40. Syntheses of Bile Pigments

Part 17[1])

Synthesis of a Non-Racemizable Urobilin Derivative

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(22.XI.93)

The optically active urobilin model compound **7** was synthesized, in which Me groups instead of H-atoms are bound to the asymmetric centers, thus preventing loss of chirality by tautomerization. The key intermediate of the eleven-step synthesis of **7** is the 1,4,5,10-tetrahydro-10-hydroxy-1-oxo-11*H*-dipyrrin-9-carboxylate *rac*-**2**, which could be resolved into enantiomers by fractional crystallization of the corresponding methyl *N*-[1-(naphth-1-yl)ethyl]carbamates **3** and **4**. The absolute configuration of enantiomerically pure (–)-**2** was determined by X-ray diffraction analysis of its camphor-10-sulfonate **5**. As the CD spectrum of the urobilin analogue **7** obtained from (–)-(*R*)-**2** displays a positive *Cotton* effect, the present results prove, in connection with previous work, that substitution of Me groups for the H-atoms bound to the asymmetric centers of a chiral urobilin chromophore do not influence the relationship between absolute configuration of the latter and its helicity.

Urobilinoids (urobilins and stercobilins) are the final products of heme catabolism in humans and mammalians [2]. This class of bile pigments deserve particular interest, however, since the existence of the urobilin chromophore was demonstrated in both algal [3] [4] and bacterial [5] accessory pigments of the phycoerythrin type, which are involved in the process of oxygenic photosynthesis in these organisms.

Both urobilins and stercobilins are characterized by the presence of two asymmetric C-atoms (C(4) and C(16) in the partially hydrogenated 22*H*-bilin-1,19(21*H*,24*H*)-dione chromophore). Thus, symmetrically substituted urobilins (4,5,15,16-tetrahydro-22*H*-bilin-1,19(21*H*,24*H*)-diones) may be either chiral or sigmasymmetric depending on whether the relative configurations at both asymmetric C-atoms are the same or opposite, respectively (*cf.* **7**). Chiral urobilins are characterized by the extremely high specific rotation of their hydrochlorides (*e.g.* $[\alpha]_D^{20} = +5000$ for natural *d*-urobilin IXα [6]), which was attributed to the presence of an inherently dissymmetric coiled dipyrrin chromophore whose chirality is preserved by intramolecular H-bonding [7]. In this model, the helicity of the dipyrrin chromophore is determined by the absolute configurations of the asymmetric atoms C(4) and C(16). Recently, it could be proved that a (*S*,*S*)-configurated urobilin molecule displays a negative *Cotton* effect in the absorption range of the dipyrrin chromophore [8]. Until now, however, the precise structure of the optically active

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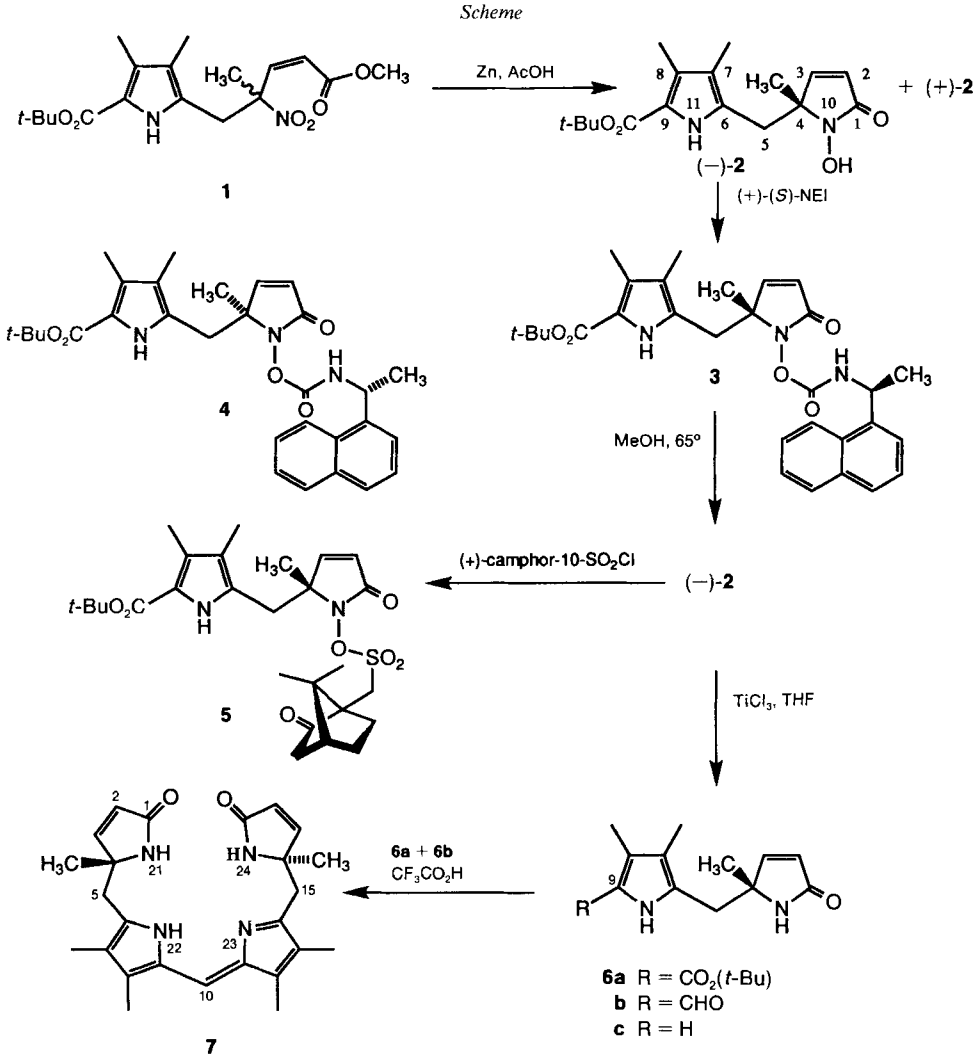
conformers cannot be deduced unequivocally from their CD spectra, since the relationship between the helicity (*P* or *M*) of the dipyrrin chromophore and the absolute configuration at C(4) and C(16) is based only on the examination of space-filling molecular models [7].

