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Intramolecular hydrogen bonding and its influence on conformation. Circular dichroism of chiral bilirubin analogs

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Abstract

Enantiopure synthetic bilirubin analogs with variously modified (e.g. alkyl for natural propionic acid or ester) C(8) and C(12) side chains and with but a single chiral center in either or both, exhibited exciton coupled circular dichroism (CD) spectra. The CD intensity is greater when the stereogenic center is in a propionic acid side chain than in an alkyl side chain. © 1999 Elsevier Science Ltd. All rights reserved.

1. Introduction

Bilirubin (Fig. 1), the neurotoxic yellow-orange pigment of jaundice, is a tetrapyrrole dicarboxylic acid formed copiously from heme proteins (mainly the hemoglobin of red blood cells).[1–3] It is surprisingly water-insoluble at physiologic pH and does extract into aqueous bicarbonate,[3] peculiarities linked to its remarkable ability to fold into a bent shape, where the component dipyrrinone chromophores rotate about the central C(10) CH_2 to produce a stable conformation shaped like a half-opened book (or ridge-tile).[4–7] Non-bonded intramolecular steric interactions are minimized in such a conformation, which is stabilized by a network of intramolecular hydrogen bonds that bridge each propionic acid group to an opposing dipyrrinone lactam and pyrrole.[8–14] Since the conformation is determined only by the presence of the two dipyrrinones conjoined to a CH_2, and two propionic acids located at C(8) and C(12), a large number of bilirubin isomers (e.g. mesobilirubin-XIIIα, Fig. 1B) differing only in the lactam ring substituents have been found to exhibit the same solution properties as bilirubin and the same stabilized conformation.[4–10,15–18]

Recent studies have shown that stereogenic centers in the propionic acid chains of mesobilirubin-XIIIα, such as β,β′-, α,α′-, α,β′-dimethyl derivatives,[9,10,16,17] dimethoxy or di(methylthio),[19] can exert control over the conformational enantiomerism depicted in Fig. 1C. By minimizing nonbonded intramolecular steric compression interactions arising from such groups in the intramolecularly hydrogen bonded conformer, it was predicted and found that, for example, the (*S*,*S*) configuration methyls act to

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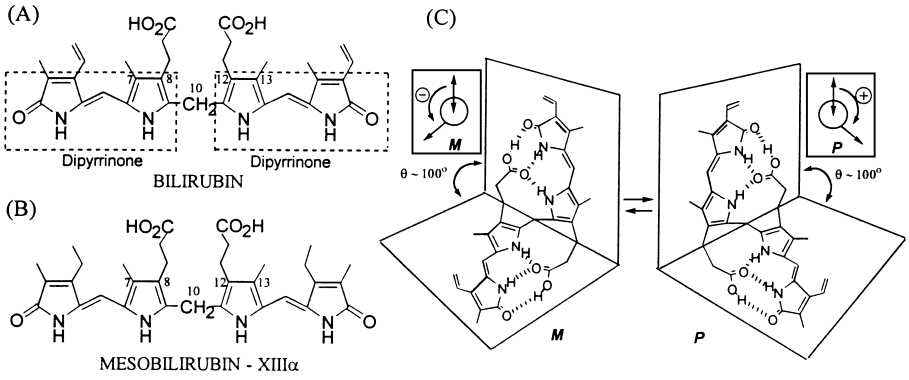
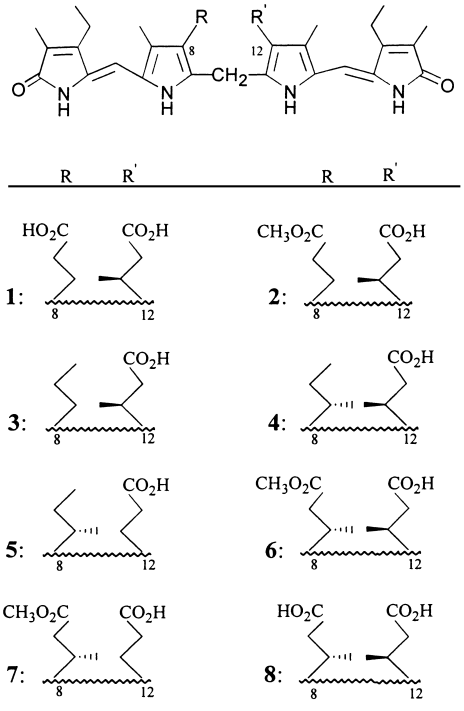


Figure 1. (A) Linear conformation of bilirubin; (B) linear conformation of mesobilirubin-XIIIα; (C) interconverting, enantiomeric ridge-tile conformations of bilirubin stabilized by intramolecular hydrogen bonding

tilt the equilibrium (Fig. 1C) decidedly toward the ***M***-helical conformer, whereas, (*R*,*R*) configuration methyls dictate a ***P***-helicity.[8–10] The (*R*,*S*) diastereomer presents an internal steric conflict, where the ***M*** and ***P*** helical conformers are either of equal energy (α,α′- and β,β′-) or of very close energy, as in α,β′-, where the α-methyl has a slightly greater steric demand than a β′-methyl.[16,17]

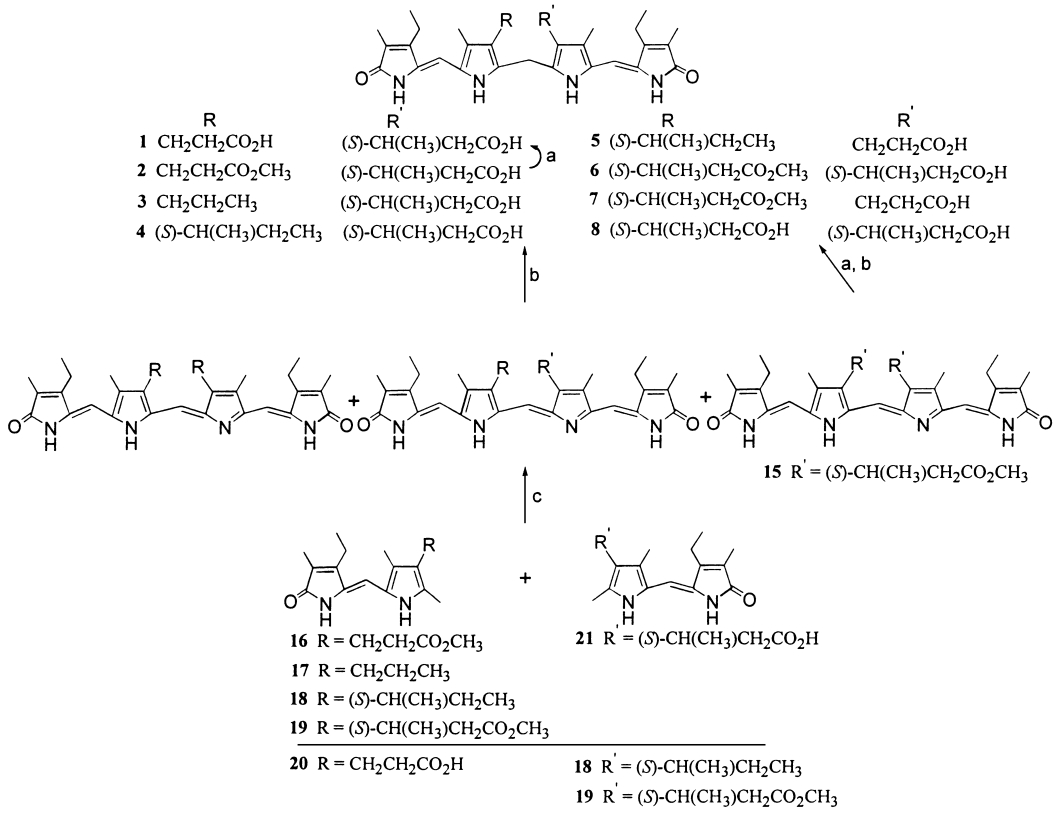
More recent studies on an optically active bilirubin analog with a single propionic acid (at C(12)) possessing a β′(*S*)-methyl indicated that a strong preference for the ***M***-helical ridge-tile conformation is still maintained.[20] In order to evaluate the importance of location of the stereogenic center in the various C(8) and C(12) substituents, we synthesized five new, optically active bilirubins (**1**, **2**, **4**, **5**, and **7**). Each has a single propionic acid group, and **1**, **2**, **5**, and **7** each have but a single stereogenic center — variously in the propionic acid, methyl ester or *sec*-butyl substituent. NMR and CD spectroscopic data for these pigments are compared to the new (**4**) and known (**6**[20] and **8**[9]) pigments with two stereogenic centers.



