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Asymmetric Synthesis of a Bacteriochlorophyll Model Compound Containing *trans*-Dialkyl Substituents in Ring D

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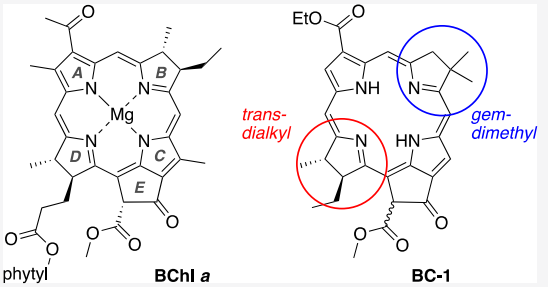
ABSTRACT: Challenges to the *de novo* synthesis of bacteriochlorophyll *a* (**BChl *a***), the chief pigment for anoxygenic bacterial photosynthesis, include creating the macrocycle along with the *trans*-dialkyl substituents in both pyrroline rings (B and D). A known route to a model bacteriochlorophyll with a gem-dimethyl group in each pyrroline ring has been probed for utility in the synthesis of **BChl *a*** by preparation of a hybrid macrocycle (**BC-1**), which contains a *trans*-dialkyl group in ring D and a gem-dimethyl group in ring B. Stereochemical definition began with the synthesis of (2*S*,3*S*)-2-ethyl-3-methylpent-4-ynoic acid, a precursor to the *trans*-dialkyl-substituted AD dihydrodipyrrin. Knoevenagel condensation of the latter and a gem-dimethyl, *β*-ketoester-substituted BC dihydrodipyrrin afforded the enone (E, 70%; Z, 3%); subsequent double-ring cyclization of the *E*-enone (via Nazarov, electrophilic aromatic substitution, and elimination reactions) gave **BC-1** (53% yield) along with a trace of chlorin byproduct (1.4% relative to **BC-1** upon fluorescence assay). **BC-1** exhibited the desired *trans*-dialkyl stereochemistry in ring D and was obtained as a 7:1 mixture of (expected) epimers owing to the configuration of the 13^2-carbomethoxy substituent. The strategy wherein *trans*-dialkyl substituents are installed very early and carried through to completion, as validated herein, potentially opens a synthetic path to native photosynthetic pigments.



1. INTRODUCTION

Bacteriochlorophylls provide the chief pigments for anoxygenic bacterial photosynthesis analogous to chlorophylls in oxygenic plant and cyanobacterial photosynthesis.[1,2] It is somewhat paradoxical that anoxygenic photosynthesis is far simpler than oxygenic photosynthesis, yet bacteriochlorophylls are structurally more complex than chlorophylls. Bacteriochlorophyll *a* (a bacteriochlorin) contains two pyrroline rings (B and D), whereas chlorophyll *a* (a chlorin) contains only a single pyrroline ring (D). Each pyrroline ring bears a *trans*-dialkyl group (Chart 1). Bacteriochlorophyll *a* absorbs in the near-infrared (771 nm) spectral region, whereas chlorophyll *a* absorbs in the red (660 nm) spectral region.[3,4] Anoxygenic photosynthesis has been the object of ongoing study for many decades now, yet bacteriochlorophylls (and their free base analogues, bacteriopheophytins) have remained outside the scope of chemical synthesis.[5]

Over the years, we have worked on developing routes to non-natural chlorins and bacteriochlorins. In these routes, we have employed a gem-dimethyl group in each pyrroline ring. The rationale[6] for the gem-dimethyl group is chiefly two-fold: (1) in the absence of gem-dimethyl groups, the pyrroline ring is susceptible to adventitious oxidative dehydrogenation, which converts the chlorin to a porphyrin, and the bacteriochlorin to a chlorin and then also the porphyrin. The progressive loss of saturation affords a commensurate loss of long-wavelength absorption. (2) Synthetic installation has proved far simpler for the gem-dimethyl moiety than the *trans*-dialkyl group (which may provide some resistance to adventitious dehydrogenation compared with the fully unsubstituted chromophore). A recent synthesis of model bacteriochlorophylls, wherein a gem-dimethyl group is positioned in each pyrroline ring is shown in Scheme 1.[7] The synthesis employs reaction of two dihydrodipyrrins: a gem-dimethyl-substituted AD half (**I**) and a gem-dimethyl-substituted BC half (**II**). A Knoevenagel condensation joins the two-halves via a propenone linkage (**III**), whereas the second set of reactions constructs the macrocycle and forms the isocyclic ring to afford the bacteriochlorophyll model compound (**IV**). The second set of reactions is carried out as a one-flask process and entails Nazarov cyclization, electrophilic aromatic substitution (S_EAr), and elimination of methanol. The conditions are relatively mild in the overall transformation. The strategy has been extended to afford chlorophyll model compounds by use of a dipyrromethane (rather than dihydrodipyrrin) as the BC half.[8]

We recently began exploring the extension of the reaction processes shown in Scheme 1 to accommodate *trans*-dialkyl rather than gem-dimethyl substitution in the pyrroline ring.

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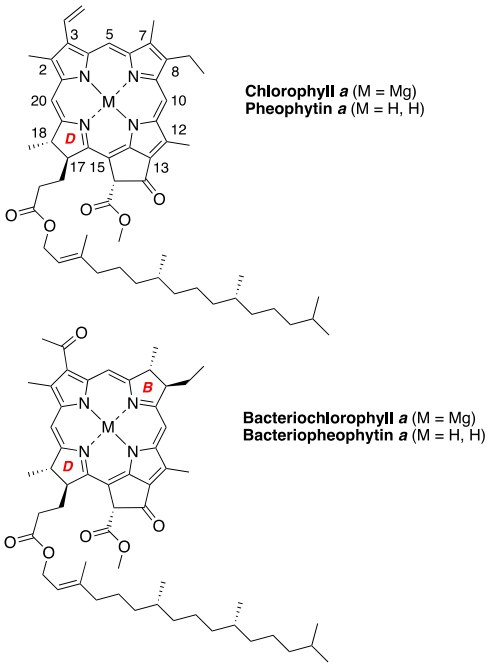
Chart 1. Native Photosynthetic Pigments



The purpose for doing so is to establish robust routes to the native photosynthetic pigments, including the bacteriochlorophylls and chlorophylls. The proposed route to **BChl *a*** (R = phytyl, R^3 = Ac, M = Mg) and analogues mirrors the synthesis of **IV**, as is shown in Scheme 2. The route installs the *trans*-dialkyl configuration at a very early stage of the synthesis (e.g., **pre-B** and **pre-D**) and carries the *trans*-dialkyl groups through multiple stages to the target macrocycle. The potential advantages of this strategy are the ability to rely on (1) well-established stereoselective synthetic methods to prepare the alkynoic acids **pre-B** and **pre-D**, (2) straightforward access to the pyrroles including **pre-C**,[9] and (3) a concise route from AD and BC dihydrodipyrrins to the target macrocycle.

