Secondary Metabolites from Basotho Medicinal Plants. I. *Bulbine narcissifolia*

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The medicinal plant *Bulbine narcissifolia* is used by the Basotho, Griqua, and whites of southern Africa for wound healing and as a mild purgative. Extraction of the powdered root has yielded acetosyringone, chrysophanol, knipholone, isoknipholone, 10,7′-bichrysophanol, and chrysalodin in addition to two new anthraquinone glycosides, knipholone-8-*O*-*β*-D-gentiobioside (**1**) and chrysalodin-10-*β*-D-gentiobioside (**2**). NMR spectroscopy was used to elucidate the structures of **1** and **2** and to show that **1** binds weakly to DNA.

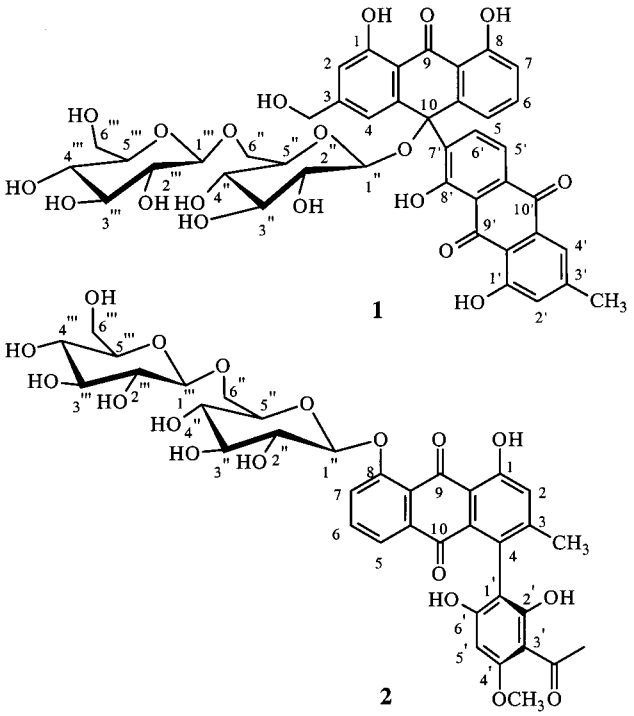
Bulbine narcissifolia Salm-Dyck, referred to in Sesotho as "khomo-ea-balisa", is an herbal medicine used in Lesotho for its laxative effect.[1] Basotho traditional doctors believe that purgation is effective in clearing gonorrhea infections, which it is believed are sometimes responsible for blockage of fallopian tubes and hence infertility in women.[2]

The genus *Bulbin*e belongs to the family Liliaceae, with *B. narcissifolia* reported to be much used as a medicine by the European South Africans, the Basotho, and the Griqua peoples.[3] The people of the Transvaal region in South Africa use the fresh sap of the plant to hasten the healing of wounds, the application of which produces smarting. In East Griqualand, the sap is used as a wart and corn remedy as well as being applied to ringworm, gravel rash, and other minor wounds.[3] An extract from a related species (*B. fruitescens*) has recently been patented as a gel to promote wound healing.[4] A decoction of the root is taken for the relief of rheumatism, and a cold infusion of the fresh leaf is used as a purgative. A related species, *B. bulosa*, is used as a food plant by Australian Aboriginals.[5]

The purgative properties of *B. narcissifolia* are considered mild enough for use by pregnant women.[2] A decoction of the roots is also given to induce pregnancy in barren women and barren cows. *Bulbine narcissifolia* is believed to have a "cleansing" effect to the womb when taken just before impregnation.[6,7]

This report represents the first chemical investigation of *B. narcissifolia*, though during the course of this research, van Wyk et al. surveyed chemotaxonomically the genus using TLC and comparative HPLC to tentatively identify chrysophanol, knipholone, isoknipholone, and 10,7′-bichrysophanol in *B. narcissifolia*.[8] In our extracts of this plant, we have isolated and confirmed the presence of the same compounds in addition to 2,6-dimethoxy-4-hydroxyacetophenone and chrysalodin. In addition, glycosides of chrysalodin and knipholone were isolated for the first time from the polar fractions.