2. Results and discussion

2.1. Synthesis

Pigments **1**–**8** were synthesized directly from known (enantiomerically pure) dipyrrinones **16**–**21**.[16,21–23] Using the well-established oxidative coupling reaction, *p*-chloranil-promoted cross-coupling[24,25] of **21** with either of **16**, **17**, **18** or **19** (or **20** with either **18** or **19**) led smoothly in each case to a ternary mixture of verdins composed of two symmetrical verdins (from unavoidable self-coupling) and the desired cross-coupled verdin, as outlined in Scheme 1. The verdin mixtures were separated by radial chromatography on silica gel, and the non-symmetrical major product was saponified (when applicable) and reduced to the corresponding rubin with sodium borohydride in methanol to afford **1**–**7**, with **8** always arising as a by-product of any attempted cross-coupling using dipyrrinone **21**.

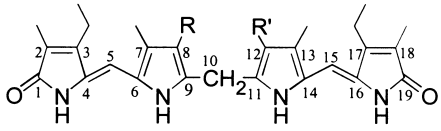


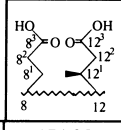
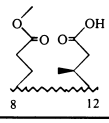
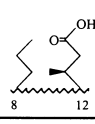
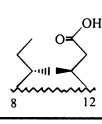
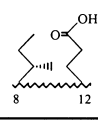
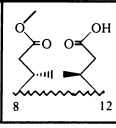
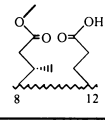
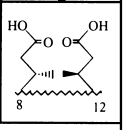
[a] 1. $NaOH/H_2O$; 2. HCl/H_2O. [b] 1. $NaBH_4/CH_3OH$; 2. CH_3CO_2H.

[c] 1. *p*-Chloranil, HCO_2H; 2. Chromatographic separation.

Scheme 1.

Table 1
Comparison of ^{13}C NMR data for mesobilirubin-XIIIα analogs **1**–**8** in 5×10^{-3} M $CDCl_3$ solutions at 25°C



Position	**1** 	**2** 	**3** 	**4** 	**5** 	**6** 	**7** 	**8** 
1,19-CONH	174.85 174.93	172.77 174.99	173.83 175.02	173.54 175.04	173.32 174.86	172.62 174.94	172.71 174.88	174.92
2,18	123.46 123.72	123.74 124.01	124.00 124.05	124.11 125.12	124.08 124.77	123.44 124.17	124.18	123.35
2,18-CH_3	7.94	7.93 8.23	7.96 8.46	7.96 8.29	7.96 8.30	7.96 8.27	7.94 8.24	7.95
3,17	148.34 148.38	147.61 148.25	148.17 148.29	148.06 148.27	147.94 148.32	147.58 148.29	147.53 148.28	148.39
3,17-CH_2CH_3	17.82	17.80 17.87	17.84 17.91	17.83 17.91	17.87 17.93	17.81 17.88	17.81 17.86	17.82
3,17-CH_2CH_3	14.88	14.72 14.84	14.77 14.89	14.60 14.87	14.52 14.86	14.75 14.87	14.76 14.84	14.90
4,16	128.21 128.37	128.38 129.68	128.13 129.89	128.43 130.61	128.47	128.31 129.40	128.54 129.14	128.25
5,15-CH=	100.33 100.44	97.68 100.65	99.32 101.38	98.60 100.93	98.34 100.82	97.49 100.25	97.43 100.53	100.29
6,14	123.32	123.43 123.53	123.10 123.51	123.76 123.91	123.28 123.65	122.56 123.39	122.46 123.41	123.31
7,13	119.28 122.34	118.55 120.42	121.38 122.02	120.65 123.29	119.96 121.54	120.56 122.21	119.56 121.89	122.42
7,13-CH_3	10.09 11.01	9.71 11.08	9.67 11.15	10.52 11.15	10.23 10.54	10.92 11.10	10.20	11.01
8,12	124.25 124.74	124.30 124.88	124.64 124.95	125.28 125.77	125.61 125.83	124.62 124.80	124.32	124.84
8^1,12^1-CH_2 or CH	18.50 26.52	19.32 26.76	25.97 27.09	26.95 32.53	18.71 32.56	26.68 27.20	18.65 27.68	26.47
8^1,12^1-CH_3	21.00	21.20	21.31	21.01 21.37	21.55	21.10 21.26	21.80	20.94
8^2,12^2-CH_2	32.63 39.55	33.95 39.45	24.46 39.52	29.65 39.56	30.20 32.61	39.51 40.24	32.66 40.93	39.56
8^3,12^3	179.25 179.39	175.67 179.23	13.99 179.12	13.12 179.02	13.18 179.23	175.60 179.36	175.65 179.46	180.14
8^4,12^4-OCH_3	—	52.62	—	—	—	52.74	53.08	—
9,11	132.71 133.40	130.34 132.86	131.01 132.10	131.26	131.15 131.32	130.64 132.70	130.91 132.12	132.93
10-CH_2	22.01	21.99	22.28	22.64	22.84	21.50	22.17	21.85

2.2. ^{13}C NMR spectra and structure

^{13}C NMR chemical shifts of **1**–**8** may be found in Table 1. Assignments for **1**, **2**, **4**, **5**, and **7** are based on those of **3**, **6**, and **8**, previously assigned by analogy and by H,C COSY and HMBC methods.[9,16,17,20] The unsymmetrical isomers show signal doubling, as expected. Interestingly, when one of the two carboxylic acids is absent (as in **3**–**5**) or present as its methyl ester (**2**, **6**, **7**), one of the lactam carbonyls becomes shielded. We assume that this lactam is the one present in the dipyrrinone to which the propionic acid is covalently attached, i.e. in the dipyrrinone that does not participate in intramolecular hydrogen bonding.

2.3. ^{1}H NMR analysis and intramolecular hydrogen bonding

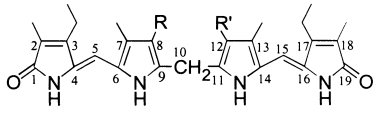
Bilirubins are thought to adopt preferentially a ridge-tile like conformation in order to minimize intramolecular non-bonded steric repulsions. Intramolecular hydrogen bonding greatly stabilizes this conformation in the crystal[4–7] and in solutions in nonpolar solvents.[8–10,13–21,26–29] It is thought to be important even in polar and hydroxylic solvents, and in bilirubin carboxylate anions.[7,13,14] Hydrogen bonding produces large deshieldings of the dipyrrinone lactam and pyrrole NHs (from ~8 ppm to ~9–11 ppm), and in bilirubins an examination of dipyrrinone NH chemical shifts have proven to be an excellent probe of the presence and nature of hydrogen bonding. Earlier studies showed that the lactam NH chemical shift lies near δ of 10.5–11.0 ppm and the pyrrole NH near δ of 9.0–9.2 ppm when the dipyrrinone is joined to a carboxylic acid group by intramolecular hydrogen bonding.[8–10,13–21] Such chemical shifts are found wherever there is a carboxylic acid group in **1**–**8** (Table 2) at C(8) or C(12). Small differences can be detected when one of the propionic acids is unsubstituted and one has a β-methyl, as in **1** (cf. **7**). When a propionic acid methyl ester is present at C(8) and a propionic acid at C(12), as in **2**, **6**, and **7**, one dipyrrinone (A/B) remains strongly hydrogen bonded (N(21)H and N(22)H) but the other dipyrrinone (C/D) is less well hydrogen bonded, as evidenced by the more shielded N(23)H and N(24)H signals. Partial hydrogen bonding must still reside with the propionate ester group because when it is replaced by an alkyl group (as in **3**–**5**) the C/D dipyrrinone NHs are even more shielded — up to the positions normally found in nonhydrogen bonded lactams and pyrroles. Yet, despite the reduced or absent intramolecular hydrogen bonding in one half of the rubin, intramolecular hydrogen bonding remains strong in the other half.