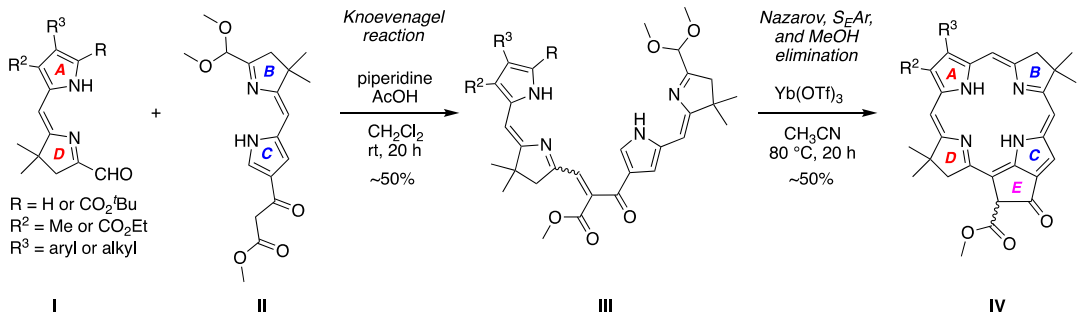
On the other hand, key issues are whether the *trans*-dialkyl group can be carried through multiple transformations with high fidelity. Racemization and epimerization are obvious concerns in essentially all syntheses using stereodefined precursors. Yet here, the dihydrodipyrrin intermediates are fraught with two potential problems of greater severity. First, tautomerization[9] in ring B or D would convert the dihydrodipyrrins **V** and **VI** to the corresponding dipyrromethanes, losing the essential saturation of the corresponding target macrocycle. Tautomerization in either ring B or D would give rise to the chlorin, not the bacteriochlorin; tautomerization in both rings would give the porphyrin. Second, beyond the little appreciated abyss upon tautomerization of dihydrodipyrrins, the problem of adventitious oxidation remains a paramount concern. Adventitious dehydrogenation of hydroporphyrins lacking stabilizing structural features (such as gem-dimethyl groups) is well known,[10−13] and the same susceptibility is expected to manifest with hydrodipyrrins. Both rings B and D of the dihydrodipyrrins are susceptible to dehydrogenation on routine handling to form the corresponding dipyrrins, which would also give the chlorin or porphyrin. The dihydrodipyrrins (**V** and **VI**), propenone derived therefrom, and target bacteriochlorin are all susceptible to adventitious dehydrogenation. In short, tautomerization and dehydrogenation must be avoided at all stages following dihydrodipyrrin construction for the success of the proposed synthetic route to *trans*-dialkyl hydroporphyrins. Stereochemical inversion would afford the undesired bacteriochlorin isomer, whereas tautomerization or dehydrogenation would cause loss of stereochemistry and loss of the bacteriochlorin chromophore. None of those problems arise with the gem-dimethyl-substituted counterparts, where extensive development chemistry has been carried out,[6−8] yet the applicability of such chemistry to the *trans*-dialkyl counterparts has heretofore not been addressed.

To ascertain the viability of the proposed synthetic route, we elected to prepare a model bacteriochlorin that contains one *trans*-dialkyl-substituted pyrroline ring (D) and one gem-dimethyl-substituted pyrroline ring (B). The resulting bacteriochlorin (**BC-1**) and precursors have one point of vulnerability toward dehydrogenation, tautomerization, and/or stereochemical inversion, and the choice of **BC-1** as a target thus enables the aforementioned issues to be singularly scrutinized. Here, we report the synthesis of **BC-1**, wherein the *trans*-dialkyl configuration of ring D is set at the earliest stage, namely, the synthesis of an alkynoic acid (analogous to **pre-D**). Six subsequent steps lead to the target **BC-1**. The stereochemistry of the two alkyl groups in **BC-1** is identical to that of the native bacteriochlorophyll macrocycles. Extensive characterization studies of intermediates and products support the validity of the synthetic strategy proposed herein.

2. RESULTS AND DISCUSSION

2.1. Synthesis of a Model AD Half. The route for the preparation of the AD-half relies on a synthetic procedure first described by Jacobi and co-workers, wherein a 2-halopyrrole is reacted with a 4-pentynoic acid.[14] As a model synthesis for bacteriochlorophyll *a*, which contains an acetyl group at the 3-position, we chose a carboethoxy analogue. Thus, van Leusen cycloaddition[15,16] of *p*-toluenesulfonylmethyl isocyanide (Tos-

Scheme 1. Established Route to Gem-Dimethyl-Substituted Model Bacteriochlorophylls



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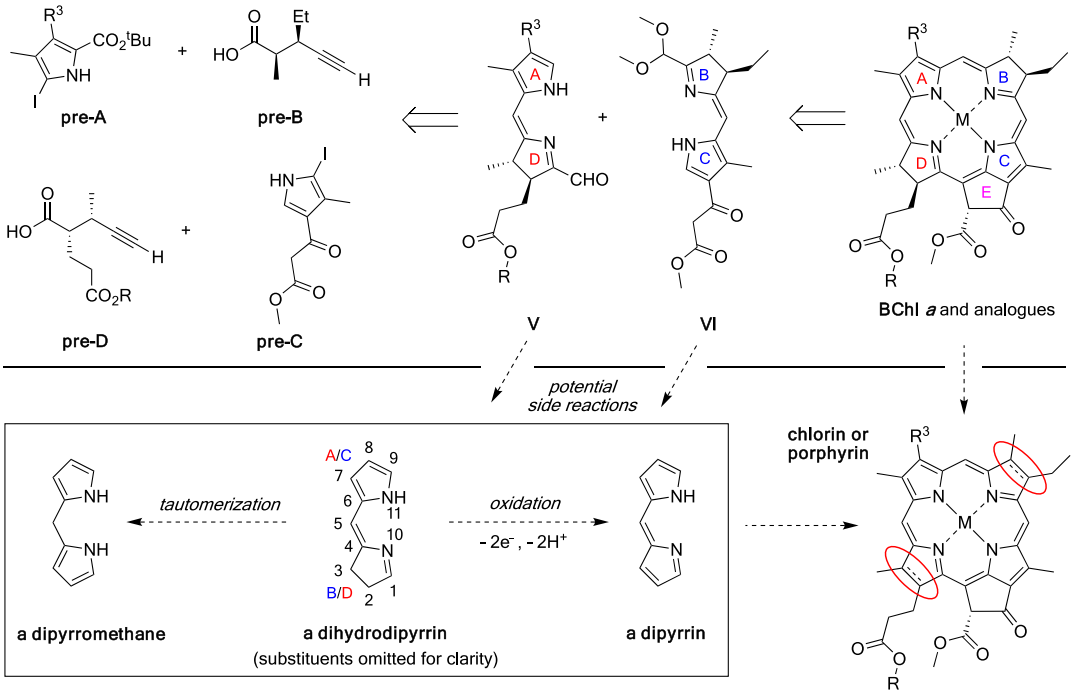
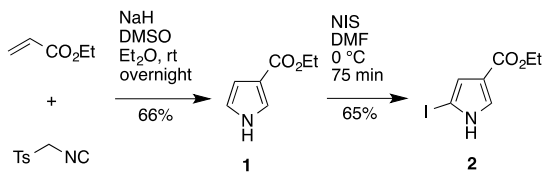
Scheme 2. Proposed Route to Native Bacteriochlorophylls (Top) and Potential Side Reactions (Bottom)



