

REGIOCONTROLLED TOTAL SYNTHESIS OF THE TROPOLO-ISOQUINOLINE ALKALOIDS IMERUBRINE AND GRANDIRUBRINE

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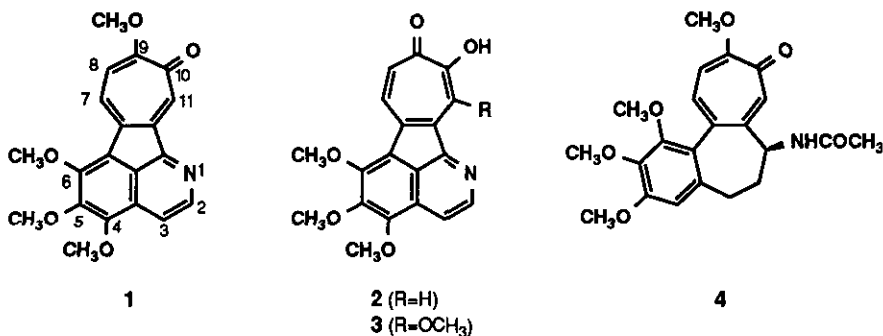
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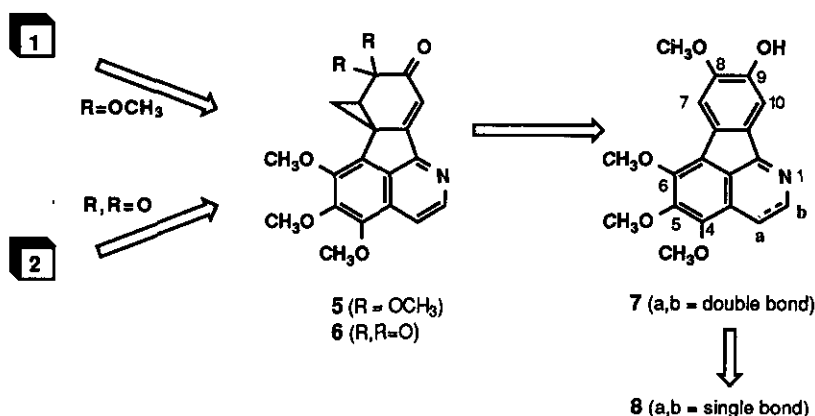
Abstract—The previously reported dihydroazafluoranthene (8) has been converted, over a number of steps, into the σ -homo- q -benzoquinone monoacetal (5). The structure of compound (5) was established by X-ray crystallographic methods and treatment of this material with trifluoroacetic acid resulted in formation of the tropoloisoquinoline alkaloid imerubrine (1). Alternatively, acetal (5) could be hydrolysed to the corresponding diketone (6) which proved to be unstable and isomerised to grandirubrine (2) on heating. Both tropoloisoquinolines (1) and (2) were tested for antitubulin activity, and weak inhibition of polymerisation was observed only with the former compound.

The tropoloisoquinoline alkaloids imerubrine (1),^{1a,2} grandirubrine (2)^{1b,2} and pareirubrine (3),³ which have been isolated from various Menispermaceae species, are constitutionally, and probably biogenetically, related to the potent antimittotic agent colchicine (4).⁴ The structures of both imerubrine (1) and pareirubrine (3) have been established unequivocally by single-crystal X-ray analysis.



Pareirubrine (3) has been shown to possess antileukaemic properties³ but biological evaluation of its simpler congeners (1) and (2) does not appear to have been undertaken. However, crude extracts of the plants from which compounds (1) and (2) have been isolated are the subject of patents as wound-healing agents.⁵ Furthermore, extracts of various parts of the shrub from which compound (3) is obtained are used in the treatment of cramps, as well as to prevent miscarriage and stop uterine haemorrhages.³