Additional evidence for intramolecular hydrogen bonding in the propionic acid groups of **1**–**8** comes from an analysis of the $^{1}H\{^{1}H\}$ coupling constants in the propionic acid chain. Thus, one finds an ABX pattern for $-CH_X(CH_3)-CH_AH_B-CO_2H$ segment of **1**, **2**, **4**, **6**, **7**, and **8** in $CDCl_3$ solvent. This is consistent with very limited segmental motion, as expected when the acid is tethered to a dipyrrinone by intramolecular hydrogen bonding, as shown in Fig. 1C and Fig. 2. If the C(8) group is a propionic acid (as in **1**), here too there is limited segmental motion, as evidenced by the ABCX coupling pattern of the $-CH_XH_A-CH_BH_C-CO_2H$. However, if the C(8) group is methyl propionate (**2**, **6**) or propyl (**3**) or *sec*-butyl (**4**, **5**) more motion in the chain is evident from the coupling pattern.

2.4. Circular dichroism and stereochemistry

Each of rubins **1**–**8** have at least one propionic acid group, and each has at least one stereogenic center — although not necessarily in a propionic acid. The propionic acids are expected to engage in intramolecular hydrogen bonding to an opposing dipyrrinone (Fig. 2), and when a β(*S*) stereogenic center is present in the propionic acid, the conformational enantiomerism shown in Fig. 1C is expected to be driven toward the ***M***-helical conformer by equilibrium-displacing steric perturbations of intramolecular origin.[8–10,16,17,19,20]

Table 2
^{1}H NMR assignments for mesobilirubin-XIIIα analogs **1**–**8** in 10^{-3} M $CDCl_3$ solutions at 25°C



Proton	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
								
21,24-NHCO	10.49 10.76	8.98 10.85	8.74 10.86	8.22 10.78	7.93 10.53	9.12 10.85	9.30 10.61	10.68
22,23-NH	9.06 9.12	8.25 9.12	8.43 8.97	7.98 8.76	7.78 9.00	8.37 9.14	8.58 9.14	9.04
8^3,12^3-COOH	13.58	13.62	13.71	13.69	13.60	13.63	13.60	13.64
2,18-CH_3	1.86	1.85 1.86	1.87 1.89	1.86	1.87	1.85 1.86	1.85 1.86	1.85
3,17-CH_2CH_3	2.47[a]	2.48[a]	2.48[o]	2.47[a] 2.49[a]	2.47[o] 2.49[o]	2.48[a]	2.48[a]	2.48[a]
3,17-CH_2CH_3	1.11[b]	1.12[b] 1.13[b]	1.13[p]	1.13[b]	1.13[p]	1.12[b] 1.13[b]	1.13[b]	1.11[b]
5,15-CH=	6.03 6.05	5.94 6.05	5.98 6.09	5.95 6.09	5.93 6.09	5.95 6.05	5.94 6.06	6.04
7,13-CH_3	2.15 2.25	2.13 2.23	2.09 2.22	2.13 2.20	2.10 2.14	2.22 2.23	2.14	2.24
8^1-CH_2	2.56[c] 2.99[d]	2.76[j]	2.44-2.49[e]	—	—	—	—	—
8^1-CH	—	—	—	2.72-2.79[e]	2.74-2.80[e]	3.41-3.47[e]	3.38-3.43[e]	3.42-3.48[e]
8^1-CH_3	—	—	—	1.29[t]	1.30[x]	1.33[f]	1.35[x]	1.35[ad]
12^1-CH	3.42-3.48[e]	3.43-3.48[e]	3.43-3.49[e]	3.42-3.48[e]	—	3.42-3.47[e]	—	3.42-3.48[e]
12^1-CH_2	—	—	—	—	2.77-2.82[e]	—	2.73-2.81[e]	—
12^1-CH_3	1.34[f]	1.32[k]	1.32[f]	1.32[f]	—	1.35[f]	—	1.35[ad]
8^2-CH_2	2.78-2.84[e] 2.84-2.90[e]	2.71-2.77[e] 2.87-2.92[e]	1.46-1.54[e]	1.64[u] 1.65[u]	1.63-1.68[e]	2.73[y] 3.00[z]	2.76-2.91[e]	2.70[ae] 3.08[af]
12^2-CH_2	2.71[g] 3.12[h]	2.72[l] 3.12[m]	2.70[q] 3.13[r]	2.70[v] 3.10[w]	2.79-2.83[e]	2.69[aa] 3.12[ab]	2.76-2.91[e]	2.70[ae] 3.08[af]
8^3 (or 12^3)-CH_3	—	—	0.93[s]	0.87[s]	0.90[s]	—	—	—
8^3 (or 12^3)-OCH_3	—	3.73	—	—	—	3.68	3.74	—
10-CH_2	4.02[i] 4.11[i]	3.94[n] 4.04[n]	4.01[n] 4.07[n]	4.01[n] 4.09[n]	4.04	3.96[ac] 4.05[ac]	4.04	4.06

[a] q, J=7.6 Hz; [b] t, J=7.6 Hz; [c] ABC*X*, ^{3}J=4.2 Hz, ^{3}J=2.3 Hz, ^{2}J=14.6 Hz; [d] *A*BCX, m; [e] m; [f] d, J=7.3 Hz; [g] *A*BX, ^{3}J=3.0 Hz, ^{2}J=18.2 Hz; [h] A*B*X, ^{3}J=12.2 Hz, ^{2}J=18.2 Hz; [i] AB, ^{2}J=15.6 Hz; [j] t, J=8.1 Hz; [k] d, J=7.2 Hz; [l] *A*BX, ^{3}J=2.1 Hz, ^{2}J=18.0 Hz; [m] A*B*X, ^{3}J=12.3 Hz, ^{2}J=18.0 Hz; [n] AB, ^{2}J=16.0 Hz; [o] q, J=7.5 Hz; [p] t, J=7.5 Hz; [q] *A*BX, ^{3}J=3.2 Hz, ^{2}J=17.7 Hz; [r] A*B*X, ^{3}J=12.3 Hz, ^{2}J=17.7 Hz; [s] t, J=7.3 Hz; [t] d, J=7.2 Hz; [u] dq, ^{3}J=7.3 Hz; [v] *A*BX, ^{3}J=3.1 Hz, ^{2}J=17.8 Hz; [w] A*B*X, ^{3}J=12.3 Hz, ^{2}J=17.8 Hz; [x] d, J=7.1 Hz; [y] *A*BX, ^{3}J=4.5 Hz, ^{2}J=16.1 Hz; [z] A*B*X, ^{3}J=12.2 Hz, ^{2}J=16.1 Hz; [aa] *A*BX, ^{3}J=2.7 Hz, ^{2}J=18.2 Hz; [ab] A*B*X, ^{3}J=12.2 Hz, ^{2}J=18.2 Hz; [ac] AB, ^{2}J=15.9 Hz; [ad] d, J=7.4 Hz; [ae] *A*BX, ^{3}J=3.0 Hz, ^{2}J=18.2 Hz; [af] A*B*X, ^{3}J=12.3 Hz, ^{2}J=18.2 Hz.

