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Characterization of Porphyrins, Chlorins, and Bacteriochlorins Formed via Allomerization of Bacteriochlorophyll *a*. Synthesis of Highly Stable Bacteriopurpurinimides and Their Metal Complexes

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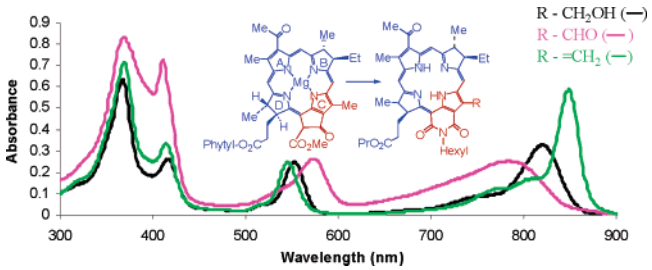

Allomerization of bacteriochlorophyll *a* (Bchl *a*) was studied under various reaction conditions. Bchl *a* on stirring with KOH/propanol produced an "unstable bacteriochlorin", which decomposed in acidic conditions to give a complex mixture containing bacteriopurpurin *a* as a principal component. The yields of other compounds varied and were found to be dependent on reaction condition. The structures of the isolated porphyrins, chlorins, and bacteriochlorins, related to Bchl *a*, were assigned on the basis of 1D, 2D NMR (ROESY), and mass spectroscopy analyses. The presence of fused anhydride rings in porphyrin, chlorin, and bacterichlorin systems showed a significant influence on their optical properties. Compared to bacteriochlorophyll *a* and bacteriopheophytin, the related structurally modified analogues, e.g., the bacteriopurpurin *a*, $13^1/15^1$-*N*-alkyl isoimide, and the imide analogues were found to be more stable with a significant difference in spectroscopic properties. Bacteriochlorins containing anhydride, imide, or isoimide cyclic rings demonstrated a significant bathochromic shift of their *Q* bands in their electronic absorption spectra. Under basic conditions the formation of the 12-hydroxymethyl, 12-formyl, and 12-methylene analogues as byproducts from the 12-methyl-bacteriopurpurin-*N*-hexylimide could be due to subsequent oxidation of the vinylogous enolate intermediates. To investigate the effect of the central metal in the electronic spectra, the stable bacteriopurpurin-18-*N*-hexylimide was converted to a series of metal complexes [Zn(II), Cd(II), and Pd(II)] by following the direct or transmetalation approaches. Compared to the free-base analogue, these complexes showed a remarkable shift in their electronic absorption spectra.

Introduction

Bacteriochlorophylls *a* and *b* and recently discovered tolyporphyn[1−3] are the only natural tetrahydroporphyrins that belong to the class bacteriochlorins (porphyrins with reduced pyrrolic rings diagonal to each other).[4] Other natural bacteriochlorophylls of *c*, *d*, and *e* types formally represent chlorins or dihydroporphyrins.[5] Among the natural pigments, bacteriochlorophyll *a* (Bchl *a*) has attracted increased attention in the past

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decade because of its unique role in bacterial photosynthetic processes.[6,7] Intensive studies on the structure of the bacterial photosynthetic reaction center (RC) have revealed that the bacteriochlorophyll moieties are assembled on a supporting protein matrix in a cascade array fashion with fixed geometries.[8,9] This specific arrangement of pigment units with precise distances and orientations defines the effectiveness of $\pi-\pi$ and $\pi-$p coupling between certain peripheral substituents during the photosynthetic act. Obviously, the presence of these functionalities is the requirement for the interaction between individual molecules.[10] Remarkably, the same Bchl *a* molecules are also employed as basic chromophore units in the antenna complexes, the crystallographic structure of which has been published recently. In these ring-constructed structures, the pigments are also oriented on a protein matrix at certain angle(s) and distance(s) to maximize the interaction, which creates the cyclic supramolecular chromophore pool to utilizes the light energy effectively.[11−16]

Compared to chlorophylls, the chemistry of Bchl *a* has not been explored extensively because of two main reasons. First, purple bacteria, which serve as a source of Bchl *a*, require special conditions for large-scale cultivation and were not commercially available in large amounts. The second reason is the chemical instability of natural bacteriochlorophylls, with their fast autoxidation into the corresponding chlorins and porphyrins[1]. A significant interest in Bchl-related compounds attracted our attention to investigate the chemistry of this novel structure. During the past decade our research has been focused on developing effective synthetic routes for the preparation of chlorins and bacteriochlorins as potential photosensitizers for photodynamic therapy (PDT) as models for photosynthetic systems.[17−21] We have shown that certain 7,8-*vic*-dihydroxybacteriochlorins had a significant antitumor activity and could be useful precursors for many chemical modifications.[22] To investigate the utility of these compounds as models for the photosynthetic reaction centers, the *vic*-dihyroxybacteriochlorins and the corresponding keto analogues formed by the pinacol reaction have also been investigated by Tamiaki and co-workers for their aggregation characteristics.[23] Following our systematic studies on reduced tetrapyrroles, we were interested in exploring the synthetic methodology of chlorophyll oxidative degradation (allomerization) in the Bchl *a* system and in developing the synthetic approaches for stable bacteriochlorins.[24]It was anticipated that these compounds will have a large red shift on their Q_x/Q_y absorption bands and a possible hyperchromic effect due to the extension of π-conjugation, while maintaining their high singlet oxygen producing efficiency.

Oxidation of chlorophylls at the C-13^2 position by triplet oxygen on standing in alcoholic solutions in atmospheric conditions is one of the reactions unique to the chemistry of tetrapyrrolic compounds.[25] Willstatter, a pioneer of chlorophyll chemistry, discovered this reaction first and introduced the special term "allomerization" to describe this process.[26] In chemical terminology it is identical to autoxidation and also known to be catalyzed in the presence of a base.[25] It is postulated that the mechanism of allomerization includes formation of an enolate ion, which on further oxidation generates a cyclic lactone structure (unstable chlorin), which on reacting with diazomethane affords 15-glyoxylic acid derivative (purpurin-7 trimethyl ester), while evaporation in diethyl ether or THF results in the formation of chlorin with a fused cyclic anhydride ring (purpurin-18). Most of the chlorins generally have green color in solutions, but the respective analogues with strong electron-withdrawing functionality at the 15-*meso* position exhibit a prominent Q_x absorption band around 550 nm, thus making the color of their solutions purple-red, which led to their generic name "purpurins".[25]

Allomerization of chlorophyll *a* has been studied in detail by several researchers[27] and became a useful methodology for the synthesis of partly oxidized chlorophyll derivatives, e.g. purpurin-18, chlorin p_6, purpurin-7, etc. Back in 1938, Fischer et al. were the first to study Bchl *a* allomerization.[28] A few years later, Scherz and co-workers reported the formation of 13^2-hydroxy-allomers on storage of Bchl *a* in methanol.[29] However, the autoxidation behavior of Bchl *a* did not receive much attention as an approach for its further modification. In this study, our objective was to extend the methodology of chlorophyll allomerization in the bacteriochlorophyll *a* system for the preparation of partly oxidized Bchl *a* derivatives, which could be important precursors as models for photosynthetic mimicking studies, as well as potential candidates for PDT.

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TABLE 1. Percentage Yields of the Allomerization Products of Bacteriochlorophyll *a*

reaction conditions[b]	yields[a] of isolated compounds (%)															
	8a	**8c**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**	**19**	**20**	**21**	**22**	**23**	**24**
a	4	31	19	0.5	1	1							0.5			
b	8	51	1	1	4	2	2	0.5	2	5	4	1	0.4			
c		39					2		1	4	16	4	0.5			
d	3										56	11				
e		57		2			1			2	8	3	0.6	4	3.5	1.5

[a] Based on the estimated amount of bacteriochlorophyll *a* using ϵ =91,000 in diethyl ether.[25] [b] Reagents and conditions: (a) PrOH/KOH/O_2, 20 min; evaporation at 55−60 °C. (b) PrOH/KOH/O_2, 1.5 h; evaporation at 55−60 °C. (c) PrOH/KOH/O_2, 1.5 h; evaporation at 85−90 °C. (d) PrOH/KOH/O_2, 1.5 h; evaporation in the presence of 2% H_2SO_4 at 85−90 °C. (e) PrOH/KOH/O_2, 1.5 h; room temperature, 10 days, 2% H_2SO_4.