Bulbine narcissifolia was collected from Thaba Putsoa in Lesotho, and the separated roots were washed and air-dried. The light petroleum and ethyl acetate extracts yielded the known compounds chrysophanol, knipholone,[9] isoknipholone,[10] chrysalodin,[11] 10,7′-bichrysophanol,[12] and 2,6-dimethoxy-4-hydroxyacetophenone.[13] These compounds were identified on the basis of published spectral data. 2,6-Dimethoxy-4-hydroxyacetophenone was isolated for the first time from *Bulbine* spp. Closely related phloroglucinols and chrysophanol are the likely biogenetic precursors of knipholone and isoknipholone.[9] The acetone/water extract yielded two new compounds, **1** and **2**.



1

2

Compound **1** was isolated from the aqueous acetone extract as an optically active yellowish brown solid with a strong positive Cotton effect in the neighborhood of 260 nm. The compound exhibited UV−vis spectra characteristic of anthraquinones. In neutral ethanol, a visible quinonoid band appeared at 409 nm and shifted to 501 nm in alkali, suggesting the presence of an anthraquinone nucleus with one H-bonded hydroxyl substituent.[14,15] This was supported by the IR spectrum, which showed two carbonyl bands at 1675 and 1623 cm^{-1} due to the stretching of non-hydrogen-bonded and hydrogen-bonded carbonyls, respectively.[16] In the ^{13}C NMR spectrum, two carbonyl signals at 187.9 and 183.5 ppm were observed, the former being due to a quinonoid carbonyl that is hydrogen-bonded to one hydroxyl, while the latter was due to a free quinonoid carbonyl.[17] However, the ^{1}H NMR spectrum showed two signals due to hydrogen-bonded phenolic hydroxyls at 14.15 and 13.09 ppm.[18] One must be H-bonded to the quinonoid

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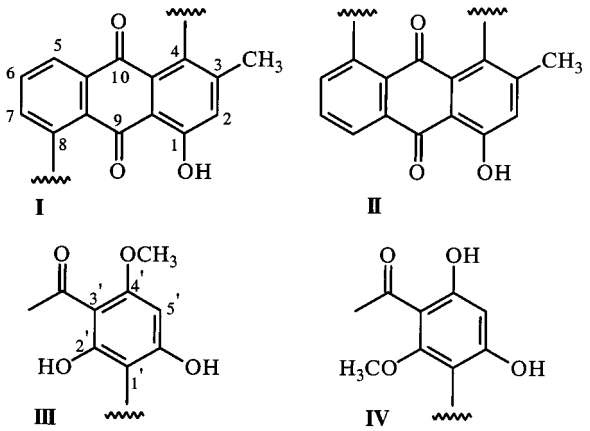
carbonyl at 187.9 ppm and the other to the aromatic ketone (δ_C 202.3). In the aromatic region of the ^{1}H NMR spectrum were five signals: an AMX system (δ 7.96, 7.82, and 7.70), a broad singlet at 7.26 ppm, and a sharp singlet at 6.23 ppm. The broad singlet was correlated to an aromatic methyl (δ_C 20.1) and the three quaternary aromatic carbons, with one (δ_C 161.6) bearing one of the H-bonded phenols (δ_H 13.09) in the HMBC spectrum. These data are consistent with partial structure **I** or **II**.



In the HMBC spectrum the "X" proton of the AMX system at δ 7.70 showed a strong ($^3J_{CH}$) correlation to the non-hydrogen-bonded quinonoid carbonyl at 182.6 ppm, thus eliminating **II** as a possibility.

The ESIMS showed a weak [M + H$^+$] ion at *m*/*z* 759 but a strong [M + Na$^+$] ion at *m*/*z* 781 corresponding to a molecular mass of 758. High-resolution electrospray, Fourier transform ion cyclotron resonance mass spectrometry (ESI-FT/ICR MS) of the [M + Na$^+$] ion suggested a molecular formula of $C_{36}H_{38}O_{18}$. The base peak of the ESIMS (*m*/*z* 435) corresponded to loss of $C_{12}H_{20}O_{10}$ (324 amu) from the [M + H$^+$], peak suggesting a disaccharide moiety.