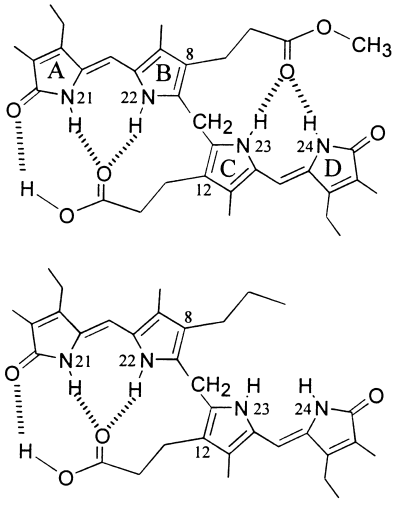


Figure 2. Intramolecular hydrogen bonding from one propionic acid to an opposing dipyrrinone in bilirubin analogs. Partial hydrogen bonding may occur from a propionate ester (upper), but no hydrogen bonding is expected from an alkyl group (lower)

According to exciton chirality theory,[30] the signed order of the bisignate CD Cotton effects may be used to predict the relative orientation of the two electric dipole transition moments, one from each dipyrrinone of the rubin. Thus, a positive exciton chirality (long wavelength (+) Cotton effect followed by a (−) short wavelength Cotton effect) corresponds to a positive torsion angle between the transition dipoles, and a negative exciton chirality (long wavelength (−) Cotton effect followed by a (+) short wavelength Cotton effect) corresponds to a negative torsion angle. The ***M***-helicity conformer of Fig. 1C is predicted to have a negative exciton chirality; the ***P***-helicity is predicted to have a positive exciton chirality.[8–10,31,32]

When the predicted forced displacement of the ***M*** ⇄ ***P*** equilibrium is achieved, the two dipyrrinone chromophores will be held in either the ***M*** or ***P*** fixed chiral geometry, depending on the *R*,*S* stereochemistry at the stereogenic center(s). The ***M*** fixed chiral geometry expected from the β(*S*) stereochemistry should lead to strong optical activity, and a negative exciton chirality circular dichroism (CD) spectrum.[8,9] This is particularly true in **8**, studied earlier, for which one observes intense bisignate CD in nonpolar and polar solvents: $\Delta\varepsilon_{433}^{max}$ −319, $\Delta\varepsilon_{392}^{max}$ +180 (CH_2Cl_2) and $\Delta\varepsilon_{429}^{max}$ −246, $\Delta\varepsilon_{386}^{max}$ +165 (DMF) (see Figs. 3 and 4) associated with the long wavelength UV-vis absorption near 420–430 nm.[9] Here there are strategically located two propionic acids, each with a β(*S*) stereogenic center. With only one stereogenic center, as in **1**, the CD intensity drops by one-half: $\Delta\varepsilon_{433}^{max}$ −227, $\Delta\varepsilon_{388}^{max}$ +132 (CH_2Cl_2) and $\Delta\varepsilon_{425}^{max}$ −132, $\Delta\varepsilon_{384}^{max}$ +89 (DMF). When one of the two propionic acids of **1** is esterified as monomethyl esters **2** and **7**, the location of the stereogenic center (whether in the acid or the ester chain) profoundly affects the CD. When in the acid chain, as in **2**, the CD in nonpolar solvents (Fig. 3, Table 3) does not differ much from **1**. But when in the ester chain, as in **7**, the CD drops to ~10% of the values found in **1** and **2**. The CD of **2** and **7** is more strongly affected by polar solvents (Fig. 4, Table 3). In contrast, when the mono-ester has a β(*S*) stereogenic center in each propionic chain, as in **6**, the CD data remain comparable to those of **1** in polar as well as in most nonpolar solvents (Figs. 3 and 4, Table 3). These data suggest that: (i) a single stereogenic center in a propionic acid has a dominant effect in displacing the ***M*** ⇄ ***P*** conformational equilibrium; (ii) whether the second group is a propionic acid or methyl ester makes little difference in nonpolar solvents, and having a stereogenic center in this group is more effective than the choice of acid or ester — especially in polar solvents; and (iii) in the mono ester, given the choice of location of a single stereogenic center, it is most important that it be in the acid chain.

For those rubins with a propionic acid at C(12) and an alkyl group at C(8), e.g. **3**–**5**, when the

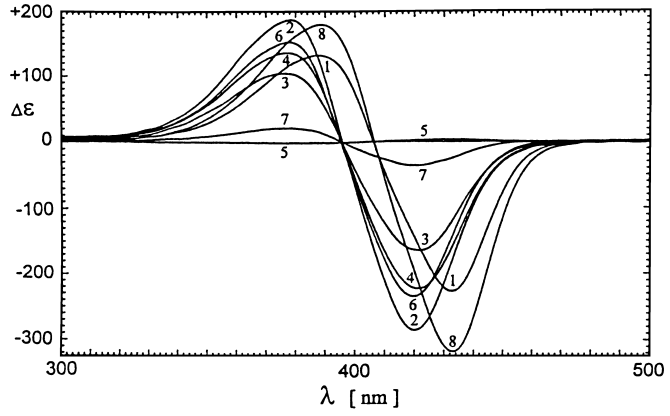


Figure 3. Bisignate circular dichroism of 1.5×10^{-5} M solutions of bilirubin analogs **1**–**8** in dichloromethane at room temperature. The compound number appears next to its CD curve

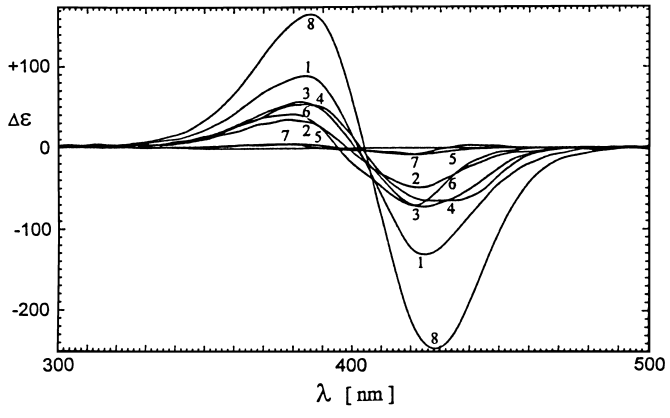


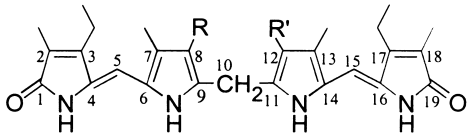
Figure 4. Bisignate circular dichroism of 1.5×10^{-5} M solutions of bilirubin analogs **1**–**8** in dimethylformamide at room temperature. The compound number appears next to its CD curve

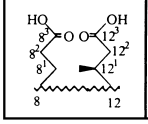
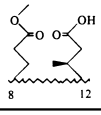
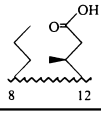
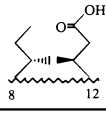
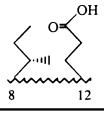
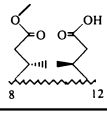
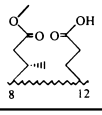
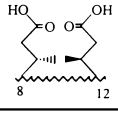
stereogenic center is in the propionic acid chain, the CD spectra remain intense in nonpolar solvents (Fig. 3, Table 3), cf. **3** and **4** to **2** and **6**. The presence of a second stereogenic center, now in the alkyl chain (as in **4**) offers added stabilization to the ***M***-helical conformer, much as was seen in the mono-ester, cf. **4** and **6** vs. **2** and **3**. The conformation determining benefits from the second stereogenic center (in **4** vs. **3**) are less apparent in polar solvents (Fig. 4, Table 3). Nevertheless, comparison of the CD data for **4** and **6**, and **2** and **3** clearly indicates that a propionic ester is more effective than an alkyl group in stabilizing conformation. This is probably due to hydrogen bonding between the ester carbonyl and the opposing dipyrrinone (Fig. 2).