Results and Discussion

Synthesis of Bacteriopurpurin *a*. Bacteriochlorophyll *a* **1** was isolated from purple bacteria such as *Rh. sphaeroides*, *Rh. Roseapersiana*, and *Rh. capsulata* using 1-propanol as the extracting solvent in an inert atmosphere. A brief treatment of **1** with 0.1 N HCl resulted in rapid demetalation to give bacteriopheophytine *a* **2**, which was purified on a silica column. Unfortunately, our attempt to produce bacteriopheophorbide *a* methyl ester **5** by a standard procedure that has been successfully used in chlorophyll chemistry, e.g., 5% sulfuric acid/methanol, resulted in the desired product **5** and a mixture of oxidation products. The oxidation products were separated by column chromatography, and the minor compound was found to be a corresponding chlorin **6** (19%, fast moving band) with long wavelength absorption λ_{max} at 699 nm. The major product was identified as a diastereomeric mixture of 13^2-(***R/S***)-hydroxy-derivatives **3** and **4** (epimers) and isolated in 54% yield. The mass spectrum of this product showed the molecular ion (*m*/*z*) at 640, indicating the incorporation of an extra oxygen atom into the structure. Its ^{1}H NMR spectrum clearly showed the presence of two isomers and the resonance of a hydroxyl group (broad singlet) observed at 5.4 ppm. Structural assignment of the major component as the (*S*)-isomer **3** was based on the anisotropic effect of the 13^2-hydroxyl functionality, which caused a characteristic downfield shift of the neighboring 17-H proton, found as a dd at 3.98 ppm, compared to the (*R*)-isomer's resonance at 4.45 ppm. Based on the integration, the ratio between (*R*) versus (*S*) isomers was 1:4. A preferential formation of (*S*)-epimer **3** was consistent with the data reported for chlorophylls 13^2-hydroxy-derivatives by Dolphin et al.[30] These investigators had shown that (*S*)-epimers in which the methoxycarbonyl moiety at position 13^2 is on the side opposite to that of bulky 17-propionic group are thermodynamically more stable in acidic conditions.[31,32] Other researchers reported difficulties working with bacteriochlorin system due to its rapid oxidation under strong acidic conditions.[28] Formation of 13^2-hydroxy-derivatives (or allomers) is one of the major problems plaguing chlorophyll and bacteriochlorophyll chemistry, because these compounds are spectroscopically indistinguishable and require a tedious chromatographic separation. Bacteriopheophorbide *a* methyl ester obtained by hydrolysis using 80% aqueous TFA under strict inert conditions[31,32] followed by treatment with diazomethanewas isolated in a total yield of 78% from starting Bchl *a* **1**. Bacteriochlorin **5** was allomerized in the presence of KOH in 1-propanol by bubbling air into the solution. Although the exact mechanism of base-promoted allomerization is not known, it is anticipated that the "unstable bacteriochlorin" **7** was possibly formed as an intermediate species. To provide evidence for the formation of the "unstable bacteriochlorin" intermediate, a part of the extract was treated with diazomethane a, which gave 13-carboxy-15-glyoxylic acid bacteriochlorin trimethyl ester **9** as a sole product. The ^{1}H NMR and LC-MS (670, M^+, 100%) spectra supported the assigned structure. The rest of the extract was subjected to evaporation under vacuum. During the evaporation of the solvent, the reaction was monitored spectrophtometrically, which revealed that, compared to purpurin-18, the anhydride ring formation in the bacteriochlorin system was much more difficult. It required more time (3−4 h) and higher temperatures (60−70 °C) yet produced lower yield (29−30%). Another problem was the low solubility of the carboxylic analogue **8a** in most organic solvents, which caused significant difficulty in purification. The presence of the cyclic anhydride ring system caused a significant red shift in its long wavelength absorption Q_y band observed at λ_{max} 813 nm. The extended conjugation also caused distinctive red shift of Soret and Q_x bands and showed a direct analogy with the spectral characteristics of purpurin-18. Due to the presence of a prominent absorbance at 545 nm, bacteriopurpurin *a* (**8a**) had a distinctive pink-rose color in contrast to the green-violet color observed for bacteriochlorins **2** and **5**. To confirm the presence of the anhydride ring system, bacteriochlorin **8a** was reacted with methanolic KOH. This caused ring opening and resulted in compound **10** with a hypsochromic shift of Q_y absorption to 765 nm due to a loss of extended conjugation. Treatment of **8a** with diazomethane produced the corresponding methyl ester **8b**, which had a far better solubility and was easier to purify.

To prevent undesirable loss of the starting materials during the preparation of bacteriopheophorbide *a*, we modified the procedure and instead of isolating bacteriopheophorbide, the Bchl *a* **1** 1-propanol extract mixture also containing bacterial carotenoids was used as such for the succeeding steps. The combined extract was treated with KOH/O_2 to give the lactone derivative, "unstable bacteriochlorin" **7**. Degradation of **7** during solvent evaporation at elevated temperatures produced bacteriopurpurin *a* (mixture of **8a** and **8c**) as a major product. Our experiments revealed that the allomerization gives a complicated mixture, with the yield of bacteriopurpurin *a* being dependent on the reaction conditions. In our present study, most of the minor byproducts formed during the allomerization process were isolated and identified, giving important information on the mechanisms of this complex process. In our attempts to optimize the reaction conditions, we varied the time of base treatment, as well as the acidity and temperature during the ring cyclization step. The conditions used are summarized in Table 1.

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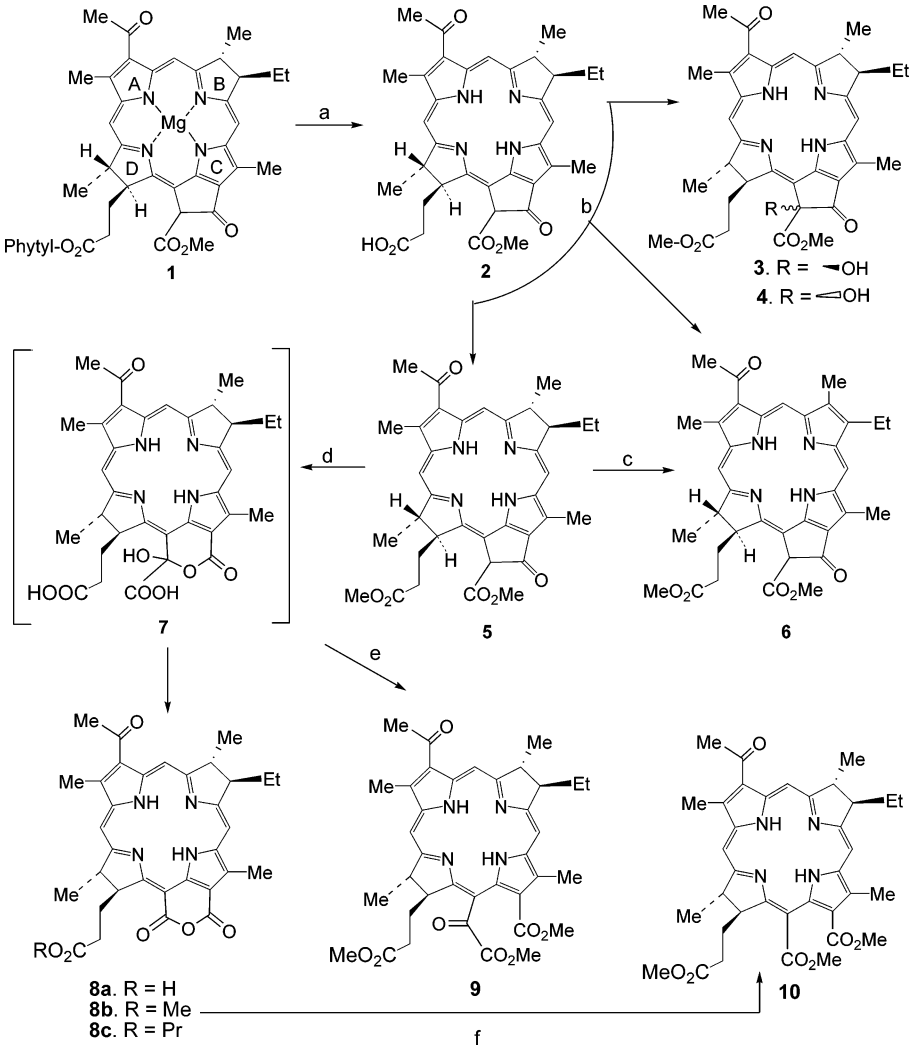
SCHEME 1. Bacteriochlorins and Chlorins Derived from Bacteriochlorophyll *a*[a]

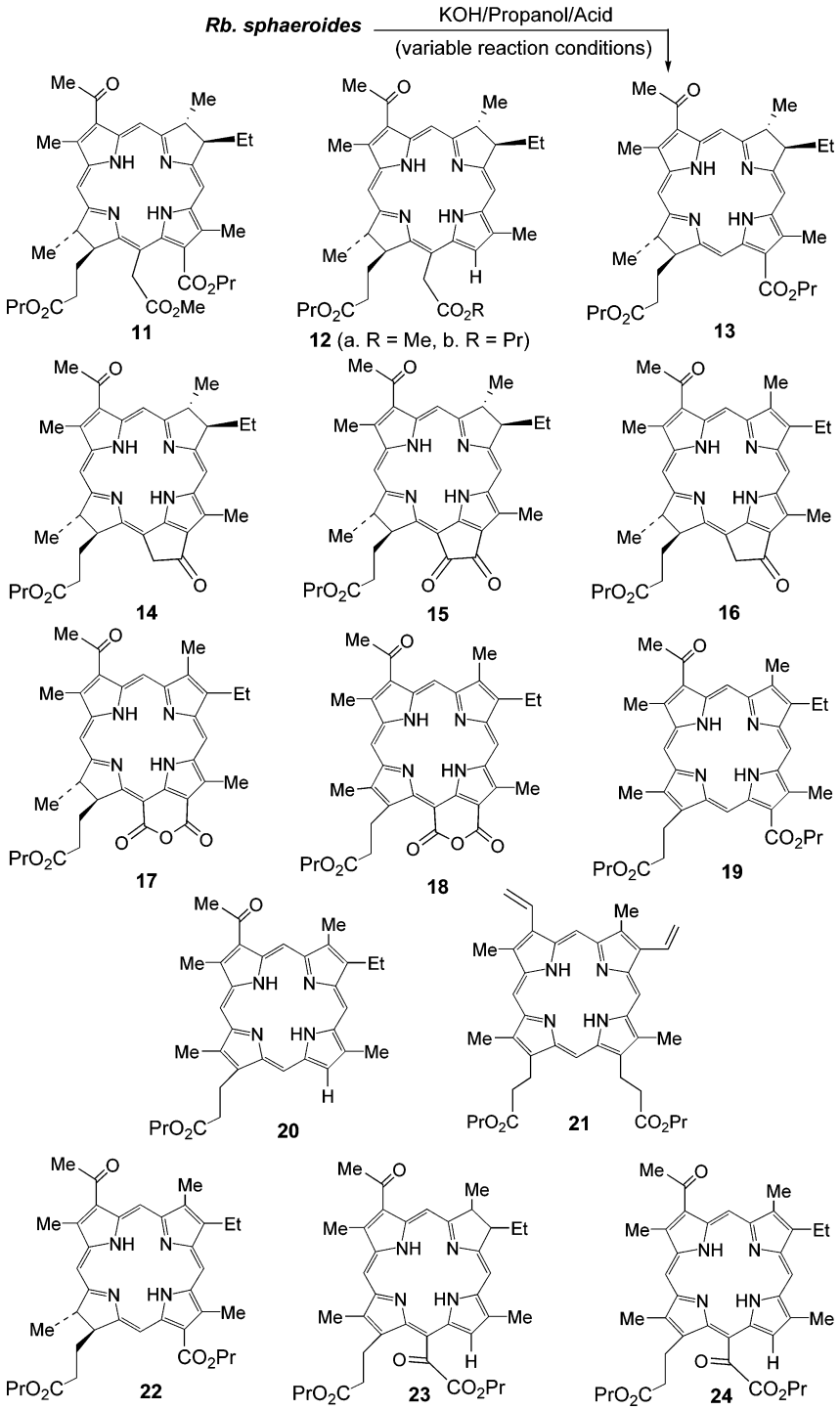


[a] Reagents and conditions: (a) 0.1% HCl; (b) 5% H_2SO_4/MeOH; (c) air at rt; (d) KOH/methanol; (e) CH_2N_2; (f) aq KOH/MeOH, CH_2N_2.