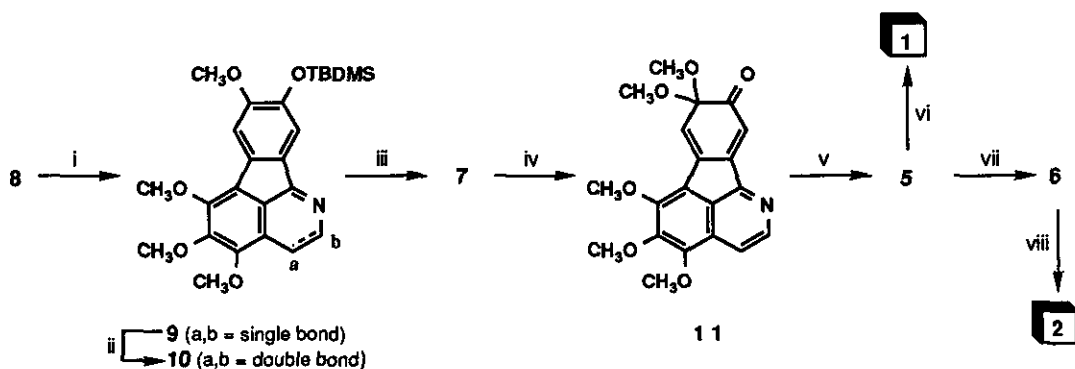
As a result of their potentially useful therapeutic properties and novel molecular architectures, compounds (1) and (2) have been the subject of at least two synthetic studies.⁶ However, no total synthesis of a tropoloisoquinoline alkaloid has been described so far. Research work from these laboratories suggests that σ -homo- α -benzoquinones⁷ and their monoacetals⁸ are excellent precursors to α -tropolones and α -tropolone \underline{Q} -alkyl ethers, respectively. On this basis, it might be expected that imerubrine (1) could be obtained by acid-catalysed rearrangement of acetal (5) (Scheme 1) while isomerisation of the related α -diketone (6) would give grandirubrine (2). Compounds (5) and (6) could, in turn, both be generated by simple elaboration of the azafluoranthene {indeno[1,2,3-*ij*]isoquinoline} (7). Since we have recently described⁹ a relatively straightforward (eight step) synthesis of the related 2,3-dihydroazafluoranthene (8) from 2-nitroisovanillin and 2-(2',3',4'-trimethoxyphenyl)ethylamine, the execution of the retrosynthetic analysis shown in Scheme 1 seemed realistic. We now describe¹⁰ the successful implementation of these ideas.



Scheme 1

SYNTHETIC STUDIES

The conversion of compound (8) into its fully dehydrogenated counterpart (7) proved to be difficult. After considerable experimentation¹¹ it was established that this could be achieved directly by heating compound (8) to 200°C in the presence of sulfur powder,¹² but under these conditions only low yields (34%) of the desired 7 were obtained. On the basis that the free phenolic unit in 7 and/or 8 could be leading to side reactions under the vigorous dehydrogenation conditions being employed, the latter compound was converted, quantitatively, into the corresponding *tert*-butyldimethylsilyl ether (9) using standard conditions.¹³



Scheme 2 Reagents and conditions: (i) TBDMSCl, DMF, $[(\text{CH}_3)_2\text{CH}]_2\text{NCH}_2\text{CH}_3$, 18°C, 5 min; (ii) 5% Pd on C, *p*-cymene, 177°C, 4 h; (iii) TBAF, THF, 0°C, 10 min; (iv) $\text{Ti}(\text{NO}_3)_3$, CH_3OH , -20°C, 30 min; (v) $\text{CH}_2\text{S}(\text{O})(\text{CH}_3)_2$, DMSO, 18°C, 24 h; (vi) $\text{CF}_3\text{CO}_2\text{H}$, CHCl_3 , 18°C, 2 h; (vii) 2 M aq. HCl, THF, 18°C, 4 h; (viii) THF, 66°C, 30 h. **Abbreviations:** TBDMS = *tert*-C₄H₉(CH₃)₂Si-; DMF = dimethylformamide; TBAF = (C₄H₉)₄N⁺F⁻; THF = tetrahydrofuran; DMSO = dimethyl sulfoxide.

Subjection of compound (9) to reaction with 5% palladium on carbon in refluxing *p*-cymene (177°C)¹⁴ then resulted in efficient dehydrogenation to produce compound (10). Removal of the silyl protecting group in azafloanthene (10) was accomplished using TBAF¹⁵ in THF and compound (7) was thereby obtained [85% overall yield from 8]. Elaboration of phenol (7) to the key acetal (5) followed from earlier work⁸ and involved, as the initial step, treatment of the former compound with thallium(III) nitrate in methanol at -20°C.¹⁶ There were no complications associated with oxidation at nitrogen, and the ring fused *o*-benzoquinone monoacetal (11) was obtained in near-quantitative yield. Due to its instability (Diels-Alder dimerisation?),¹⁶ compound (11) was immediately subjected to nucleophilic cyclopropanation with

dimethyl sulfoxonium methylide $[\text{CH}_2 \text{S}(\text{O})(\text{CH}_3)_2]^{17,18}$ and the required σ -homo-*o*-benzoquinone monoacetal (**5**) was obtained, albeit in modest (38%) yield. The spectral data acquired for compound (**5**) were in accord with expectation, but final confirmation of structure was obtained by single-crystal X-ray analysis (see Figure 1 and Tables 1-3).

