74. Structure Determination of New Isomeric Naphtho[2,3-*b*]furan-4,9-diones from *Tabebuia avellanedae* by the Selective-INEPT Technique

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Beside the known naphthoquinones, dehydro-α-lapachone (**17**) and lapachol (**20**), four new naphtho[2,3-*b*]-furan-4,9-diones, *i.e.* the 2-acetyl-5-hydroxy, 2-acetyl-8-hydroxy, (–)-5-hydroxy-2-(1′-hydroxyethyl), and (±)-8-hydroxy-2-(1′-hydroxyethyl) derivatives **16**, **15**, **12**, and **13**, respectively, and the new compound benzo[*b*]furan-6-carboxaldehyde (**8**) have been isolated from a $CHCl_3$ extract of the inner stem bark of *Tabebuia avellanedae* LORENTZ *ex* GRISEB., together with four known naphthofurandiones, a dihydroisocoumarin derivative, (–)-6-hydroxymellein, and five benzoic-acid and three benzaldehyde derivatives which have not been reported previously from this plant. Structure determination of the isomeric 5- and 8-hydroxynaphtho[2,3-*b*]furan-4,9-diones was carried out unambiguously by a combination of selective-INEPT experiments and X-ray crystallographic analysis.

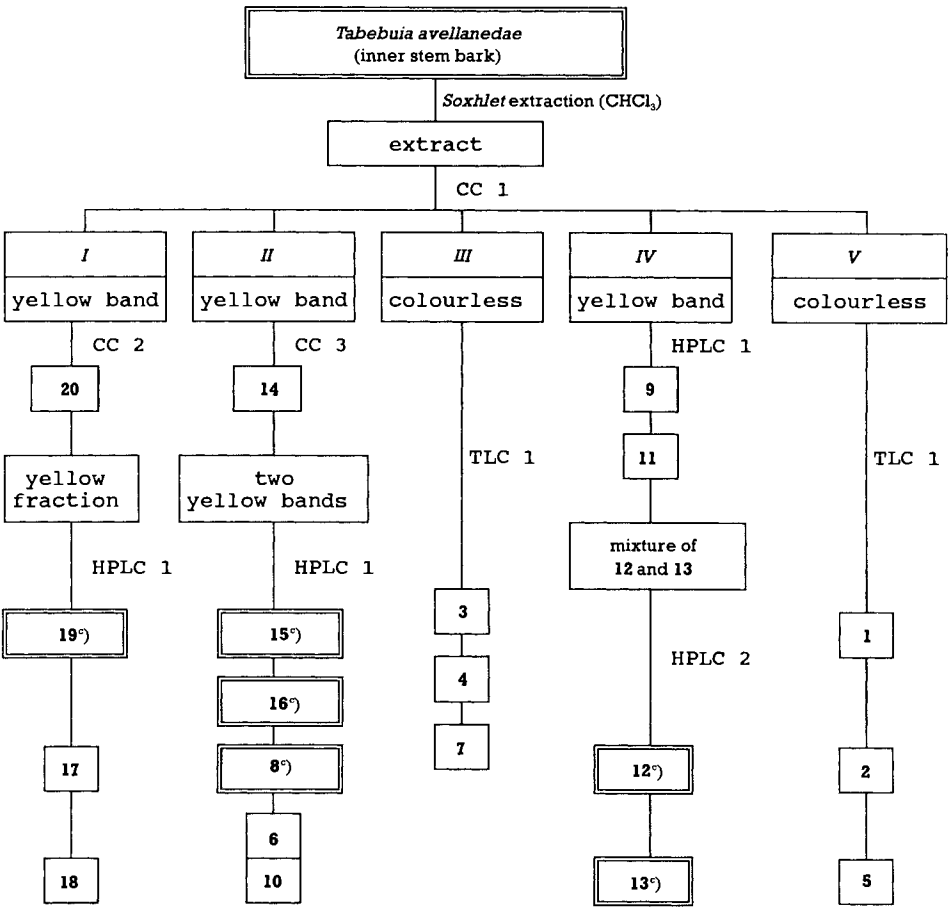
Introduction. – The stem bark of the South-American tree *Tabebuia avellanedae* LORENTZ *ex* GRISEB., Bignoniaceae (syn. *T. impetiginosa* MART. *ex* DC., *T. heptaphylla* VELL. TOLEDO, and *T. ipé* MART. *ex.* SCHUM. [1]), known in folk medicine as Pau d'Arco, Ipé Roxo, Taheebo, and Lapacho, is used in North and South America for many years as an anticancer, antifungal, antibacterial, and antiinflammatory drug [2] [3].

Lapachol and dehydro-α-lapachone, the major naphthoquinones of the heartwood of *Tabebuia avellanedae* LORENTZ *ex* GRISEB., were found to be active against different types of tumors [4–7]. *In vivo* antitumor effects were observed using *p.o.* administration of 20–30 mg/kg lapachol [8], and *de Santana et al.* [9] demonstrated *in vivo* antineoplastic effects on *Yoshida*'s sarcoma and *Walker 256* carcinosarcoma for the lipophilic extract of the inner bark of *T. avellanedae* LORENTZ *ex* GRISEB.

These pharmacological activities, and recent results from our laboratory (Munich) concerning the immunostimulating effects of various naphthoquinones when applied in minute doses [10] prompted us to initiate a new, thorough investigation of the lipophilic extract of the inner bark of *T. avellanedae* LORENTZ *ex* GRISEB.