We observed that KOH-catalyzed allomerization of Bchl *a* **1** requires more time (1.2−1.5 h) for completion than that reported for chlorophyll *a* (10−20 min). This could be due to the presence of carotenoids in the reaction mixture, which are known to decrease the rate of allomerization by acting as free-radical scavengers.[27] We observed that treatment of **1** with KOH for 20 min resulted in isolation of a substantial amount (18−23%) of bacteriochlorin **11** with an acetic acid functionality at position 15. Its structure was assigned by ^{1}H NMR, showing a distinctive quartet for CH_2 protons at 5.39 ppm and resonances for two propyl and one methyl ester groups. The mass spectral data (*m*/*z* 712) also supported the assigned structure, which obviously formed through a ring-opening reaction involving attack by propyloxy anion on starting Bchl *a* **1**. Further oxidation transformed **11** into bacteriopurpurin *a*. Increasing the time for KOH-catalyzed allomerization to 1.2−1.5 h resulted in almost complete disappearance of bacteriochlorin **11**. As we observed for purpurin-18, the presence of a catalytic amount of acid and propanol also helped in esterification of the 17-propionic acid side chain to the corresponding propionic ester functionality, and the corresponding bacteriopurpurin *a* propyl ester analogue **8c** was isolated in 50−51% yield (*m*/*z* 624). Along with the desired product **8c**, some other products were also obtained from the reaction mixture, including a number of bacteriochlorins, chlorins, and porphyrins (Scheme 2). The NMR, mass, and UV−vis spectral analyses confirmed structural assignments of the isolated products. In brief, the two fastest moving bands were assigned as bacteriochlorin **12** and bacteriochlorin **13** (as propyl esters), isolated in approximately 1% and 4% yields (starting from Bchl *a*), respectively. Both compounds exhibited a typical bacteriochlorin UV−vis spectrum with long wavelength absorption at 744 (**12**) and 753 nm (**13**). The blue shift of the Q_y band in the electron the absorption spectrum of **12** indicated that one of the electron-attracting substituents, located on the diagonal opposite rings A and C, was absent. The ^{1}H NMR of this compound showed a singlet for an acetyl group at 3.18 ppm, and an additional singlet at 8.5 ppm attributed to 13-H at the pyrrolic β-free position. The resonances observed at 5.0 ppm assigned to the 15-CH_2 protons with a characteristic AB were splitting due to the effect of the neighboring chiral center present at position 17.

Compared to compound **12**, the bacteriochlorin **13,** bearing two electron-withdrawing funcionalities at the pyrrolic rings A and C, exhibited long wavelength absorption at 756 nm. The 2D ROESY and COSY data for bacteriochlorins **12** and **13** confirmed the assigned structures, demonstrating the distinctive

SCHEME 2. Allomerization Products of Bacteriochlorophyll *a*



through-space $^1H-^1H$ dipolar couplings. More specifically, compound **12** exhibited the key interactions (ROESY cross-peaks) of the 13-H resonance (8.5 ppm) with both the 12-methyl singlet (3.76 ppm) and the 15-CH_2 quartet (5.2 ppm). The ROESY spectrum of bacteriochlorin **13** showed a dipolar interaction between the 15-*meso* proton and the neighboring 17-H atom. Two other bacteriochlorins, more polar compared to **8c**, were also isolated from the reaction mixture. One of them (a violet band with λ_{max} at 753 nm) was identified as bacteriopyropheophorbide *a* propyl ester **14** (*m*/*z* 594). This compound had a characteristic ABX pattern observed at 5.0 ppm for the 13^2-protons. The other bacteriochlorin exhibited a long-wavelength absorption at 768 nm, suggesting the possibility of an additional electron-withdrawing functionality. Indeed, compared to **14**, the mass spectrum of this compound (*m*/*z* 608) demonstrated the presence of an additional oxygen atom. The NMR spectrum revealed that the signals for 13^2-H disappeared, while the signals characteristic of the17-H proton shifted to a lower field region at 5.08 ppm. From these data for compound **15**, we concluded the presence of a keto-functionality at the 13^2-position. Evidently, bacteriochlorin **14** could be formed by decarboxylation of Bchl *a* during base treatment, which on

TABLE 2. Comparative Optical Properties of Certain Tetrapyrrole-Based Compounds

chromophore	compound	Soret band		Q_x		Q_y λ_{max}, nm ($\epsilon \times 10^3$)
porphyrin	**19**	408 (179.0)	520 (12.2)	558 (18.5)	582 (12.7)	622 (4.6)
	18	429 (146.0)	550 (8.2)	573 (11.5)	612 (16.5)	660 (19.4)
	22	408 (118.5)		509 (12.0)		681 (44.2)
chlorin	**17**	411 (115.3)		552 (24.6)		720 (46.6)
	13	446 (81.6)	387 (49.3)	519 (14.1)		759 (54.1)
bacteriochlorin	**8c**	463 (85.3)	410 (53.1)	543 (32.7)		813 (56.5)

further oxidation resulted into the 13^2-oxo derivative **15**. A similar phenomenon was recently reported from our laboratory in the related chlorin analogue.[33]

Among the products of allomerization, two chlorins were isolated in minor yields. These were identified as 3-acetyl-3-devinyl-pyropheophorbide *a* **16** and 3-acetyl-3-devinylpurpurin-18 **17** propyl esters. Both compounds showed the typical chlorin-type UV−vis spectra. ^{1}H NMR spectra of these chlorins exhibited a quartet for the methylene protons of the 8-ethyl substituent of aromatic pyrrolic ring B at 3.75 ppm. Compared to **16**, chlorin **17** showed a distinctive downfield shift of the 17-H proton (dd at 5.2 ppm), demonstrating the presence of the anhydride ring system. On the other hand, compound **16** showed a characteristic ABX-type pattern for the 13^2-CH_2 protons. Obviously these compounds are the products of 7,8-dehydrogenation (oxidation) of the corresponding bacteriochlorins **14** and **8c** and are formed during the allomerization process. In general it has been observed that Bchl *a* and its derivatives oxidize regioselectively at pyrrolic ring B first, while ring D remains intact.[27] In our study, allomerization of Bchl *a* also produced certain porphyrins. One product, a polar green band, was identified as 3-acetylporphyrin **18.** It contained an anhydride ring system (emeraldin) and had a specific "reverse-etio"-type electronic absorption spectrum with strongest peak observed at 663 nm. Its ^{1}H NMR spectrum demonstrated the presence of the anhydride ring, which deshielded the 12-methyl substituent to 3.82 ppm. Obviously porphyrin **18** is a product of further oxidation of bacteriopurpurin *a* **8c** through the intermediate chlorin **17**. Similar compounds are reported in the literature and were formed by oxidation of bacteriopurpurin *a* **8b** with DDQ.[34]

Porphyrin **19** exhibited a distinctive oxo-rhodo type of spectrum due to the presence of two strong electron-withdrawing substituents located at the diagonal positions. Another porphyrin with a rhodo-type spectrum was identified as 13-decarboxylated derivative **20**. Compared to **19**, its ^{1}H NMR spectrum showed an additional singlet at 8.95 ppm corresponding to the β-free pyrrolic proton at position 13. The molecular ion at *m*/*z* 550 confirmed the structure as 3-acetylpyrroporphyrin XV propyl ester. The yields of porphyrins **19** and **20** varied, depending on the reaction conditions used (Table 1).

To understand the reaction mechanism(s) of the decarboxylation/oxidation step, the "unstable bacteriochlorin" **7** was treated with 2% sulfuric acid in 1-propanol/THF at room temperature. On keeping the reaction mixture for several days (7−8 days), three new compounds (chlorins **22** and **23** and porphyrin **24**) were isolated. In both chlorins, the ring D had oxidized. These structures were confirmed by proton NMR studies (1D and 2D ROESY). These findings were in contrast to the previous reports where regioselective oxidation of ring B was reported.[27]

The oxidative decarboxylation of chlorins in acidic conditions is well-known in chlorophyll chemistry; however, the mechanism of this transformation is not clear. Acid-catalyzed migration of the protons at positions 17 and 18, suggested recently by Dolphin[30] in certain chlorins, could be a plausible mechanism of regioselective ring D oxidation. Thus, the migration of the 17-H atom could led to the formation of unstable bacteriochlorin-type intermediates, as was earlier proposed by Kenner et al.,[35] which were oxidized in situ and led to subsequent loss of a proton at position 18 giving ring D oxidized products. In contrast, the oxidation of bacteriochlorin **13** with DDQ afforded ring-B oxidized chlorin **22** as a major product. ^{1}H NMR of **22** showed a distinctive quartet of the 8-ethyl substituent at 3.7 ppm, and the signals for the propionic acid group were observed at 2.4−2.7 ppm. These chlorins could be intermediate species between the bacteriochlorin **7** and porphyrins **19** and **20**. The structures of intermediate chlorins allow us to propose the decomposition pathways for "unstable bacteriochlorin" **7**. Initially the decarboxylation and subsequent oxidation of the neighboring ring D could produce intermediate chlorins **22** and **23**. Because of their instability, these compounds could further oxidize at higher temperature to yield the corresponding porphyrins **19** and **24,** respectively. Porphyrin **24**, containing a glyoxylic function at the 15-*meso* position, in acidic conditions on further decarboxylation could produce porphyrin **20**.

From the reaction mixture, we also isolated a minor porphyrin with a characteristic etio-type UV−vis spectrum, which was identified as protoporphyrin IX dipropyl ester **21**. Porphyrin **21** has no chemical connection with oxidative degradation of BChl *a*, and its presence could possibly be attributed to protoporphyrin IX, a biosynthetic precursor of Bchl, which is always present in purple bacteria.