Two anomeric protons appeared at δ 4.42 (J_{HH} 7.87 Hz) and 5.02 (d, J_{HH} 7.74 Hz) in the ^{1}H NMR spectrum. The trans-diaxial coupling constants suggested two β-hexose linkages.[19] NMR assignment of the dihexose (Table 1) was achieved through HSQC and TOCSY spectra. The strong H−H coupling between adjacent protons suggested a Glc-Glc structure (gentiobiose). This was supported by comparison of ^{1}H and ^{13}C chemical shifts of a large range of disaccharides.[20,21] The HMBC spectrum (Figure S1; Supporting Information) indicated that the anomeric proton at 5.03 ppm (H-1″) was coupled to C-8 of the anthraquinone nucleus, confirming the site of attachment. In addition, both H-6″ protons were coupled to the other anomeric carbon (C-1‴; δ_C 103.5). A (1→6) linkage was also indicated by the chemical shift (^{13}C) of C-6″, which was 7.6 ppm downfield of that for glucose, indicative of etherification at C-6″.[22] An 8-*O*-β-D-gentiobioside unit was confirmed by comparison of ^{13}C NMR chemical shifts with those of the *O*-β-D-gentiobioside of the flavonoid quercetin.[23]

From the mass spectrum, the remaining mass (181 amu) would be a fragment $C_9H_9O_4$. This is composed of one strongly hydrogen-bonded phenolic proton (δ_H 14.15), an acetyl group (δ_H 2.62, δ_C 32.6, 202.3), a methoxyl group (δ_H 3.96 and δ_C 55.58), a non-hydrogen-bonded phenolic proton (δ_H 9.16, broad), and an aromatic singlet (δ_H 6.23). This suggested a phloroglucinol group either of the knipholone (**III**) or isoknipholone (**IV**) type. Briefly, the chemical shift of the aromatic singlet (δ_H 6.23) in the ^{1}H NMR spectrum suggested that it lies between two oxygen-bearing carbons to explain the upfield shift. The acetyl group and one of the phenolic protons must be *ortho* (hydrogen-bonding) and the other *meta* or *para*. As there is only one hydrogen-bonded phenolic proton, the methoxy must also be *ortho* to the acetyl group as shown in **III** and **IV**.

Table 1. ^{1}H and ^{13}C NMR Data of Knipholone-8-*O*-β-D-gentiobioside (**1**) Measured in Acetone-d_6 at 150.92 MHz

position	δ_C (**1**) (ppm)	δ_H (**1**) (ppm)	HMBC correlations
1	161.6	13.09(OH)	C-1, C-2, C-3
2	124.3	7.26	C-1, C-4, C-8a, ArMe
3	149.3		
4	126.6		
5	122.1	7.70	C-7, C-9a
6	136.1	7.82	C-8, C-10a
7	124.2	7.96	C-6, C-8, C-5, C-9a
8	157.5		
9	187.9		
10	182.6		
10a	135.6		
9a	120.3		
8a	115.6		
4a	131.0		
ArMe	20.1	2.08	C-2, C-3, C-4
ArOMe	55.6	3.96	C-4′
ArC(O)*Me*	32.6	2.61	C-3′, Ar*C*(O)Me
Ar*C*(O)Me	202.3		
1′	107.2[a]		
2′	163.1	14.15(OH)	
3′	104.7[a]		
4′	162.3		
5′	91.0	6.23	C-1′, C-3′, C-4′, C-6′, C-4, Ar*C*(O)Me
6′	161.1	9.16 (OH)	C-1′, C-5′, C-6′
1″	100.6	5.03	C-8
2″	73.3	3.65	C-1″
3″	76.5	3.83	
4″	70.1	3.32	C-5″
5″	76.8	3.34	C-4″
6″a	68.6	3.78	C-1‴, C-5″
6″b	68.6	4.19	C-1‴
1‴	103.5	4.43	C-6″
2‴	73.6	3.21	C-1‴
3‴	76.6	3.23	
4‴	69.8	3.43	
5‴	76.2	3.54	C-2‴
6‴a	61.0	3.81	
6‴b	61.0	3.62	

[a] Assignments of C-1′ and C-3′ have been interchanged in ref 24.