The conversion of acetal (**5**) into compound (**1**) was readily achieved by treating the former compound with a large excess of trifluoroacetic acid in chloroform at room temperature for 2 h. Under these conditions imerubrine was obtained in 70% yield as dark-red crystals. The physical and spectral data obtained on this synthetic material (see Experimental) were in good agreement with those reported in the literature for the natural product. Furthermore, the $\{^1\text{H}\}^{13}\text{C}$ nmr spectrum of synthetic **1** displayed the expected sixteen sp^2 -carbon and four methoxy methyl carbon resonances.

The synthesis of grandirubrine (**2**) proved unexpectedly straightforward. Thus, reaction of acetal (**5**) with 2 M aqueous hydrochloric acid in THF produced an orange product which is tentatively assigned as the expected diketone (**6**). Attempts to purify this material by recrystallisation resulted in the deposition of deep-red crystals which proved to be grandirubrine (**2**). The most efficient method for effecting the conversion of **6** into **2** involved heating the former compound in refluxing THF for 30 h. In this way an 87% yield of tropoloisoquinoline (**2**) was realised. Once again, the spectral and physical data obtained on synthetic **2** were in excellent agreement with those reported^{1b,2} in the literature for the natural product. In addition, the $\{^1\text{H}\}^{13}\text{C}$ nmr spectrum of synthetic grandirubrine revealed the expected sixteen sp^2 -carbon as well as three methoxy methyl carbon resonances.

TUBULIN BINDING STUDIES ON COMPOUNDS (1) AND (2)

The close structural relationship between the title compounds (**1**) and (**2**) and colchicine (**4**) prompted an examination of the tubulin binding properties of the synthetically derived tropoloisoquinolines. Tubulin binding activity has frequently been used as a relatively simple prescreen to identify colchicinoids more likely to possess useful *in vivo* antitumour activity.¹⁹ Both imerubrine and grandirubrine were examined at several concentrations in a highly sensitive polymerisation system.²⁰ Compound (**2**) proved to be inactive ($\text{IC}_{50} > 100 \mu\text{M}$), but imerubrine (**1**) had weak activity, yielding an IC_{50} value of $46 \mu\text{M}$. For comparison, under this reaction condition IC_{50} values of $0.80 \mu\text{M}$ and $0.46 \mu\text{M}$ had been obtained for

colchicine and podophyllotoxin²⁰ respectively. Imerubrine (1) was also examined for potential inhibition of cell growth, using a Burkitt lymphoma cell line but the agent was without effect at the highest concentration examined (10 μ M; IC₅₀ value of colchicine about 50 nM). The significantly reduced activity of 1 (by comparison with 4) is presumably the result of the highly rigid nature of the former compound and its consequent inability to adopt a suitable (non-planar) conformation for initial binding to tubulin.²¹

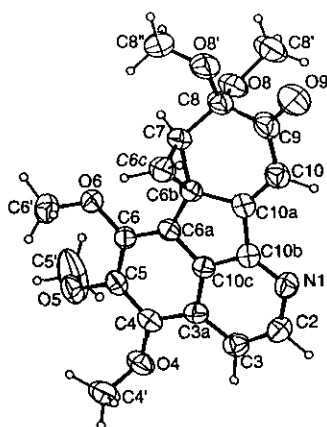


Figure 1: ORTEP Drawing of Compound (5) with 50% Ellipsoids.

Table 1: X-ray Crystallographic Data for Compound (5).