Until now, attempts to obtain crystalline samples of optical active urobilins have been hampered in part, by their facile racemization, caused by the acidity of the H-atoms bound to the chiral centers C(4) and C(16). Thus, the synthesis of an urobilin analogue, in which these positions are substituted by Me groups was envisaged to prevent the loss of chirality by tautomerization. An appropriate intermediate for this purpose *rac*-**6a**, was prepared before, starting with the pyrrole derivative **1**, by *Battersby* and coworkers [9] in

Scheme



the scope to study the mechanism of action of the enzyme uroporphyrinogen-III cosynthetase.

To resolve *rac*-**6a** into its enantiomers, the conventional method, consisting in the liberation of the carboxylic-acid function on C(9) and subsequent fractional crystallization of a derivative of the latter obtained by reaction with an optically active amine (*cf.* [8] [10]), could not be taken into consideration because liberation of pyrrolecarboxylic acids from the corresponding *tert*-butyl esters by means of CF_3COOH is accompanied by decarboxylation, even at low temperature (*cf.* [11]). However, advantage was taken from the fact that reduction of the nitro group of **1** with Zn in AcOH at 0° in the absence of $TiCl_3$ leads to the hydroxamic acid *rac*-**2** instead of lactam *rac*-**6a** (*cf.* [12]; see *Scheme*). Esterification of the OH group of *rac*-**2** with an optically active acid derivative should allow the resolution of the racemic mixture into the individual enantiomers. Thus, according to a method developed for the resolution of chiral tertiary alcohols [13], reaction of *rac*-**2** with (+)-(*S*)-1-(naphth-1-yl)ethyl isocyanate ((+)-(*S*)-NEI) yielded the corresponding carbamate as a mixture of diastereoisomers which were separated by fractional crystallization. According to ^{1}H-NMR analysis of the resonances of the well discernible *tert*-Bu and both Me groups on C(4) and C(10[4]), the enantiomeric purity of **3**, which was obtained by this procedure, amounts to 98 ± 2%. Alternatively, reaction of *rac*-**2** with (−)-(*R*)-1-(naphth-1-yl)ethyl isocyanate afforded a mixture of the corresponding diastereoisomeric carbamates, from which **4**, the enantiomer of **3**, could be isolated in 98 ± 2% enantiomeric purity by fractional crystallization from benzene.

Unfortunately, however, the quality of the obtained crystals of **3** or **4** was insufficient for X-ray diffraction studies and thus for the determination of the absolute configuration on the basis of the known absolute configuration of (−)-1-(naphth-1-yl)ethyl isocyanate which was correlated with (+)-1-(naphth-1-yl)ethylamine [14], the absolute configuration of which was previously established to be (*R*) [15] [16].

Therefore, carbamate **3** was cleaved into (−)-(*S*)-methyl *N*-[(naphth-1-yl)ethyl]carbamate and (−)-**2** by treatment with boiling MeOH for 24 h, and thereon the latter was reacted with (+)-camphor-10-sulfonyl chloride to yield sulfonic ester **5** in 83% overall yield. After recrystallization from MeOH, a sample of **5** in a quality suitable for X-ray diffraction studies[2]) could be obtained. Since (+)-camphor-10-sulfonyl chloride is prepared from (+)-camphor-10-sulfonic acid [17], which in turn is obtained by sulfonation of (+)-camphor [18], the absolute configuration of which is known [19], the absolute configuration of C(4) in **5** can be inferred to be (*R*) from the relative configuration of the chiral C-atoms of the latter, which is available from the X-ray diffraction pattern (see *Fig. 1*).