The non-hydrogen-bonded OH (δ_H 9.16) showed a strong correlation to C-5′ (δ_H 91.0, δ_H 6.23) in the HMBC spectrum. H-5′ in turn was coupled to C-3′, C-1′, C-4′, and C-6′. These correlations would be observed for both **III** and **IV**. However, the methoxyl was coupled only to C-4′, eliminating **IV**. H-5′ also showed a weak coupling to C-4 of the anthraquinone nucleus. In addition, the chemical shifts of the methoxyl (δ_H 3.96, δ_C 55.6) are typical of knipholone (δ_H 3.90. δ_C 55.6), but not isoknipholone (δ_H 3.29, δ_C 60.7),[24] thus establishing the structure as knipholone-8-*O*-β-D-gentiobioside (**1**). In addition, the ^{13}C (Table 1) and ^{1}H NMR chemical shifts of **1** matched those of knipholone.[24] Notable exceptions were C-8, C-9, and C-9a, which are shifted by the 8-*O*-β-D-gentiobioside unit.

The stereochemistry about the biaryl bond was determined as *M* based on comparisons of our CD spectrum with (+)-knipholone and the calculated spectrum for *M*-knipholone.[25] The spectra for **1** and (+)-knipholone are quite similar and correlate well with the calculated CD of *M*-knipholone (Figure S2; Supporting Information). Specifically, the negative peaks at 215 and 250 nm and the

positive Cotton effect at 200 nm are convincing. In contrast, the negative Cotton effect at 260 nm, so characteristic of these compounds, was much more intense than that predicted in the calculated spectrum. The calculated spectrum for *P*-knipholone did not correlate at all with the experimental spectrum.

The second new compound (**2**) was isolated as a light-brown amorphous solid showing UV−vis spectra characteristic of anthraquinones. The spectrum recorded in neutral ethanol showed the characteristic anthraquinonoid bands of 1,8-dihydroxy-substituted anthraquinones at 291 and 433 nm[15] as well as two benzenoid bands at 262 and 369 nm and was essentially identical to that of chrysalodin.[15] In alkaline ethanol the visible quinonoid band shifted to 530 nm, characteristic of 1,8-dihydroxy-substituted anthraquinone nuclei.[26]

In the ^{1}H NMR spectrum, a broad singlet integrating for two protons appeared at δ_H 4.78. A similar signal (δ_H 4.60) was assigned to the benzylic methylene of chrysalodin. The aromatic region was similar to those of 10,7′-bichrysophanol, chryslandicin, or chrysalodin, showing nine signals: an AB system at δ 8.83 and 7.97 (J = 7.68 Hz) and an ABX system at 7.59 and 7.18 ppm. The remaining signals appeared as broad singlets at 7.34, 7.11, 6.97, and 6.89 ppm. Four H-bonded phenolic protons (δ_H 15.12, 12.84, 12.22, and 11.64) confirmed the presence of two 1,8-dihydroxy anthroquinones. However, the presence of only one aromatic methyl (δ_H 2.42, δ_C 20.9) and a benzylic alcohol or ether (δ_H 4.78, δ_C 61.0) ruled out a 10,7′-bichrysophanol and chryslandicin-type skeletons and confirmed a chrysalodin skeleton.[11]

The negative-ion ESIMS showed an $[M - H]^-$ ion at *m/z* 847 and a fragmentation peak at *m/z* 521 corresponding to loss of a dihexose unit of formula $C_{12}H_{22}O_{10}$ (326 amu). No molecular ion was observed in the positive ion spectrum, but a peak at *m/z* 871 was confirmed as the $[M + Na]^+$ ion by addition of potassium chloride, whereupon another ion at 887 amu appeared. MS-MS of the *m/z* 871 ion showed loss of 326, as was observed in the negative-ion spectrum. ESI-FT/IRC MS of the $[M + Na]^+$ ion confirmed a molecular formula of $C_{42}H_{40}O_{19}$, which minus $C_{12}H_{22}O_{10}$ (the disaccharide) left a bianthraquinone fragment.

The point of attachment of the disaccharide was obtained from evidence present in the ^{1}H NMR spectrum. The presence of four H-bonded phenols excluded attachment at any of the phenolic oxygens, leaving only the two alcoholic carbons. Of particular note was the chemical shift and redundancy of the protons on benzylic methylene. Attachment of a disaccharide at this carbon would result in hindered rotation about the glycoside and splitting of the methylene protons. Typically, these are observed at 4.8/5.0 ppm (J = 13 Hz).[27,28] In contrast, unsubstituted hydroxymethylenes are singlets at 4.5−4.8 ppm.[29,30] For **2**, a broad singlet, integrating for two protons at 4.77 ppm, was observed. In addition, the tertiary hydroxy proton observed at δ_H 2.88 in the spectrum of chrysalodin was absent, leading to the conclusion that the disaccharide was attached at C-10. The CD spectrum of **2** and chrysalodin exhibited the same negative Cotton effect in the neighborhood of 260 nm, indicating that the stereochemistry at C-10 was identical in both compounds.