Results and Discussion. – Column chromatography (CC) of the $CHCl_3$ extract of *T. avellanedae* LORENTZ *ex* GRISEB. stem bark on silica gel, using $CHCl_3$ as eluent resulted in five fractions (*Fr. I–V*) which afforded twenty compounds by further CC, prep. HPLC, and prep. TLC (see *Scheme*). In the present publication, we describe the structure elucidation of the new compounds **8**, **12**, **13**, **15**, **16**, and **19** which were obtained from *Fr. I, II,* and *IV*. The other compounds and their biological activities will be described in a forthcoming paper.

Scheme. *Separation of Inner Stem Bark Compounds from* Tabebuia avellanedae LORENTZ ex GRISEB.[a][b])

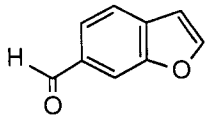


[a]) Column chromatography (CC) 1: silica gel, $CHCl_3$; CC 2: silica gel, toluene; CC 3: silica gel, toluene/$CHCl_3$ (gradient); HPLC 1: *ODS*, MeCN/H_2O (gradient); HPLC 2: *ODS*, THF/H_2O (gradient); TLC 1: silica gel (0.5 mm), $CHCl_3$/MeOH (1% HCOOH).

[b]) The sequence of the compounds given in the *Scheme* is equivalent to the sequence on HPLC spearation (*ODS*, MeCN/H_2O) of the $CHCl_3$ extract of *Tabebuia avellanedae* LORENTZ *ex* GRISEB. **1** = 4-hydroxybenzoic acid; **2** = 4-hydroxy-3-methoxybenzoic acid (vanillic acid); **3** = 4-hydroxy-3-methoxybenzaldehyde (vanillin); **4** = 3,4-dimethoxybenzoic acid (veratric acid); **5** = 3,4,5-trimethoxybenzoic acid; **6** = 3,4-dimethoxybenzaldehyde (veratric aldehyde); **7** = 4-methoxybenzoic acid (anisic acid); **8** = benzo[*b*]furan-6-carboxaldehyde[c]); **9** = (−)-3,4-dihydro-6,8-dihydroxy-3-methylisocoumarin ((−)-6-hydroxymellein); **10** = 4-methoxybenzaldehyde (anisaldehyde); **11** = (+)-2-(1′-hydroxyethyl)naphtho[2,3-*b*]furan-4,9-dione; **12** = (−)-5-hydroxy-2-(1′-hydroxyethyl)naphtho[2,3-*b*]furan-4,9-dione[c]); **13** = (±)-8-hydroxy-2-(1′-hydroxyethyl)naphtho-[2,3-*b*]furan-4,9-dione[c]); **14** = 2-acetylnaphtho[2,3-*b*]furan-4,9-dione; **15** = 2-acetyl-8-hydroxynaphtho-[2,3-*b*]furan-4,9-dione[c]); **16** = 2-acetyl-5-hydroxynaphtho[2,3-*b*]furan-4,9-dione[c]); **17** = 2,2-dimethylnaphtho[2,3-*b*]pyran-5,10-dione (dehydro-α-lapachone); **18** = (−)-2,3-dihydro-2-(1′-methylethenyl)naphtho-[2,3-*b*]furan-4,9-dione ((−)-dehydro-iso-α-lapachone); **19** = 2,3-dihydro-5-hydroxy-2-(1′-methylethenyl)-naphtho[2,3-*b*]furan-4,9-dione (dehydro-5-hydroxy-iso-α-lapachone)[c]); **20** = 3-(3′,3′-dimethylallyl)-2-hydroxynaphthalene-1,4-dione (lapachol).

[c]) Compounds described in the present publication.

Compounds **12** and **13** were isolated as a yellow crystalline mixture which revealed in the MS a molecular ion M^+ at 258. The UV (λ_{max} = 232, 247, 295, and 407 nm) and IR spectra (C=O absorption at 1642 and 1676 cm^{-1}) displayed absorption maxima typical of a napthoquinone structure [11]. In the ^{1}H-NMR spectrum, all signals appeared in pairs with different intensity, indicating that the isolate was a mixture of two isomers in the approximate ratio 2:1. The most conspicuous spectroscopic difference between **12** and **13** was the δ shift of a signal (1H) from 12.17 to 12.02 ppm. Separation of these isomers was successfully achieved by prep. HPLC on an *ODS*-phase column using THF/H_2O. Based on the results of different NMR studies (1D SPT, 1D INEPT), the optically active compound **12** was identified as *(−)-5-hydroxy-2-(1′-hydroxyethyl)naphtho[2,3-b]furan-4,9-dione* and thus **13** as *(±)-8-hydroxy-2-(1′-hydroxyethyl)naphtho[2,3-b]furan-4,9-dione.*



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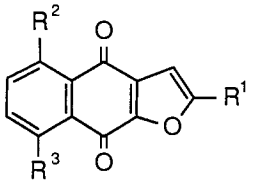
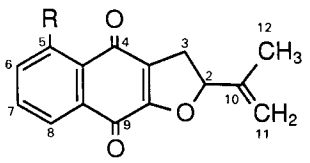


11 R^1 = CH(OH)–CH_3, R^2 = R^3 = H
12[1]) R^1 = CH(OH)–CH_3, R^2 = OH, R^3 = H
13[1]) R^1 = CH(OH)–CH_3, R^2 = H, R^3 = OH
14 R^1 = $COCH_3$, R^2 = R^3 = H
15[1]) R^1 = $COCH_3$, R^2 = H, R^3 = OH
16[1]) R^1 = $COCH_2$, R^2 = OH, R^3 = H



