Journal of Natural Products
Vol. 58, No. 12, pp. 1945-1949, December 1995

TELISATIN A, TELISATIN B, AND TELITOXINONE, THREE NEW APORPHINOIDS FROM *TELITOXICUM PERUVIANUM*

MARY D. MENACHERY,* GEOFFREY W. BLAKE, ROBERT C. GOURLEY,

Penn State-Altoona, 3000 Ivyside Park, Altoona, Pennsylvania 16601-3760

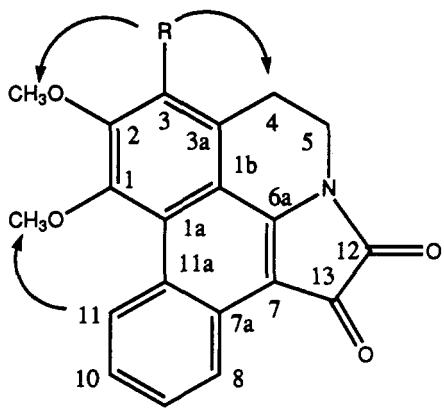
and ALAN FREYER

SmithKline Beecham Pharmaceuticals, UW-2940, 709 Swedeland Road, P.O. Box 1539, King of Prussia, Pennsylvania 19406

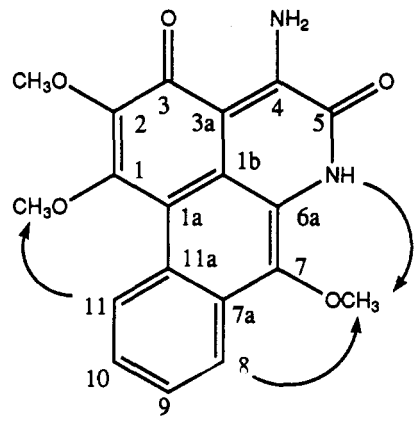
ABSTRACT.—The structures of telisatins A [**1**] and B [**2**] and telitoxinone [**3**], three new aporphinoid alkaloids isolated from a neutral fraction of *Telitoxicum peruvianum*, were determined by spectral data interpretation.

Telitoxicum peruvianum Moldenke (Menispermaceae) was used by the Huitoto Indians of Peru as an ingredient of curare (1). We have reported previously the alkaloids isolated from the basic fraction of this species as azafluoranthenes, oxoaporphines, and other aporphinoids (2). The presence of additional alkaloids in a neutral fraction of *T. krukovii* (3) led us to investigate the same fraction of *T. peruvianum*, which yielded telisatins A [**1**] and B [**2**] (both of which are reported here from a plant source for the first time), along with two oxoaporphines, lysicamine and *O*-methylmoschatoline, as well as telitoxinone [**3**].

Telisatin A [**1**] was crystallized from CH_2Cl_2/MeOH as wine-red crystals. Hreims established the molecular formula as $C_{20}H_{15}NO_4$. The carbonyl peaks at 1751 and 1695 cm^{-1} and the M^+-28 peak (loss of CO) followed by a second loss of 28 atomic mass units in the ms suggested the presence of a dioxo group. The uv spectrum showed an extended conjugation which included two carbonyl groups. In the downfield portion of the ^{1}H-nmr spectrum, four multiplets appeared between δ 9.43 and δ 7.52, along with one narrow triplet at δ 7.18 (exhibiting a 1.0 Hz coupling). The upfield portion of the spectrum consisted of two methoxy singlets at δ 4.11 and δ 3.96 and two methylene multiplets at δ 3.99 (a triplet) and δ 3.36 (a triplet of doublets). The ^{1}H-nmr chemical shift assignments presented in Table 1 are based on homo-decoupling and nOe data. Decoupling experiments established that the four most downfield aromatic mul-



1 R=H
2 R=MeO



3

TABLE 1. ^{1}H- and ^{13}C-Nmr Assignments for Compounds **1–3** in $CDCl_3$.