Formula	C ₂₁ H ₂₁ NO ₆	λ , Å	1.5418
<i>M</i>	383.4	μ (Cu K α), cm ⁻¹	7.85
Crystal colour	pale orange-brown	Scan mode	ω -2 θ
Crystal form	tabular	2 θ max, °	130
Crystal size, mm	0.052x0.205x0.218	<i>h, k, l</i>	$\pm 12, 10, 25$
Lattice	monoclinic	No. of reflections	3354
Space group	<i>P</i> 2 ₁ / <i>c</i>	No. of terms ($F_0 > 3\sigma(F_0)$)	2429
D_c , g cm ⁻³	1.349	Diffractometer	Rigaku-AFC
<i>Z</i>	4	No. of variables	270
$F(000)$	808	<i>S</i>	2.64
<i>a</i> , Å	10.313(2)	<i>R</i> (<i>F</i>)	0.051
<i>b</i> , Å	8.844(1)	<i>R_w</i> (<i>F</i>)	0.074
<i>c</i> , Å	21.231(7)	ρ_{min} , e Å ⁻³	-0.21
β , °	102.91(2)	ρ_{max} , e Å ⁻³	+0.39
<i>V</i> , Å ³	1887(1)	Max. shift-to-error	0.008
Temperature, K	291(1)	Weights, ($\sigma^2 F_0 + m F_0 ^2$)-1	$m=0.0005$

Table 2: Fractional Coordinates and Equivalent Isotropic Temperature Factors (\AA^2) with Estimated Standard Deviations in Parentheses for Compound (5).

Atoms	x	y	z	B_{eq}
N1	0.4951(2)	0.7797(2)	0.0410(1)	3.85(5)
C2	0.5405(3)	0.7109(3)	0.0991(1)	4.02(6)
C3	0.4945(2)	0.5771(3)	0.1194(1)	3.68(6)
C3a	0.3907(2)	0.4984(3)	0.0772(1)	3.00(5)
C4	0.3291(2)	0.3594(3)	0.0864(1)	3.34(6)
O4	0.3749(2)	0.2823(2)	0.1430(1)	4.39(5)
C4'	0.2845(3)	0.2670(5)	0.1845(1)	5.56(9)
C5	0.2317(2)	0.3007(3)	0.0364(1)	3.34(6)
O5	0.1755(2)	0.1630(2)	0.0444(1)	4.36(5)
C5'	0.2411(5)	0.0396(4)	0.0247(3)	9.98(19)
C6	0.1858(2)	0.3786(3)	-0.0233(1)	3.33(6)
O6	0.0922(2)	0.3156(2)	-0.0724(1)	4.23(5)
C6'	-0.0389(3)	0.2972(5)	-0.0613(2)	6.59(10)
C6a	0.2439(2)	0.5128(3)	-0.0331(1)	3.12(5)
C6b	0.2199(2)	0.6295(3)	-0.0849(1)	3.20(5)
C6c	0.0791(3)	0.6875(4)	-0.1130(1)	4.46(7)
C7	0.1560(3)	0.6122(3)	-0.1556(1)	3.83(6)
C8	0.2062(3)	0.7106(3)	-0.2036(1)	4.08(6)
O8	0.2935(2)	0.6154(2)	-0.2285(1)	4.72(5)
C8'	0.3515(4)	0.6796(5)	-0.2785(2)	6.58(10)
O8'	0.1014(2)	0.7645(3)	-0.2525(1)	5.32(5)
C8''	0.0079(4)	0.6516(5)	-0.2824(2)	6.65(10)
C9	0.2831(3)	0.8534(4)	-0.1735(1)	4.70(7)
O9	0.2892(3)	0.9634(3)	-0.2072(1)	7.64(8)
C10	0.3557(3)	0.8486(3)	-0.1059(1)	4.05(7)
C10a	0.3296(2)	0.7425(3)	-0.0656(1)	3.29(5)
C10b	0.3988(2)	0.7054(3)	0.0012(1)	3.15(5)
C10c	0.3453(2)	0.5677(3)	0.0170(1)	2.90(5)

Table 3: Distances (Å) and Angles (°) in Compound (5).