Noteworthy, H–C(2) of enantiomerically pure (−)-**2** and of the racemic mixture *rac*-**2** absorbs at different resonance frequencies (see *Fig. 2*), presumably because intermolecular association between molecules of opposite configuration occurs in solution. This peculiarity enables to determine the enantiomeric purity (e.e.) of (−)-**2** and of its enan-

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[2]) Crystal-structure analysis of **5**: Data collected at 87 K; space group $P2_1$, $Z = 2$ for $C_{27}H_{38}N_2O_7S$; $a = 11.158$ (3) Å, $b = 11.884$ (3) Å, $c = 11.582$ (4) Å, $\beta = 118.75°$, $V = 1346$ (2) Å^3, $d_{calc} = 1.319$ g/cm^3. Intensity data collected with Ni-filtered CuK_α radiation ($\lambda = 1.54178$ Å) for two octants or reciprocal space with $5.5° \leq 2\theta \leq 115°$, 2318 total and 2269 significant ($I_{obs} > 2\sigma(I)$) reflections. Structure refined (against F^2) with anisotropic atomic displacement parameters for all non-H-atoms, H-atoms at calculated positions; $R = 0.08$ for 343 parameters and 2269 observations. Atomic coordinates were deposited at the *Cambridge Crystallographic Data Centre*.

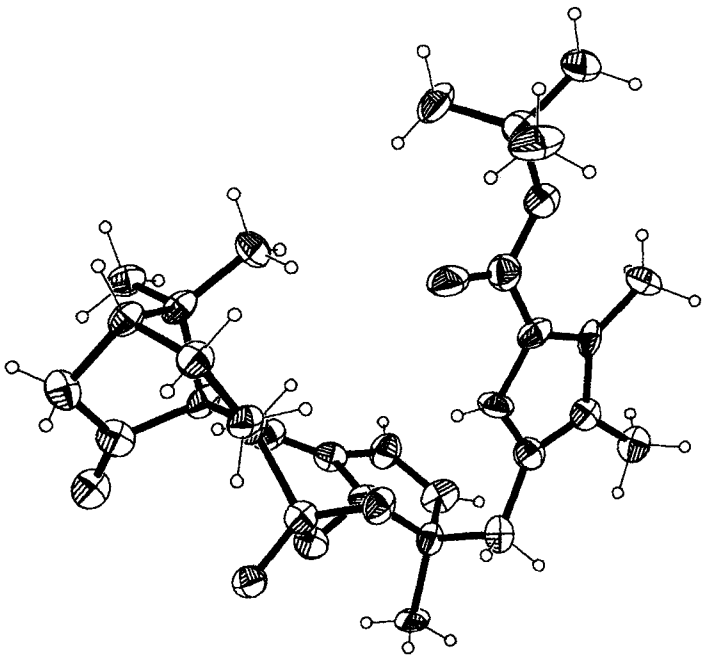

Fig. 1. *Structure of* tert-*butyl (−)-(*R*)-10-(camphersulfonyloxy)-1,4,5,10-tetrahydro-4,7,8-trimethyl-1-oxo-1*H*-dipyrrin-9-carboxylate* (**5**)

tiomer (+)-**2**, which was obtained by methanolysis of **4**, with an accuracy of 99 ± 1% without use of any chiral additive. Accordingly, solid *rac*-**2** is a crystalline compound of m.p. 162 ± 0.5°, whereas until now all attempts to obtain sharp-melting samples of the pure enantiomers failed.

With a view to the synthesis of the desired optically active urobilin analogue **7**, a sample of the enantiomerically pure hydroxamic acid (−)-**2** was reacted with $TiCl_3$ in AcOH/THF to yield the corresponding dipyrrin-1(10*H*)-one derivative **6a** in 93% yield. Then the latter was transformed into the corresponding aldehyde **6b** by means of trimethyl orthoformate in the presence of CF_3COOH (*cf.* [20]). In the absence of trimethyl orthoformate, *tert*-butyl ester **6a** was cleaved by CF_3COOH yielding the corresponding decarboxylated product **6c**, which was readily oxidized by air. Therefore, **6a** was dissolved in CF_3COOH and condensed *in situ* with aldehyde **6b** to afford the desired optically active urobilin analogue **7** in 35% yield. Like other urobilinoids, **7** forms a zinc chelate which displays a strong fluorescence at λ_{max} 530 nm ($\Phi = 0.23$). As expected, the 4,16-dimethyl derivative **7** is markedly more stable than usual urobilins bearing tertiary asymmetric C-atoms. Thus, it could be purified and characterized as free base rather than as hydrochloride which is otherwise the more handy form of this sensitive class of compounds.

As other enantiomerically pure dissymmetric urobilin hydrochlorides, **7** · HCl is characterized by its extremely high specific optical rotation ($[\alpha]_D^{20} = +4900 \pm 50$ in CH_2Cl_2). Moreover, the dipyrrin chromophore of this (4*R*,16*R*)-configurated urobilin analogue gives rise to a positive *Cotton* effect ($\Theta = 227\,700$ at λ_{max} 496 nm), the sign of which is