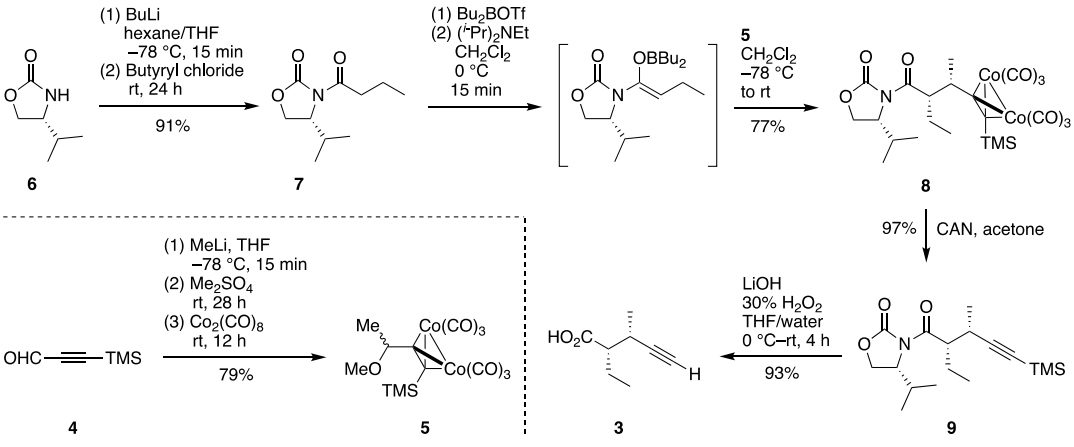
MIC) with ethyl acrylate followed by treatment of the resulting pyrrole **1**[17] with *N*-iodosuccinimide (NIS) gave pyrrole **2** (Scheme 3) in 5-fold larger quantity than in the prior reports.[18]

Scheme 3. Synthesis of Pyrrole Ring A



The two vicinal stereocenters in the substituted pentynoic acid (**3**) can be simultaneously assembled via the Nicholas reaction,[19] where a hexacarbonyldicobalt-stabilized propargylic ether is reacted with the boron enolate of the appropriate *N*-acyl oxazolidinone.[20] The appropriate choice of the enantiomerically pure oxazolidinone (Evans chiral auxiliary) results in the predominant formation of an adduct possessing the desired stereocenters, which can be easily purified from its diastereomeric byproduct through chromatography. A one-flask conversion of commercial 3-(trimethylsilyl)propynal (**4**) to (±)hexacarbonyldicobalt-stabilized propargylic ether **5**[21] was developed (Scheme 4). Thus, reaction of MeLi with **4** at low temperature followed by treatment with dimethyl sulfate and then $Co_2(CO)_8$ at room temperature gave **5**, which was easily purified by column chromatography. Treatment of the lithium salt of (*R*)-4-isopropyloxazolidin-2-one (**6**) with butyryl chloride afforded (*R*)-3-butyryl-4-isopropyloxazolidin-2-one (**7**[22]). Reaction of the latter with Bu_2BOTf in a basic medium generated the corresponding boron enolate, which underwent an aldol-type condensation with **5** to exclusively form **8** containing the expected two *S*-stereocenters. Mild oxidation with ceric ammonium nitrate (CAN) caused removal of the hexacarbonyldicobalt moiety. In the final step, treatment[23] of **9** with LiOOH (generated from LiOH and

Scheme 4. Synthesis of a Stereodefined Precursor to Ring D



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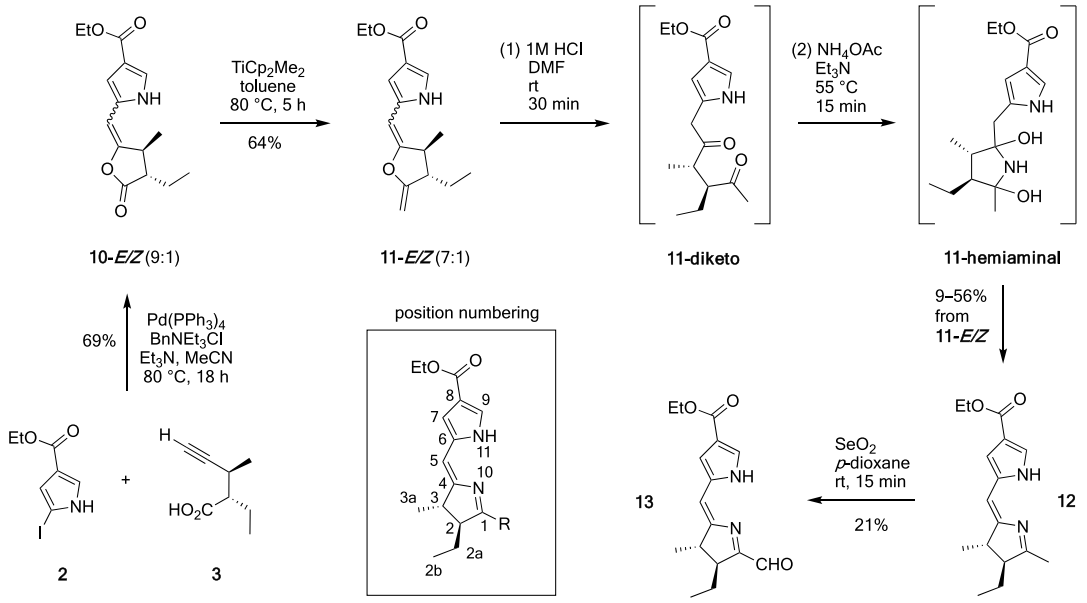
Scheme 5. Synthesis of a Model AD Half



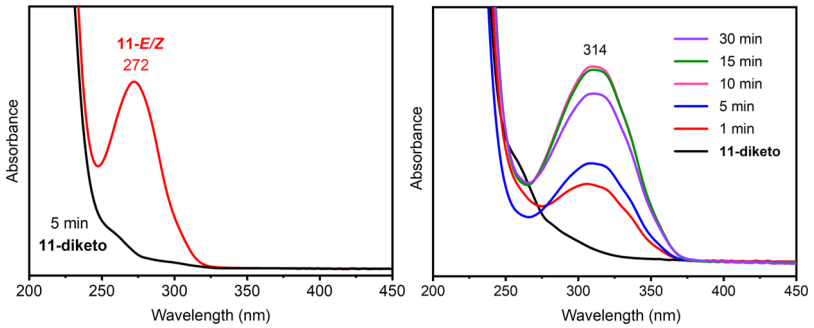


Figure 1. Absorption spectra (in acetonitrile at room temperature) for the hydrolysis of **11-*E*/*Z*** (left), followed by Paal–Knorr cyclization of **11-diketo** to form **12** (right).