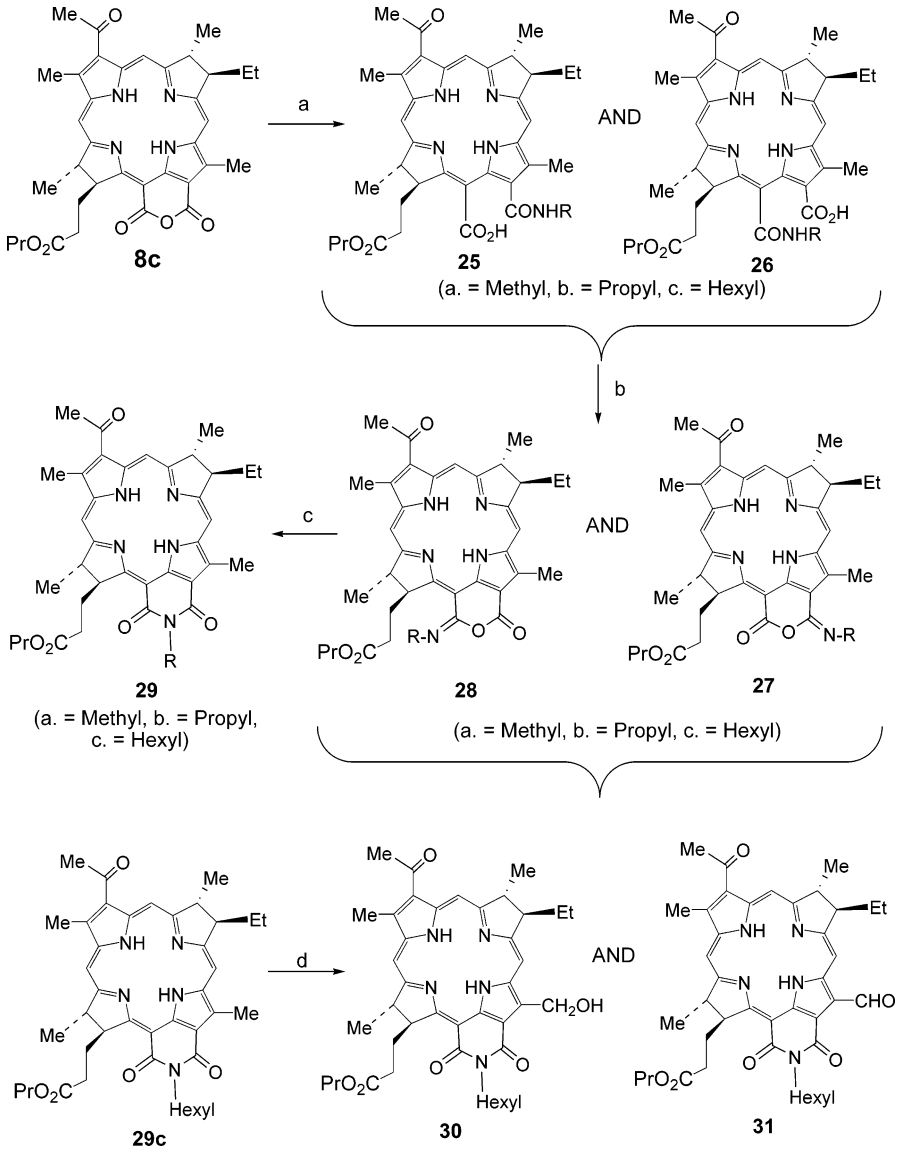
Influence of the Anhydride Ring System in the Spectral Properties of Porphyrin, Chlorin, and Bacteriochlorin Systems. In the present study, the wide range of the compounds isolated from the reaction mixture, related to Bchl *a*, gives a unique opportunity to observe the effect of progressive changes in chemical structures on the absorption spectra of various chromophores. We found that the presence of a cyclic anhydride ring system had a significant influence on the absorption spectra of the tetrapyrrole-based compounds. For a comparative spectroscopic study, porphyrin **19**, chlorin **22**, and bacteriochlorin **13** were selected as reference chromophores without conjugated isocyclic ring versus emeraldin **18** and bacteriopurpurin **8c**, bearing a cyclic anhydride ring system. The presence of a fused anhydride ring system showed a significant changes in the electronic absorption spectra of these compounds, which are summarized in Table 2. For example, in porphyrin pair **18**/**19** spectral characteristics changed drastically from rhodo- for porphyrin **19** to the unusual "reverse-etio"-type for emeraldin **18**. Compared to **19**, the Soret band of emeraldin **18** was

(33) Kozyrev, A. N.; Dougherty, T. J.; Pandey, R. K. *Chem. Commun.* **1998**, 481−482.

(34) Mironov, A. F.; Efremov, A. V.; Efremova, O. A.; Bonnett, R.; Martinez, G. *J. Chem. Soc., Perkin Trans. 1* **1998**, 3601−3608.

(35) Kenner, G. W.; McCombie, S. W.; Smith, K. M. *J. Chem. Soc., Perkin Trans. 1* 1**973**, 2517.

SCHEME 3. Stable Bacteriochlorins Derived from Bacteriopurpurin-18 Propyl Ester[a]



[a] Reagents and conditions: (a) NH_2R (R = methyl, propyl, or *N*-hexyl); (b) DCC and DMAP; (c) KOH/MeOH; (d) KOH/MeOH.

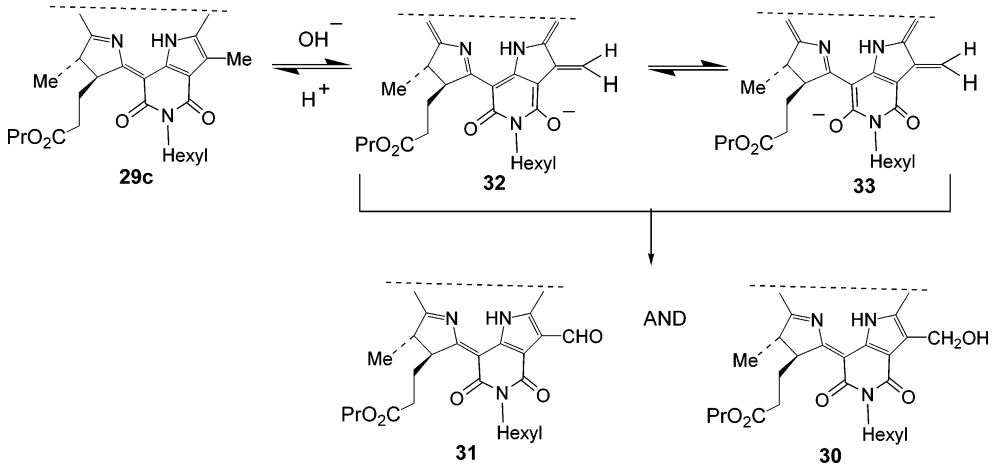
significantly red-shifted ($\Delta\lambda$ = 21 nm). Because of a strong absorption at 622 nm, porphyrin with anhydride ring **19** showed an intense green color in solution, similar to those derived from chlorophyll *a*.[24] For chlorin pair **22**/**17** the Soret bands were found to be almost identical. However, compared to chlorin **22**, the Q_x and Q_y absorption bands of 3-acetyl-3-devinyl-purpurin-18 **17** demonstrated significant bathochromic shifts (Table 2).

Compound **17** showed a distinctive characteristic of purpurin-based compounds, exhibiting an intensive Q_x absorption at 552 nm (red color in solution) and a red-shifted long wavelength band (Q_y) at 720 nm. Similar spectral changes were also observed for the bacteriochlorin pair **13**/**8c**. The presence of the anhydride ring caused bathochromic shifts in B_2, Q_x, and Q_x bands. Compared to the bacteriochlorin **13**, bacteriopurpurin *a* **8c** exhibited an intense absorption at 543 nm, which contributes for the distinctive pink-red color of its solution. The most profound effect was observed for the Q_y band that shifted to the near-IR region with λ_{max} at 813 nm ($\Delta\lambda$ = 54 nm).

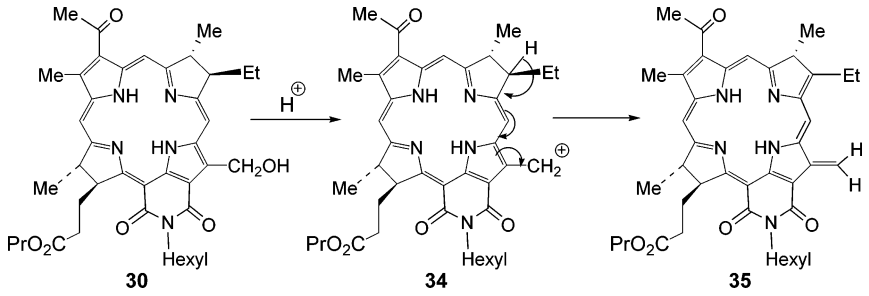
Synthesis of Bacteriopurpurin *a* Imide and Isoimide Derivatives. In our previous studies with a series of 7,8-dihydroxy-bacteriopurpurins, we observed that the fused anhydride ring present in the molecule cleaves rapidly in vivo resulting in a ring-opened product. To prepare a product that is stable in vivo we decided to convert the cyclic anhydride ring system into the cyclic-imide derivative, which is known for its remarkable stability in chlorin systems.[36,37] Bacteriopurpurin **8c**, on reacting with selected amines gave the corresponding amides as a mixture of two isomers, **25** and **26**. ^{1}H NMR data indicated that 13-amide derivatives **25a**−**c** were a major product (85−90%) and 15-amide analogues **26a**−**c** were formed in smaller amounts (10−15%), which was in agreement with data reported for the related chlorin analogues.[37] Treatment of the isomeric mixture of amides with DCC afforded bacteriopurpurin *a*

(36) Lee, S. H.; Jagerovich, N.; Smith, K. M. *J. Chem. Soc., Perkin Trans. 1* **1993**, 2369−2377.
(37) Kozyrev, A. N.; Zheng, G.; Zhu, C.; Dougherty, T. J.; Smith, K. M.; Pandey, R. K. *Tetrahedron Lett.* **1996**, 3781−3784.
(38) Zheng, G.; Potter, W. R.; Camacho, S. H.; Missert, J. R.; Wang, G.; Bellnier, D. A.; Henderson, B. W.; Rodgers, M. A. J.; Dougherty, M. A. J.; Pandey, R. K. *J. Med. Chem.* **2001**, *44*, 1540−1559.