N1-C2	1.361(3)	C6a-C10c	1.401(3)
N1-C10b	1.326(3)	C6b-C6c	1.530(4)
C2-C3	1.379(4)	C6b-C7	1.506(4)
C3-C3a	1.416(3)	C6b-C10a	1.496(3)
C3a-C4	1.417(4)	C6c-C7	1.487(4)
C3a-C10c	1.400(3)	C7-C8	1.517(4)
C4-O4	1.371(3)	C8-O8	1.419(4)
C4-C5	1.389(3)	C8-O8'	1.404(3)
O4-C4'	1.425(4)	C8-C9	1.550(4)
C5-O5	1.375(3)	O8-C8'	1.446(5)
C5-C6	1.427(3)	O8'-C8'	1.434(5)
O5-C5'	1.396(5)	C9'-O9	1.218(4)
C6-O6	1.371(3)	C9-C10	1.464(3)
C6-C6a	1.366(4)	C10a-C10a	1.337(4)
O6-C6'	1.433(4)	C10a-C10b	1.475(3)
C6a-C6b	1.488(3)	C10b-C10c	1.408(4)
C2-N1-C10b	114.4(2)	C7-C6b-C10a	118.4(2)
N1-C2-C3	126.4(2)	C6b-C6c-C7	59.9(2)
C2-C3-C3a	119.2(2)	C6b-C7-C6c	61.5(2)
C3-C3a-C4	129.6(2)	C6b-C7-C8	118.2(2)
C3-C3a-C10c	114.4(2)	C6c-C7-C8	117.4(2)
C4-C3a-C10c	115.9(2)	C7-C8-O8	104.4(2)
C3a-C4-O4	118.4(2)	C7-C8-O8'	111.7(2)
C3a-C4-C5	119.3(2)	C7-C8-C9	114.2(2)
O4-C4-C5	122.2(2)	O8-C8-O8'	111.9(2)
C4-O4-C4'	116.4(2)	O8-C8-C9	109.2(2)
C4-C5-O5	119.3(2)	O8'-C8-C9	105.5(2)
O5-C5-C6	118.1(2)	C8-O8-C8'	116.2(2)
C5-O5-C5'	114.2(2)	C8-O8'-C8''	114.9(2)
C5-C6-O6	121.1(2)	C8-C9-O9	119.8(2)
C5-C6-C6a	119.0(2)	C8-C9-C10	118.9(2)
O6-C6-C6'	119.7(2)	O9-C9-C10	121.1(2)
C6-O6-C6a	116.8(2)	C9-C10-C10a	121.1(2)
O6-C6a-C6b	135.1(2)	C6b-C10a-C10	122.8(2)
C6-C6a-C10c	117.6(2)	C6b-C10a-C10b	106.8(2)
C6b-C6a-C10c	107.1(2)	C10-C10a-C10b	130.4(2)
C6a-C6b-C6c	120.5(2)	N1-C10b-C10a	129.4(2)
C6a-C6b-C7	129.0(2)	N1-C10b-C10c	106.9(2)
C6c-C6b-C7	58.6(2)	C3a-C10c-C6a	125.6(2)
C6c-C6b-C10a	118.5(2)	C3a-C10c-C10b	121.8(2)
C4-C5-C6	122.6(2)	C6a-C10c-C10b	112.6(2)

EXPERIMENTAL

Unless otherwise specified, all nmr spectra were recorded in CDCl_3 on a Varian Unity 300 spectrometer. General experimental procedures have been described elsewhere.⁹

4,5,6,8-Tetramethoxyindeno[1,2,3-*ij*]isoquinolin-9-ol (7).

Dihydroazafluoranthene (8)⁹ (90 mg, 0.26 mmol) and sulfur powder (18 mg, 0.56 mmol) were ground together in a mortar and pestle to form a fine powder. The resulting mixture was placed in a round-bottomed flask and heated at 200°C (oil bath) for 20 min under N_2 . Gas evolution was observed during this time. The reaction mixture was cooled to room temperature, treated with water (20 ml) and extracted with CHCl_3 (3 x 40 ml). The combined organic phase was dried (MgSO_4), filtered and concentrated under reduced pressure to give a black oil. This material was subjected to preparative tlc (ethyl acetate elution) and the resulting chromophoric band (R_f 0.5) extracted (ethyl acetate) to afford the title compound (7) (30 mg, 34%) as a pale orange solid, mp 170-171°C (lit.,⁹ mp 170-171°C). This material was identical in all other respects with samples of compound (7) obtained previously.⁹

9-tert-Butyldimethylsilyl-4,5,6,8-tetramethoxy-2,3-dihydroindeno[1,2,3-*ij*]isoquinoline (9).