When the stereogenic center is in the alkyl chain and not in the propionic acid (**5**), the CD magnitudes drop to very small values and are typically signed opposite to those of the other entries of Table 3. The situation is comparable to that of mono ester **7**, where the stereogenic center is in the ester chain and not the acid, but here the CD intensities, while small, are much larger than those of **5**, and the Cotton effect signs are not inverted. These data (compared to those of **5**) also suggest a role for the carbomethoxy group in intramolecular hydrogen bonding.

The CD data from $(CH_3)_2SO$ solvent are distinct from those of other solvents, as has been noted previously.[8–10,13–21] The bisignate CD are typically very weak, and the Cotton effect signs are often inverted (Table 3). $(CH_3)_2SO$ is thought to insert into the matrix of intramolecular hydrogen bonds in bilirubins, with the propionic acid groups being tethered to opposing dipyrrinones through solvent

Table 3
Comparison of circular dichroism and ultraviolet–visible spectral data from ~1.5×10^{-5} M solutions of mesobilirubin-XIIIα analogs **1**–**8** at 22°C



1	**2**	**3**	**4**	**5**	**6**	**7**	**8**
							

Pigment	Solvent	ϵ^a	CD			UV	
			$\Delta\epsilon^{max}(\lambda_1)$	λ_2 at $\Delta\epsilon=0$	$\Delta\epsilon^{max}(\lambda_3)$	ϵ^{max}	λ (nm)
1	Benzene	2.3	–250 (434)	407	+137 (391)	49 400	434
2			–213 (421)	395	+134 (379)	52 100	415
3			–178 (423)	396	+111 (379)	41 900	415
4			–195 (422)	396	+114 (378)	44 200	415
5			+11 (431)	404	–6 (387)	46 800	417
6			–254 (420)	395	+152 (380)	52 700	416
7			–92 (420)	398	+55 (381)	55 500	415
8			–362 (434)	406	+191 (390)	60 000	432
1	$CHCl_3$	4.7	–266 (434)	407	+147 (389)	50 300	431
2			–236 (422)	397	+156 (379)	52 800	417
3			–178 (424)	398	+115 (379)	43 800	416
4			–200 (423)	397	+126 (379)	46 100	417
5			+7 (431)	414	–3 (395)	48 500	418
6			–238 (422)	397	+153 (380)	53 300	418
7			–32 (425)	398	+19 (381)	54 200	416
8			–337 (434)	407	+186 (389)	55 800	431
1	THF	7.3	–238 (432)	406	+134 (388)	51 600	431
2			–173 (419)	395	+117 (377)	48 300	413
3			–179 (420)	396	+127 (379)	44 400	412
4			–174 (419)	396	+117 (379)	43 900	412
5			—	—	—	—	—
6			–210 (419)	395	+133 (378)	53 000	414
7			–54 (419)	395	+33 (378)	52 600	414
8			–338 (433)	406	+188 (390)	57 900	431
1	CH_2Cl_2	8.9	–227 (433)	406	+132 (388)	49 800	431
2			–286 (420)	396	+187 (378)	52 100	412
3			–165 (422)	395	+105 (377)	42 300	415
4			–224 (421)	396	+136 (377)	44 800	416
5			+3 (428)	—	—	46 200	417
6			–235 (420)	395	+153 (378)	52 300	415
7			–36 (421)	396	+21 (377)	55 000	415
8			–319 (433)	407	+180 (392)	56 400	430

Table 3 (continued)

1	$(CH_3)_2CO$	20.7	–216 (430)	404	+125 (386)	49 700	427
2			–197 (419)	394	+134 (377)	50 600	412
3			–168 (420)	396	+120 (377)	42 800	411
4			–164 (419)	395	+116 (377)	43 300	408
5			—	—	—	—	—
6			–209 (418)	394	+134 (376)	50 300	412
7			–21 (421)	395	+12 (376)	53 500	412
8			–322 (430)	404	+182 (387)	57 100	427
1	CH_3OH	32.6	–130 (430)	404	+86 (385)	50 600	425
2			–91 (424)	400	+71 (382)	57 700	424
3			–74 (427)	402	+56 (382)	51 000	425
4			–54 (427)	403	+49 (383)	50 900	424
5			+2 (428)	404	–3 (394)	58 100	429
6			–153 (424)	398	+98 (380)	55 800	422
7			–29 (423)	398	+16 (383)	59 700	423
8			–285 (431)	405	+177 (386)	60 800	425
1	CH_3CN	36.2	–211 (428)	403	+125 (384)	49 000	425
2			–195 (417)	393	+129 (374)	49 400	410
3			–163 (419)	394	+114 (375)	42 000	404
4			–161 (418)	394	+114 (376)	42 100	402
5			—	—	—	—	—
6			–210 (417)	392	+134 (375)	50 000	410
7			–14 (422)	393	+7 (378)	51 300	410
8			–315 (429)	403	+181 (384)	56 700	423
1	$(CH_3)_2NCHO$	36.7	–132 (425)	402	+89 (384)	49 100	423
2			–49 (423)	398	+35 (378)	55 400	422
3			–73 (424)	401	+57 (382)	47 300	395
4			–65 (427)	403	+54 (383)	47 100	420
5			—	—	—	—	—
6			–70 (421)	395	+43 (379)	53 400	420
7			–8 (421)	388	+4 (379)	57 100	421
8			–246 (429)	404	+165 (386)	54 000	421
1	$(CH_3)_2SO$	46.5	–3 (436)	413	+7 (381)	52 500	428
2			–2 (433)	414	+8 (380)	60 700	428
3			–2 (432)	413	+8 (384)	55 500	429
4			—	—	+5 (387)	54 200	429
5			–9 (426)	391	+5 (376)	61 900	431
6			+18 (427)	393	–11 (378)	56 000	426
7			+4 (428)	397	–3 (374)	60 800	428
8			+23 (425)	385	–6 (369)	56 700	425
1	CH_3NHCHO	182.4	–183 (427)	400	+113 (383)	54 700	427
2			–119 (423)	397	+76 (379)	56 500	424
3			–103 (425)	398	+64 (381)	50 200	425
4			–73 (425)	398	+45 (380)	49 800	424
5			—	—	—	—	—
6			–152 (422)	395	+85 (379)	55 600	421
7			–27 (419)	392	+14 (379)	57 500	423
8			–359 (427)	400	+200 (383)	66 000	426

[a] Dielectric constant from Gordon, A.J.; Ford, R.A. *The Chemist's Companion*, Wiley, NY 1972, pp 4–8.

molecules.[26–29,33,34] Such loosening of hydrogen bonds tends to remove the intramolecular nonbonded steric interactions that together with stereogenic centers in the propionic acid chains govern the position of the ***M*⇄*P*** conformational equilibrium (of Fig. 1C). By relaxing these stereochemical criteria, **1**–**4** and **6**–**8** in $(CH_3)_2SO$ behave more like **5** in all solvents, and thus the CD spectra are correspondingly weak. We assume that the very weak CD spectra originate from only a slight displacement of a 1:1 ***M*⇄*P*** conformational equilibrium. However, yet a different explanation might come from a reorienting

of the dipyrrinone electric transition dipole moments (Fig. 1C) to nearly parallel (and hence to very weak bisignate Cotton effects) due to a more open dihedral angle (θ) forced by intercalating molecules of $(CH_3)_2SO$.

3. Concluding comments

Intramolecular hydrogen bonding, which has a dominating influence on stabilizing the conformation of bilirubin as a ridge-tile shape, is present and very effective even when the rubin has only one propionic acid. The current study shows that a single propionic acid group at C(8) or C(12) is sufficient for conformational stabilization. When a stereogenic center is present in the lone propionic acid chain, the intramolecular nonbonded interactions are sufficient to displace the ***M*⇄*P*** conformational equilibrium in a major way toward either ***M*** (β*S* configuration) or ***P*** (β*R* configuration). When a stereogenic center is absent from the propionic acid chain, e.g. at C(12), but present in a methyl propionate chain at C(8), the unbalancing of the conformational equilibrium is much less effective. When the stereogenic center is in a C(8) alkyl chain, the influence is marginal.