SCHEME 4 Possible Mechanism for the Formation of 12-Hydroxymethyl and 12-Formyl Bacteriochlorins under Basic Conditions from Bacteriopurpurin-18-*N*-hexylimide



SCHEME 5. Acid-Catalyzed Transformation of 12-Hydroxymethyl-bacteriochlorin to the Corresponding 12-Methylene Analogue



N-alkylisoimides **27a**−**c** and **28a**−**c** in the same ratio (7:1), which were separated by column chromatography and isolated as individual compounds. As for corresponding chlorin 13^1/15^1-isoimide derivatives, ^{1}H NMR analysis played a crucial role for their structural assignment. The 15^1-isoimides **27a**−**c** showed a distinctive downfield-shifted resonance of the17-H proton as a dd at 5.18 ppm and nonequivalent signals of the α−CH_2 protons in the *N*-alkyl chain as a multiplet at 3.91 ppm. In contrast, 13^1-*N*-alkylisoimides **28a**−**c** have a signal of the 17-H at 5.35 ppm and α-CH_2 as well-resolved triplet at 4.11 ppm.

Treatment of the isoimides **27a**−**c** and/or **28a**−**c** with methanolic KOH resulted into the related *N*-alkylimides **29a**−**c** via C−N rearrangement in 55−65% yields. Reacting **29** under basic conditions (in the presence of air) produced the corresponding 12-formyl- bacteriopurpurin imide **31** (4−6%) and 12-hydroxymethyl analogue **30** (2−4%) as minor products. Compared to **29**, the ^{1}H NMR spectra of the byproducts demonstrated a disappearance of the 12-methyl singlet at 3.71 ppm and exhibited new signals for the 12-*CH*$_2$OH for compound **30** at 6.00 ppm, and the singlet observed at 11.70 ppm was assigned to 12-formyl proton in bacteriopurpurin **31**. A similar selective oxidation of the 12-methyl substituent in chlorin analogues (purpurinimides) has been attributed to unique vinylogous enolization of this group. The enolization is a consequence of the powerful electron-withdrawing effect of the neighboring isocyclic system.[33] As shown in Scheme 4, the tautomerization in bacteriopurpurinimides could lead to the formation of enolates **32** and **33.** The UV−vis spectra of the enolic forms demonstrated profound changes on the Soret band and a large bathochromic shift of the long wavelength Q_y band observed at 846 nm. These enolates were found to be extremely susceptible to oxidation, producing the 12-hydroxymethyl- and 12-formyl derivatives, **30** and **31**, respectively.

From the reaction mixture, we also isolated an unusual bacteriochlorin (0.5−1%) with a unique absorption spectrum. In this compound, the Soret and Q_x band regions were similar to those of the bacteriopurpurin **29**, except the Q_y band had shifted to 849 nm. The mass spectrum gave the molecular ion peak (*m*/*z* at 705.39). The 2D ROESY data provided valuable information for the structural assignment. A singlet for the 10-*meso* proton showed the only dipolar coupling with the resonances of the 12-methylene group and the 8-H proton was found to be missing. Based on these and other results, the structure of this novel bacteriochlorin was assigned as bacteriopurpurinimide **35**. As shown in Scheme 5, the acid-promoted elimination of the hydroxyl group in **30** could produce a carbocation **34**, which is stabilized by the loss of a proton at position 8 followed by the structural rearrangement of double bonds within bacteriochlorin macrocycle. To prove the structure, the 12-hydroxymethyl derivative **30** was refluxed in *o*-dichlorobenzene (acidic) for 20 min, and the bacteriochlorin **35** was isolated as a major product (∼50%), supporting the proposed mechanism. The structure of this novel structure was confirmed by 2DROESY NMR studies (see Supporting Information).

The electronic absorption spectra of bacteriopurpurinimide and isoimide derivatives exhibited remarkably intense long wavelength absorptions (Q_y band) in the near-IR region and showed some resemblance to those of purpurin-18 analogues. The isoimide derivatives show small hypsochromic shifts compared to the parent bacteriopurpurin *a* **8c**. However, the most profound hyperchromic effect for Q_y-absorption was observed for 13^1-*N*-alkyl isoimides **28**, where electron-

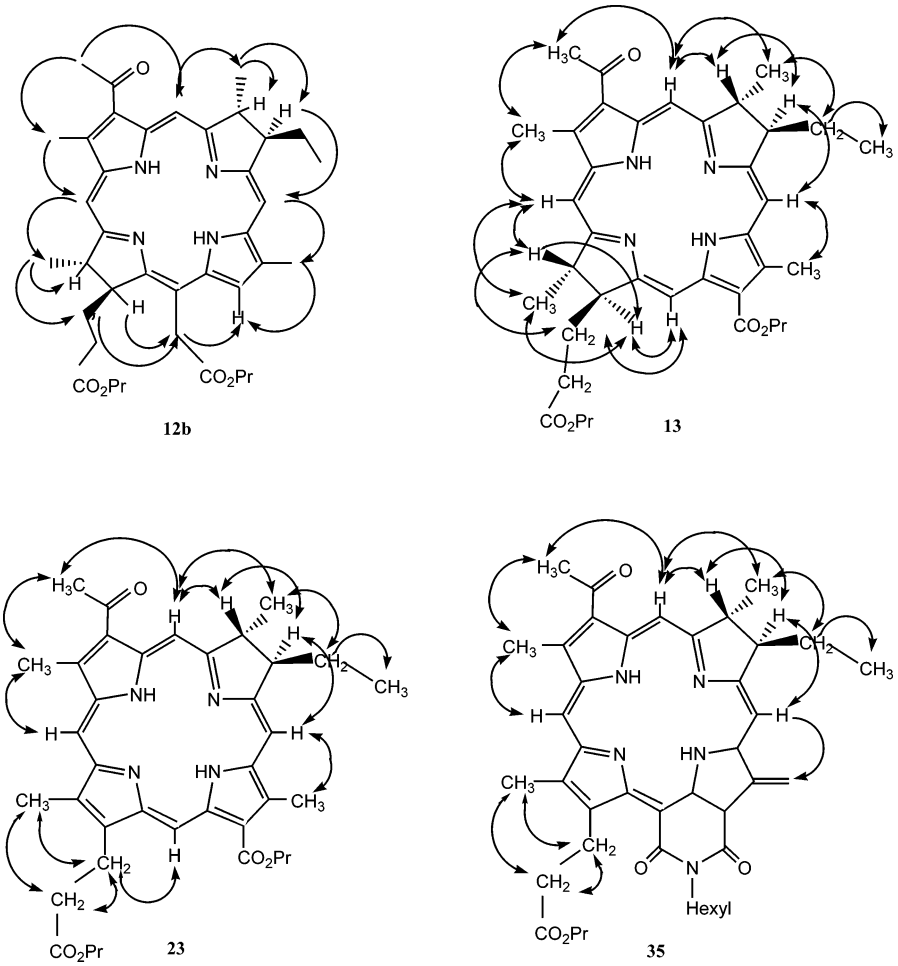


FIGURE 1. Characterization of the bacteriochlorin analogues by NOE studies.

TABLE 3. Comparative Optical Properties of the Bacteriopurpurins

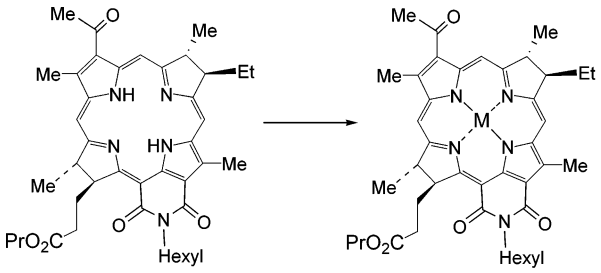
compd	Soret		Q_x	Q_y λ_{max}, nm ($\epsilon \times 10^3$)
8c	363 (85.3)	410 (53.1)	543 (32.7)	813 (56.5)
28c	363 (84.5)	405 (62.0)	537 (28.2)	798 (67.3)
27c	360 (85.6)	408 (53.1)	534 (31.8)	804 (78.5)

withdrawing groups are located along a NH−NH axis, providing a maximum *trans*-diagonal effect for the transition dipole moments.

Synthesis of Bacteriopurpurinimide Metal Complexes. In contrast to porphyrins and chlorins, metalation of bacteriochlorin is difficult. Recently, a series of metal-substituted bacteriochlorophylls (Cu, Zn, Cd) were prepared using direct and transmetalation (Pd, Co, Mn) methods.[29] We appplied this methodology for the preparation of metal-substituted bacteriopurpurinimides to study the effect of metal complexation on their optical properties and photodynamic efficacy. For the treatment of cancer by photodynamic therapy (PDT), it is now well established that, besides light and oxygen, the nature of the photosensitizer and its photophysical, electrochemical, and photobleaching characteristics play an important role in overall efficacy. In general, the presence of a central metal in porphyrn-based macrocycles generally shows strong influence on such characteristics.

Our attempts to metalate bacteriopurpurin *a* **8c** by following the standard DMF and AcOH methods were unsuccessful and

SCHEME 6. Conversion of Free Base Bacteriochlorin to Various Metal Complexes



resulted in a mixture of oxidized and ring-opened products. In contrast, under similar conditions, the imide analogue **29c** was found to be stable and the corresponding Zn(II) (**36**) and Cd-(II) (**78**) complexes were obtained in good yields (∼70%) [Scheme 6].

The Cd(II)-substituted tetrapyrroles are known to be good precursors for preparation of other metal complexes, which are otherwise difficult to synthesize by direct metalation. Therefore, the Cd(II) bacteriochlorin was reacted with palladium chloride and the corresponding Pd(II)bacteriopupurin *a* imide **38** was obtained in 78% yield. Compared to the Cd(II)complex, the Zn-(II) and Pd(II) bacteriopupurins were found to be more stable. The spectral characteristics of the metal-substituted pigments are presented in Table 4. As expected, compared to the nonmetal

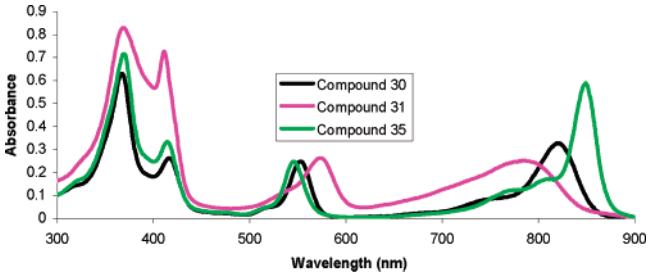


FIGURE 2. Electronic absorption spectra of the bacteriopurpurinimides (**30**, **31**, and **35**) in dichloromethane at equimolar concentration (10 μM).