A mixture of dihydroazafluoranthene (8)⁹ (150 mg, 0.44 mmol) in anhydrous DMF (5 ml) was treated sequentially with *N,N*-diisopropylethylamine (120 μl , 0.66 mmol) and TBDMSCl (80 mg, 0.53 mmol), and the resulting mixture was stirred under N_2 for 5 min. The ensuing yellow solution was poured into water (20 ml) and extracted with CH_2Cl_2 (3 x 30 ml). The combined organic phase was dried (MgSO_4), filtered and concentrated under reduced pressure to give the title compound (9) (205 mg, 96%) as a pale yellow and waxy solid (Found: M^+ , 455.2122. $\text{C}_{25}\text{H}_{33}\text{NO}_5\text{Si}$ requires M^+ , 455.2128). ν_{max} (KBr)/ cm^{-1} 2933, 1453, 1416, 1393, 1316, 1258, 1091, 826, 783; ^1H nmr δ 7.31 (br s, 1H), 7.27 (s, 1H), 4.15 (t, \downarrow 8.1 Hz, 2H, H2), 3.99 (s, 3H, OCH_3), 3.97 (s, 3H, OCH_3), 3.91 (s, 3H, OCH_3), 3.87 (s, 3H, OCH_3), 2.76 (t, \downarrow 8.1 Hz, 2H, H3), 1.00 (s, 9H), 0.17 (s, 6H); ^{13}C nmr δ 164.8, 153.7, 150.6, 150.1, 146.8, 144.4, 136.3, 129.3, 125.6, 123.8, 121.8, 115.0, 106.9, 61.0, 60.8, 60.7, 55.5, 49.5, 25.6,

18.3, 18.1, -4.8; m/z (70 eV) (%) 455 (29) (M^+), 398 (11) ($M^+ - C_4H_9$), 383 (100) ($M^+ - C_4H_9 - CH_3$). This material was used, without purification, in the next reaction.

9-*tert*-Butyldimethylsilyl-4,5,6,8-tetramethoxyindeno[1,2,3-*ij*]isoquinoline (10).

A mixture of 9-*tert*-butyldimethylsilyl-4,5,6,8-tetramethoxy-2,3-dihydroindeno[1,2,3-*ij*]isoquinoline (9) (50 mg, 1.1 mmol), anhydrous *p*-cymene (5 ml) and 5% palladium on carbon (80 mg) was heated at reflux under N_2 for 4 h. The cooled reaction mixture was filtered through a 2 cm deep pad of Celite™ and the filtrate concentrated under reduced pressure to afford the title compound (10) (49 mg, 100%) as a pale yellow oil (Found: M^+ , 453.1958. $C_{25}H_{31}NO_5Si$ requires M^+ , 453.1971). ν_{max} (NaCl)/ cm^{-1} 2928, 1472, 1458, 1262, 1136, 975, 826, 784; 1H nmr δ 8.50 (d, \downarrow 5.9 Hz, 1H, H2), 7.57 (s, 1H), 7.53 (d, \downarrow 5.9 Hz, 1H, H3), 7.46 (s, 1H), 4.14 (s, 3H, OCH₃), 4.09 (s, 3H, OCH₃), 4.02 (s, 3H, OCH₃), 3.95 (s, 3H, OCH₃), 1.03 (s, 9H), 0.21 (s, 6H); ^{13}C nmr δ 159.4, 152.8, 150.8, 149.6, 148.9, 144.9, 144.3, 132.9, 131.4, 125.8, 123.6, 122.1, 115.1, 112.7, 108.1, 62.1, 61.3 (two signals superimposed), 55.8, 25.7, 18.5, -4.6; m/z (70 eV) (%) 453 (22) (M^+), 396 (18) ($M^+ - C_4H_9$), 381 (100) ($M^+ - C_4H_9 - CH_3$). This material was used, without purification, in the next reaction.

4,5,6,8-Tetramethoxyindeno[1,2,3-*ij*]isoquinolin-9-ol (7).

A solution of compound (10) (720 mg, 1.6 mmol) in anhydrous THF (10 ml), maintained under N_2 , was cooled to 0°C and treated dropwise with TBAF (1.8 ml of a 1 M solution in THF, 1.8 mmol). The resulting purple solution was stirred for 10 min at 0°C, poured onto water (50 ml) and extracted with CH_2Cl_2 (5 x 50 ml). The combined organic phase was dried ($MgSO_4$), filtered and concentrated under reduced pressure to give a bright orange solid. This material was subjected to chromatographic filtration (2 cm deep pad of tic grade silica gel, ethyl acetate elution) and concentration of the yellow fractions afforded the title compound (7) (480 mg, 89%) as an orange solid, mp 170-171°C (lit.,⁹ mp 170-171°C). This material was identical in all respects with samples of compound (7) isolated previously.⁹

8,9-Dihydro-4,5,6,8,8-pentamethoxyindeno[1,2,3-*i*]isoquinolin-9(9H)-one (11).