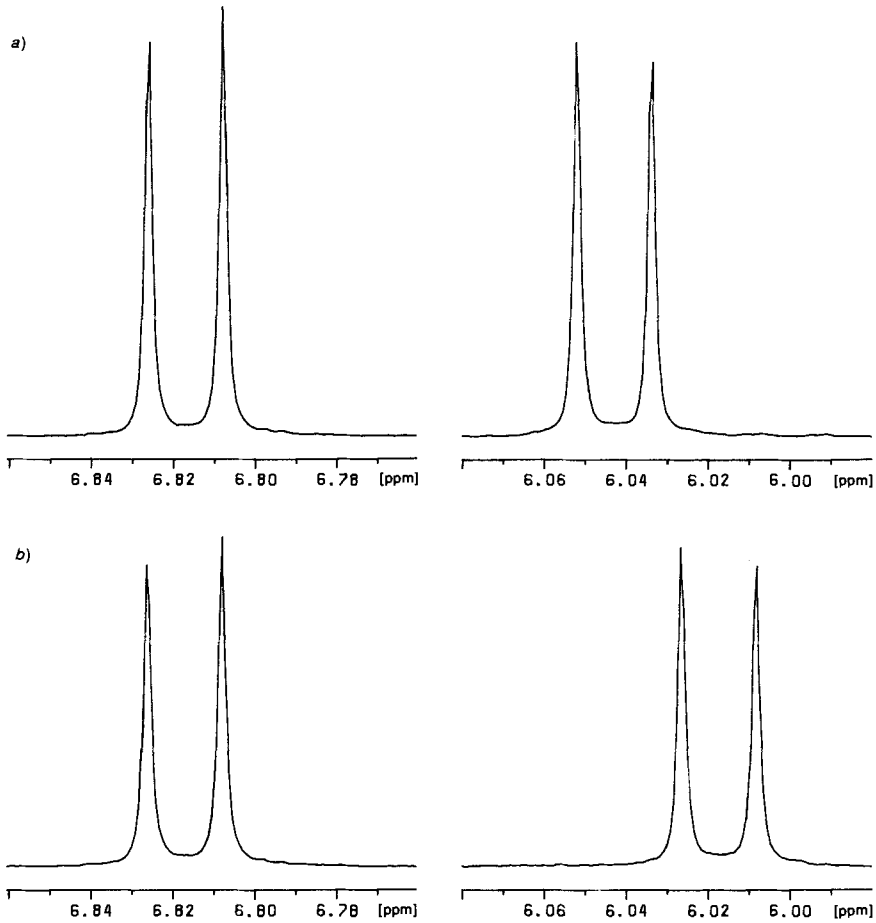

Fig. 2. *Detail of the* ^{1}*H-NMR spectrum* ($1.1 \cdot 10^{-2}$ M in $CDCl_3$) *of* a) rac-**2** *and* b) *(−)-(*R*)-***2** *in the range of absorption of H−C(3) and H−C(2), showing the characteristic down-field displacement of the latter in the racemate*

opposite to that of a previously synthesized urobilin derivative of (*S*)-configuration at both chiral C(4) and C(16) atoms [8]. This result supports, therefore, the generality of the relationship between helicity of the urobilin chromophore and absolute configuration of the chiral C-atoms in the same molecule, even though Me groups instead of H-atoms are present on both chiral C-atoms. As a matter of fact, however, the helicity (*P* or *M*) of the optically active chromophores cannot be inferred from these experimental results, and further work has to be done to obtain crystalline urobilinoid molecules, the shape of which can be determined by X-ray diffraction analysis. It is to be expected that the availability of urobilin analogues of the type represented by **7** will smooth away this difficulty.

Financial support of this work by the *Swiss National Science Foundation* (project No. 20-25575.88), the *Austrian Science Foundation* (project No. 8371), and the Agro Division of *Ciba Geigy AG*, Basle, is gratefully acknowledged.

Experimental Part

General. See [8]. Solvents for chemical reactions and chromatography were generally dried and distilled prior to use. (−)-(*R*)- and (+)-*S*-1-(Naphth-1-yl)ethyl isocyanate ((*R*)- and (*S*)-NEI, resp.) were purchased from *Fluka Chemie AG*. Reactions were monitored by TLC. TLC: *E. Merck* silica gel $60F_{254}$ (0.2 mm) precoated aluminium foils. Column chromatography (CC): *E. Merck* silica gel *60* (230–400 mesh). Flash chromatography (FC [21]) and medium-pressure chromatography (MPLC [22]): *E. Merck* silica gel *60* (40–63 mm). CD: *Jobin-Yvon-Auto-Dichrograph-Mark-V*; band amplitudes are given as specific ellipticity Θ and wavelengths λ in nm. Emission spectra: *Perkin-Elmer-MPF-4* fluorescence spectrophotometer. NMR Spectra: recorded by *F. Fehr* with a *Bruker-AM-360* instrument equipped with a data system *Aspect 3000*. FAB-MS: at 6 kV in 3-nitrobenzyl alcohol with Ar at 8 kV. Elemental analyses: *Ciba-Geigy AG*, Forschungszentrum, CH–1723 Marly.