The disaccharide signals in the ^{13}C and ^{1}H NMR spectra were virtually identical to those of **1**, leading to the conclusion that both contained β-D-gentiobioside. These data identified this new compound as chrysalodin-3-*O*-β-D-gentiobioside (**2**).

It has been suggested that the cytotoxicity and hence anticancer activity of anthraquinone glycosides is a result of formation of anthraquinone-DNA complexes.[31] Investigation of the DNA binding properties of knipholone-8-*O*-β-D-gentiobioside (**1**) with the oligonucleotide [d(ATATGCATAT)]$_2$ was carried out. Upon addition of excess oligonucleotide, H-6 and H-7 suffered substantial broadening and shifted upfield. In contrast, H-2 shifted only slightly and was not broadened (Figure 3S; Supporting Information). These data are consistent with shielding of ring A by the DNA due to groove binding or intercalation.[32] However, a 2D NOESY spectrum (Figure 4S; Supporting Information) of the mixture revealed no intermolecular NOEs, but it was noted that the anthraquinone showed many intramolecular NOEs that were strong and positive. In contrast, free anthraquinone gave only weak negative NOEs, indicating that in the presence of DNA the correlation time of the anthraquinone had increased, indicative of an association.[32]

Experimental Section

General Experimental Procedures. Melting points were obtained on a Gallenkamp melting point apparatus and were not corrected. Optical rotation measurements were obtained from 1,4-dioxane solutions using an AA-10 automatic polarimeter. UV spectra were recorded in 100% ethanol on a Cary 1E UV−vis spectrometer using Varian software. Circular dichroism measurements were obtained on a JASCO Model J-500C spectropolarimeter in acetone or methanol solutions. IR spectra (KBr disk) were recorded on a Paragon Perkin-Elmer 1000FT IR spectrometer using Grams Analyst software. 1D ^{1}H NMR spectra were obtained on a Varian XL400 400 MHz NMR spectrometer using VXR series software. 2D NMR spectra were obtained on a Bruker DMX600 600 MHz NMR spectrometer. All spectra were recorded in deuterated solvent (Aldrich 99.99%) using the solvent as internal reference, with chemical shifts recorded in ppm relative to TMS. For HMBC experiments, multiple bond coherence was optimized for a coupling constant of 8.3 Hz and direct (HSQC) spectra were optimized for a coupling ($^1J_{CH}$) of 145 Hz. The LC/MS spectra were recorded on a Fisons Instruments Quattro II triple quadrupole mass spectrometer using electrospray and electron impact ionization techniques. In most cases, both the $[M + H]^+$ peak and a fragmentation pattern of the molecular ion peak (MS/MS) were obtained. High-resolution electrospray ionization Fourier transform ion cyclotron resonance mass spectrometry (HRESI-FT/ICR MS) was performed on a Bruker Daltonics (Billerica, MA) BioAPEX II FT/ICR mass spectrometer equipped with a 7 T superconducting magnet (Magnex, Yarnton, UK) and an Analytica (Branford, CT) external ESI source. An rf/dc hexapole ion trap at the end of the ESI source was used to accumulate ions before they were pulsed out as an ion packet to the ICR cell.

Column chromatography was performed on Si gel type 60 (Merk 230−400 Mesh ASTM). HPLC was performed using a Waters 600 solvent delivery system equipped with a Millipore SDM-Milligen/Biosearch solvent programmer, a Knauer variable wavelength monitor, and a Shimadzu C-R6A chromatopac recorder. Analytical separations were performed on Alltech Econosphere columns (250 × 4.6 mm Si gel 5 μm or C_{18} 5 μm) with a flow rate of 1 mL/min. Preparative separations were then performed on the corresponding Alltech Econosil columns (250 × 22.5 mm, 10 μm) using a flow rate of 10 mL/min. All solvents were AR grade and redistilled before use. Ethyl acetate and methanol were distilled from anhydrous potassium carbonate and petroleum ether from sodium wire. Petroleum ether had a boiling point of 65−69 °C. Water was distilled and then deionized using a Milli-Q system. Acetonitrile (190 nm HPLC grade) was not purified before use.