A solution of thallium trinitrate (18 mg, 0.046 mmol) in anhydrous methanol (2 ml) was added dropwise to a solution of azafuoranthene (7) (14 mg, 0.04 mmol) in anhydrous methanol (4 ml) maintained at -20°C under N₂. The resulting dark brown solution was stirred at -20°C for 30 min then treated with pentane (5 ml). The reaction mixture was subjected to chromatographic filtration (3 cm deep pad of tlc grade basic alumina topped with a plug of cotton wool, CH₂Cl₂ elution). Concentration of the yellow eluant afforded the title compound (11) (15 mg, 98%) as a yellow oil (Found: M⁺, 369.1218. C₂₀H₁₉NO₆ requires M⁺, 369.1212). ν_{max} (NaCl)/cm⁻¹ 2949, 1734, 1671, 1579, 1453, 1408, 1364, 1105, 1080; ¹H nmr δ 8.59 (d, \downarrow 5.9 Hz, 1H, H2), 7.69 (d, \downarrow 5.9 Hz, 1H, H3), 7.04 (s, 1H), 6.91 (s, 1H), 4.11 (s, 6H, 2 x OCH₃), 4.05 (s, 3H, OCH₃), 3.47 (s, 6H, 2 x OCH₃); ¹³C nmr δ 194.5, 155.6, 150.7, 150.4, 149.9, 148.2, 145.6, 134.8, 132.0, 127.0, 126.7, 119.8, 117.3, 115.2, 92.5, 61.7, 61.5, 61.0, 50.6, 50.5; *m/z* (70 eV) (%) 369 (85) (M⁺), 354 (63) (M⁺-CH₃), 298 (100). This material was used, without purification, in the next reaction.

6b,7,8,9-Tetrahydro-4,5,6,8,8-pentamethoxyindeno[1,2,3-*i*]cyclopropa[1,2-*c*]isoquinolin-9(9H)-one (5).

A solution of trimethyl sulfoxonium iodide¹⁷ (110 mg, 0.5 mmol) in DMSO (3 ml) was treated with 100% NaH (12 mg, 0.52 mmol), and the resulting mixture was stirred at room temperature until gas evolution had ceased. An aliquot (270 μ l) of this solution was added dropwise to a stirred solution of enone 11 (14 mg, 0.038 mmol) in anhydrous DMSO (2 ml). The reaction mixture was protected from light and stirred at room temperature for 24 h then poured into water (20 ml) and extracted with CH₂Cl₂ (3 x 50 ml). The combined organic phase was dried (MgSO₄), filtered and concentrated under reduced pressure to a bright yellow oil. Subjection of this material to preparative tlc (ethyl acetate elution) resulted in one major chromophoric band (R_f 0.55) which was extracted (1:1 CHCl₃/methanol) to give a pale orange-brown solid. Recrystallisation (hexane/CHCl₃) of this material then gave the title compound (5) (5.5 mg, 38%) as orange-brown prisms, mp 167.5-169°C (Found: M⁺, 383.1365. C₂₁H₂₁NO₆ requires M⁺, 383.1369). ν_{max} (NaCl)/cm⁻¹ 2947, 2837, 1680, 1460, 1453, 1408, 1123, 1105; ¹H nmr δ 8.60 (d, \downarrow 5.9 Hz, 1H), 7.69 (d, \downarrow 5.9 Hz, 1H), 6.69 (s, 1H), 4.04 (s, 3H, OCH₃), 4.02 (s, 3H, OCH₃), 3.99

(s, 3H, OCH₃), 3.63 (s, 3H, OCH₃), 3.20 (s, 3H, OCH₃), 3.16 (dd, \downarrow 9.1 and 6.6 Hz, 1H), 2.07 (dd, \downarrow 9.1 and 4.9 Hz, 1H), 1.33 (dd, \downarrow 6.6 and 4.9 Hz, 1H); ¹³C nmr δ 189.5, 162.0, 157.1, 150.8, 147.1, 146.1, 145.6, 131.7, 127.0, 125.9, 115.1, 114.0, 97.0, 62.0, 61.8, 61.3, 50.8, 49.5, 31.0, 26.8, 25.3; *m/z* (70 eV) (%) 383 (100) (M⁺), 368 (63) (M⁺-CH₃), 352 (10) (M⁺-OCH₃).

4,5,6,9-Tetramethoxyazuleno[1,2,3-*ij*]isoquinolin-9(10H)-one (Imerubrine) (1).