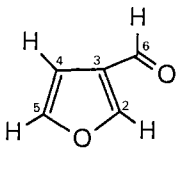
18 R = H
19[1]) R = OH

The ^{1}H-NMR spectrum of **12** showed an aromatic *ABX* system (7.75, 7.60, and 7.26 ppm) and a *s* at 12.17 ppm which disappeared on D_2O exchange and could be readily assigned to a chelated OH group. The position of this proton *peri* to a carbonyl group was confirmed by the bathochromic shift of 88 nm observed in the UV spectrum on addition of $AlCl_3$/HCl. A br. *s* (1H) was observed at 2.30 ppm. The ^{13}C-NMR spectrum showed resonances for 8 quaternary C-atoms, 5 CH, and 1 CH_3 as well as for 2 C=O groups at 173.2 and 187.0 ppm, the latter signal being deshielded by the H-bond of an OH group *peri* to C=O. Three O-bearing aromatic C-atoms appeared at 162.8, 165.9, and 152.5 ppm. The sp^3-C resonances at 64.4 and 22.68 ppm were readily assigned to an O-bearing aliphatic C-atom and a CH_3 group, respectively. The remaining H-bearing C-atoms were assigned by one-dimensional heteronuclear correlation using the CSCM 1D pulse sequence [12]. Selective population transfer (SPT) from the high-field ^{13}C satellites of *H*–C(6) and *H*–C(8) gave negative resonances at 125.84 (C(6)) and 120.54 ppm (C(8)), and magnetization transfer from the downfield ^{13}C satellites of *H*–C(3), *H*–C(6), and *H*–C(8) gave rise to positive signals at 103.98 (C(3)), 125.84 (C(6)), and 120.54 ppm (C(8)), respectively.

This combined spectral evidence identified the compound as a naphthofurandione bearing a hydroxyethyl side chain at C(2). However, it was not possible to distinguish with certainty between the two possible positional isomers **12** and **13**. Due to the scarcity of material (4 mg), a sensitive and non-destructive way for the unambiguous assignment was needed. Based on the following rationale, one-dimensional heteronuclear correlation using the selective INEPT pulse sequence [13] was thought to be the most suitable approach. Theoretically, a distinction between the isomers **12** and **13** should be possible utilizing the three-bond coupling of the two *C*=O with *H*–C(3) and *H*–C(8) or *H*–C(5), respectively. Polarization transfer from *H*–C(3) to C(4) would identify the nature of the C=O (chelated or not chelated), thereby indirectly defining the substituent at C(5). On the other hand, polarization transfer from *H*–C(5) to C(4) (or from *H*–C(8) to *H*–C(9)) would define the substitution pattern and preclude erroneous conclusions that might be induced by an unwanted four-bond correlation from *H*–C(3) to C(9).

The coupling constants ^3J(C,H) and ^4J(C,H) were estimated with the aid of suitable model compounds. Based on a ^{13}C-NMR study of quinones [14], ^3J(C(9),8) or ^3J(C(4),5) was estimated at 4 Hz. Indeed, polarization transfer from the ^{1}H resonance at 7.75 ppm to the *C*=O resonance at 173.24 ppm of **12** occurred in a selective-

[1]) The numbering of the side chain is arbitrary.



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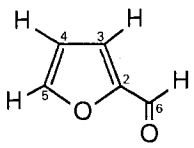

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INEPT experiment with delays optimized for 4 Hz. For the estimation of ^3J(C(4),3) and the unwanted ^4J(C(9),3), the furan derivatives **21** and **22** were chosen as models. In $CDCl_3$ solution, furan-3-carboxaldehyde (**21**) predominantly occurs as the O,O-*trans* conformer, with a ^3J(C(6),4) of 1.26 Hz. The O,O-*cis* form of furan-2-carboxaldehyde (**22**), on the other hand, exhibits a ^4J(C(6),4) of 0.47 Hz [15]. With delays set for J=1.5 Hz, selective polarization transfer from H–C(3) to the downfield C=O at 187.0 ppm (C(4)) of **12** was observed *(Fig. 1)*.

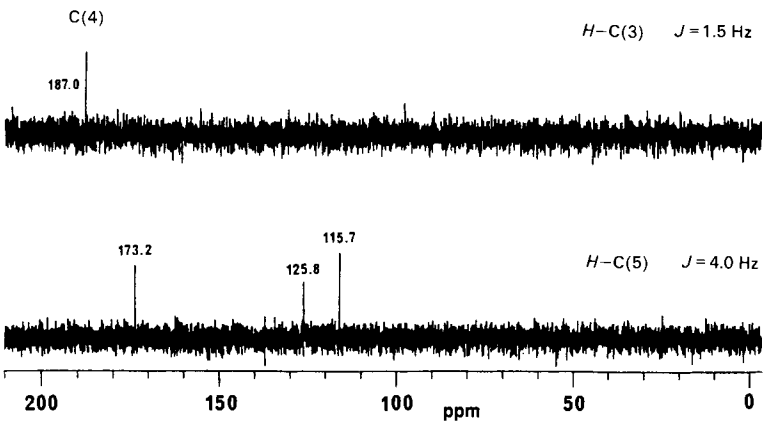
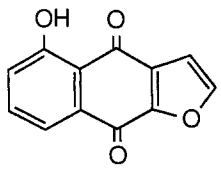