Plant Material. *Bulbine narcissifolia* was collected from the north-facing slope of Thaba Putsoa, Lesotho, in Novem-

ber 1992. The plant sample was identified by Prof. C. R. Nagendran at the National University of Lesotho, Roma, Lesotho. A specimen sample is kept at the Roma Herbarium (Lesotho), specimen number B. Hargreaves 3453.

Extraction and Isolation. The fresh roots of *B. narcissifolia* were washed with water and left to air-dry at room temperature (indoors). The air-dried roots (365 g) were ground into a powder and subjected to sequential extraction (Soxhlet) with petroleum ether, ethyl acetate, and 95% acetone/water. The extracts were evaporated in vacuo, and any residual water was removed by lyophilization. The petroleum ether extract (0.55 g) was dissolved in toluene and chromatographed (Biobeads S-X3; toluene) to yield three fractions. The third (low molecular weight) fraction from gel filtration was flash chromatographed on reversed-phase (C_{18}) Si gel eluting with 60% aqueous acetonitrile and by Si gel HPLC (petroleum ether/chloroform) to yield chrysophanol as a yellow amorphous solid (5 mg; 0.001% dry weight): mp 195−196 °C (lit.[15] mp 196 °C).

The ethyl acetate extract (1.25 g) was partitioned between ethyl acetate and water, and the organic phase was chromatographed (Sephadex LH-20; methanol) to yield three fractions. The highest molecular weight fraction was flash chromatographed on Si gel (30−100% ethanol/dichloromethane) and finally purified by preparative HPLC (C_{18}; 70−100% acetonitrile) to yield (−)-10,7′-bichrysophanol (44 mg; 0.012% dry weight), as a pale yellow powder: mp 225−226 °C (dec), $[\alpha]^{18}_D$ −41° (*c* 0.0024, dioxane) (lit.[12] 225−227 °C (dec) $[\alpha]^{25}_D$ −30°). A more polar flash chromatography fraction yielded after HPLC (C_{18}, 80−90% methanol) chrysalodin (7 mg; 0.002% dry weight): mp 223−226 °C; lit.[11] 223−228 °C. The second fraction from gel filtration was flash chromatographed on reversed-phase Si gel (C_{18}; 50−70% methanol/water) to yield two fractions. The more polar was rechromatographed on reversed-phase Si gel (C_{18}, 65% methanol) to yield isoknipholone (4 mg; 0.001% dry weight), mp 251.5 °C; lit.[10] 251.5 °C, and knipholone (1.5 mg; 0.0004% dry weight), mp 251.5 °C; lit.[9] 251.5 °C. The last fraction from gel filtration upon chromatography (C_{18}, aqueous ethanol) and refrigeration yielded a fraction that precipitated 2,6-dimethoxy-4-hydroxyacetophenone as colorless crystals (300 mg; 0.082% dry weight): mp 76−78 °C, lit.[33] mp 76−78 °C.

The 95% acetone/water extract (9.75 g) was partitioned between ethyl acetate and water and the organic phase was chromatographed (Sephadex LH-20; methanol) to yield two fractions. The higher molecular weight fraction was chromatographed over Si gel, and an orange fraction, eluting with 100% ethanol, was purified by HPLC (C_{18}, 50−100% methanol) to yield knipholone-8-*O*-*β*-D-gentiobioside (**1**) (60 mg; 0.016% dry weight) as an orange powder and chrysalodin-*β*-D-gentiobioside (**2**) as a pale yellow amorphous solid (18.1 mg; 0.049% dry weight).