Position	Compound					
	1		**2**		**3**	
	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C
1		146.6		152.1		160.3
1a		129.3		126.6		113.5
1b		112.2		114.1		119.5
2		157.1		152.0		144.7
3	7.18 (1H, t, J=1.0)	112.3		150.1		179.4
3a		130.7		121.1		106.7
4	3.36 (2H, td, J=6.4, 1.0)	27.6	3.33 (2H, t, J=6.4)	21.2	11.83 (1H, d, J=5.5, NH-4) 7.61 (1H, d, J=5.5, NH-4)	156.8
5	3.99 (2H, t, J=6.4)	36.5	3.91 (2H, t, J=6.4)	36.1		148.6
6					9.88 (1H, s, NH-6)	
6a		153.3		152.6		116.5
7		103.2		104.2		142.0
7a		127.5		126.4		125.0
8	8.65 (1H, dd, J=8.3, 1.8)	123.7	8.61 (1H, dd, J=8.3, 1.8)	123.7	8.17 (1H, dd, J=8.2, 2.1)	121.6
9	7.65 (1H, ddd, J=8.3, 8.3, 1.8)	129.2	7.61 (1H, ddd, J=8.3, 8.3, 1.8)	128.7	7.61 (1H, m)	125.5
10	7.52 (1H, ddd, J=8.3, 8.3, 1.8)	125.6	7.51 (1H, ddd, J=8.3, 8.3, 1.8)	125.8	7.61 (1H, m)	126.9
11	9.43 (1H, dd, J=8.3, 1.8)	128.3	9.37 (1H, dd, J=8.3, 1.8)	127.5	9.44 (1H, dd, J=8.2, 2.1)	127.0
11a		125.8		126.1		129.1
12		160.3		159.8		
13		180.0		180.7		
MeO-1	3.96 (3H, s)	60.0	4.02 (3H, s)	60.5	4.18 (3H, s)	61.1
MeO-2	4.11 (3H, s)	56.6	4.15 (3H, s)	61.4	4.08 (3H, s)	60.5
MeO-3			4.04 (3H, s)	61.3		
MeO-7					4.10 (3H, s)	62.9

tiplets were located contiguously about a single ring and that the narrow triplet at δ 7.18 shared a 1.0 Hz coupling with the δ 3.36 methylene multiplet. NOe saturation of the H-11 doublet of doublets at δ 9.43 enhanced the OMe-1 signal at δ 3.96. Saturation of the H-3 triplet at δ 7.18 enhanced the OMe-2 singlet at δ 4.11 as well as the H-4 methylene multiplet at δ 3.36. The nOe and coupling network together suggested arrangement of the protons in **1** as shown, which was further confirmed by ^{13}C-nmr experiments.

In the downfield portion of the ^{13}C-nmr spectrum of **1**, there were eleven quaternary resonances between δ 180.0 and δ 103.2, along with five protonated signals between δ 129.2 and δ 112.3. In the upfield portion of the spectrum there were two methoxy resonances at δ 60.0 and δ 56.6 and two methylene resonances at δ 36.5 and δ 27.6. The ^{13}C-nmr assignments presented in Table 1 are based on the ^{1}H-nmr assignments and correlation results.

The HMBC data of **1** provided long-range correlations supporting these assignments. Of particular interest were the correlations between H-3 at δ 7.18 and C-2 at δ 157.1, C-1 at δ 146.6, C-3a at δ 130.7, and C-1b at δ 112.2 and C-4 at δ 27.6. The methoxy protons at δ 4.11 and δ 3.96, which were assigned by nOe observations, correlated to the corresponding ring carbons to which they were attached, C-2 at δ 157.1 and C-1 at δ 146.6, respectively. Additionally, the C-4 methylene protons at δ 3.36 correlated to C-3a at δ 130.7, C-3 at δ 112.3, C-1b at δ 112.2, and C-5 at δ 36.5, while the C-5 methylene protons at δ 3.99 correlated to C-6a at δ 153.3, C-3a at δ 130.7, and C-4 at δ 27.6. The aromatic H-11 at δ 9.43 correlated to C-1a at δ 129.3, C-9 at δ 129.2, and C-7 at δ 127.5. In

similar fashion, H-8 at δ 8.65 correlated to C-11a at δ 125.8, C-10 at δ 125.6, and C-7 at δ 103.2.