4. Experimental

4.1. General

All circular dichroism spectra were recorded on a Jasco J-600 spectropolarimeter, and all UV–vis spectra were recorded on a Perkin–Elmer lambda 12 or a Cary 219 spectrophotometer. Optical rotations were measured on a Perkin–Elmer model 141 polarimeter. NMR spectra were obtained on a GN-300 or Varian Unity Plus spectrometers at 300 and 500 MHz ^{1}H frequency, respectively. Deuteriochloroform solvent was used throughout and chemical shifts were reported in δ ppm referenced to residual $CHCl_3$ ^{1}H signal at 7.26 ppm and $CDCl_3$ ^{13}C signal at 77.00 ppm. J-modulated spin-echo experiments (attached proton test) were used to obtain the ^{13}C NMR assignments. HPLC analyses were carried out on a Perkin–Elmer series 410 high-pressure liquid chromatograph with a Perkin–Elmer LC-95 UV–vis spectrophotometric detector (set at 420 nm for rubinoid compounds) equipped with a Beckman Altex ultrasphere IP 5 μm C-18 ODS column (25×0.46 cm) kept at 34°C. The flow rate was 1 mL per minute, and the mobile phase was 0.1 M di-*n*-octylamine acetate buffer in 5% H_2O in methanol (v/v) with pH 7.7 at 22°C. Radial chromatography was carried out on Merck silica gel PF_{254} with $CaSO_4$ binder preparative layer grade, using a Chromatotron (Harrison Research, Inc., Palo Alto, CA) with 1, 2, or 4 mm thick rotors. Melting points were determined on a Mel-Temp capillary apparatus and are uncorrected. High resolution FAB mass spectra were obtained at the Midwest Center for Mass Spectrometry, University of Nebraska, Lincoln, for samples which were >95% pure by HPLC and ^{13}C NMR.

The spectral data were obtained in spectral grade solvents (Aldrich or Fischer). HPLC grade solvents were dried and purified following standard procedures.[35]

4.2. The starting compounds

3-Ethyl-8-(2-(methoxycarbonyl)ethyl)-2,7,9-trimethyl-1,10-dihydro-(11*H*)-dipyrrin-1-one (**16**, xanthobilirubic acid methyl ester),[23] 3-ethyl-8-propyl-2,7,9-trimethyl-1,10-dihydro-(11*H*)-dipyrrin-1-one (**17**),[20] 8-(2-carboxyethyl)-3-ethyl-2,7,9-trimethyl-1,10-dihydro-(11*H*)-dipyrrin-1-one (**20**, xanthobilirubic acid),[23] and optically pure (+)-(*S*)-3-ethyl-8-(1-methylpropyl)-2,7,9-trimethyl-1,10-

dihydro-(11*H*)-dipyrrin-1-one (**18**),[21] (+)-(*S*)-3-ethyl-8-(2-methoxycarbonyl-1-methylethyl)-2,7,9-trimethyl-1,10-dihydro-(11*H*)-dipyrrin-1-one (**19**),[9] (−)-(*S*)-3-ethyl-8-(2-carboxy-1-methylethyl)-2,7,9-trimethyl-1,10-dihydro-(11*H*)-dipyrrin-1-one (**21**)[36] were synthesized as described previously.

4.3. Mesobiliverdin-XIIIα analogs. General procedure

A mixture of 1 mmol of each corresponding dipyrrinone, 1.23 g (5 mmol) of *p*-chloranil, 440 mL of CH_2Cl_2 and 22 mL of formic acid (97%) was heated at reflux for 24 h. The volume of the mixture was reduced by distillation to one half, and reflux was continued for 6 h. Then the mixture was chilled overnight at −20°C. The separated solid was filtered, and the blue filtrate was washed with water (3×200 mL). The organic layer was dried over anhydrous Na_2SO_4, filtered, and the solvent was evaporated under vacuum. The crude mixture of three verdins was redissolved in minimum volume of CH_2Cl_2, filtered from unreacted *p*-chloranil and separated by radial chromatography on silica gel eluting with gradient CH_3OH:CH_2Cl_2=1.5:98.5 to 6:94 (v/v). Only the pure fractions containing medium polarity blue band were collected, combined and evaporated under vacuum to afford unsymmetric mesobiliverdin-XIIIα analogs.

*4.4. (*S*)-3,17-Diethyl-8-(2-methoxycarbonylethyl)-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(21*H*,24*H*)-bilin-1,19-dione (**9**, β′S-methylmesobiliverdin-XIIIα 8-methyl ester)*

The title compound was obtained in 68% yield, mp 154–157°C. ^{1}H NMR: δ 1.12 (3H, d, J=6.9 Hz), 1.25 (3H, t, J=7.4 Hz), 1.26 (3H, t, J=7.4 Hz), 1.84 (3H, s), 1.86 (3H, s), 2.14 (3H, s), 2.16 (3H, s), 2.46 (4H, q, J=7.4 Hz), 2.48 (2H, t, J=7.9 Hz), 2.57 (2H, t, J=7.9 Hz), 2.91 (2H, *ABX*, ^{3}J=7.3, 9.8 Hz), 3.16–3.22 (1H, m), 3.61 (3H, s), 6.01 (1H, s), 6.18 (1H, s), 6.76 (1H, s), 8.38 (1H, br. s), 9.74 (1H, br. s), ~11 (1H, very br. s) ppm; and ^{13}C NMR: δ 8.07, 8.22, 9.25, 10.82, 14.50, 17.82, 17.95, 19.80, 20.51, 27.64, 35.36, 39.60, 51.64, 96.69, 98.36, 114.12, 126.53, 127.47, 128.29, 128.80, 135.02, 136.54, 137.75, 141.15, 144.84, 145.39, 146.71, 148.01, 171.59, 172.80, 175.30, 176.69 ppm. HRMS (3-NBA): calcd for $C_{35}H_{43}N_4O_6$ $(M+H)^+$ 615.3183; found 615.3175, error 1.2 ppm, Δ0.7 mDa.

*4.5. (*S*)-3,17-Diethyl-8-propyl-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(21*H*,24*H*)-bilin-1,19-dione (**10**)*

The title compound was obtained in 62% yield. For spectral data see Boiadjiev et al.[20]

*4.6. (*S,S*)-3,17-Diethyl-8-(1-methylpropyl)-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(21*H*,24*H*)-bilin-1,19-dione (**11**)*

The title compound was obtained in 59% yield, mp 216–218°C. ^{1}H NMR: δ 0.80 (3H, t, J=7.3 Hz), 1.15 (3H, d, J=6.8 Hz), 1.26 (6H, t, J=7.5 Hz), 1.32 (3H, d, J=7.0 Hz), 1.61–1.68 (2H, m), 1.84 (3H, s), 1.86 (3H, s), 2.14 (3H, s), 2.18 (3H, s), 2.46–2.51 (2H, m), 2.56 (4H, q, J=7.5 Hz), 2.82–2.89 (1H, m), 3.19–3.25 (1H, m), 6.02 (1H, s), 6.19 (1H, s), 6.86 (1H, s), 8.38 (1H, br. s), 9.46 (1H, br. s) ppm; and ^{13}C NMR: δ 8.11, 8.28, 10.43, 10.88, 12.78, 14.54, 17.88, 18.00, 20.52, 20.72, 27.85, 29.94, 32.95, 39.84, 96.83, 98.38, 114.91, 125.77, 127.24, 128.13, 128.51, 137.26, 137.32, 137.47, 140.78, 141.66, 144.46, 144.96, 146.79, 148.09, 171.52, 175.00, 176.64 ppm. HRMS (3-NBA): calcd for $C_{35}H_{45}N_4O_4$ $(M+H)^+$ 585.3441; found 585.3467, error −4.5 ppm, Δ−2.6 mDa.