Fig. 1. *Polarization transfer from* H–*C(3)* (6.84 ppm) *and* H–*C(5)* (7.75 ppm) *of* **12**

Other quaternary C-atoms of **12** were assigned with the aid of additional selective-INEPT experiments. On irradiation of H–C(8) (J = 4 Hz), polarization transfer also occurred to C(4a) (115.73 ppm) and C(6) (125.84 ppm). Two-bond coupling from H–C(3) to C(3a) (131.54 ppm) and three-bond coupling to C(9a) (165.90 ppm) were observed with delays set for 4 Hz and 6 Hz, respectively. Unambiguous ^{13}C assignments are given in the *Exper. Part.*

According to [16], compound **12** is a new naphthoquinone. However, there exists some uncertainty concerning the structure of several previously isolated naphthofurandiones. *E.g.*, the positional isomer of **12**, compound **13**, has been reported from *Kigelia pinnata* DC. [17], *Tabebuia cassinoides* (LAM.) DC. [18], and *Crescentia cujete* L. [19] and was named 'kigelione'. The structure, however, was proposed without convincing spectral or chemical evidence. The fact, that the ^{1}H-NMR data of **12** and kigelione are identical might indicate kigelione to be **12** rather than the isomer **13**. The structure of kigelione should, therefore, be reinvestigated using long-range heteronuclear-correlation spectroscopy. The above described methodology might also be successfully applied to other naphthofurandiones, the structures of which have not been established unambiguously, *e.g.* diodantunezone (**23** or **23a**) [20], 8-hydroxy-2-isoprenylnaphtho[2,3-*b*]furan-4,9-dione (**24**) [21], and the dihydro derivatives **25**–**29** [21] [22].

The selective-INEPT technique was found to be an elegant and effective method for distinguishing the positional isomers **12** and **13** without resorting to synthesis. Scarcity of



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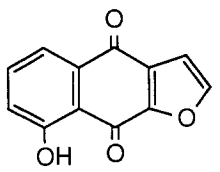
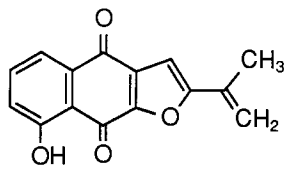


23a



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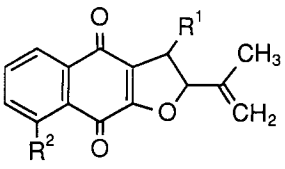
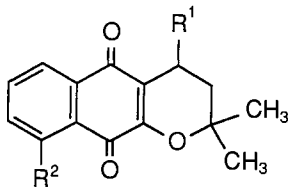
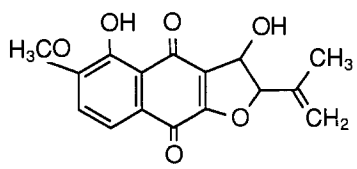


25 $R^1 = H$, $R^2 = OH$
26 $R^1 = R^2 = OH$



27 $R^1 = H$, $R^2 = OH$
28 $R^1 = R^2 = OH$



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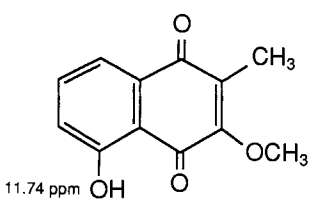
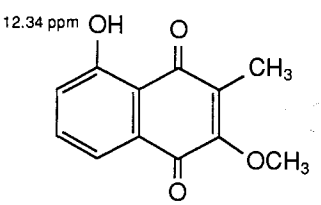
material and the small ^3J(C(4),3) precluded the use of 2D heteronuclear long-range correlation experiments. Three- and four-bond couplings, most importantly ^3J(C(4),3), could be quite well predicted with the aid of the chosen model compounds. In this context, it was of interest to compare the results obtained by selective-INEPT experiments with the empirical prediction of chemical shifts of chelated OH protons with the help of data, recently reported by *Musgave et al.* [23], who observed that the chemical shift of the OH proton of 5-hydroxy-1,4-naphthoquinones (juglone type) is influenced by different substituents (in position C(2) and C(3)) such as OH, CH_3, *etc.*, independent of the sample concentration. The authors found that these effects are additive and calculated substituent-dependent chemical shifts ($\Delta\delta$). These findings have been applied to the structure elucidation of some naphthoquinones [17] [19]. The calculated chemical shifts of the OH group of the model compounds **30** and **31** using these values are 11.74 (**30**) and 12.34 ppm (**31**), respectively. As expected, the value for **30** is approximately consistent with the chemical shifts of the phenolic OH group of **13**, and the value of isomer **31** is more similar to the one of **12**. Thus, the validity of the prediction of structures of naphthofurandione derivatives by this method was proved by selective-INEPT spectroscopy, and an empirical rule can be formulated: OH protons of aryl-unsubstituted 8-hydroxynaphtho[2,3-*b*]furan-4,9-diones appear in the ^{1}H-NMR spectrum more upfield than OH protons of the corresponding 5-hydroxy isomer.

Compounds **15** and **16** were also isolated together by CC as a sharp single yellow band. High-resolution MS revealed the molecular ion M^+ 256 to have the molecular



30 (*cf.* **13**)



31 (*cf.* **12**)

formula $C_{14}H_8O_5$. The UV and IR spectra were similar to those of **12** and **13**, and again the ^{1}H-NMR spectrum showed signals with different integrations and slightly shifted apart, indicating the presence of two isomers of a hydroxynaphthofurandione. Separation of these isomers was successfully achieved using prep. HPLC on an *ODS* column and MeCN/H_2O. Fortunately, since we had the opportunity to examine both isomers, and with the help of the spectroscopic data and selective-INEPT experience obtained from **12** and **13**, we were able to define the structure of **15** as *2-acetyl-8-hydroxynaphtho[2,3-*b*]-furan-4,9-dione* (OH at 11.95 ppm), and of **16** as its 5-hydroxy isomer (OH at 12.13 ppm). The occurrence of these quinones which are new structures and reported here for the first time with spectral proof was predicted by *Inouye et al.* [24].