*(±)-*tert*-Butyl 1,4,5,10-Tetrahydro-10-hydroxy-4,7,8-trimethyl-1-oxo-1*H*-dipyrrin-9-carboxylate* (*rac*-**2**). Zn dust (1.9 g) was added in 3 portions within 1.5 h to a stirred soln. of **1** [9] (5.4 g, 14.7 mmol) in AcOH/THF 1:1 (120 ml; *v/v*). The mixture was stirred at 0° for further 1.5 h and then allowed to warm up to r.t. Thereon, the remaining Zn dust was filtered off and the soln. diluted with CH_2Cl_2 (120 ml) before it was neutralized with sat. aq. $NaHCO_3$ soln. The aq. layer was extracted with CH_2Cl_2 (2 × 60 ml), the combined org. phase washed with H_2O, dried ($MgSO_4$), and evaporated, and the residue crystallized from AcOEt/hexane: 4.3 g (91%) of *rac*-**2**. M.p. 161.5–162.5°. IR: 3420*m*, 3930*m*, 1685*s*, 1610 (sh), 1425*m*, 1365*m*, 1295*m*, 1280*m*, 1245*m*, 1165*m*. UV: 284 (3.98), 250 (sh, 3.31). ^{1}H-NMR: 9.34 (br. *s*, NH); 6.82 (*d*, *J* = 6.5, H–C(3)); 6.04 (*d*, *J* = 6.5, H–C(2)); 3.07, 2.97 (2*d*, *J* = 14.8, CH_2(5)); 2.16 (*s*, Me–C(8)); 1.88 (*s*, Me–C(7)); 1.53 (*s*, *t*-Bu); 1.46 (*s*, Me–C(4)). ^{13}C-NMR: 167.4 (*s*, C(1)); 161.4 (*s*, COO); 149.1 (*d*, C(3)); 126.7, 125.5, 119.3, 117.9 (4*s*, C(6), C(7), C(8), C(9)); 124.7 (*d*, C(2)); 80.2 (*s*, Me_3C); 69.0 (*s*, C(4)); 31.9 (*t*, C(5)); 28.5 (*q*, Me_3C); 21.4 (*q*, Me–C(4)); 10.7 (*q*, Me–C(8)); 9.2 (*q*, Me–C(7)). MS: 320 (2, M^+), 264 (12), 247 (9), 209 (5), 208 (67), 153 (29), 152 (100), 151 (6), 135 (8), 134 (63), 108 (7), 106 (19), 96 (5), 79 (19), 77 (8), 57 (7), 44 (20). FAB-MS: 321 (MH^+). Anal. calc. for $C_{17}H_{24}N_2O_4$ (320.38): C 63.72, H 7.55, N 8.74; found: C 63.85, H 7.66, N 8.80.

*(−)-(*R*)-Ester* (−)-**2**. A soln. of **3** (0.95 g, 1.83 mmol) in MeOH (25 ml) was refluxed for 24 h. After evaporation, the residue was purified by CC (silica gel, CH_2Cl_2/MeOH 98:2): *(−)-(*S*)-methyl* N-*[1-(naphth-1-yl)ethyl]carbamate* (0.35 g, 83%), m.p. 87.3–87.9° (from Et_2O/hexane), $[\alpha]_D^{22} = -18.1$ (*c* = 0.41, MeOH), and 0.52 g (88%) of (−)-**2** (99% e.e.), $[\alpha]_D^{20} = -144.3$ (*c* = 0.401, MeOH).

*(+)-(*S*)-Ester* (+)-**2**. Methanolysis of **4** (0.50 g, 0.96 mmol) as described for (−)-**2** gave *(+)-(*R*)-methyl* N-*[1-(naphth-1-yl)ethyl]carbamate* (0.21 g, 95%) and (+)-**2** (0.28 g, 88%), $[\alpha]_D^{20} = +143.7$ (*c* = 0.259, MeOH).

tert-*Butyl (−)-(4*R*,10⁴*S*)-1,4,5,10-Tetrahydro-4,7,8-trimethyl-10-{*N*-[1-(naphth-1-yl)ethyl]carbamoyloxy}-1-oxo-1*H*-dipyrrin-9-carboxylate* (**3**). To a soln. of *rac*-**2** (1.00 g, 3.12 mmol) in dry benzene (30 ml), (+)-(*S*)-NEI (0.62 g, 3.14 mmol) was added and the mixture heated under reflux for 2 h. Then *ca.* 20 ml of solvent were evaporated, and after addition of dry hexane (20 ml), the mixture was allowed to stand overnight. The white product which separated (792 mg, 49%) reached, after 3 recrystallizations from benzene, a constant $[\alpha]_D^{20}$ of −183.1 (*c* = 0.605, MeOH) corresponding to 99% e.e. M.p. 169–170° (from benzene). IR: 3403*m*, 3325 *m*, 3000*m*, 2974*m*, 2932*w*, 1755*s*, 1727*s*, 1686*s*, 1498*s*, 1450*s*, 1368*m*, 1282*s*, 1165*m*, 1110*s*, 1055*s*. ^{1}H-NMR: 10.57 (br. *s*, NH); 8.12–7.49 (*m*, 7 arom. H); 6.87 (*d*, *J* = 6.5, H–C(3)); 5.91 (*d*, *J* = 6.5, H–C(2)); 5.78 (5 lines, *J* = 6.8–7.5, H–C(10⁴)); 5.68 (*d*, *J* = 7.6, H–N(10³)); 2.90, 2.82 (2*d*, *J* = 15.0, CH_2(5)); 2.21 (*s*, Me–C(8)); 1.90 (*s*, Me–C(7)); 1.78 (*d*, *J* = 6.6, Me–C(10⁴)); 1.54 (*s*, *t*-Bu); 1.37 (*s*, Me–C(4)). ^{13}C-NMR: 170.2 (*s*, C(1)); 160.9 (*s*, CO, ester); 154.7 (*s*, CO, carbamate); 153.5 (*d*, C(3)); 137.9 (*s*), 133.91 (*s*), 130.7 (*s*), 130.0, 128.4 (2C), 126.06, 123.8, 122.8, 122.4 (6*d*, arom. C, naphthyl); 125.4 (*d*, C(2)); 126.5, 125.8, 119.0, 116.6 (4*s*, C(6), C(7), C(8), C(9)); 79.7 (*s*, Me_3C); 68.9 (*s*, C(4)); 47.7 (*d*, C(10⁴)); 31.7 (*t*, C(5)); 28.5 (*q*, Me_3C); 22.8 (*q*, *Me*–C(10⁴)); 21.4 (*q*, *Me*–C(4)); 10.4 (*q*, *Me*–C(8)); 9.0 (*q*, *Me*–C(7)). FAB-MS: 540 ($[M + Na]^+$), 518 (MH^+). Anal. calc. for $C_{30}H_{35}N_3O_5$ (517.6): C 69.61, H 6.81, N 8.11; found: C 69.72, H 6.84, N 8.30.

