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ISOFLAVONOID CONSTITUENTS OF THE WEST AFRICAN RED WOOD *BAPHIA NITIDA*

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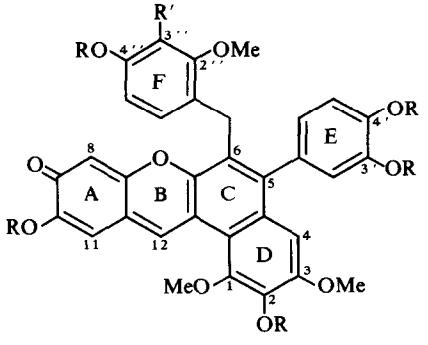
Key Word Index—*Baphia nitida*; Leguminosae; structural determination; santarubin C; isoflavene.

Abstract—The red heartwood of the West African tree *Baphia nitida* (camwood) contains the new pigment santarubin C and a dimethoxytrihydroxyisoflavene; both compounds have been identified on the basis of spectroscopic analysis of derivatives.

The so-called 'insoluble red woods' form a well-known group of dyewoods, consisting of some Asian *Pterocarpus* species (padauk, sandalwood, narrawood), and the West African redwoods, camwood and barwood. The structure of santalins A and B from Asian *Pterocarpus* species and of santarubins A and B from the African species *Pterocarpus osun* and *P. soyauxii* have been recently elucidated [1, 2]. Another West African species, *Baphia nitida*, which is known as camwood, usually produces a yellow wood, but in some parts, notably Sierra Leone, it develops a red heartwood [3]. This paper reports the results of the investigation of some constituents from the red heartwood of *B. nitida*, collected in Sierra Leone.

Extraction of the wood with ethyl acetate and/or methanol afforded a red pigment (**1a**), which could be purified by chromatography on polyamide. Inspection of spectroscopic data (UV and IR) suggested a strong similarity of this pigment to the santalins or santarubins, which was confirmed by more detailed comparison of the fully alkylated derivatives, prepared with MeI (**1b**) or CD_3I (**1c**) and K_2CO_3 in acetone. Mass spectral and analytical data for **1b** and **1c** are consistent with a $C_{33}H_{26}O_{11}$ formula for **1a**, i.e. a santalin or santarubin with three OMe groups and six OH groups. The attribution of the structure **1a**, i.e. a 3″-hydroxy-santarubin, to the pigment resulted from the detailed analysis of the ^1H and ^{13}C NMR spectra of **1b** and **1c**, in comparison with those of santarubin permethyl ether (**1d**). The following discussion takes into account the extensive spectroscopic data already obtained for santalins and santarubins [1, 2, 4].



1a R = H; R′ = OH
1b R = Me; R′ = OMe
1c R = CD_3; R′ = OCD_3
1d R = Me; R′ = H

The similarity of the spectra of **1b** and **1d** leaves no doubt that compound **1b** has the carbon skeleton of the santalins and santarubins. The difference between these two classes of pigments is in the substitution of rings E and F. In the ^1H NMR spectrum of compound **1b** the presence of a coupling of 0.3 Hz between the protons of the benzylic CH_2 group and H-6″, which is coupled with an adjacent proton, and the 3,4-substitution of ring E establishes that **1a** is a santarubin, which we name santarubin C. However, **1a** shows an additional OH or OMe group at C-3″ of ring F. The presence of this group at C-3″ is consistent with the appearance of *two* low field OMe-bearing carbons in the ^{13}C NMR spectrum of the hexatrideuteriomethyl ether (**1c**), attributed to the 2-OMe carbons of 1,2,3-trisubstituted benzenes (Table 1) [4]. Having established the substitution on the aromatic rings, the location of the OH and OMe groups had to be found. Irradiation of the OMe protons decoupled only one proton signal ($J = 0.1$ Hz), namely H-4, thus locating one OMe at C-3 and excluding the presence of a OMe at C-4″ (no decoupling from H-5″). The position of the other OMe groups was found by preparing the hexatrideuteriomethyl ether (**1c**) of **1a** and comparing the lanthanide-induced shifts (LIS) of the OMe groups with those of santarubin permethyl ether (**1d**). These shifts have already been found to be diagnostically useful [2]. Comparison of the LIS for the OMe groups of **1c** and **1d** (Table 2) rules out the presence of OMe groups on carbons 10, 2, 3′, 4′ and 3″.

As one group is certainly located at C-3, the remaining positions for two methoxyls are 1, 2″ or 4″. The value of

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Table 1. ^{13}C NMR of **1d** in comparison with **1c**

Carbon	**1d** δ	**1c** δ
1″	120.9	126.2
2″	159.0	152.0*
3″	98.0*d*	137.2
4″	157.7	151.5*
5″	103.9*d*	107.0*d*
6″	128.5*d*	122.4*d*
1-OMe*		60.8*q*
2″-OMe*		60.5*q*
3-OMe		55.7*q*

* May be interchanged. All the other C signals of both compounds are similar [4] (within ±0.4 ppm).