Compound **19** was obtained in a small amount (*ca.* 1 mg) as crystallized plates. With the help of the empirical rule and the selective-INEPT experience, the OH group could be

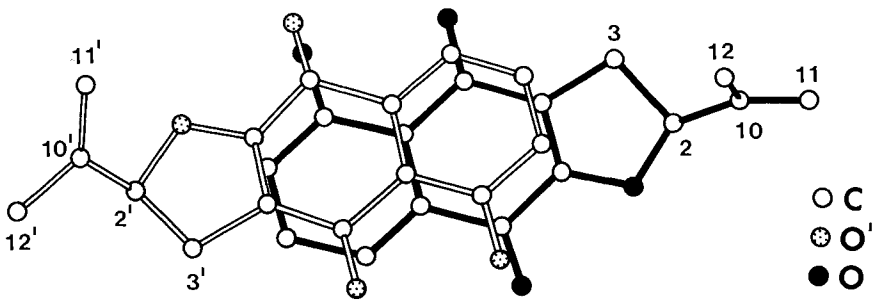


Fig. 2. *Two molecules of compound* **19** *crystallized together* (in projection to the crystallographic *xz* plane)[2])

predicted to be most probably at C(5). The complete structure of **19** was determined by X-ray analysis[2]) as *2,3-dihydro-5-hydroxy-2-(1'-methylethenyl)naphtho[2,3-*b*]furan-4,9-dione* (= dehydro-5-hydroxy-iso-α-lapachone), which was isolated initially from *Cescentia cujete* [19] and was reported with the same spectral data. Due to the scarcity of material, we were not able to define the optical activity of our isolate.

Again, the UV and IR spectra of **19** indicated the presence of a naphthoquinoid structure, and the ^{1}H-NMR spectrum defined it as a 2,3-dihydronaphtho[2,3-*b*]furan-4,9-dione (3.03 (*dd*, H–C(3)), 3.35 (*dd*, H–C(3)), and 5.45 ppm (*dd*, H–C(2))) with a 1-methylethenyl side chain (1.81 (*s*, CH_3), 5,02 (*m*, vinyl H *trans* to CH_3), and 5.14 ppm (*s*, vinyl H *cis* to CH_3)) and an aromatic *ABX* system (7.63, 7.54, and 7.26 ppm). A signal at 12.24 ppm (1H) again indicated the presence of a chelated OH group at C(5) or C(8).

Compound **8** formed yellow waxy plates with a complex UV spectrum λ_{max} = 222, 241, 285, 305 (sh), 320 (sh), and 431 nm). The ^{1}H-NMR data, long range couplings, and the chemical shifts of the C-atoms were in good agreement with the data obtained for benzo[*b*]furan derivatives, presented by *Benassi et al.* [26]. The structure of **8** was defined as *benzo[*b*]furan-6-carboxaldehyde* which is a new structure (synthetic or natural).

The EI-MS of **8** showed the characteristic pattern of aldehydes $[M-1]^+$ at 145 (100%); CI-MS: 147 (100, $[M+H]^+$), 164 (3, $[M+NH_4]^+$). The ^{1}H-NMR spectrum (80.13 MHz) in $CDCl_3$ was rather simple with two downfield 1-H *s* at 9.91 and 9.21 (br.) ppm which were not exchangeable with D_2O, and 4 arom.-H signals at 7.88

[2]) The atomic coordinates are deposited at the *Cambridge Crystallographic Data Centre.*

(*d*), 7.80 (*d*), 7.31 (*dd*), and 6.56 ppm (*dd*). In order to carry out irradiation experiments, chemical-shift dispersion of the arom.-H signals was enhanced, using (D_6) benzene as the solvent. Resonances in the 360-MHz spectrum now appeared at 9.96 (*s*), 9.01 (br. *s*), 7.36 (*d*), 6.64 (*d*), 6.39 (*dd*), and 6.17 (*dd*) ppm. Couplings between the signals at 7.36 and 6.17 ppm ($J = 3.1$ Hz) and between the signals at 6.64 and 6.39 ppm ($J = 5.3$ Hz) were established by irradiation experiments. Small couplings of the signals at 6.17 ($J = 0.9$ Hz) and 6.39 ppm ($J = 0.8$ Hz) disappeared when the *s* at 9.01 ppm was irradiated, and reverse irradiation was focusing the *s* at 9.01 ppm which is unusually broad.