H_2O_2) was required to provide[24] alkynoic acid **3**, which was obtained without stereochemical scrambling of the established chiral centers. The base LiOOH is weaker than LiOH, thus reducing the possible deprotonation at the α-carbonyl stereocenter.[25,26]

Pd-mediated coupling of **2** and **3** gave the lactone-pyrrole **10-*E*/*Z*** as a relatively inseparable mixture of isomers in 9:1 ratio (E/Z, unknown assignment) along with deiodinated pyrrole **1** as determined by ^{1}H NMR spectroscopy (Scheme 5). The product **10-*E*/*Z*** was isolated in crude form via column chromatography (estimated 87% **10-*E*/*Z***, 13% pyrrole **1**). Formation of such deiodinated pyrroles in Pd(0)-mediated reactions is commonly noted.[14,27] Preparative TLC of a small sample afforded **10-*E***. For synthetic purposes, the crude mixture of **10-*E*/*Z*** and **1** was carried forward by reaction with the Petasis reagent to give **11-*E*/*Z*** (wherein impurity **1** was removed upon chromatography). The selective reactivity of the lactone carbonyl group versus that of the carboethoxy group despite an excess of the Petasis reagent is attributed to the lessened reactivity of the latter owing to conjugation with the pyrrole ring.[28]

Compound **11-*E*/*Z*** was hydrolyzed with aqueous HCl in DMF at room temperature, followed by amination at 55 °C (Scheme 5). The two steps were carried out as a one-flask transformation with monitoring by absorption spectroscopy. The absorption of **11-*E*/*Z*** (272 nm) disappeared within 5 min because of formation of the putative intermediate diketone (**11-diketo**). Upon addition of ammonium acetate and triethylamine, the absorption (314 nm) steadily grew in over the course of 10 min because of amination yielding dihydrodipyrrin **12** (Figure 1). The conversion of **11-*E*/*Z*** to **12** can be regarded as a type of Paal–Knorr reaction, which dates to the late-19th century conversion of a 1,4-diketone to the corresponding furan. A comprehensive review[29] of the Paal–Knorr reaction shows the well-known extension beyond furans to access diverse pyrroles[30] and also thiophenes, but to our knowledge, the extension to form a pyrroline has been limited (with rare exceptions[31]) to the cases of hydrodipyrrin precursors leading to hydroporphyrins.[6] Mechanistic considerations indicate the Paal–Knorr reaction leading to pyrroles proceeds via a hemiaminal rather than an imine intermediate.[29] A similar route is expected to be followed here from **11-diketo**, via putative **11-hemiaminal**, to form the dihydrodipyrrin **12**, as is illustrated in Scheme 5.

Unlike in the classical Paal–Knorr process leading to the pyrrole, several distinct side reactions can occur on the path from **11-*E*/*Z*** to dihydrodipyrrin **12**. Such reactions include epimerization, particularly for **11-diketo** in the presence of ammonium acetate and triethylamine, and two-fold tautomerization leading to the dipyrromethane **12-DPM**. To prepare **12**

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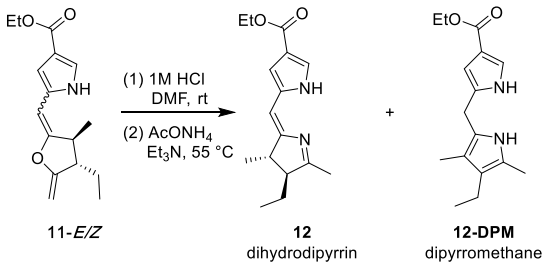
with intact stereochemical integrity, we examined a variety of procedures for amination of **11-diketo**. Key observations are as follows:

(i) Reaction of **11-diketo** at 55 °C for 4 h as reported for gem-dimethyl analogues[18] afforded dipyrromethane **12-DPM** as the predominant product (Scheme 6, top). The dipyrromethane is an isomer of **12** and originates via tautomerization, not oxidation. No such side product can occur for analogous ene−lactones with a gem-dimethyl substituent.[18,32] Ene−lactones lacking a gem-dimethyl moiety (**VII**) have been reported to give dipyrromethanes (**VIII-DPM**) in yields enhanced by a basic medium at elevated temperature (Scheme 6, bottom). On the other hand, reactions at room temperature or without base selectively gave the desired dihydrodipyrrin (**VIII**) versus the dipyrromethane (**VIII-DPM**).[14]

Scheme 6. Paal−Knorr Transformation of 11 (Top) and Data[14] for Analogues (Bottom)



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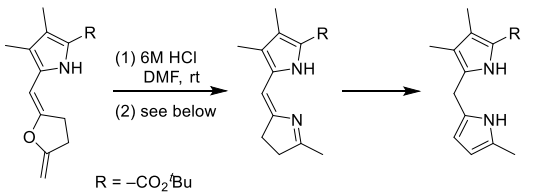

Condition for step 2:		
$AcONH_4/Et_3N$, 55 °C:	0%	59%
$AcONH_4/Et_3N$, 25 °C:	53%	< 2%
$EtCO_2NH_4$, 0 °C:	42%	trace

(ii) Upon shortening the reaction time with **11-diketo** to 15 min, dihydrodipyrrin **12** was formed preferentially; however, the isolated yield was inconsistent. We found that minor variations during the work-up and chromatography could engender tautomerization of dihydrodipyrrin **12** to dipyrromethane **12-DPM**. In an extreme case, **12** and **12-DPM** were obtained in yields of 9 and 72%, respectively, after concentration of the crude sample in a warm water bath, drying overnight under vacuum, and chromatography on silica.

(iii) Cooling the reaction mixture before quenching with cold aqueous saturated KH_2PO_4, concentrating the sample in a cooled water bath, and chromatographing on deactivated silica pretreated with Et_3N limited the unwanted tautomerization process.

In summary, the process of reaction of **11-diketo** with NH_4OAc and Et_3N at 55 °C for 15 min, rapid quenching at 0 °C, and purification by chromatography on deactivated silica gave **12** in 56% yield, which is comparable to yields recorded for unsubstituted ene−lactones.[14] While the dipyrromethane is believed to be the thermodynamic product, the conditions identified for the Paal−Knorr reaction with **11-*E*/*Z*** enabled formation of the dihydrodipyrrin **12** with limited or no competing formation of the dipyrromethane.