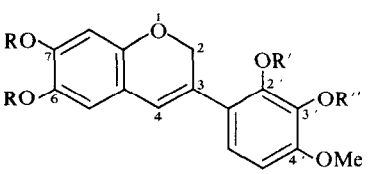
Table 2. Lanthanide-induced spectral shifts

OMe	Δδ* for (**1d**)	Δδ* for (**1c**)
1	3.3	3.22
2	10.3	
3	2.7	3.02
10	10.4	
3′	7.5	
4′	6.2	
2″	0.6	1.85
4″	0.1	

* Shifts induced by adding $Eu(dpm)_3$ (4 mol equiv.) to 20 mg of compound in $CDCl_3$.

the LIS is consistent with a position 1 (or 3) of a 1,2,3-trimethoxybenzene, and thus it does not discriminate among the three possibilities. However, the location at C-4″ of a OMe is excluded by decoupling experiments described above, and in addition the chemical shift of 60 ppm for another OMe group (which is not C-3) can be only explained by location at C-1 [4]. The structure (**1a**) is therefore assigned to santarubin C.

The chloroform extract of the heartwood also contains a colourless substance, **2a**, isolated in small amounts. This compound could be isolated by preparative TLC and



2a R = H; R′ = H or Me; R″ = Me or H
2b R = Ac; R′ = Ac or Me; R″ = Me or Ac
3 R = Ac; R′ = Ac or Me; R″ = Me or Ac 3,4-dihydro

gave a well-characterized triacetate (**2b**). MS data indicate the formula $C_{17}H_{16}O_6$ for (**2a**). ^1H NMR spectra data, particularly the presence of two OMe groups, of two *para* and two *ortho* aromatic protons, and the peculiar CH_2 signal at *ca* δ 5 indicate without doubt that **2a** is a dimethoxytrihydroxyisoflavene. Confirmation of this hypothesis was obtained by catalytic hydrogenation of **2b** to the corresponding isoflavan (**3**). Isoflavenes have been rarely found in natural sources, possibly because of their low stability, only a few examples of natural occurrence being known [5, 6].

The substitution pattern (2′, 3′, 4′, 6, 7) of **2a** was established from the observation of a small coupling constant between H-4 and one of the *ortho* aromatic protons, which must be H-6′. The two *para* aromatic protons are therefore H-5 and H-8. It was more difficult to establish the location of the OH and the OMe groups, due to the very small amount of wood and consequently of **2a** available. The presence of a peak at *m/e* 180 in the mass spectrum of the isoflavan (**3**), consistent with a retro-Diels–Alder reaction, suggests that both the OMe groups must be on ring C, therefore leaving two OH groups at C-6 and C-7. One of the methoxyls is certainly at C-4′, as it shows a coupling with H-5′. The definitive assignment of the structure, i.e. the choice between 2′-OH, 3′-OMe or 2′-OMe, 3′-OH substitution could not be effected, as lack of material prevented the preparation of suitable derivatives, or the measurement of the ^{13}C NMR spectrum. However, structure **2a** has a great similarity with the 'lower' half of santarubin C (**1a**) and also with santarubins A and B [2]. Biogenetic hypotheses have been put forward [1, 7], also supported by model synthetic experiments [8], which indicate an isoflavene as a building moiety of the santarubin pigments. On this basis, we consider that **2a** most probably has the 2′-OMe, 3′-OH substitution. Attempts to synthesize **2a** via the Tl(III) oxidative chalcone–isoflavone conversion [9, 10] failed at this stage, as oxidations with various Tl(III) salts of 2,4-dimethoxy-3,3′,4′-tribenzyloxychalcone gave poor yields of mixtures of uncyclized or partially debenzylated products.

EXPERIMENTAL

Mps are uncorr. UV spectra were measured in 95% EtOH (λ_{max} in nm). Unless stated otherwise, column chromatography was performed with Merck Si gel (0.05–0.20 mm) and TLC with Merck HF_{254} Si gel. Unless otherwise indicated, the purity of the products was checked by TLC, ^1H NMR and MS and deemed sufficient for structural elucidation purposes. *Baphia nitida* heartwood was collected by the Forest Officer at Kenema, Sierra Leone, whom we thank for supplying it.