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Experimental Part

General. Column chromatography (CC): silica gel *60* (< 63 μm, *Merck*). TLC: pre-coated TLC plates, silica gel *60 F-254* (0.5 mm, *Merck*). Prep. HPLC (*ODS* phase): *Hibar® (Merck)*, pre-packed column *RT-250-7, LiChrosorb RP-18* (7 μm, *Merck*, 4 ml/min solvent flow); apparatus *Waters* with pump *501, Wisp 710 B*, data module *740*, system controller *720*, and, detector *Lambda Max 481*; fraction controller *201-202 (Gilson)*. M.p.: *Kofler* hot-plate (*Reichert*, Wien). Optical rotation: polarimeter *241 (Perkin-Elmer)*. UV spectra (λ_{max} in nm (log ε in mol^{-1} l cm^{-1})): spectrophotometer *550 SE UV/VIS (Perkin Elmer)*; 1 mg/100 ml MeOH *p.a.*; recorder *561 (Perkin-Elmer)*. IR spectra ($\tilde{\nu}_{max}$ in cm^{-1}): spectrometer *Acculab 1 (Beckman)*; 1 mg/100 mg KBr. FT-IR spectra: spectrophotometer *PE 1750 (Perkin-Elmer)*; 50 μg/20 mg KBr. ^{1}H- and ^{13}C-NMR spectra: *Bruker WP-80* (80.13 or 20.15 MHz, resp.) or *Bruker AM-360* (360.13 or 90.56 MHz, resp.), δ [ppm] relative to internal Me_4Si (= 0 ppm) and J in Hz. Selective-INEPT experiments (compound **12**): *Nicolet NMC 1280* (361.08 or 90.8 MHz, resp.); calibration of proton pulse width by AcOH in C_6D_6 [22]; radiofrequency field strength of the proton soft pulse was on the order of 25 Hz; parameters for the CSCM 1D experiments [12] were as follows: ^{13}C: 32 K data points, 8000 transients, 32 180° ^{13}C pre-saturation pulses of 18 μsec, spaced by a delay of 44 ms (2.2 × ^{1}H proton soft pulse); ^{1}H: 180° proton soft pulse = 20 ms, average ^1J(C,H) values of 160 Hz for arom. C, H were used; delays D_3 and $D_4 = 1/2$ J(C,H), selective-INEPT experiments were performed according to [13]: ^{13}C: 32 K data points, 26000 transients; ^{1}H: 90° soft proton pulse = 10 ms, delay $D_3 = (1/4\,J(C,H)) = 10$ ms, $D_4 = D_3$. MS (*m/z* (rel. int. [%])): *M* 80 RFA (*Kratos*); CI-MS, reactant NH_3. X-Ray: *Nicolet-R-3M* diffractometer, CuK_α radiation, Ni filter, *Data General Nova 3*.

Plant Material. The stem bark of *Tabebuia avellanedae* Lorentz *ex* GRISEB. was obtained from *Kenneth Corwin (Dr. Meyer's Lapacho Co.*, Santa Monica, California), collected in Argentina, and was taxonomically identified by Dr. *A. H. Gentry* (Missouri Botanical Gardens, St. Louis, Missouri, U.S.A.). Authenticity of the plant material was confirmed by comparison of the HPLC fingerprint with vouchers from the 'Missouri Botanical Gardens' and the 'Instituto de Botanica del Nordeste', Corrientes, Argentina.

Extraction and Isolation. The powdered inner stem bark of *Tabebuia avellanedae* LORENTZ *ex* GRISEB. (800 g) was extracted with $CHCl_3$ in a *Soxhlet* for 72 h. The crude $CHCl_3$ extract (28.5 g) was separated by CC on a 85 mm × 70 cm column (silica gel) with $CHCl_3$. Five fractions (*Fr. I*, 300 ml; *Fr. II*, 500 ml; *Fr. III*, 600 ml; *Fr. IV*, 900 ml; *Fr. V*, 1500 ml) were collected. *Fr. I* (150 mg dissolved in $CHCl_3$) was further purified on a silica-gel column (40 mm × 60 cm, toluene) and prep. HPLC on *ODS* phase ($MeCN/H_2O$, gradient 10–70% MeCN in 20 min, detection 250 nm) to afford **19** (1 mg). *Fr. II* (225 mg dissolved in toluene/$CHCl_3$) was purified by CC (40 mm × 60 cm, toluene/$CHCl_3$, gradient 0–60% $CHCl_3$, discontinuous) and prep. HPLC (*ODS* phase, $MeCN/H_2O$, gradient 30–60% MeCN in 30 min, detection 240 nm) to supply **15** (3.5 mg), **16** (3 mg), and **8** (4 mg). *Fr. IV* was purified by two-step prep. HPLC (HPLC 1: *ODS* phase, $MeCN/H_2O$, 20–60% MeCN in 20 min, detection 250 nm; HPLC 2: *ODS* phase, THF/H_2O, 10–40% THF in 30 min, detection 280 nm) to give **12** (8 mg) and **13** (3 mg).

(−)-5-Hydroxy-2-(1'-hydroxyethyl)naphtho[2,3-b]furan-4,9-dione (**12**). Yellow needles. M.p. 156–158° (m.p. of **12**/**13**: 129–131°). $[\alpha]_D^{20}$ −16.6 (0.1%, MeOH). UV: 233 (sh, 4.34), 247 (4.42), 300 (3.84), 396 (3.75). IR: 1674, 1640, 1600, 1583, 1539, 1452, 1370, 1310, 1224. ^{1}H-NMR ($CDCl_3$, 361.08 MHz): 12.17 (*s*, OH–C(5)); 7.75 (*dd*, *J* = 7.5, 1.2, H–C(8)); 7.60 (*dd*, *J* = 8.3, 7.7, H–C(7)); 7.26 (*dd*, *J* = 8.3, 1.2, H–C(6)); 6.84 (*d*, *J* = 0.7, H–C(3)); 5.05 (*m*, H–C(10)); 2.03 (*br. s*, OH–C(10)); 1.65 (*d*, *J* = 6.5, 3 H–C(11)). ^{13}C-NMR ($CDCl_3$, 90.80 MHz): 187.01 (C(4)); 173.24 (C(9)); 165.90 (C(9a)); 162.85 (C(5)); 152.57 (C(2)); 136.81 (C(7)); 133.21 (C(8a)); 131.54 (C(3a)); 125.84 (C(6)); 120.54 (C(8)); 115.73 (C(4a)); 103.98 (C(3)); 64.39 (C(10)); 22.08 (C(11)). HR-MS: 258.0534 ($C_{14}H_{10}O_5$, calc. 258.05282). MS: 258 (98, M^+), 243 (100), 216 (83), 215 (86), 187 (48), 159 (12), 123 (30), 121 (38).