*(+)-(4*S*,10⁴*R*)-Enantiomer* **4** was obtained as described for **3** from *rac*-**2** (181 mg, 0.55 mmol) and (−)-(*R*)-NEI (134 mg, 0.68 mmol). $[\alpha]_D^{20} = +183.9$ (*c* = 0.530, MeOH).

tert-*Butyl (−)-(*R*)-10-(Camphersulfonyloxy)-1,4,5,10-tetrahydro-4,7,8-trimethyl-1-oxodipyrrin-9-carboxylate* (**5**). A soln. of (+)-camphor-10-sulfonyl chloride (160 mg, 0.63 mmol) in dry pyridine (3 ml) was added to a soln. of **2** (100 mg, 0.31 mmol) in dry pyridine (5 ml) previously cooled to 5°. The mixture was stirred for 6 h, then diluted with H_2O (100 ml), and finally extracted with CH_2Cl_2. The combined extract was washed with 2N HCl and H_2O, dried ($MgSO_4$), and evaporated: 158 mg (94%) of **5**. Crystals suitable for X-ray diffraction analysis were obtained by recrystallization from MeOH. M.p. 154° (dec.). $[\alpha]_D^{20} = -120.3$ (*c* = 0.593, $CHCl_3$). IR ($CHCl_3$): 3450*w*, 3410*m*, 3030*w*, 2980*m*, 2930*m*, 1645*s*, 1680*s*, 1455*m*, 1380*m*, 1275*m*, 1174*m*, 1120*m*. ^{1}H-NMR: 9.10 (br. *s*, NH); 7.02 (*d*, *J* = 6.5, H–C(3)); 5.95 (*d*, *J* = 6.5, H–C(2)); 4.06, 3.68 (2*d*, *J* = 14.9, CH_2(1′)); 3.11, 2.96 (2*d*,

J = 15.1, CH_2(5)); 2.54–2.33 (*m*, 2 H); 2.16 (*s*, Me–C(8)); 2.15–2.01 (*m*, 2 H); 2.01–1.87 (*m*, 2 H); 1.88 (*s*, Me–C(7)); 1.53 (*s*, Me–C(4)); 1.52 (*s*, *t*-Bu); 1.52–1.44 (*m*, H–C(4′)); 1.13, 0.91 (2*s*, 2 Me–C(7′)). ^{13}C-NMR: 213.11 (*s*, C(2′)); 171.53 (*s*, C(1)); 160.93 (*s*, COO); 155.04 (*d*, C(3)); 125.71, 125.15, 119.47, 117.84 (4*s*, C(6), C(7), C(8), C(9)); 123.01 (*d*, C(2)); 79.99 (*s*, Me_3C); 70.61 (*s*, C(4)); 58.29 (*s*, C(1′)); 49.76 (*t*, C(1′^{1})); 48.38 (*s*, C(7′)); 42.92 (*d*, C(4′)); 42.46 (*t*, C(3′)); 32.52 (*t*, C(5)); 28.48 (*q*, Me_3C); 27.06 (*t*, C(6′)); 25.28 (*t*, C(5′)); 22.82 (*q*, *Me*–C(4)); 19.72, 19.66 (2*q*, *Me*–C(7′)); 10.50 (*q*, *Me*–C(8)); 9.33 (*q*, *Me*–C(7)). FAB-MS: 557 ([M + Na]$^+$), 535 (MH$^+$). Anal. calc. for $C_{27}H_{38}N_2O_7S$ (534.6): C 60.65, H 7.16, N 5.23, S 5.99; found: C 60.52, H 7.17, N 5.01, S 6.10.