The Riley oxidation[33,34] of **12** with SeO_2 to form aldehyde **13** was monitored by absorption spectroscopy. The absorbance of **12** (314 nm) decreased, whereas that of aldehyde **13** (423 nm) increased over time (Figure 2). The reaction proceeded slowly in dry dioxane but more rapidly with the addition of a small quantity of water[33] (5 μL with respect to 3 mL of dioxane). Purification on a short column packed with deactivated silica (pretreated with Et_3N/hexanes) gave **13** as a dark yellow paste, which was fairly stable for weeks at −20 °C. The transformation proceeded in low yield (21%), the origin of which is unknown, but the remediation of which will be required to streamline this synthetic route.

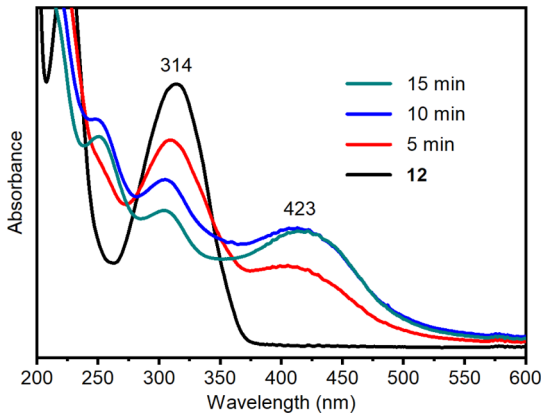

Figure 2. Absorption spectra (in acetonitrile) for the conversion of **12** to aldehyde **13** in wet dioxane.

2.2. Stereochemical Features of Dihydrodipyrrins. The integrity of the desired *trans*-dialkyl configuration was examined in dihydrodipyrrins **12** and **13**. The stereochemical relationship of H^2 and H^3 in the pyrroline ring was probed by analysis of the coupling constants in one-dimensional ^1H NMR and correlations in NOESY spectroscopy. The protons H^2 and H^3 of aldehyde **13** gave similar chemical shifts (δ 2.72−2.78 ppm) and hence a strong second-order coupling effect, complicating analysis. On the other hand, the precursor **12** gave distinct chemical shifts for the same protons and was used for the following analysis. The assignment of H^2, H^3, and two H^{2a} atoms to the observed resonances was easily achieved by COSY analysis (see the Supporting Information, Figure S1). In Figure 3 panel A, the two vicinal protons H^3 and H^2 appear as first-order multiplets, which was further analyzed as a quartet of doublets of doublets ($J_{H^3-H^{3a}}$ = 7.4 Hz, $J_{H^2-H^3}$ = 4.0 Hz, and long-range $J_{H^3-H^5}$ = 1.6 Hz) and a doublet of doublets of doublets ($J_{H^2-H^{2a}}$ = 8.6 Hz, $J_{H^2-H^{2a}}$ = 4.3 Hz, and $J_{H^2-H^3}$ = 4.0 Hz), respectively. The experimental $J_{H^2-H^3}$ value (4.0 Hz) in this case coheres with those reported for trans proton−proton coupling in other well-studied, rigid, five-membered cyclic systems (Figure 3 panel B).[35] The 2- and 3-positions of the dihydrodipyrrin correspond to the respective 17- and 18-positions of the chlorin macrocycle. The $J_{H^{17}-H^{18}}$ values of chlorophyll *a* (in acetone-d_6 or THF-d_8), pheophytin *a* (in $CDCl_3$), and methyl pheophorbide *a* (in $CDCl_3$) are 1.8, 2.0,

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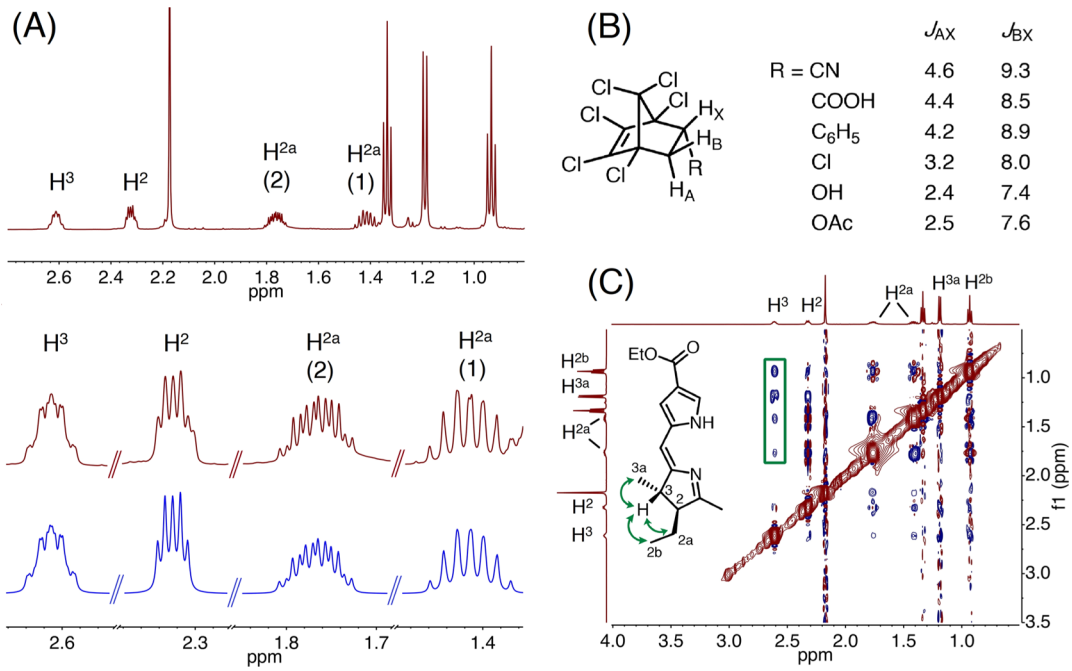
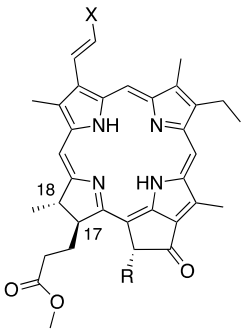


Figure 3. (A) Observed (red) and simulated (blue) ^{1}H NMR patterns of selected protons of **12**. (B) Vicinal H−H coupling constants in hexachlorobicyclo[2.2.1]heptenes.[35] (C) NOESY correlations of selected protons of **12**.

and 1.6 Hz, respectively (Chart 2).[36] On the basis of the derived experimental coupling constants, the simulated multiplets (via MestreNova software) corresponding to H^2, H^3, and two diastereotopic H^{2a} protons were constructed (Figure 3 panel A), which essentially recapitulated the experimental spectrum except for a shoulder on the high-field edge of the H^2 multiplet at 2.30 ppm.