Knipholone-2-*O*-*β*-D-gentiobioside (1): orange powder from water; UV (ethanol) λ_{max} 252, 284, 332, 409 nm; UV (alkaline ethanol) λ_{max} 244, 252, 329, 500 nm; IR (KBr) ν_{max} 3380, 2925, 1675, 1623 cm^{-1}; ^1H NMR (acetone-d_6, 600.13 MHz) δ 14.15 (1H, s, OH-2′), 13.09 (1H, s, OH-1′), 9.16 (1H, brs, OH-6′), 7.96 (1H, dd, J = 8.1, 1.1 Hz, H-7), 7.82 (1H, dd, J = 8.1, 7.7 Hz, H-6), 7.70 (1H, dd, J = 7.7, 1.1 Hz, H-5), 7.26 (1H, brs, H-2), 6.23 (1H, s, H-5′), 5.03 (1H, d, J = 7.74 Hz, H-1″) 4.43 (1H, d, J = 7.87 Hz, H-1‴), 4.19 (1H, m, H-6″), 3.96 (3H, s, OMe), 3.83 (1H, m, H-3″), 3.81, (1H, m, H-6‴), 3.78 (1H, m, H-6″), 3.65 (1H, m, H-2″), 3.62 (1H, m, H-6‴), 3.54 (1H, m, H-5‴), 3.43 (1H, m, H-4‴), 3.34 (1H, m, H-5″), 3.32 (1H, m, H-4″), 3.23 (1H, m, H-3‴), 3.21 (1H, m, H-2‴), 2.61 (3H, s, COMe), 2.08 (3H, brs, Ar-Me); ^{13}C NMR (acetone-d_6, 125.8 MHz), see Table 1; ESIMS *m*/*z* (positive ion) 759 (18, $[M + H^+]$), 435 (100); ESI-FT/ICR MS) *m*/*z* 781.1959 $[M + Na^+]$ (calcd for $C_{36}H_{38}O_{18}Na$, 781.1950).

Chrysalodin-10-*β*-D-gentiobioside (2): pale yellow amorphous solid from water (18.1 mg; 0.015% dry weight); UV (ethanol) λ_{max} 227, 262, 291, 369, 433 nm; UV (alkaline ethanol) λ_{max} 223, 424, 530 nm; IR (KBr) ν_{max} 3441, 1684, 1670, 1217 cm^{-1}; ^1H NMR (acetone-d_6, 600.13 MHz) δ 15.12 (1H, s, OH, OH-8), 12.84 (1H, s, OH-1), 12.22 (1H, s, OH-8′), 11.64 (1H, s, OH-1′), 8.83 (1H, d, J = 7.68 Hz, H-6′), 7.97 (1H, d, J = 7.68, H-5′), 7.59 (1H, m, H-4′), 7.34 (1H, brs, H-6), 7.18 (1H, brs, H-2′), 7.11 (1H, brs, H-7), 6.97 (1H, brs, H-5), 6.89 (1H, brs, H-2), 6.10 (1H, brs, H-4), 4.97 (1H, d, J = 7.77 Hz, (H-1″/H-1‴), 4.78 (2H, brs, Ar−CH_2O), 4.6−3.1 (20H, H-2″-H-6″/H-2‴−H-6‴), 2.42 (3H, s, ArMe′); ^{13}C NMR (acetone-d_6, 125.8 MHz) δ 181.9 (C-10′), 161.1 (C−O), 157.6 (C−O), 151.0 (C-3/3′), 135.2, 133.0, 132.7, 132.6, 132.5, 124.0, 121.59, 120.1, 119.0, 115.9 (Ar−CH), 115.7, 114.6, 113.9, 103.0, 103.4 (C-1‴), 100.9 (C-1″), 76.8, 76.6, 76.3, 76.1, 73.5 (C-2‴), 73.4 (C-2″), 70.1 (C-4″), 69.8 (C-4‴) 68.5 (C-6″), 62.3 (C-6‴), 61.0 (Ar−CH_2O), 22.0 (ArMe′); ESIMS *m*/*z* (negative ion) 847 ($[M - H^+]$, 32), 585 (12), 553 (45), 521 (84); positive ion ESIMS *m*/*z* (positive ion) 871 (25) $[M + H + Na]^+$, 529 (20), 347 (100); ESI-FT/ICR MS *m*/*z* 871.2057 $[M + Na^+]$ (calcd for $C_{42}H_{40}O_{19}Na$, 871.2056).

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Supporting Information Available: A detail from the HMBC spectrum of **1**, CD spectra for knipholone and **1** compared to a calculated spectrum for (+)-knipholone, ^1H proton NMR spectrum of **1** with the oligonucleotide $[d(ATATCGATAT)]_2$, a NOESY spectrum of **1** with this oligonucleotide, and full spectral data for all known compounds isolated are supplied. This information is available free of charge via the Internet at http://pubs.acs.org.

References and Notes

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