tert-*Butyl (−)-(*R*)-1,4,5,10-Tetrahydro-4,7,8-trimethyl-1-oxo-11*H*-dipyrrin-9-carboxylate* (**6a**). To a soln. of (−)-**2** (0.50 g, 1.56 mmol) in AcOH/THF 1:1 (25 ml) containing NH_4^+ (AcO^-) (0.29 g, 3.8 mmol) a 15% aq. $TiCl_4$ soln. (2.3 ml) was added at once. The mixture was stirred for 4 h and then diluted with H_2O (20 ml) before it was alkalinized to pH 8 with 10% aq. K_2CO_3 soln. and extracted with CH_2Cl_2 (3 × 25 ml). The org. layer was dried ($MgSO_4$) and evaporated and the residue purified by CC (CH_2Cl_2/MeOH 97:3): 0.44 g (93%) of **6a**. $[\alpha]_D^{20} = -147.5$ (c = 0.333, MeOH). UV (MeOH): 282 (4.23). IR: 3330*m*, 3225*m*, 2985*m*, 2935*m*, 1694*s*, 1675*s*, 1505*m*, 1460*m*, 1438*m*, 1322*m*, 1312*s*, 1286*s*, 1246*s*, 1189*s*, 1172*s*, 1138*s*, 1122*s*, 1112*s*, 813*s*. ^{1}H-NMR: 9.25 (br. *s*, H–N(11)); 7.10 (br. *s*, H–N(10)); 7.00 (*dd*, J = 5.6, 1.4, H–C(3)); 5.96 (*dd*, J = 5.6, 1.4, H–C(2)); 2.88, 2.82 (2*d*, J = 14.6, CH_2(5)); 2.18 (*s*, Me–C(8)); 1.90 (*s*, Me–C(7)); 1.51 (*s*, *t*-Bu); 1.36 (*s*, Me–C(4)). ^{13}C-NMR: 173.3 (*s*, C(1)); 161.7 (*s*, COO); 154.6 (*d*, C(3)); 125.7, 125.6, 119.2, 118.0 (4*s*, C(6), C(7), C(8), C(9)); 125.6 (*d*, C(2)); 82.0 (*s*, Me_3C); 65.2 (*s*, C(4)); 35.0 (*t*, C(5)); 28.4 (*q*, Me_3C); 23.7 (*q*, *Me*–C(4)); 10.8 (*q*, *Me*–C(8)); 9.3 (*q*, *Me*–C(7)). EI-MS: 304 (1, $M^{+\cdot}$), 231 (8), 208 (35), 153 (18), 152 (100), 135 (6), 134 (52), 108 (5), 107 (6), 106 (12), 96 (9), 79 (9), 57 (6), 42 (6), 41 (10).

*(−)-(*R*)-1,4,5,10-Tetrahydro-4,7,8-trimethyl-1-oxo-11*H*-dipyrrin-9-carbaldehyde* (**6b**). A soln. of **6a** (304 mg, 1 mmol) in CF_3CO_2H (25 ml) was stirred under Ar for 15 min at r.t. before it was cooled in an ice-bath. Methyl orthoformate (10 ml) was added at once and the mixture stirred for further 30 min at r.t. After evaporation, the residue was dissolved in CH_2Cl_2, the soln. washed with aq. $NaHCO_3$ soln. and H_2O, dried ($MgSO_4$) and evaporated, and the residue crystallized from CH_2Cl_2/hexane: 182 mg (78%) of **6b**. M.p. 196–198°. $[\alpha]_D^{20} = -258.0$ (c = 0.189, MeOH). UV (MeOH): 312 (4.26), 278 (sh, 3.74). IR: 3300*m*, 3240*m*, 2977*m*, 2860*w*, 1677*s*, 1500*w*, 1480*w*, 1450*m*, 1428*m*, 1395*w*, 1365*m*, 1310*m*, 1285*s*, 1240*m*, 1220*w*, 1185*m*, 1164*s*, 1130*s*, 1115*s*, 818*s*. ^{1}H-NMR: 10.23 (br. *s*, H–N(11)); 9.43 (*s*, CHO); 7.41 (br. *s*, H–N(10)); 7.01 (*dd*, J = 5.5, 1.2, H–C(3)); 5.95 (*dd*, J = 5.5, 1.2, H–C(2)); 2.98, 2.90 (2*d*, J = 14.3, CH_2(5)); 2.24 (*s*, Me–C(8)); 1.92 (*s*, Me–C(7)); 1.41 (*s*, Me–C(4)). ^{13}C-NMR: 176.5 (*d*, J = 105.0, CHO); 173.4 (*s*, C(1)); 154.5 (*d*, J = 89.2, C(3)); 133.8, 132.1, 128.7, 119.3 (4*s*, C(6), C(7), C(8), C(9)); 125.9 (*d*, J = 83.6, C(2)); 65.1 (*s*, C(4)); 35.0 (*t*, C(5)); 24.0 (*q*, Me_3–C(4)); 9.0 (*q*, 2 Me). EI-MS: 232 (47, $M^{+\cdot}$), 137 (13), 136 (100), 135 (4), 108 (6), 97 (19), 77 (7), 69 (11), 54 (4), 42 (8).