A solution of pentacycle (5) (4 mg, 0.01 mmol) in anhydrous CHCl₃ (5 ml) was treated with trifluoroacetic acid (1 ml, 12.9 mmol) and the reaction mixture stirred at ambient temperature for 2 h, poured onto water (30 ml) and basified with sodium bicarbonate (5 ml of a saturated aqueous solution). The resultant yellow solution was extracted with CH₂Cl₂ (3 x 20 ml) and the combined organic extracts were dried (MgSO₄), filtered and concentrated under reduced pressure to give a pale orange solid. Subjection of this material to preparative tlc (acetone elution) afforded one major chromophoric band (*R_f* 0.5) which was extracted (acetone) to give imerubrine (1) (2.6 mg, 70%) as a bright-orange solid, mp 181.5-183°C (lit.,^{1a,2} mp 183-185°C) (Found: M⁺, 351.1099. C₂₀H₁₇NO₅ requires M⁺, 351.1106). *vmax* (NaCl)/cm⁻¹ 1585, 1460, 1375, 1240, 1170, 1083 1005; ¹H nmr (400 MHz) δ 8.69 (d, \downarrow 5.7 Hz, 1H, H2), 8.30 (s, 1H, H11), 8.07 (d, \downarrow 10.3 Hz, 1H, H7), 7.76 (d, \downarrow 5.7 Hz, 1H, H3), 6.87 (d, \downarrow 10.3 Hz, 1H, H8), 4.15 (s, 3H, OCH₃), 4.14 (s, 3H, OCH₃), 4.07 (s, 3H, OCH₃), 4.02 (s, 3H, OCH₃); ¹³C NMR (100 MHz) δ 179.4, 164.1, 157.6, 151.0, 150.2, 148.8, 145.4, 145.2, 136.7, 128.4, 126.3, 126.2, 126.1, 121.9, 115.0, 112.0, 61.9, 61.4, 61.2, 56.3; ¹³C nmr (100 MHz) δ (DMSO-d₆) 177.7, 163.7, 156.7, 150.6, 150.1, 148.2, 145.2, 144.0, 135.1, 126.8 (two signals superimposed), 125.5, 125.2, 120.9, 115.3, 112.5, 61.9, 61.4, 61.2, 56.1; *m/z* (70 eV) (%) 351 (15) (M⁺), 323 (3) (M⁺-CO), 322 (5), 308 (6), 224 (49), 211 (100).

10-Hydroxy-4,5,6-trimethoxyazuleno[1,2,3-*ij*]isoquinolin-9(9H)-one

(Grandirubrine) (2).

A solution of compound (5) (35 mg, 0.09 mmol) in THF (2 ml) was treated in one portion with HCl (3 ml of a 10% aqueous solution) and stirred for 4 h. The resultant red solution was poured onto water (5 ml), basified with sodium bicarbonate (saturated aqueous solution) and extracted with CH₂Cl₂ (3 x 20 ml). The

combined organic extract was concentrated under reduced pressure to afford an orange solid [presumed to contain diketone (6)] which was not purified due to its instability. This material was taken up in THF (10 ml) and the solution heated at reflux under N₂ for 30 h. Concentration of the reaction mixture gave a solid which upon recrystallisation (hexane/CHCl₃) afforded grandirubrine (2) (27 mg, 87%) as deep-red plates, mp 200-201.5°C (lit.,^{1b,2} mp 201-203°C) (Found: M⁺, 337.0937. C₁₉H₁₅NO₅ requires M⁺, 337.0950). ν_{max} (KBr)/cm⁻¹ 1536, 1453, 1408, 1324, 1226, 1013; ¹H nmr (400 MHz) δ 8.75 (d, \downarrow 5.9 Hz, 1H, H2), 8.43 (s, 1H, H11), 8.35 (d, \downarrow 10.8 Hz, 1H, H7), 7.80 (d, \downarrow 5.9 Hz, 1H, H3), 7.43 (d, \downarrow 10.8 Hz, 1H, H8), 4.19 (s, 3H, OCH₃), 4.16 (s, 3H, OCH₃), 4.05 (s, 3H, OCH₃), OH not observed; ¹³C nmr δ 173.4, 167.6, 157.5, 152.4, 150.0, 149.8, 146.1, 145.6, 137.1, 131.2, 126.1, 124.4, 121.6, 120.0, 119.8, 115.7, 62.0, 61.5, 61.4; m/z (70 eV) (%) 337 (100) (M⁺), 322 (22) (M⁺-CH₃), 309 (6) (M⁺-CO), 294 (23).

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