The assignment of the C-12 and C-13 carbonyls to δ 160.3 and δ 180.0, respectively, was supported by chemical shifts reported for putisatin [**4**], a metabolite of *Streptomyces albus*, which has a similar functionality (4). For putisatin the corresponding resonances were observed at δ 160.79 and δ 182.82 respectively.

A synthetic sample of telisatin A, which has been prepared previously by reaction of dehydronuciferine with oxalyl chloride (5), was found to be identical to **1** by co-tlc, mixed mp, and spectral data. This compound, upon further oxidation with H_2O_2 in the presence of NaOH, gave the oxoaporphine lysicamine as reported in the literature (6).

Telisatin B [**2**] crystallized from CH_2Cl_2/MeOH as reddish-purple needles. The ir spectrum was similar to that of telisatin A [**1**] showing the presence of two carbonyl groups. Telisatin B [**2**] possessed spectral characteristics which were very similar to those of telisatin A [**1**]. The ^{1}H-nmr spectrum of **2** indicated the presence of four aromatic multiplets between δ 9.37 and δ 7.51 (the H-3 aromatic triplet observed in **1** was missing), three methoxy groups (only two were observed for telisatin A), and two methylene triplets at δ 3.91 and δ 3.33 (this resonance in **1** was a triplet of doublets). The additional methoxy group was located at C-3 by mutual nOe enhancements between the H-4 triplet at δ 3.33 and MeO-3 at δ 4.04. The ^{1}H-nmr assignments for **2** are summarized in Table 1.

In the downfield portion of the ^{13}C-nmr spectrum of **2**, there were 12 quaternary resonances between δ 180.7 and δ 104.2, along with four protonated signals between δ 128.7 and δ 123.7. The upfield region consisted of three methoxy resonances at δ 61.4, δ 61.3, and δ 60.5 and two methylene resonances at δ 36.1 and δ 21.2. The ^{13}C-nmr resonances of **2** reflected the presence of one additional methoxy group and one fewer protonated aromatic signal than were observed for **1**. Once again, correlation experiments were employed in making the nmr assignments, which were analogous to those for **1** (see Table 1).

Telitoxinone [**3**], $C_{19}H_{16}N_2O_5$, crystallized from CH_2Cl_2/MeOH as ruby-red needles. The ir spectrum showed the presence of two carbonyl groups at 1670 and 1600 cm^{-1}, noticeably different from the carbonyl values for **1** and **2**. In the downfield portion of the ^{1}H-nmr spectrum there appeared three exchangeable resonances at δ 11.83 (1H, br d, J=5.5 Hz), δ 9.88 (1H, br s), and δ 7.61 (1H, br d) which were not previously observed for **1** or **2** and four aromatic multiplets between δ 9.44 and δ 7.61, reminiscent of the ring-D signals in **1** and **2**. The upfield portion of the ^{1}H-nmr spectrum consisted of three methoxy signals at δ 4.18, δ 4.10, and δ 4.08. Taking into account the molecular formula and the thirteen ring- and double-bond equivalents present, telitoxinone [**3**] appeared likely to be a four-ring dioxoaporphine with three methoxy, two oxo, and one primary amine substituent on rings A–C and four contiguous hydrogens on ring D.

The four ring-D aromatic multiplets were shown to be contiguous by homodecoupling nmr experiments. The H-11 doublet of doublets at δ 9.44 in ring D shared mutual nOes with H-10 at δ 7.61 and MeO-1 at δ 4.18 in ring A, establishing the presence of a methoxy substituent at C-1. Likewise, saturation of the H-8 doublet of doublets at δ 8.17 in ring D enhanced H-9 at δ 7.61 and MeO-7 at δ 4.10 establishing this methoxy group as the sole substituent in ring C. Irradiation of the broad NH-6 singlet at δ 9.88 in ring B also enhanced this same C-7 methoxy group.

