temperature : Value-

Optimum- 28 °C

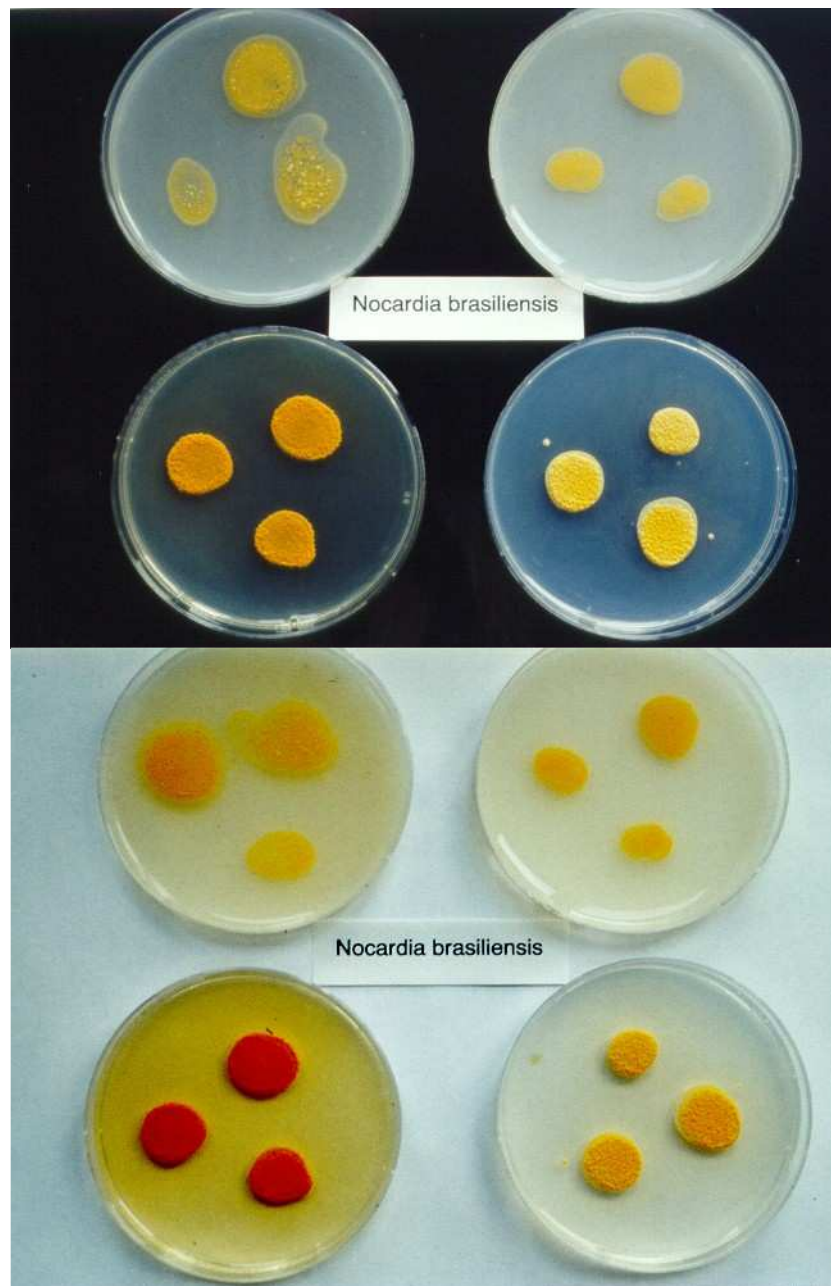
Carbon utilization:

Glu	Ara	Suc	Xyl	Ino	Man	Fru	Rha	Raf	Cel
+	-	-	-	+	-	(+)	-	-	-

Enzymes:

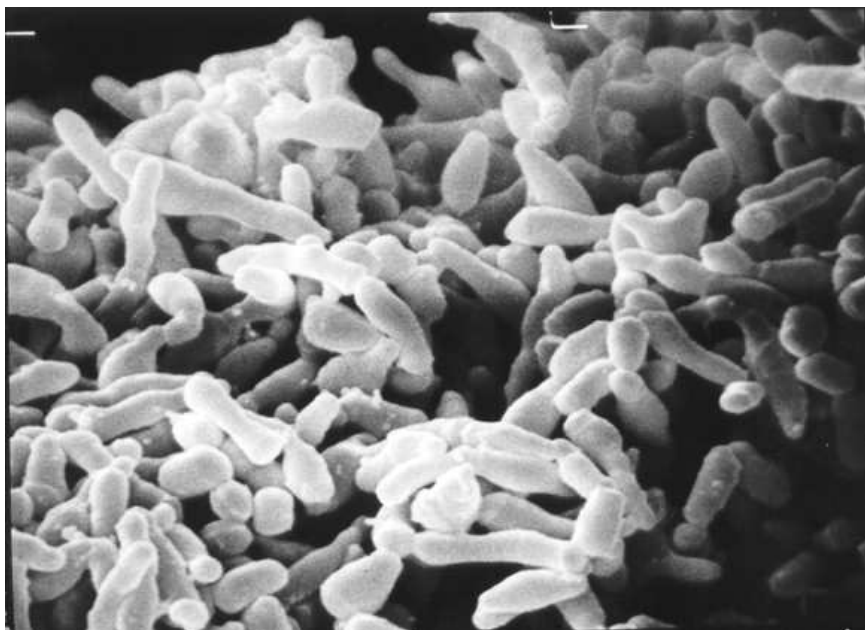
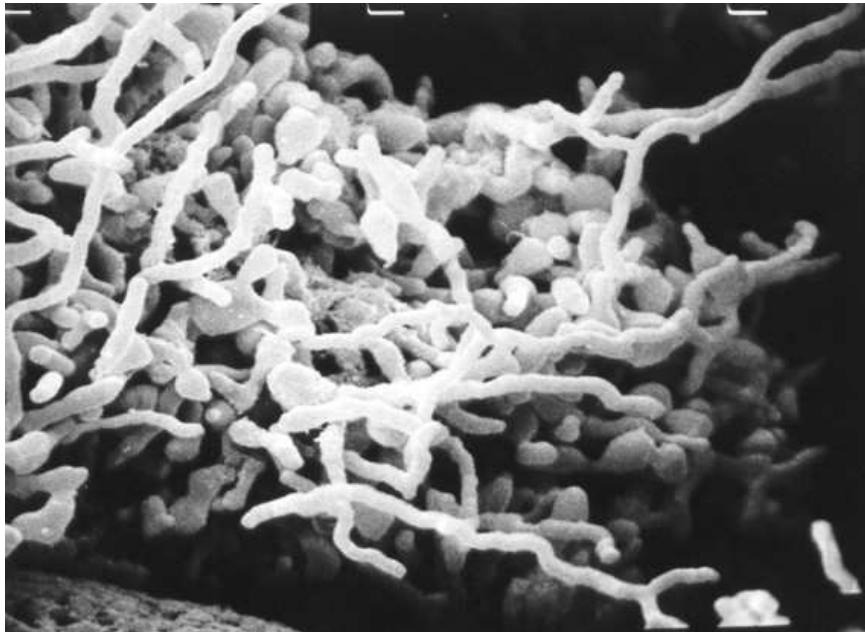
Gel	Cit	Ure	Arg	Onp	Trp	Lys	Odc	VP	Ind	H ₂ S
+	+	+	-	-	-	-	-	-	-	-

Comments:



Nocardia brasiliensis

A and B – Agar plates medium 5317, 5315, 5265 and 5323



Nocardia brasiliensis

Fragmentation of vegetative aerial mycelium in SEM

C x 7.500 D x 10.000

Brasilibactin A, a Cytotoxic Compound from Actinomycete *Nocardia brasiliensis*

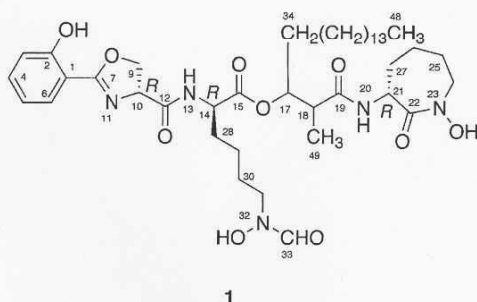
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Received November 12, 2004

A new cytotoxic compound, brasilibactin A (**1**), has been isolated from the actinomycete *Nocardia brasiliensis* IFM 0995, and the structure was elucidated on the basis of spectroscopic data and chemical means.

During our search for bioactive substances from actinomycetes of the genus *Nocardia*, we previously isolated a 32-membered macrolide possessing immunosuppressive and antifungal activity,¹ three biogenetically unique benz[*a*]-anthraquinones with cytotoxic and antibacterial activity,² a cytotoxic indole alkaloid with an isonitrile group,³ a tricyclic terpenoid with immunosuppressive and cytotoxic activity,⁴ and three cytotoxic benzenoid metabolites.⁵ Our recent investigation on extracts of *Nocardia brasiliensis* IFM 0995 resulted in the isolation of brasilibactin A (**1**), a new cytotoxic compound. Here we describe the isolation and structure elucidation of **1**.