*4.7. (*S,S*)-3,17-Diethyl-8-(2-methoxycarbonyl-1-methylethyl)-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(21*H,*24*H)*-bilin-1,19-dione (**12**, β*S,β′*S*-dimethylmesobiliverdin-XIIIα 8-methyl ester)*

The title compound was obtained in 69% yield. For spectral data see Boiadjiev et al.[20]

*4.8. (*S*)-3,17-Diethyl-8-(2-carboxyethyl)-12-(1-methylpropyl)-2,7,13,18-tetramethyl-(21*H,*24*H)*-bilin-1,19-dione (**13**)*

The title compound was obtained in 44% yield, mp 176–179°C. ^{1}H NMR: δ 0.86 (3H, t, J=7.3 Hz), 1.24 (2×3H, 2×t, J=7.5 Hz), 1.32 (3H, d, J=7.0 Hz), 1.67–1.72 (2H, m), 1.83 (3H, s), 1.85 (3H, s), 2.00 (3H, s), 2.14 (3H, s), 2.15 (2H, t, J=7.5 Hz), 2.52 (2H, q, J=7.5 Hz), 2.55 (2H, q, J=7.5 Hz), 2.69 (2H, t, J=7.5 Hz), 2.78–2.86 (1H, m), 5.95 (1H, s), 6.08 (1H, s), 6.80 (1H, s), 8.71 (2H, br. s), ~9.3 (1H, very br. s) ppm; and ^{13}C NMR: δ 8.16, 8.20, 9.54, 10.29, 12.85, 14.48, 17.81, 17.85, 19.80, 20.97, 30.10, 33.25, 35.25, 97.00, 97.69, 115.03, 126.66, 127.84, 128.13, 128.62, 138.77, 139.45, 140.06, 142.47, 143.11, 146.77, 147.33, 172.71, 174.22, 176.30 ppm. HRMS (3-NBA): calcd for $C_{34}H_{43}N_4O_4$ $(M+H)^+$ 571.3284; found 571.3282, error 0.4 ppm, Δ0.2 mDa.

*4.9. (*S*)-3,17-Diethyl-8-(2-carboxyethyl)-12-(2-methoxycarbonyl-1-methylethyl)-2,7,13,18-tetramethyl-(21*H,*24*H)*-bilin-1,19-dione (**14**, β′S-methylmesobiliverdin-XIIIα 12-methyl ester)*

The title compound was obtained in 89% yield, mp 181–183°C. ^{1}H NMR: δ 1.22 (6H, t, J=7.5 Hz), 1.35 (3H, d, J=7.1 Hz), 1.81 (3H, s), 1.82 (3H, s), 2.02 (3H, s), 2.16 (3H, s), 2.24 (2H, t, J=7.4 Hz), 2.52 (2×2H, 2×q, J=7.5 Hz), 2.68 (2H, d, J=7.7 Hz), 2.75 (2H, t, J=7.4 Hz), 3.46–3.53 (1H, m), 3.59 (3H, s), 5.94 (1H, s), 6.01 (1H, s), 6.83 (1H, s), 8.79 (2H, br. s), ~9.1 (1H, very br. s) ppm; and ^{13}C NMR: δ 8.05, 8.06, 9.40, 10.24, 14.35, 17.67, 17.71, 19.65, 20.69, 27.77, 35.21, 41.11, 51.56, 97.02, 97.34, 114.96, 126.75, 127.87, 128.11, 128.63, 138.77, 138.92, 139.12, 140.04, 140.70, 142.18, 146.74, 147.08, 147.35, 172.60, 173.06, 174.04, 176.03 ppm. HRMS (3-NBA): calcd for $C_{35}H_{43}N_4O_6$ $(M+H)^+$ 615.3183; found 615.3167, error 2.6 ppm, Δ1.6 mDa.

4.10. Mesobilirubin-XIIIα analogs. General procedure

To a cooled (ice bath) solution of the corresponding mesobiliverdin (0.1 mmol) in THF (10 mL) and CH_3OH (30 mL, both dry and deoxygenated) was added $NaBH_4$ (378 mg, 10 mmol) over 15 min under N_2. The cooling bath was removed and after a further 30 min of stirring, the yellow green mixture was diluted with ice/water (100 mL) and acidified with CH_3COOH. The product was extracted with $CHCl_3$ (4×50 mL), washed with H_2O (3×70 mL), dried over anhydrous Na_2SO_4, filtered, and the solvent was evaporated under vacuum. The crude material was purified by preparative TLC (3% CH_3OH/CH_2Cl_2) or radial chromatography on silica gel (1–3% CH_3OH/CH_2Cl_2 v/v). The isolated yellow pure mesobilirubin fraction was recrystallized from $CHCl_3/CH_3OH$.

*4.11. (*S*)-3,17-Diethyl-8-(2-carboxyethyl)-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(10*H,*21*H,*23*H,*24*H)*-bilin-1,19-dione (**1**, β′S-methylmesobilirubin-XIIIα)*

The mesobilirubin **2** (61.7 mg, 0.1 mmol) was dissolved in THF:CH_3OH=1:1 (30 mL, both deoxygenated) and 5 mL of 1 M aqueous NaOH was added. The mixture was stirred under N_2 at 50°C for 3 h. The THF and CH_3OH solvents were evaporated under vacuum, water (100 mL) was added followed by 7 mL of CH_3COOH and 1–2 mL of 2% HCl (pH<7), and the product was extracted with $CHCl_3$ (3×25

mL). The extracts were washed with water (2×100 mL), dried over anhydrous Na_2SO_4, filtered, and the solvent was evaporated under vacuum. The crude product was purified by preparative TLC on silica gel (4% CH_3OH/CH_2Cl_2 v/v) and the pure yellow fraction was recrystallized from $CHCl_3/CH_3OH$ to afford 32 mg (53%) of mesobilirubin **1**. Mp 294–298°C (decomp.). $[\alpha]_D^{20}$ −4460 (c 4.7×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1. HRMS (3-NBA): calcd for $C_{34}H_{42}N_4NaO_6$ $(M+Na)^+$ 625.3002; found 615.3001, error 0.2 ppm, Δ0.1 mDa.

*4.12. (*S*)-3,17-Diethyl-8-(2-methoxycarbonylethyl)-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(10*H*,21*H*,23*H*,24*H*)-bilin-1,19-dione (**2**, β′*S*-methylmesobilirubin-XIIIα 8-methyl ester)*

The title compound was obtained in 67% yield, mp 230–232°C (decomp.). $[\alpha]_D^{20}$ −4130 (c 6.5×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1. HRMS (3−NBA+NaI): calcd for $C_{35}H_{44}N_4NaO_6$ $(M+Na)^+$ 639.3159; found 639.3137, error 3.4 ppm, Δ2.2 mDa.

*4.13. (*S*)-3,17-Diethyl-8-propyl-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(10*H*,21*H*,23*H*,24*H*)-bilin-1,19-dione (**3**)*

The title compound was obtained in 54% yield,[20] mp 191–194°C (decomp.). $[\alpha]_D^{20}$ −3870 (c 7.5×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1.

*4.14. (*S,S*)-3,17-Diethyl-8-(1-methylpropyl)-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(10*H*,21*H*,23*H*,24*H*)-bilin-1,19-dione (**4**)*

The title compound was obtained in 39% yield, mp 242–244°C (decomp.). $[\alpha]_D^{20}$ −3870 (c 4.8×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1. HRMS (3−NBA+NaI): calcd for $C_{35}H_{46}N_4NaO_4$ $(M+Na)^+$ 609.3417; found 609.3415, error 0.2 ppm, Δ0.2 mDa.