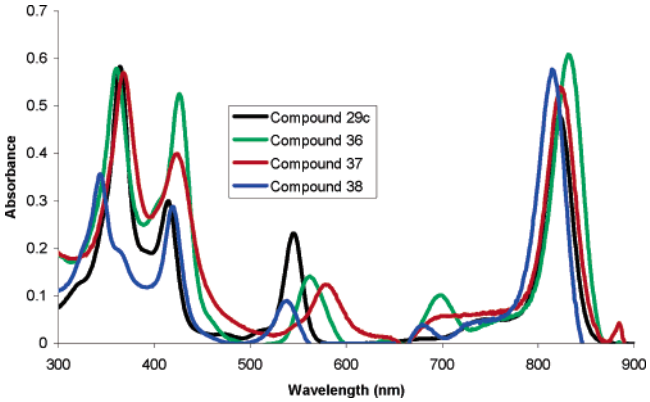


FIGURE 3. Electronic absorption spectra of bacteriopurpurinimide **29c** and the corresponding metal complexes, **36** [Zn(II)], **37** [Cd(II)], and **38** [Pd(II)], in dichloromethane at equimolar concentration (10.0 μM).

TABLE 4. Optical Properties of 29c and the Corresponding Zn(II), Cd(II), and Pd(II) Complexes

compd	Soret		Q_x	Q_y λ_{max}, nm ($\epsilon \times 10^3$)
29c	365 (60.6)	414 (36.4)	543 (25.7)	822 (48.5)
36	363 (58.0)	425 (52.1)	562 (14.1)	831 (61.0)
37	368 (58.6)	422 (42.4)	578 (14.8)	823 (56.1)
38	344 (35.8)	419 (28.9)	538 (9.02)	814 (67.9)

analogue **29c** the Zn(II) and Cd(II) complexes exhibited bathochromic shift, whereas the Pd(II) complex showed hypsochromic shifts. Because of their intense long-wavelength absorptions in the near-IR region, these compounds could be attractive candidates for PDT. The electronic absorption spectra of free base bacteriopurpurinimides and the metal complexes are shown in Figures 2 and 3.

Conclusion

We have developed an effective in situ transformation of bacteriochlorophyll *a* present in *Rh. sphaeroides* into bacteriopurpurin-18 via allomerization approach. This methodology has a significant advantage for the preparation of a variety of bacteriochlorins. The NMR and mass spectrometry analyses confirmed the structures of all allomerized products. We have also shown that the regioselective dehydrogenation of bacteriochlorins at pyrrolic ring D could be achieved to produce the corresponding chlorins in high yields. Our study indicates that the presence of a fused cyclic anhydride or imide ring has a significant impact on optical characteristics of the cyclic tetrapyrrolic system. The 3-acetyl-bacteriopurpurin-18-*N*-hexylimide was converted to the corresponding metalated analogues by following the direct or transmetalation approaches. Because of their strong absorptions in their near-IR regions, the highly stable bacteriopurpurinimides (free base and the corresponding metal complexes) have a distinct advantage over other naturally occurring bacteriochlorins as photosensitizers for photodynamic therapy. These compounds could also be useful precursors for developing more stable models to understand the natural bacterial photosynthetic reaction centers. The detailed photophysical and electrochemical studies with these novel structures are currently in progress.

Experimental Section

All reactions and operations were carried out with protection from direct light. Preparative thin-layer chromatography was performed on 1 mm silica gel plates. Column chromatography was carried out using silica gel 60 (70−230 mesh). Chemical shifts are reported in ppm (δ) downfield from tetramethylsilane as an internal standard.

General Procedure for Bacteriochlorophyll *a* Allomerization. Method A. Bacteriochlorophyll *a* (**1**) was extracted from *Rh. sphaeroides* (300 mL, ca. 20% dry weight) with propanol (800 mL) on stirring at room temperature for 12 h and under constant nitrogen bubbling. The deep blue-green extract was filtrated on a paper filter, and the residue was washed with propanol (3 × 200 mL). KOH (10 g) was added to the combined extract, and air was bubbled into the solution for 20 min with intensive stirring (room temperature). The solution was treated with 20% sulfuric acid to pH 1.5 and "unstabile bacteriochlorin" was extracted with a dichloromethane/THF mixture (1:1, v/v, 3 × 200 mL). The combined extracts without washing were evaporated in a vacuum at 55−60 °C. To the residue was added hexane (300 mL), and the resulting precipitate was filtrated and washed with hexane to remove most of the carotenoids. The precipitate was dissolved in dichloromethane/THF and chromatographed on a silica column to give fractions, which were purified using preparative TLC. The yields of isolated products (**8**−**24**) are reported in Table 1.

Method B. Same as method A, except air was bubbled into the reaction mixture for 1.5 h.

Method C. Same as method B, except the combined extract was evaporated at 85−90 °C.

Method D. Same as method B, except the combined extract was treated with sulfuric acid (2% v/v) and evaporated at 85−90°C.

Method E. Same as method B, except the combined extract was treated with sulfuric acid (2% v/v) and stirred at room temperature for 7 days.

Bacteriopheophorbide *a* (*2*) [Method A]. *Rh. sphaeroides* (300 mL) was added to a 2-L Erlenmeyer flask with 1-propanol (1.5 L), and nitrogen was bubbled into the suspension with stirring at room temperature for 12 h. The mixture was filtered under vacuum through Whiteman no. 1 filter paper, and the residue was washed with 1-propanol (2 × 300 mL) until the filtrate was colorless. To the combined extract was added concentrated HCl (20 mL), and the solution was stirred for 20 min and then poured into aqueous 5% NaCl (1.5 L). Bacteriopheophorbide *a* was extracted with CH_2Cl_2 (3 × 250 mL), and the extract washed with brine (3 × 500 mL). The extract was dried over sodium sulfate, and solvent was evaporated under vacuum. The resulting residue was purified by column chromatography on silica and recrystallized from CH_2Cl_2/hexane to give the title compound as a violet-black powder (850 mg, 96%): mp 191−197 °C; UV−vis, λ_{max} (CH_2Cl_2), nm (ϵ x 10^4), 756 (10.7), 543 (3.21), 410 (5.48) and 363 (7.76); ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 9.22 (s, 1H, 5-H), 8.80 (s, 1H, 10-H), 8.62 (s, 1 H, 20-H), 6.40 (s, 1H, 13^2-H), 5.14 (d, 1H, J =9.1 Hz, 17-H), 4.30 (m, 2H, 7-H, 18-H), 4.08 (m, 1H, 8-H), 3.66 (s, 3H, 12-Me), 3.60 (s, 3H, OMe), 3.55 (s, 3H, 2-Me), 3.18 (s, 3H, 3-$COCH_3$), 2.73 (m, 1H, 17^1-H), 2.41 (m, 3H, 8^1-CH_2 + 17^2-H), 2.14 (m, H,

17^{1a}-H), 1.98 (m, H, 17^2-H), 1.81, 1.73 (each d, 6H, $J = 7.5$ Hz, 18-Me, 7 CH_3), 1.11 (t, 3H, $J = 7.5$ Hz, 8-CH_3), 0,30 and −0.67 (each br s, 21, 23-NH); HRMS calcd for $C_{35}H_{38}N_4O_6$ 610.2791, found 610.2711.

Treatment of 2 with Methanol/Sulfuric Acid. Bacteriopheophorbide *a* (150 mg) was dissolved in 2% sulfuric acid in methanol (250 mL), and the reaction mixture stirred under nitrogen for 12 h. It was then poured into ic− water (500 mL) and extracted with CH_2Cl_2 (3 × 200 mL). The combined extract was dried over sodium sulfate, and the solvent was evaporated under vacuum. The residue was chromatographed on silica column, and two main fractions were collected.

The product obtained from the most polar band was identified as **13^2-*(R,S)*-hydroxy-bacteriopheophorbide *a* methyl ester** (**3**, **4**) (84.9 mg) as a dark-violet powder (from CH_2Cl_2/hexane), obtained as 1:4 mixture of diastereomers: UV−vis, λ_{max} (CH_2Cl_2), nm ($\epsilon \times 10^4$), 756 (10.7), 543 (3.21), 410 (5.48) and 363 (7.76); ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm). *Major diastereomer* (*S*): 9.12 (s, 1H, 5-H), 8.59 (s, 1H, 10-H), 8.52 (s, 1H, 20-H), 5.39 (br s, 1H, 13^2-OH), 4.38 (m, 2H, 7,8-H), 4.08 (m, 1H, 18-H), 3.98 (dd, 1H, $J = 8.0$, 1.3 Hz, 17-H), 3.66 (s, 3H, CH_3), 3.61 (s, 3H, CH_3), 3.47 (s, 3H, CH_3), 3.46 (s, 3H, CH_3), 3.18 (s, 3H, 3-$COCH_3$), 2.86 (m, 1H, 17^1-H), 2.54 (m, 1H, 17^1-H), 2.34 (m, 4H, 2H for 8-CH_2CH_3, 2H for 17^2-H,), 1.84 (d, 3H, $J = 8.0$ Hz, 18-CH_3), 1.58 (d, 3H, $J = 8.0$ Hz, 7-CH_3), 1.23 (t, 3H, $J = 7.6$ Hz, 8-CH_3), 0.20 and −0.19 (each br s, 21, 23-NH). *Minor diastereomer* (*R*): 9.10 (s, 1H, 5-H), 8.57 (s, 1H, 10-H), 8.49 (s, 1H, 20-H), 5.46 (br s, 1H, 13^2-OH), 4.45 (dd, 1H, $J = 8.0$, 1.3 Hz, 17-H), 4.38 (m, 2H, 7,8-H), 4.06 (m, 1H, 18-H), 3.66 (s, 3H, CH_3), 3.59 (s, 3H, CH_3), 3.45 (s, 3H, CH_3), 3.43 (s, 3H, CH_3), 3.16 (s, 3H, 3-$COCH_3$), 2.86 (m, 1H, 17^1-H), 2.54 (m, 1H, 17^1-H), 2.34 (m, 4H, 2H for 8-CH_2-CH_3, 2H for 17^2-H,), 1.78 (d, 3H, $J = 8.0$ Hz, 18-CH_3), 1.61 (d, 3H, $J = 8.0$ Hz, 7-CH_3), 1.23 (t, 3H, $J = 7.6$ Hz, 8-CH_3), 0.28 and −0.16 (each br s, 21, 23-NH); HRMS (FAB) calcd for $C_{36}H_{40}N_4O_7$ 640.2897, found 640.2888