Chart 2. Derivatives of Chlorophyll *a*



methyl pheophorbide *a*
(R = CO_2Me, X = H)

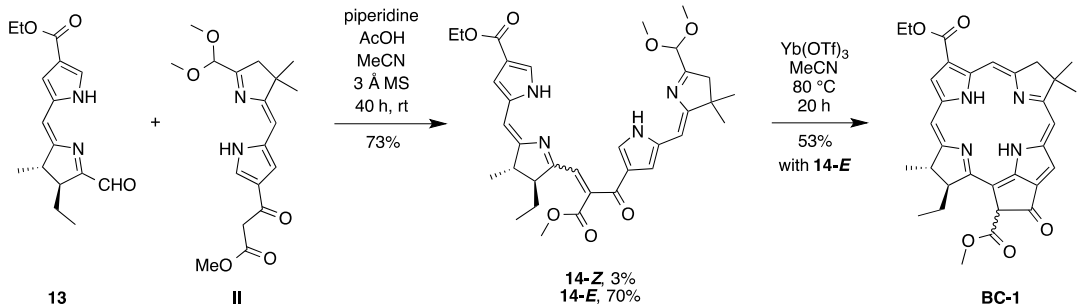
methyl pyropheophorbide *a*
(R = H, X = H)

IX (R = H, X = CHCHCO$_2$Me)

Finally, the NOESY spectrum of **12** shows a strong correlation of H^3 with the ethyl group bearing two H^{2a} and three H^{2b} atoms, in addition to the inevitable correlation of H^3 with the vicinal methyl group (Figure 3, panel C). This observation implies that H^3 and the adjacent ethyl substituent must be on the same side of the five-membered ring. Analogous NOE correlations were observed for H^{18} and the methylene groups in the ester chain at C^{17} in the NOESY spectrum of a 3-substituted analogue of methyl pyropheophorbide *a* (**IX**, Chart 2).[37] In summary, all of the spectroscopic results are consistent with a *trans*-dialkyl configuration in dihydrodipyrrin **12**.

2.3. Macrocycle Formation. We next examined the reaction of dihydrodipyrrin−carboxaldehyde **13** in the formation of a bacteriochlorophyll model compound (Scheme 7). Thus, the Knoevenagel reaction[38−40] of **13** and **II** proceeded under mild conditions at room temperature to furnish two separately isolated enone isomers in a total yield of 73% (3 and 70%). The E or Z configuration of each enone (**14**) could not be assigned by NOESY spectroscopy (Figures S2−S5); however, in each isomer, the multiplicities of protons at or near the key stereocenters in the pyrroline ring resembled those found in the precursor **12**, indicating successful retention

Scheme 7. Bacteriochlorin Macrocycle Formation



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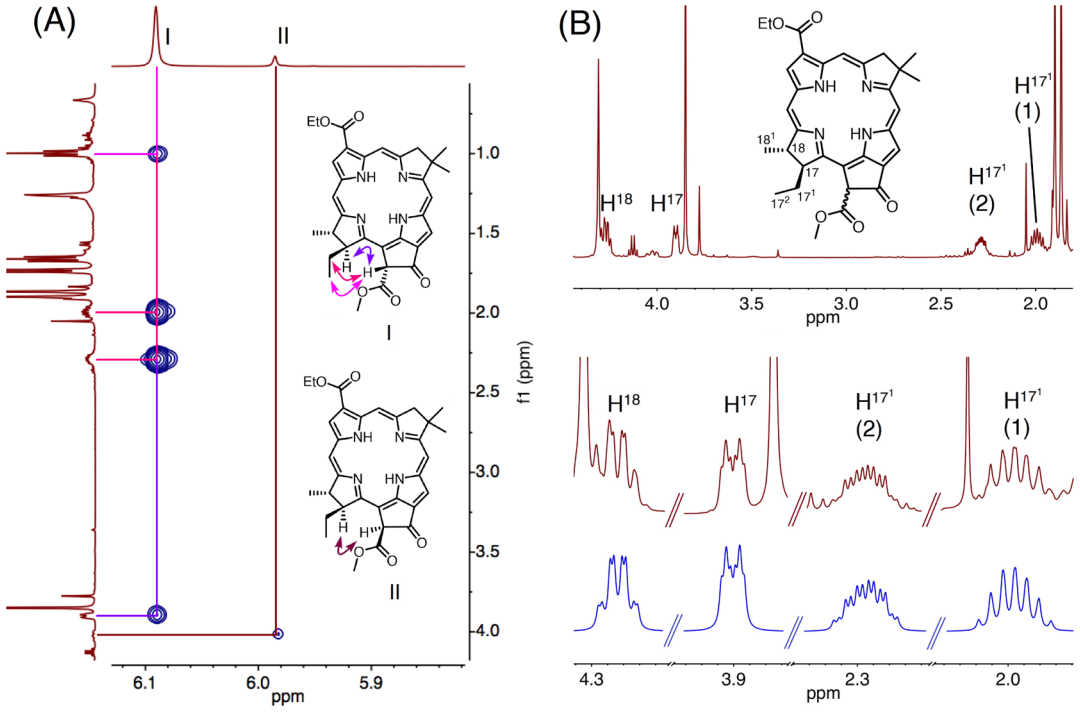

Figure 4. (A) Selected NOESY correlations supporting the assignments and ratio of two epimeric forms of bacteriochlorin **BC-1**. (B) Observed (red) and simulated (blue) ^{1}H NMR patterns of selected protons of **BC-1**. The vicinal H^{17}–H^{18} coupling constant was 2.5 Hz, consistent with the trans configuration.

of the preinstalled *trans*-dialkyl stereochemistry. A single-crystal X-ray structure determination of the major isomer (from acetonitrile/diethyl ether) was carried out (Figures S6–S11). The structure clearly revealed the E configuration of the enone and the *trans*-dialkyl (17-*S*, 18-*S*) configuration in the pyrroline ring, despite considerable positional disorder at several sites such as the carboethoxy group (which can assume multiple orientations). To our knowledge, only one single-crystal X-ray structure has been reported for a Knoevenagel enone derived from a heteroaromatic β-keto ester [Ar-C(O)CH_2CO_2Me], which was also the E isomer.[41] The E and Z isomers of related enones are known to interconvert under the conditions of the Nazarov reaction.[41−45] Here, we employed the major Knoevenagel enone (**14-*E***, previously referred to as a hydrobilin)[7] in the subsequent one-flask reaction to form the desired bacteriochlorin **BC-1**.