*4.15. (*S*)-3,17-Diethyl-12-(2-carboxyethyl)-8-(1-methylpropyl)-2,7,13,18-tetramethyl-(10*H*,21*H*,23*H*,24*H*)-bilin-1,19-dione (**5**)*

The title compound was obtained in 37% yield, mp 219–223°C (decomp.). $[\alpha]_D^{20}$ −205 (c 4.4×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1. HRMS (3−NBA): calcd for $C_{34}H_{44}N_4O_4$ $(M)^+$ 572.3362; found 572.3348, error 2.5 ppm, Δ1.4 mDa.

*4.16. (*S,S*)-3,17-Diethyl-8-(2-methoxycarbonyl-1-methylethyl)-12-(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(10*H*,21*H*,23*H*,24*H*)-bilin-1,19-dione (**6**, β*S*,β′*S*-dimethylmesobilirubin-XIIIα 8-methyl ester)*

The title compound was obtained in 67% yield,[20] mp 237–239°C (decomp.). $[\alpha]_D^{20}$ −4170 (c 7.0×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1.

*4.17. (*S*)-3,17-Diethyl-12-(2-carboxyethyl)-8-(2-methoxycarbonyl-1-methylethyl)-2,7,13,18-tetramethyl-(10*H*,21*H*,23*H*,24*H*)-bilin-1,19-dione (**7**, β′*S*-methylmesobilirubin-XIIIα 12-methyl ester)*

The title compound was obtained in 56% yield, mp 243–247°C (decomp.). $[\alpha]_D^{20}$ −1000 (c 4.1×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1. HRMS (3−NBA): calcd for $C_{35}H_{44}N_4O_6$ $(M)^+$ 616.3261; found 616.3279, error −2.9 ppm, Δ−1.8 mDa.

*4.18. (*S,S*)-3,17-Diethyl-8,12-bis(2-carboxy-1-methylethyl)-2,7,13,18-tetramethyl-(10*H,*21*H,*23*H,*24*H)-bilin-1,19-dione (**8**, β*S*,β′*S*-dimethylmesobilirubin-XIIIα)*

The title compound was obtained in 70% yield from saponification followed by $NaBH_4$ reduction of the corresponding β*S*,β′*S*-dimethylmesobiliverdin-XIIIα dimethyl ester (**15**),[9] mp 292–294°C (decomp.). $[\alpha]_D^{20}$ −4730 (c 8.6×10^{-3}, $CHCl_3$); ^{1}H NMR in Table 2 and ^{13}C NMR in Table 1.

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References

1. Chowdury, J. R.; Wolkoff, A. W.; Chowdury, N. R.; Arias, I. M. Hereditary Jaundice and Disorders of Bilirubin Metabolism. In *The Metabolic and Molecular Bases of Inherited Disease*; Scriver, C. R.; Beaudet, A. L.; Sly, W. S.; Valle, D., Eds., McGraw-Hill: New York, 1995; Vol. 2, Chapter 67, pp. 2161–2208.
2. Berk, P. D.; Noyer. C. *Seminars Liver Dis.* **1994**, *14*, 323–394.
3. McDonagh, A. F. Bile Pigments: Bilatrienes and 5,15-Biladienes. In *The Porphyrins*; Dolphin, D., Ed.; Academic Press: New York, 1979; Vol. 6, pp. 293–491.
4. Bonnett, R.; Davies, J. E.; Hursthouse, M. B.; Sheldrick, G. M. *Proc. R. Soc. London, Ser. B* **1978**, *202*, 249–268.
5. LeBas, G.; Allegret, A.; Mauguen, Y.; DeRango, C.; Bailly, M. *Acta Crystallogr., Sect. B* **1980**, *B36*, 3007–3011.
6. Becker, W.; Sheldrick, W. S. *Acta Crystallogr., Sect. B* **1978**, *B34*, 1298–1304.
7. Mugnoli, A.; Manitto, P.; Monti, D. *Acta Crystallogr., Sect. C* **1983**, *38*, 1287–1291.
8. Person, R. V.; Peterson, B. R.; Lightner, D. A. *J. Am. Chem. Soc*. **1994**, *116*, 42–59.
9. Boiadjiev, S. E.; Person, R. V.; Puzicha, G.; Knobler, C.; Maverick, E.; Trueblood, K. N.; Lightner, D. A. *J. Am. Chem. Soc*. **1992**, *114*, 10123–10133.
10. Puzicha, G.; Pu, Y.-M.; Lightner, D. A. *J. Am. Chem. Soc*. **1991**, *113*, 3583–3592.
11. Shelver, W. L.; Rosenberg, H.; Shelver, W. H. *Intl. J. Quantum Chem*. **1992**, *44*, 141–163.
12. Shelver, W. L.; Rosenberg, H.; Shelver, W. H. *J. Molec. Struct*. **1994**, *312*, 1–9.
13. Dörner, T.; Knipp, B.; Lightner, D. A. *Tetrahedron* **1997**, *53*, 2697–2716.
14. Nogales, D.; Lightner, D. A. *J. Biol. Chem*. **1995**, *270*, 73–77.
15. Kar, A.; Lightner, D. A. *Tetrahedron* **1998**, *54*, 5151–5170.
16. Boiadjiev, S. E.; Lightner, D. A. *Tetrahedron: Asymmetry* **1997**, *8*, 2115–2129.
17. Boiadjiev, S. E.; Lightner, D. A. *Tetrahedron: Asymmetry* **1996**, *7*, 1309–1322.
18. Xie, M.; Lightner, D. A. *Tetrahedron* **1993**, *49*, 9235–9250.
19. Boiadjiev, S. E.; Lightner, D. A. *J. Org. Chem*. **1998**, *63*, 6220–6228.
20. Boiadjiev, S. E.; Lightner, D. A. *Tetrahedron: Asymmetry* **1997**, *8*, 3603–3615.
21. Boiadjiev, S. E.; Pfeiffer, W. P.; Lightner, D. A. *Tetrahedron* **1997**, *53*, 14547–14564.
22. Shrout, D. P.; Lightner, D. A. *Synthesis* **1990**, 1062–1065.
23. Lightner, D. A.; Ma, J.-S.; Adams, T. C.; Franklin, R. W.; Landen, G. L. *J. Heterocyclic Chem*. **1984**, *21*, 139–144.
24. Boiadjiev, S. E.; Lightner, D. A. *Synlett* **1994**, 777–785.
25. Trull, F. R.; Rodriguez, M.; Lightner, D. A. *Synthetic Commun*. **1993**, *23*, 2771–2783.
26. Kaplan, D.; Navon, G. *Isr. J. Chem*. **1983**, *23*, 177–186.
27. Kaplan, D.; Navon, G. *Org. Magn. Res*. **1981**, *17*, 79–87.
28. Kaplan, D.; Navon, G. *Biochem. J*. **1982**, *201*, 605–613.
29. Navon, G.; Frank, S.; Kaplan, D. *J. Chem. Soc., Perkin Trans. 2* **1984**, 1145–1149.
30. Harada, N.; Nakanishi, K. *Circular Dichroic Spectroscopy — Exciton Coupling in Organic Stereochemistry*; University Science Books: Mill Valley, CA, 1983.
31. Lightner, D. A.; Gawroński, J. K.; Wijekoon, W. M. D. *J. Am. Chem. Soc*. **1987**, *109*, 6354–6362.

32. Boiadjiev, S. E.; Lightner, D. A. *Tetrahedron: Asymmetry* **1999**, *10*, 607–655.
33. Gawroński, J. K.; Poloński, T.; Lightner, D. A. *Tetrahedron* **1990**, *46*, 8053–8066.
34. Trull, F. R.; Shrout, D. P.; Lightner, D. A. *Tetrahedron* **1992**, *48*, 8189–8198.
35. Perrin, D. D.; Armarego, W. L. F. *Purification of Laboratory Chemicals*; 3rd ed.; Pergamon Press: England, 1988.
36. Boiadjiev, S. E.; Anstine, D. T.; Lightner, D. A. *J. Am. Chem. Soc*. **1995**, *117*, 8727–8736.