Isolation of the constituents. The heartwood shavings (40 g) were extracted with hexane, then with $CHCl_3$, EtOAc and MeOH. The EtOAc and MeOH extracts were mixed, concentrated and chromatographed through a Macherey and Nagel polyamide column with $CHCl_3$–MeOH mixtures to give a red crude product, of which santarubin C was the major constituent. Santarubin C (**1a**), 120 mg, crystallized from Me_2CO in orange needles, mp 225–227°. λ_{max} 240, 270, 279, 306, 319, 443sh, 473 and 505 nm (ε 39 000, 31 000, 31 500, 12 500, 13 000, 14 500, 24 000, 24 000); ν_{max} (KBr) 3400 (OH), 1640 (conj. CO) cm^{-1}. The $CHCl_3$ extract was concentrated and the residue taken up with C_6H_6, and submitted to prep. TLC ($CHCl_3$–MeOH 9:1), to give **2a**, 30 mg, mp 165–170°, λ_{max} 296, 338 and 435 nm (ε 11 000, 11 500, 15 000), *m/e* 316, 301, 285, 180, 168; ^1H NMR (Me_2CO-d_6): δ 3.78 and 3.84 (OMe), 4.88 (C-2), 6.2–7.0 (5 aromatic protons), 7.63 (OH).

Santarubin C hexamethyl ether (**1b**). Santarubin C (50 mg) in dry Me_2CO (10 ml) was refluxed 6 hr with MeI (1 ml) and K_2CO_3 (200 mg). Filtration, evapn and crystallization from

MeOH gave santarubin C hexamethyl ether (**1b**), mp 222–223°, λ_{max} 235, 269, 277, 300, 308sh, 322, 394sh, 418, 447, 475 and 507 nm (ε 52000, 40500, 43000, 12500, 12000, 14500, 10000, 14000, 21000, 34000, 28000), ν_{max} (KBr) 1620 cm^{-1} (conj. CO), *m/e* 682. (Found: C, 68.87; H, 5.66. $C_{39}H_{38}O_{11}$ requires: C, 68.61; H, 5.60%).

Santarubin C hexatrideuteriomethyl ether (**1c**). Santarubin C (50 mg) was methylated as described above with CD_3I. Filtration, evapn and prep. TLC with $CHCl_3$–MeOH (15:1) gave 2,10,3′,4′,3″,4″-hexakis-*O*-trideuteriomethyl santarubin C (**1c**), *m/e* 700, ^1H NMR (C_6H_6-d_6): δ 3.34, 3.66 and 3.94 (OMe), 4.26 (aryl–CH_2–aryl, *J* = 0.3), 6.28 (C-6″, *J* = 8.5 and 0.3), 6.31 (C-11), 6.48 (C-5″, *J* = 8.5), 6.55 (C-8, *J* = 1.3), 6.71 (C-5′, *J* = 8), 6.82 (C-2′, *J* = 1.8), 6.91 (C-6′, *J* = 8 and 1.8), 6.98 (C-4, $J_{4,OMe}$ = 0.1), 9.42 (C-12, *J* = 1.3).

6,7,2′-*Triacetoxy*-3′,4′-*dimethoxyisoflav*-3-*ene* or 6,7,3′-*triacetoxy*-2′,4′-*dimethoxyisoflav*-3-*ene* (**2b**). **2a** (20 mg) in 0.5 ml dry pyridine and 0.5 ml Ac_2O were left at room temp. for 8 hr. Addition of $CHCl_3$, washing with satd $NaHCO_3$, water, satd $KHSO_4$ and water, evapn and prep. TLC (hexane–EtOAc, 1:1) gave the triacetate (**2b**) mp 52–55°, λ_{max} 291 and 327 nm (ε 9500, 11500), ν_{max} (nujol) 1770 cm^{-1}, *m/e* 442, 400, 358, 316, ^1H NMR ($CDCl_3$): δ 2.26, 2.36 (MeCO), 3.73 and 3.84 (OMe), 5.03 (C-2, *J* = 2), 6.58 (C-4), 6.71 (C-5 or C-8), 6.75 (C-5′, *J* = 8, coupled with OMe), 6.89 (C-5 or C-8), 7.14 (C-6′, *J* = 8, coupled with H-4).

6,7,2′-*Triacetoxy*-3′,4′-*dimethoxyisoflavan or* 6,7,3′-*triacetoxy*-2′,4′-*dimethoxyisoflavan* (**3**). 5 mg of **2b** in 5 ml MeOH were hydrogenated in the presence of 10% Pd/C. Filtration and evapn gave **3**, mp 145–150°, λ_{max} 279, 289sh, 312 and 326 nm (ε 3000, 2500, 600, 600), *m/e* 444.145 ± 0.004 (calc. for $C_{23}H_{24}O_9$ 444.142), 402, 360, 318.1087 ± 0.004 (calc. for $C_{17}H_{18}O_6$ 318.1103), 209, 180, 168.

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