(±)-8-Hydroxy-2-(1'-hydroxyethyl)naphtho[2,3-b]furan-4,9-dione (**13**). Yellow needles. M.p. 145–147°. $[\alpha]_D^{20} = \pm 0$ (0.07%, MeOH). UV: 234 (4.25), 247 (4.24), 295 (3.76), 419 (3.72). FT-IR: 1678, 1646, 1600, 1583, 1534, 1454, 1383, 1308, 1277. ^{1}H-NMR ($CDCl_3$, 360.13 MHz): 12.02 (*s*, OH–C(8)); 7.72 (*dd*, *J* = 7.2, 1.3, H–C(5)); 7.59 (*dd*, *J* = 8.1, 7.7, H–C(6)); 7.26 (*dd*, *J* = 8.3, 1.2, H–C(7)); 6.86 (*d*, *J* = 0.7, H–C(3)); 5.05 (*m*, H–C(10)); 2.03 (br. *s*, OH–C(10)); 1.65 (*d*, *J* = 6.5, 3H–C(11)). ^{13}C-NMR ($CDCl_3$, 90.55 MHz): 180.36 (C(4)); 178.94 (C(9)); 166.23 (C(9a)); 163.13 (C(8)); 151.77 (C(2)); 136.81 (C(6)); 133.81 (C(4a)); 132.42 (C(3a)); 125.76 (C(7)); 120.69 (C(5)); 115.28 (C(8a)); 104.69 (C(3)); 64.45 (C(10)); 22.13 (C(1)). HR-MS: 258.0534 ($C_{14}H_{10}O_5$, calc. 258.05282). MS: 258 (88, M^+), 243 (100), 216 (28), 215 (36), 187 (14), 159 (8), 123 (2), 121 (10).

2-Acetyl-8-hydroxynaphtho[2,3-b]furan-4,9-dione (**15**). Yellow-orange needles. M.p. 212–215°. UV: 220 (4.35), 255 (4.54), 274 (sh, 4.42), 305 (sh, 3.81), 422 (3.92). FT-IR: 1683, 1646, 1598, 1582, 1453, 1360, 1292, 1257, 1190, 1071, 1025. ^{1}H-NMR ($CDCl_3$, 360.13 MHz): 11.95 (*s*, OH–C(8)); 7.79 (*dd*, *J* = 7.5, 1.2, H–C(5)); 7.68 (*dd*, *J* = 8.4, 7.6, H–C(6)); 7.33 (*dd*, *J* = 8.5, 1.1, H–C(7)); 7.60 (*s*, H–C(3)); 2.67 (*s*, 3 H–C(11)). ^{13}C-NMR ($CDCl_3$, 90.55 MHz): 178.87 (C(4)); 178.81 (C(9)); 173.15 (C(10)); 162.97 (C(9a)); 155.79 (C(8)); 152.29 (C(2)); 137.02 (C(6)); 133.22 (C(4a)); 131.29 (C(3a)); 125.53 (C(7)); 120.55 (C(5)); 115.27 (C(8a)); 112.44 (C(3)); 26.77 (C(11)). MS: 256 (94, M^+), 241 (100), 226 (6), 213 (10), 188 (7), 173 (8), 157 (9), 149 (10), 129 (22).

2-Acetyl-5-hydroxynaphtho[2,3-b]furan-4,9-dione (**16**). Yellow needles. M.p. 218–221°. UV: 213 (4.32), 255 (4.64), 275 (sh, 4.47), 305 (sh, 3.96), 413 (3.82). FT-IR: 1695, 1673, 1646, 1572, 1449, 1370, 1239, 1225, 1202. ^{1}H-NMR ($CDCl_3$, 360.13 MHz): 12.13 (*s*, OH–C(5)); 7.82 (*dd*, *J* = 7.5, 1.0, H–C(8)); 7.67 (*dd*, *J* = 8.2, 7.7, H–C(7)); 7.33 (*dd*, *J* = 8.7, 1.0, H–C(6)); 7.60 (*s*, H–C(3)); 2.67 (*s*, 3H–C(11)). ^{13}C-NMR ($CDCl_3$, 90.55 MHz): 174.88 (C(4)); 173.17 (C(10)); 173.01 (C(9)); 162.65 (C(9a)); 155.66 (C(5)); 152.90 (C(2)); 136.64 (C(7)); 132.74 (C(8a)); 130.52 (C(3a)); 125.88 (C(6)); 120.43 (C(8)); 115.27 (C(4a)); 111.84 (C(3)); 26.77 (C(1)). MS: 256 (79, M^+), 241 (100), 227 (3), 213 (10), 186 (5), 173 (5), 157 (10), 145 (5), 129 (24), 119 (8).