The reaction of enone **14-*E*** to form the bacteriochlorin was carried out under the conditions employed previously,[7] namely, reaction of the enone (~0.2 mM) in the presence of 10 equiv of Yb$(OTf)_3$ in acetonitrile at 80 °C for 20 h under argon. The overall reaction entails Nazarov cyclization[46,47] to form the isocyclic ring, S_EAr to form the macrocycle, and elimination of MeOH to achieve aromaticity; the order of the reaction steps is not known. The reaction was carried out with 24 mg of **14-*E***, affording a purple reaction mixture after 20 h. Standard work-up followed by purification by preparative TLC gave the bacteriochlorophyll model compound **BC-1** in 53% yield.

2.4. Macrocycle Characterization. The product **BC-1** exhibited the expected protonated molecular ion peak $[M + H]^+$ at m/z 555.2591 upon LC-HRMS analysis with an identical isotopic distribution pattern to theoretical calculation for $C_{32}H_{35}N_4O_5$ at m/z 555.2602. The ^{1}H NMR spectrum revealed peaks due to the distinct NH protons in the upfield region (−0.67 and 0.66 ppm), while signals due to the two methine protons in ring D and two methylene protons in ring B were downfield (3.89–4.33 ppm) with respect to those in the precursor **14-*E*** (2.47–2.62 ppm) due to the ring current of the aromatic macrocycle. The vicinal H^{17}–H^{18} coupling constant was 2.5 Hz, in accord with the trans configuration[35] and consistent with the spectra for chlorophyll and derivatives.[36] The existence of two epimers, which is well documented in natural chlorophylls and bacteriochlorophylls,[48−50] was apparent in the ^{1}H NMR spectrum: two peaks (6.09 and 5.99 ppm) with a 7:1 ratio were observed for the 13^2 proton, reflecting the major and minor epimers. NOESY spectroscopy led to assignment of the major epimer with trans–trans configuration with respect to three methine protons in rings D and E (i.e., 17–18 and 18–13^2) and hence the trans–cis configuration for the minor epimer (Figure 4). Such epimers are known to interconvert via an intermediate enol of the β-ketoester with rate that depends strongly on the structure and electronic properties of the macrocycle,[48−50] as well as the nature and polarity of the medium.[48] The reported ratio of 13^2-epimers for bacteriochlorophyll *a* ranges from 7:3[51] to 3:1[49] to >10:1,[52] which encompass the observed ratio of 7:1 for **BC-1**. The rates for bacteriopheophytins are apparently not known, but pheophytins generally epimerize 20–40 times faster than chlorophylls.[49] Fundamental studies to explore this process, including the effects of conditions and coordinated metals, are now possible but are beyond the scope of the present paper. The more pressing issue here concerned the stereochemical integrity of the alkyl groups at positions 17 and 18. The NMR results indicate that the two alkyl groups in the pyrroline ring are present in the expected trans stereochemistry.

Bacteriochlorin **BC-1** in toluene at room temperature exhibits absorption bands (at 356, 379, 536, and 749 nm)

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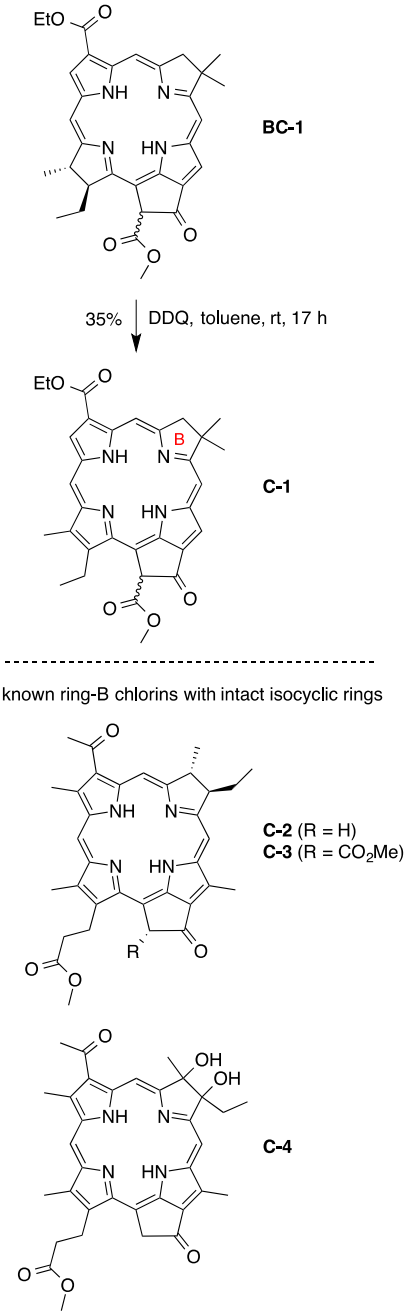
characteristic of a bacteriopheophytin π system.[3] The long-wavelength (Q_y) band (749 nm) exhibited intensity versus the near-UV (B) band [I_{Q_y}/I_B] of 0.94, a full width at half-maximum of 22 nm, and a molar absorption coefficient (ε) of 72,100 M^{-1} cm^{-1} (Figure S12). The fluorescence properties, also in toluene at room temperature, include λ_{em} at 753 nm (Stokes shift of 4 nm) and fluorescence quantum yield (Φ_f) of 0.17. The spectral properties can be compared with those of bacteriopheophytin *a*, the free base ligand of bacteriochlorophyll *a* (Chart 1), which exhibits λ_{abs} = 748 nm, ε = 67,600 M^{-1} cm^{-1},[3,4] [I_{Q_y}/I_B] = 0.64, λ_{em} at 765 nm, and Φ_f of 0.126 (in diethyl ether)[53] or 0.094 (in acetone/methanol 7:3).[54,55]

2.5. Macrocycle Dehydrogenation. Treatment of hydroporphyrins with the high-potential quinone DDQ[56] is well established,[57] and with native bacteriochlorophylls and derivatives causes dehydrogenation of pyrroline ring B preferentially over ring D.[10,58−60] Here, the *trans*-dialkyl-substituted ring D is susceptible to dehydrogenation, whereas the gem-dimethyl-substituted ring B is not. The reaction of bacteriochlorin **BC-1** with DDQ at room temperature afforded the chlorin **C-1** in 35% yield (Scheme 8, top). The **C-1** contains a saturated ring B, in contrast to native chlorophylls, wherein ring D is saturated. Although obtained in tiny quantity (1.4 mg), the chlorin **C-1** was characterized by HRMS, ^1H and ^{13}C NMR spectroscopy, and absorption and fluorescence spectroscopy (Figure 5, bottom panel), including fluorescence quantum yield determination (Φ_f = 0.41). Although **BC-1** is composed of a mixture of diastereomers (epimers at the 13^2-position), **C-1** is expected as a pair of enantiomers. Relatively few ring-B chlorins containing an intact isocyclic ring are known (Scheme 8, bottom), all of which have been prepared by semisynthesis beginning with bacteriochlorophyll *a* (by selective dehydrogenation with $FeCl_3$)[59,60] or a porphyrin derived from chlorophyll *a* (by selective vicinal hydroxylation with OsO_4).[61] The known ring-B chlorins exhibit the following red-region absorption properties: **C-2**, λ = 691 nm, ε_{691nm} = 4.31×10^4 M^{-1} cm^{-1} (CH_2Cl_2);[59] **C-3**, λ = 682 nm, ε_{682nm} = 3.65×10^4 M^{-1} cm^{-1} (CH_2Cl_2/THF);[60] and **C-4**, λ = 683 nm (CH_2Cl_2).[61]