The mycelium of *N. brasiliensis* IFM 0995 obtained from 2 L of the culture broth was extracted with MeOH, and the diethyl ether-soluble parts of the extract were subjected to C₁₈ HPLC (CH₃CN/H₂O, 80:20) to yield brasilibactin A (**1**, 33 mg) as a colorless amorphous solid.

The molecular formula of **1** was established to be C₄₂H₆₇N₅O₁₀ by HRFABMS [*m/z* 802.4953, (M + H)⁺, Δ −1.3 mmu]. IR absorptions of **1** indicated the presence of OH and/or NH (3298 cm^{−1}), ester (1738 cm^{−1}), and amide carbonyl (1644 cm^{−1}) groups. UV absorptions [λ_{max} (MeOH) 306 (ε 3000), 260 (sh 2500), 250 (sh 6000), and 244 nm (7200)] of **1** were indicative of a substituted benzenoid chromophore. The ¹H and ¹³C NMR spectra of **1** in DMSO-*d*₆ (see Experimental Section) revealed the presence of signals due to seven sp² quaternary carbons including four

carbonyls, one *N*-formyl, four sp² methines, five sp³ methines, three sp³ methylenes adjacent to heteroatoms, two methyls, and several methylenes in a long alkyl chain. Since nine out of 12 unsaturations were accounted for, **1** was inferred to possess three rings. In the ¹³C NMR spectrum, the *N*-formyl (C-33: δ 161.5, 156.9) and four methylene carbons (C-28: δ 30.12, 30.13; C-29: δ 22.5, 22.3, C-30: δ 26.45, 26.44; C-31: δ 45.4, 48.9) were observed as a set of signals in the ratio of 6:4, indicating the presence of two rotation isomers of the *N*-CHO bond.

¹H–¹H COSY, HSQC, CH- and CH₂-selected editing-HSQC, and CH- and CH₂-selected editing-HSQC-TOCSY spectra revealed six ¹H–¹H connectivities from H-3 to H-6, from H₂-9 to H-10, from NH-13 to H₂-31, from H₃-49 to H₂-36, from H₂-46 to H₃-48, and from NH-20 to H₂-24 (Figure 1). The coupling patterns of H-3 (δ 7.00, d, *J* = 8.3 Hz), H-4 (δ 7.46, brt, *J* = 8.0 Hz), H-5 (δ 6.94, brt, *J* = 8.0 Hz), and H-6 (δ 7.63, d, *J* = 7.7 Hz) and their ¹³C chemical shifts (C-3, δ 116.6; C-4 δ 134.0; C-5 δ 119.0; C-6 δ 128.0) indicated the presence of a 1,2-disubstituted benzene ring. HMBC correlations for H-3/C-1 (δ 109.8), H-4/C-2 (δ 159.1), and H-5/C-1 suggested that the benzene ring possessed a phenolic hydroxyl group at C-2. HMBC correlations of H-6/C-7 (δ 165.9), H₂-9 (δ 4.65 and 4.47)/C-7, and H-10 (δ 5.02)/C-7 implied that the benzenoid ring (C-1 to C-6) was connected to C-9 and C-10 through C-7 and some heteroatoms. The chemical shifts of C-7, C-9 (δ 69.2), and C-10 (δ 67.0) corresponded well to those of a dihydrothiazole ring rather than those of a dihydrothioxazole ring.⁶ The HMBC correlation for H₂-31/C-33 and the NOESY cross-peak for H₂-31/H-33 suggested that the *N*-formyl group was attached to C-31 through N-32. The existence of an ε-aminocaprolactam ring was deduced from HMBC correlations from H-21 and H₂-24 to C-22 (δ 168.9). HMBC correlations for H-10/C-12 (δ 169.9), NH-13/C-12, NH-20/C-19 (δ 172.1), and H₃-49/C-19 indicated that C-10 and C-18 were connected to N-13 and N-20, respectively, through amide carbonyls (C-12 and C-19, respectively), while the presence of an ester bond between C-14 and C-17 was suggested by HMBC correlations for H-14/C-15 (δ 171.5) and H-17/C-15. A long alkyl chain was assigned as a linear C₁₅ chain by FABMS/MS fragment ions as shown in Figure 2. The presence of two hydroxyl groups on N-23 and N-32 were indicated by FABMS/MS fragment ions at *m/z* 145 and 273. The relative stereochemistry at C-17–C-18 was assigned as *erythro* on the basis of *J*-based

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