The most nonpolar band was identified as **3-acetyl-3-devinyl-pheophorbide *a* methyl ester 6** (2.9 mg) as brown needles recrystallized from CH_2Cl_2/hexane: mp 191−197 °C; UV−vis, λ_{max} (CH_2Cl_2), nm ($\epsilon \times 10^4$), 754 (10.8), 546 (3.24), 410 (5.40) and 360 (7.74); ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 10.02 (s, 1H, 5-H), 9.70 (s, 1H, 10-H), 8.82 (s, 1H, 20-H), 6.39 (s, 1H, 13^2-H), 4.54 (dd, 1H, $J = 7.8$, 1.2 Hz, 17-H), 4.28 (dq, 1H, $J = 7.5$, $J = 1.2$ Hz, 18-H), 3.86 (s, 3H, 13^2-CO_2CH_3), 3.73 (s, 3H, CH_3), 3.69 (q, 2H, 8-CH_2), 3.67 (s, 3H, CO_2CH_3), 3.62 (s, 3H, CH_3), 3.34 (s, 6H, 2 x CH_3), 2.72 (m, 1H, 17^1-H), 2.49 (m, 1H, 17^1-H), 2.34 (m, 2H, 17^2-H), 1.82 (d, 3H, $J = 7.8$ Hz, 18-CH_3), 1.61 (t, 3H, $J = 7.6$ Hz, 8-CH_3), −1.60 and −1.79 (each br s, 21, 23-NH); MS−FAB *m*/*z* 622.3 (M^+, 100%), 567 (67), 532 (37), 508 (23); HRMS (FAB) calcd for $C_{36}H_{38}N_4O_6$ 622.2791, found 622.2796.

By following Method B, the slowest moving band was characterized as bacteriopurpurin *a* carboxylic acid (**8a**): NMR similar to **8b**, except the resonance for the CO_2CH_3 was missing. Anal. Calcd for $C_{33}H_{34}N_4O_6$: C, 68.03; H, 5.88; N, 9.62. Found: C, 68.36; H, 5.73; N, 9.51.

Bacteriopurpurin *a* Methyl Ester (8b). Compound **2** (100 mg, 0.17 mmol) was dissolved in ethyl ether (200 mL), KOH (1 g, 21 mmol) in propanol (50 mL), and pyridine (10 mL). Air was bubbled into the solution for 0.5 h with intensive stirring at room temperature. The solution was diluted with water (300 mL), and the ether layer was removed. The water layer was treated with 1.0 N HCl until pH 4.0 and extracted with dichloromethane.THF (1:1, 5 × 100 mL). Combined extracts were washed with water (2 × 300 mL) and treated with diazomethane, and reaction progress was monitored by TLC. The reaction mixture was evaporated in a vacuum to give a residue, which was purified on a silica column and recrystallized from dichloromethane/hexane to give the title compound as fine purple crystals (28.3 mg, 29%): mp 272 °C; UV−vis λ_{max} (CH_2Cl_2) 364 nm (ϵ 8.91 × 10^4), 412 nm (ϵ 5.36 × 10^4), 545 nm (ϵ 3.4 × 10^4), 815 (ϵ 5.53 × 10^4); ^{1}H NMR (400 MHz, $CDCl_3$) δ (ppm) 9.21(s, 1H, 5-H), 8.78(s, 1H, 10-H), 8.62-(s, 1H, 20-H), 5.13(m,1H, 17-H), 4.30(m, 2H, 1H for 7-H, 1H for 18-H), 4.08(m, 1H, 8-H), 3.63(s, 3H, 12-CH_3), 3.57(s, 3H, -$COOCH_3$), 3.52(s, 3H, 2-CH_3), 3.15(s, 3H, CH_3C), 2.70(m, 1H, -C*H*HCH_2COOCH_3), 2.42(m, 2H, 8-CH_2CH_3), 2.35(m, 1H, -CH*H*CH_2COOCH_3), 2.00(m, 2H, -$CH_2CH_2COOCH_3$), 1.80(d, $J = 7.17$ Hz, 3H, 7-CH_3),1.70(d, $J = 6.82$ Hz, 3H, 18-CH_3), 1.1(t, $J = 6.46$ Hz, 3H, 8-CH_2CH_3), −0.30(s, 1H, NH), −0.65(s, 1H, NH) Anal. Calcd for $C_{34}H_{36}N_4O_6$: C, 68.44; H, 6.08; N, 9.36. Found: C, 68.16; H, 6.03; N, 9.11. Mass calcd for $C_{34}H_{36}N_4O_6$: 596.3. Found: *m*/*e* 596.8 (M + 1). HRMS: calcd 596.2635, found 596.2615.

Note. For the characterization of compounds **8c**, **9**−**11**, **12c**, and **13**−**24**, see Supporting Information.

General Procedure for Synthesis of Isoimide Derivatives. DCC (5−10 equiv) was added to a solution of amide **26/27a**−**c** (1 equiv) in dichloromethane. The reaction mixture was stirred at room temperature for 24−48 h, with progress monitored by UV−vis spectra. After the completion of reaction, the solution was cooled to −10 °C and precipitated urea was filtered off. The solvent was removed in a vacuum, and the residue was chromatographed on silica using a gradient of 2−6% acetone/dichloromethane as eluent. Isolated bands were purified using preparative TLC.

Bacteriopurpurin *a* 13^1-*N*-Hexylisoimide Propyl Ester (27c). Prepared from a mixture of **25c** and **26c** (100 mg, 0.137 mmol) using the general procedure as described above. After evaporation of the solvents the residue was recrystallized from dichloromethane/hexane to give the title compound (48.7 mg, 50%): UV−vis λ_{max} (CH_2Cl_2), see Table 3; ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) 9.38 (s, 1H, 5- H), 8.88 (s, 1H, 10-H), 8.73 (s, 1 H, 20-H), 5.46 (m, 1H, NHCO), 5.18 (d, 1H, $J = 8.0$, 17-H), 4.34 (m, 2H, 7-H and 18-H), 4.17 (m, 1H, 8-H), 3.91 (m, 2H, hexylamide *a*-CH_2), 4.06 (t, 2H, CO_2CH_2), 3.68 (s, 3H, 12-Me), 3.59 (s, 3H, 2-Me), 3.19 (s, 3H, 3-Me), 2.73 (m, H, 17b-H), 2.42 (m, 5H, $CH_2CH_2CH_3$ and 8a-CH_2 and 7b′-H), 2.14 (m, H, 17a-H), 2.08 (m, 5H, hexylamide-*b*,*c*-CH_2 and 17a′-H), 2.01, 1.93 (each d, 3H, $J = 8.0$ and 18-Me, 7-Me), 1.57 (m, 4H, hexylamide-*d*,*e*-CH_2 and cyclohexyl-CH), 1.25 (m, 20H, cyclohexyl-CH_2), 1.12 (t, 3H, $J = 7.8$, 3b-Me), 0.96 (t, 3H, hexylamide-*f*-CH_3), 0.87 (t, 3H, $J = 8.2$, $CH_2CH_2CH_3$), −0.86 and −1.13 (each br s, 2H, 21 and 23-NH).

Note. For the characterization of compound **28c**, see Supporting Information.

Base-Catalyzed Rearrangement of Isoimides to Imide. A mixture of isoimide **27c** and **28c** (50 mg) in dichloromethane was treated with a catalytic amount of KOH/methanol, and the reaction mixture was stirred for 10 min. The completion reaction was monitored spectrophotometrically (new peak at 822). The reaction mixture was washed with water. The organic layer was separated and dried over anhydrous sodium sulfate, and the solvent was evaporated. The residue after crystallization from dichloromethane/hexane afforded **29c**, **30**, and **31**.

Bacteriopurpurin *a* *N*-Hexylimide 17-Propyl Ester (29c). Mp > 260 °C (dec); UV−vis λ_{max} (CH_2Cl_2) 364 nm (ϵ 6.06 × 10^4), 413 nm (3.64 × 10^4), 545 nm (2.57 × 10^4), 822 (4.85 × 10^4); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) 9.23(s, 1H, 5-H), 8.80(s, 1H, 10-H), 8.62(s, 1H, 20-H), 5.32(m,1H, 17-H), 4.43(t, $J = 6.03$ Hz, 2H, -$NCH_2(CH_2)_4CH_3$), 4.3(m, 2H, 1H for 7-H, 1H for 18-H), 4.10-(m, 1H, 8-H), 3.90(m, 2H, $COOCH_2CH_2CH_3$), 3.71(s, 3H,12-CH_3), 3.54(s, 3H, 2-CH_3), 3.16(s,3H, CH_3C), 2.65(m, 1H, -$CHHCH_2$-$COOC_3H_7$), 2.36(m, 3H, 1H for -$CHHCH_2COOC_3H_7$, 2H for 8-CH_2-CH_3), 1.97(m, 2H, -$NCH_2CH_2(CH_2)_3CH_3$), 1.82(d, $J = 6.90$ Hz, 3H, 7-CH_3), 1.74(d, $J = 6.91$ Hz, 3H, 18-CH_3), 1.60(m, 2H, -CH_2-$CH_2COOC_3H_7$), 1.50(m,2H, -$COOCH_2CH_2CH_3$), 1.42(m, 4H, -NCH_2-$CH_2CH_2CH_2CH_2CH_3$), 1.30(m, 2H−$(CH_2)_4CH_2CH_3$), 1.10(t, $J = 6.89$ Hz, 3H, 8-CH_2CH_3), 0.95(t, $J = 7.60$ Hz, 3H, $N(CH_2)_5CH_3$), 0.85(t, $J = 7.30H_z$, 3H, -$COOCH_2CH_2CH_3$), −0.5(s, 1H, NH), −0.72(s, 1H, NH). Mass calcd for $C_{42}H_{53}O_5N_5$ 707.4, found 707.9 (M + 1). HRMS: calcd 707.4046, found 707.4051.