2,3-Dihydro-5-hydroxy-2-(1'-methylethenyl)naphtho[2,3-b]furan-4,9-dione (**19**). Yellow-orange plates. M.p. 118–121°. $[\alpha]_D^{20}$ not measured (lack of material). UV: 223 (4.25), 245 (4.23), 291 (4.05), 409 (3.74). FT-IR: 2957, 2918, 2850, 1711, 1678, 1638, 1619, 1465, 1455, 1228. ^{1}H-NMR ($CDCl_3$, 360.13 MHz): 12.24 (*s*, OH–C(5)); 7.64 (*dd*, *J* = 7.3, 0.8, H–C(8)); 7.54 (*dd*, *J* = 8.0, 7.3, H–C(7)); 7.25 (*dd*, *J* = 8.0, 0.8, H–C(6)); 5.45 (*dd*, *J* = 11.0, 8.7, H–C(2)); 5.14 (*s*, 1 H, H–C(11), *cis* to CH_3); 5.02 (*m*, 1H, H–C(11), *trans* to CH_3); 3.35 (*dd*, *J* = 17.4, 11.0, H–C(3)); 3.03 (*dd*, *J* = 17.4, 8.7, H–C(3)); 1.81 (*s*, 3H–C(12)). ^{13}C-NMR ($CDCl_3$, 90.55 MHz): 176.97 (C(4)); 172.35 (C(9)); 161.11 (C(5)); 160.81 (C(9a)); 141.40 (C(10)); 135.09 (C(7)); 131.75 (C(8a)); 125.28 (C(6)); 123.53 (C(3a)); 119.53 (C(8)); 114.79 (C(4a)); 114.22 (C(11)); 88.99 (C(2)); 31.57 (C(3)); 16.87 (C(12)). MS: 256 (100, M^+), 241 (36), 228 (19), 213 (76), 210 (28), 121 (47), 120 (59).

X-Ray Structure Analysis. The compound was crystallized from a mixture of $CHCl_3$/MeOH/H_2O 100:10:1 to give thin transparent yellow plates, size 1.0 × 0.5 × 0.02 mm, in the orthorhombic space group $P2_12_12_1$ with unit cell constants *a* = 6.51, *b* = 7.88, *c* = 47.99 Å. The density measured by flotation in KI/H_2O was 1.40 g cm^{-3} ($d_{calc.}$ = 1.39 g cm^{-3}) and showed *z* = 8 molecules in the unit cell, that means two molecules in the asymmetric unit. A total of 1868 unique reflections were measured on a *Nicolet-R-3M* diffractometer (Ω-scan, scan speed 4° min^{-1}), 1526 were observed ($I > 3(\sigma)I$). An empirical absorption correction was applied to the measurements. The structure was solved by direct methods using SHELXTL [25] and subsequent difference *Fourier* synthesis. The refinement with anisotropic thermal vibrations converged without H-atoms at 8.3%. The two molecules in the asymmetric unit are slightly different with regard to the positioning of the methylethenyl group to the basic planar ring skeleton, *e.g.* the dihedral angle in the first molecule O(1)–C(2)–C(10)–C(11) is +127°, whereas in the other molecule, the angle O(1')–C(2')–C(10')–C(11') is −14° (see *Fig. 2*). *Fig. 2* also gives an explanation for the crystallization of two formula units in one asymmetric unit. The naphtho[2,3-*b*]furan ring systems are nearly antiparallel and the molecules lie staggered with their naphtho ring systems, showing a distance of 3.3 Å. The angle between these planes is 4.7°.

Benzo[b]furan-6-carboxaldehyde (**8**). Yellow waxy material. M.p. ~ 60°. UV: 223 (3.64), 241 (3.58), 287 (3.50), 308 (sh, 3.35), 323 (sh, 3.11), 430 (3.12). ^{1}H-NMR ($CDCl_3$, 80.13 MHz): 9.91 (br. *s*, CHO–C(6)); 9.21 (br. *s*,

H–C(7)); 7.88 (*d*, J = 3.5, H–C(2)); 7.80 (*d*, J = 5.5, H–C(4)); 7.31 (*dd*, J = 5.5, 1.0, H–C(5)); 6.56 (*dd*, J = 3.5, 1.0, H–C(3)). ^{1}H-NMR (C_6D_6, 360.13 MHz): 9.96 (*s*, CHO–C(6)); 9.01 (br. *s*, H–C(7)); 7.36 (*d*, J = 3.1, H–C(2)); 6.64 (*d*, J = 5.3, H–C(4)); 6.39 (*dd*, J = 5.3, 0.8, H–C(5)); 6.17 (*dd*, J = 3.1, 0.9, H–C(3)). ^{13}C-NMR ($CDCl_3$, 90.55 MHz): 184.9 (CHO–C(6)); 151.2 (C(7a)); 146.3 (C(2)); 143.3 (C(3a)); 128.1 (C(6)); 127.8 (C(5)); 127.5 (C(4)); 111.2 (C(7)); 110.6 (C(3)). ^{13}C-NMR (C_6D_6, 90.55 MHz): 176.8 (CHO–C(6)); 151.2 (C(7a)); 145.4 (C(2)); 142.6 (C(3a)); 128.3 (C(6)); 128.0 (C(5)); 127.7 (C(4)); 110.7 (C(7)); 110.2 (C(3)). MS: 146 (93, M^+), 145 (100), 117 (72), 89 (79), 63 (77). CI-MS (NH_3): 164 (21, $[M + NH_4]^+$), 147 (100, $[M + H]^+$), 117 (2), 89 (2).

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