The two primary amine doublets at δ 11.83 and δ 7.61 shared a 5.5 Hz coupling constant but exhibited no nOes

with any of the methoxy groups or the NH-6 proton, indicating that the primary amine was probably flanked by two carbonyl substituents. This proposal was buttressed by the noticeably downfield shift of one of the amine protons (δ 11.83) due to strong hydrogen bonding. The arrangement of atoms proposed could be accomplished by placing the two oxo groups at C-3 and C-5, with the primary amine sandwiched between them at C-4 and the residual methoxy group at C-2 in **3** or by placing the two oxo groups at C-2 and C-4 with the primary amine at C-3 and the residual methoxy at C-5. The former proposed structure [**3**] agreed well with the observed nOe data, but the latter proposed structure could not be drawn in a fashion that was consistent with the spectral data. Additionally, the latter structure would give rise to an nOe between the C-5 methoxy protons at δ 4.08 and NH-6 which would be more intense than that observed between NH-6 and MeO-7 because the C-5 methoxy protons and NH-6 would be closer together. No such effect was observed. With the two oxo groups at C-3 and C-5, the C-4 primary amine and the C-2 methoxy group, one would not expect such an nOe enhancement. The C-1 methoxy group had a chemical shift similar to that of the C-2 methoxy group, so mutual effects would not be observed.

The ^{13}C-nmr spectrum of **3** indicated the presence of twelve quaternary resonances between δ 179.4 and δ 106.7, along with four protonated signals between δ 127.0 and δ 121.6. In the upfield portion of the spectrum there were three methoxy resonances at δ 62.9, δ 61.1, and δ 60.5. The observed chemical shifts, HMQC correlations, and HMBC correlations were consistent with the structure suggested by the ^{1}H-nmr data. In a closely related 3,5-dioxoaporphine, telikovinone (3), which had a C-7 proton instead of a methoxy group, the ^{13}C-nmr chemical shifts for C-2, C-3, C-3a, C-4, and C-5 were δ 145.7, 179.3, 106.7, 157.3, and 149.8, respectively. The observed shifts for the corresponding carbon resonances in **3** were, in turn, δ 144.7, 179.4, 106.7, 156.8, and 148.6 The similarity in chemical shifts firmly established the positions of the two oxo groups as well as those of the remaining methoxy and amine functionalities.

To further support these assignments, HMBC correlations were observed between the NH_2-4 protons at δ 11.83 and δ 7.61 and C-4 at δ 156.8 and between the NH-6 proton at δ 9.88 and the C-5 oxo resonance at δ 148.6 and C-6a at δ 116.5 at the B/C ring junction. There were additional long-range correlations of interest. The methoxy protons at δ 4.18, 4.10, and 4.08 (two of which were assigned by nOe enhancements) correlated to the corresponding ring carbons to which they were attached, C-1 at δ 160.3, C-7 at δ 142.0, and C-2 at δ 144.7, respectively. H-8 at δ 8.17 correlated to C-7 at δ 142.0, C-11a at δ 129.1, C-10 at δ 126.9, and C-7a at δ 125.0. H-11 at δ 9.44 correlated to C-11a at δ 129.1, C-9 at δ 125.5, C-7a at δ 125.0, C-1a at δ 113.5, and C-1b at δ 119.5.

It is interesting to note that the telisatins (**1** and **2**) may be the precursors of the corresponding oxoaporphines which we have isolated from this plant. Telitoxinone [**3**] was an analogue of telikovinone with the methoxy group at C-7 (3). In order to determine whether telitoxinone was an artifact of the isolation procedure, 20 g of the small amount of the remaining plant material were extracted using MeOH. Tlc of the resulting crude extract did not show the presence of telitoxinone.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mps are reported uncorrected. ^{1}H- and ^{13}C-nmr spectra were collected using a Bruker AMX400 system at normal probe temperature in $CDCl_3$. Ir spectra were recorded on a Mattson Galaxy series Ft-ir 3000 spectrophotometer and uv spectra on a Hewlett Packard 5842 diode-array uv/visible spectrophotometer. Mass spectra were recorded

on a Kratos-MS 9/50 mass spectrometer at 70 eV.