Note. For the characterization of compounds **30** and **31**, see Supporting Information.

12-Demethyl-12-methylene-bacteriopurpurin-18-*N*-hexylimide Propyl Ester (35). Bacteriopurpurinimide **30** (10 mg) was dissolved in *o*-dichlorobenzene. The reaction mixture was refluxed for 1 h and monitored spectrophotometrically (disappearance of the original peak at 821 nm and an appearance of a new peak at 849 nm). It was then washed with water. The organic layer was separated and dried over anhydrous sodium sulfate. Evaporation of the solvent gave a residue, which was purified by preparative chromatography. Yield: 30%. UV−vis λ_{max} (CH_2Cl_2) 849 nm (ϵ 5.88 × 10^4), 546 nm (ϵ 2.47 × 10^4), 414 nm (ϵ 3.33 × 10^4), 369 (ϵ 7.15 × 10^4); ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) 9.30 (s, 1H, 10-H), 9.06 (s, 1H, 5-H), 8.54(s, 1H, 20-H), 5.35 (m,1H, 17-H), 5.00 (s, 2H, 12-C=CH_2), 4.70 (t, J = 6.03 Hz, 2H, -NCH_2(CH_2)$_4$$CH_3$), 4.25 (m, 1H, 7-H), 4.15 (m, 1H, 18-H), 3.95 (m, 4H, 2H for $COOCH_2CH_2CH_3$, 2H for 8-CH_2CH_3), 3.50 (s, 3H, 2-CH_3), 3.11 (s,3H, CH_3CO-), 2.40 (m, 2H, -C$H_2CH_2COOC_3H_7$), 2.15 (m, 2H, -$NCH_2CH_2(CH_2)_3CH_3$), 2.00 (m, 2H, -$CH_2CH_2COOC_3H_7$), 1.75 (d, J = 6.90 Hz, 3H, 7-CH_3), 1.70 (d, J = 6.91 Hz, 3H, 18-CH_3), 1.50 (m, 6H, 4H for -$NCH_2CH_2CH_2CH_2CH_3$, 2H for -CH_2CH_2-$COOC_3H_7$), 1.25 (m, 5H, 2H for -$(CH_2)_4CH_2CH_3$, 3H for 8-CH_2CH_3), 0.98 (t, J = 7.60 Hz, 3H, N$(CH_2)_5CH_3$), 0.86 (t, J = 7.30H_z, 3H, -$COOCH_2CH_2CH_3$), −0.57 (s, 1H, NH), −0.82 (s, 1H, NH); HRMS calcd for $C_{42}H_{51}O_5N_5$ 705.3890, found 705.3824.

Zn(II) Complex of 3-Acetyl-bacteriopurpurin-18-*N*-hexylimide Propyl Ester (36). 3-Acetyl-bacteriopurpurin-18-*N*-hexylimide 1-propyl ester **29C** (50 mg, 0.070) was dissolved in 40 mL of anhydrous *N*,*N*-dimethyl formide (DMF), and then dry zinc acetate (600 mg) was added. Under a continuous flow of dry argon, the mixture was heated at 140 °C for 0.5 h. After all of the DMF was removed by high vacuum (bath temperature 60 °C), the residue was chromatographed over a silica column eluting with methanol/CH_2Cl_2 (5% v/v). The appropriate fractions were combined. The residue obtained after removing the solvents was crystallized from CH_2Cl_2/*n*-hexane as dark red crystals in 68% yield (37 mg): mp 180−182°C. UV−vis λ_{max} (in CH_2Cl_2), see Table 4; ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) 8.57 (two singlet overlapped, 2H, 5-H and 10-H), 8.35(s, 1H, 20-H), 5.11(m,1H, 17-H), 4.20(m, 1H, -NC*H*H-$(CH_2)_4CH_3$), 4.05(m, 3H, 1H, -NCH*H*$(CH_2)_4CH_3$, 1H for 7-H, 1H for 18-H), 3.95(m, 1H, 8-H), 3.80 (m, 2H, $COOCH_2CH_2CH_3$), 3.50 (s, 3H,12-CH_3), 3.00 (s, 3H, 2-CH_3), 2.50 (s,3H, CH_3CO-), 2.30 (m, 2H, -C$H_2CH_2COOC_3H_7$), 2.00(m, 4H, 2H for 8-CH_2CH_3, 2H for-$NCH_2CH_2(CH_2)_3CH_3$), 1.80 (2H, for -$CH_2CH_2COOC_3H_7$), 1.65-(d, J = 6.84 Hz, 3H, 7-CH_3), 1.53 (d, J = 6.88 Hz, 3H, 18-CH_3), 1.46 (m,2H, -$COOCH_2CH_2CH_3$), 1.38−1.15 (m, 9H, 6H for -NCH_2-$CH_2CH_2CH_2CH_2CH_3$, 3H for 8-CH_2CH_3), 0.87 (t, J = 7.58 Hz, 3H, N$(CH_2)_5CH_3$), 0.75 (t, J = 7.30H_z, 3H, -$COOCH_2CH_2CH_3$); HRMS calcd for $C_{42}H_{51}O_5N_5Zn$ 769.3181, found 769.3114.

Cd(II) Complex of 3-Acetyl-bacteriopurpurin-18-*N*-hexylimide Propyl Ester (37). 3-Acetyl-bacteriopurpurin-18-*N*-hexylimide propyl ester **29C** (50 mg, 0.070 mmol) was dissolved in 40 mL of anhydrous *N*,*N*-dimethyl formamide (DMF), and then cadmium acetate (600 mg) was added. Under a continuous flow of dry argon, the mixture was heated at 140 °C for 0.5 h. All of the DMF was removed by high vacuum and heat (60 °C). After the completion of the reaction, the residue was chromatographed over a silica column eluting with methanol/CH_2Cl_2 (10% v/v). The appropriate fractions were combined. The product obtained after removing the solvents was crystallized from CH_2Cl_2/*n*-hexane as dark red crystals in 35% yield (20 mg): mp > 260 °C; UV−vis λ_{max} (CH_2Cl_2), see Table 4; ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) 8.44 (s, 1H, 5-H), 8.26 (two singlet overlapped, 2H, 10-H and 20-H), 4.85 (m,1H, 17-H), 4.05 (m, 5H, 2H for -NCH_2(CH_2)$_4$$CH_3$, 1H for 7-H, 1H for 18-H, 1H for 8-H), 3.79 (m, 2H, $COOCH_2CH_2CH_3$), 3.51 (s, 3H,-12-CH_3), 3.02 (s, 3H, 2-CH_3), 2.48 (s,3H, CH_3CO-), 2.31 (m, 2H, -C$H_2CH_2COOC_3H_7$), 2.08 (m, 4H, 2H for 8-CH_2CH_3, 2H for-$NCH_2CH_2(CH_2)_3CH_3$), 1.70 (m,5H, 2H for -$CH_2CH_2COOC_3H_7$, 3H for 7-CH_3), 1.50 (d, J = 6.88 Hz, 3H, 18-CH_3), 1.47 (m,2H, -$COOCH_2CH_2CH_3$), 1.30−1.10 (m, 9H, 6H for -NCH_2-$CH_2CH_2CH_2CH_2CH_3$, 3H for 8-CH_2CH_3), 0.85 (t, J = 7.58 Hz, 3H, N$(CH_2)_5CH_3$), 0.75 (t, J = 7.30H_z, 3H, -$COOCH_2CH_2CH_3$); HRMS calcd for $C_{42}H_{51}O_5N_5Cd$ 819.2925, found 819.2976

Pd(II) Complex of 3-Acetyl-bacteriopurpurin-18-*N*-hexylimide Propyl Ester (38). The Cd(II) complex of bacteriopurpurinimide **37** (18 mg, 0.0021 mmol) was dissolved in anhydrous acetone (40 mL), and then palladium chloride (600 mg) was added. Under a continuous flow of dry argon, the mixture was refluxed for 0.5 h. After evaporation of the solvent, the residue was chromatographed over a silica column eluting with acetone/CH_2Cl_2 (1% v/v). The appropriate fractions were combined. The residue obtained after removing the solvents was crystallized from CH_2Cl_2/*n*-hexane as dark red crystals in about 50% yield (24 mg): mp 220−222°C; UV−vis λ_{max} (in CH_2Cl_2), see Table 4; ^{1}H NMR (400 MHz, $CDCl_3$, δ ppm) 9.27(s, 1H,5-H), 8.60 (s, 1H,10-H), 8.54 (s, 1H, 20-H), 5.44 (m,1H, 17-H), 4.38 (m, 4H, 2H for -NCH_2(CH_2)$_4$$CH_3$, 1H for 7-H, 1H for 18-H), 4.05 (m, 1H, 8-H), 3.80 (m, 2H, $COOCH_2$-CH_2CH_3), 3.60 (s, 3H,12-CH_3), 3.45 (s, 3H, 2-CH_3), 3.05 (s,3H, CH_3CO-), 2.65(m, 2H, -C$H_2CH_2COOC_3H_7$), 2.35 (m, 4H, 2H for 8-CH_2CH_3, 2H for-$NCH_2CH_2(CH_2)_3CH_3$), 2.00(2H, for -CH_2CH_2-$COOC_3H_7$), 1.90 (d, J = 6.84 Hz, 3H, 7-CH_3), 1.80(d, J = 6.88 Hz, 3H, 18-CH_3), 1.47 (m,2H, -$COOCH_2CH_2CH_3$), 1.50−1.20 (m, 9H, 6H for -$NCH_2CH_2CH_2CH_2CH_2CH_3$, 3H for 8-$CH_2CH_3$), 0.90 (t, J = 7.58 Hz, 3H, N$(CH_2)_5CH_3$), 0.75 (t, J = 7.30H_z, 3H, -$COOCH_2CH_2CH_3$); HRMS calcd for $C_{42}H_{51}O_5N_5Pd$ 811.2919, found 811.2998.

Supporting Information Available: Characterization data of compounds **8c**, **9**−**11**, **12b**, **13**−**24**, **28c**, **30**, and **31**. This material is available free of charge via the Internet at http://pubs.acs.org.

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