*(+)-(4*R*,16*R*)-4,5,15,16-Tetrahydro-4,7,8,12,13,16-hexamethyl-22*H*-bilin-1,19(21*H*,24*H*)-dione* (**7**). A soln. of **6a** (46 mg, 0.151 mmol) in CF_3CO_2H (5 ml) was stirred under Ar at r.t. for 15 min before a soln. of **6b** (36 mg, 0.155 mmol) in CF_3CO_2H (5 ml) was added at once. After 15 min, the mixture was cooled in an ice-bath, MeOH (1 ml) added, and stirring continued for 1 h at r.t. Thereafter, the mixture was diluted with CH_2Cl_2 (100 ml), the soln. washed with sat. aq. $NaHCO_3$ soln. and H_2O, dried ($MgSO_4$) and evaporated, and the residue submitted to prep. TLC (CH_2Cl_2/MeOH 9:1). The component of R_f 0.30 was purified by MPLC (CH_2Cl_2/MeOH 9:1), and the yellow residue obtained after evaporation (22 mg, 35%) was recrystallized from CH_2Cl_2/hexane: pure **7**. M.p. 215–218°. $[\alpha]_D^{20} = +1960 \pm 20$ ($c = 4.8 \cdot 10^{-3}$, CH_2Cl_2). CD ($2.05 \cdot 10^{-5}$ M, MeOH): 469 (60010). CD ($1.15 \cdot 10^{-4}$ M, CH_2Cl_2): 465 (+56100). UV/VIS (CH_2Cl_2): 458 (4.40); UV/VIS (MeOH containing $Zn(AcO)_2$): 511 (4.67), 372 (3.62). Emission spectrum (MeOH containing $Zn(AcO)_2$): $\lambda_{max.}$ (exc.) 380.8; $\lambda_{max.}$ (em.) 529.8. $\Phi = 0.23$, relative to fluorescein ($\Phi = 0.77$ in aq. soln. of pH 9.6) as standard (*cf.* [26]). IR ($CHCl_3$): 3691*m*, 3608*w*, 3017*m*, 2921*w*, 2862*w*, 1691*s*, 1613*s*, 1367*w*, 1269*w*, 1226*w*, 934*w*. ^{1}H-NMR (CD_2Cl_2): 8.91 (br. *s*, H–N(21),H–N(24)); 6.98 (*dd*, J = 5.6, 1.5, H–C(3), H–C(17)); 6.68 (*s*, H–C(10)); 5.82 (*dd*, J = 5.6, 1.5, H–C(2), H–C(18)); 2.99, 2.77 (2*d*, J = 13.9, CH_2(5), CH_2(15)); 2.09 (*s*, Me–C(8), Me–C(12)); 1.91 (*s*, Me–C(7), Me–C(13)); 1.43 (*s*, Me–C(4), Me–C(16)). ^{13}C-NMR: 174.3 (*s*, C(1), C(19)); 154.6 (*d*, C(3), C(17)); 150.1, 137.4, 134.3, 124.0 (4*s*, C(6), C(7), C(8), C(9)); 126.3 (*d*, C(2), C(18)); 117.0 (*d*, C(10)); 65.0 (*s*, C(4), C(16)); 36.7 (*t*, C(5), C(15)); 23.3 (*q*, *Me*–C(4), *Me*–C(16)); 9.65 (*q*, 4 Me). FAB-MS: 441 ([M + Na]$^+$), 419 (MH$^+$).

*(+)-(4*R*,16*R*)-4,5,15,16-Tetrahydro-4,7,8,12,13,16-hexamethyl-22*H*-bilin-1,19(21*H*,24*H*)-dione Hydrochloride* (**7**·HCl). A soln. of **7** (10 mg, 23.9 μmol) in CH_2Cl_2 was treated with 10^{-2} M HCl in CH_2Cl_2. After evaporation, the residue was crystallized from CH_2Cl_2/hexane: **7**·HCl; orange crystals. M.p. 250° (dec.). $[\alpha]_D^{20} = +4900 \pm 50$ ($c = 1.4 \cdot 10^{-3}$, CH_2Cl_2). CD ($3.52 \cdot 10^{-5}$ M, CH_2Cl_2): 496 (+227700). UV/VIS (CH_2Cl_2): 496 (4.78). ^{1}H-NMR (CH_2Cl_2): 12.95 (br. *s*, NH); 7.83 (br. *s*, H–N(21), H–N(24)); 7.08 (*s*, H–C(10)); 7.01 (*dd*, J = 6.0, 1.5, H–C(3), H–C(17)); 5.76 (*dd*, J = 6.0, 1.5, H–C(2), H–C(18)); 3.56, 3.20 (2*d*, J = 13.8, CH_2(5),

CH_2(15)); 2.20 (*s*, Me–C(7), Me–C(13)); 1.91 (*s*, Me–C(8), Me–C(13)); 1.57 (*s*, Me–C(4), Me–C(16)). HR-FAB-MS: 419.2449 ($[C_{25}H_{13}N_4O_2]^+$; calc. 419.2447).

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