PLANT MATERIAL.—As described previously (2).

EXTRACTION AND ISOLATION.—The extraction procedure for the plant material was described previously (2). The neutral fraction (42 g) was chromatographed over Si gel-60 by elution with hexane, hexane/EtOAc mixtures, EtOAc, and EtOAc/MeOH. All the alkaloids were obtained in pure form only after a second and/or third chromatographic separation and recrystallization.

Telisatin A [**1**].—Wine-red crystals (14 mg); mp 238–239° (dec) (CH_2Cl_2/MeOH), [lit. (5) mp 233–234°]; ir (KBr) ν max 1751, 1695 cm^{-1}; eims *m/z* $[M]^+$ 333 (100), 305 (50), 277 (11), 219 (17), 190 (12), 163 (13); hreims *m/z* calcd for $C_{20}H_{15}NO_4$ 333.0997, found 333.0971; uv λ max (EtOH) 258 (log ϵ 4.11), 284 sh (3.48), 324 sh (3.61), 336 (3.68), 352 sh (3.45), 504 (3.13) nm; ^{1}H- and ^{13}C-nmr data, see Table 1.

Telisatin B [**2**].—Reddish-purple needles (4.3 mg); mp 221–222° (CH_2Cl_2/MeOH); ir (KBr) ν max 1750, 1703 cm^{-1}; eims *m/z* $[M]^+$ 363 (100), 335 (59), 307 (7), 276 (8), 234 (10); hreims *m/z* calcd for $C_{21}H_{17}NO_5$ 363.1102, found 363.1116; uv λ max (EtOH) 258 (log ϵ 4.66), 318 sh (4.09), 330 (4.19), 344 sh (3.91), 406 (3.48), 502 (3.56) nm; ^{1}H- and ^{13}C-nmr data, see Table 1.

Telitoxinone [**3**].—Ruby-red needles (11.6 mg); mp 247–248° (dec) (CH_2Cl_2/MeOH); ir (KBr) ν max 3420, 3380, 1670, 1600 cm^{-1}; eims *m/z* $[M]^+$ 352 (100), 337 (82), 322 (15), 309 (52), 294 (26), 266 (21), 251 (17); hreims *m/z* calcd for $C_{19}H_{16}N_2O_5$ 352.1055, found 352.1048; uv λ max (EtOH) 242 (log ϵ 4.01), 262 (4.31), 284 (3.91), 308 (3.56), 324 (3.61), 388 (3.59), 406 (3.83), 426 (3.88), 470 (3.35) nm; ^{1}H- and ^{13}C-nmr data, see Table 1.

IDENTIFICATION OF KNOWN COMPOUNDS.—The structures of lysicamine (0.7 mg) and *O*-methylmoschatoline (1.0 g) were confirmed by comparison (^{1}H-nmr, ir, uv, ms, and co-tlc) with authentic samples.

ACKNOWLEDGMENTS

We are grateful to Drs. Michael P. Cava and M.V. Lakshmikantham, University of Alabama, for the supply of the plant extract and the known compound sample. M.D.M. would like to acknowledge the generous support of the Altoona Campus Advisory Board for the Endowment Fund.

LITERATURE CITED

1. B.A. Krukoff and R.C. Barneby, *Mem. N.Y. Bot. Gard.*, **20**, 1 (1970).
2. M.D. Menachery and M.P. Cava, *J. Nat. Prod.*, **44**, 320 (1981).
3. M.D. Menachery, G.W. Blake, C. Beiswenger, and A. Freyer, *Heterocycles*, **41**, 1425 (1995).
4. U. Grufe and L. Radics, *J. Antibiot.*, **39**, 162 (1986).
5. J.M. Saá and M.P. Cava, *J. Org. Chem.*, **43**, 1096 (1978).
6. L. Castedo, C. Saá, J.M. Saá, and R. Suau, *J. Org. Chem.*, **47**, 513 (1982).

Received 2 June 1995