The availability of **C-1** provided a valuable marker to address one of the key questions posed at the outset of this research, namely, does adventitious dehydrogenation occur during the course of macrocycle formation leading to a chlorin byproduct accompanying the bacteriochlorin **BC-1**? The chlorin **C-1** was used as an authentic standard to search for chlorin contamination via fluorescence spectroscopy. A series of mock samples was prepared by mixing **BC-1** and **C-1**, with the latter in serially diluted quantities (i.e., $1:10^{-1}$, $1:10^{-2}$, $1:10^{-3}$, and $1:10^{-4}$), and fluorescence spectra (650−800 nm) were collected upon illumination at 406, 420, 430, 440, and 553 nm; the wavelengths were chosen where the chlorin absorbs strongly but the bacteriochlorin absorbs weakly (Figures S13 and S14). Excitation at 406 nm proved superior, where the limit of detection of the chlorin was found to be 0.1% of the bacteriochlorin (2 μM), as shown in Figure 6. With these calibrants in hand, the reaction of **14-*E*** was repeated to form bacteriochlorin **BC-1**. Following reaction for 21 h at 80 °C, a sample from the crude reaction mixture—without any workup other than dilution in toluene—gave the absorption spectrum and the fluorescence spectrum (λ_{ex} 406 nm) shown in Figure 6. Comparison of the fluorescence spectra shows that the amount of chlorin present corresponds to 0.014 times that of the bacteriochlorin (i.e., 1:71 ratio). The presence of such a small quantity of impurity is likely undetectable by most methods but is observable by careful fluorescence spectroscopy; indeed, there is no chlorin detectable by absorption spectroscopy at 692 nm, a wavelength where the chlorin absorbs strongly compared with that of the bacteriochlorin.

Scheme 8. Ring-B Chlorins: Formation Here (Top) and Known Previously (Bottom)



3. OUTLOOK

The work described herein is part of a program focused on developing a *de novo* synthesis of native bacteriochlorophylls. The installation of *trans*-dialkyl groups in the two pyrroline rings is essential to such a synthesis. The strategy we have employed installs the groups at a very early stage of the synthesis, in an acyclic precursor to the pyrroline ring, and has

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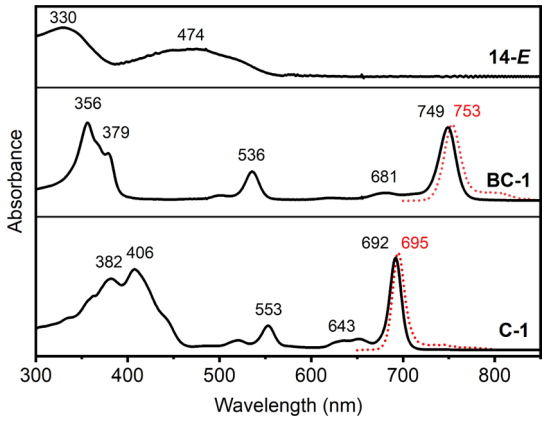

Figure 5. Spectra in toluene at room temperature. Absorption of **14-*E*** (top); and absorption and fluorescence (red dotted lines) of bacteriochlorin **BC-1** (middle) and chlorin **C-1** (bottom).

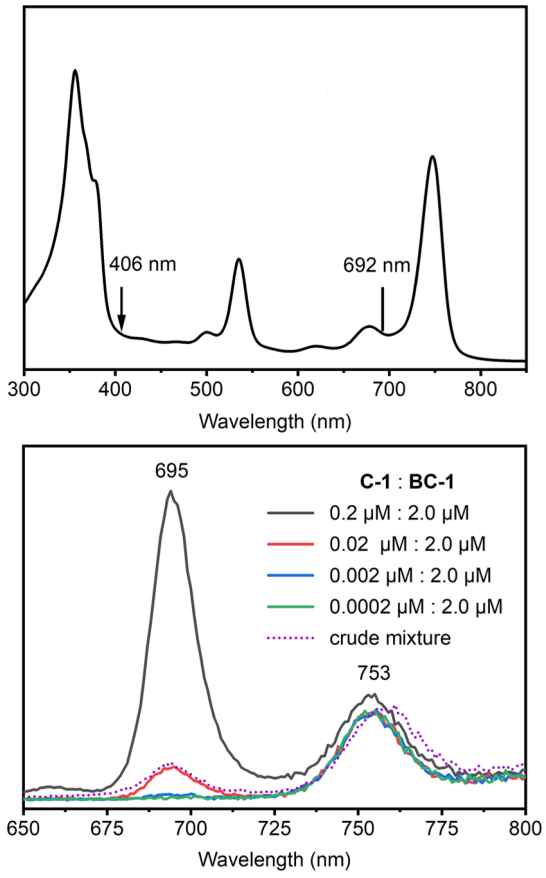

Figure 6. Absorption spectrum (toluene, room temperature) of the final (21 h) crude mixture upon reaction of **14-*E*** (top). Fluorescence spectra upon excitation at 406 nm of (i) standard samples containing **BC-1** and serially diluted **C-1** (solid lines) overlaid with (ii) the final crude reaction mixture of **14-*E*** (dotted line).

been demonstrated for methyl and ethyl substituents in ring D. The *trans*-dialkyl configuration is achieved by Nicholas reaction to afford a 2,3-disubstituted pent-4-ynoic acid. Subsequent reactions (conditions) include TMS cleavage (LiOOH, 0 °C—rt, 4 h), Pd-mediated coupling (Et_3N in MeCN, 80 °C, 18 h), Petasis olefination (toluene, 80 °C, 5 h), aqueous hydrolysis (1 M HCl in DMF, rt, 30 min), Paal–Knorr-like reaction (NH_4OAc and Et_3N, 55 °C, 15 min), Riley oxidation (SeO_2, rt, 15 min), Knoevenagel condensation (piperidine/AcOH in MeCN, rt, 40 h), and a one-flask domino process of Nazarov cyclization, S_EAr, and elimination of methanol ($Yb(OTf)_3$ in MeCN, 80 °C, 20 h). The *trans*-dialkyl configuration is retained to a high degree over the course of the synthesis. The Riley oxidation of 1-methyldihydrodipyrrin **12** affords dihydrodipyrrin–carboxaldehyde **13** in a yield that is less than half that of many gem-dimethyl-substituted dihydrodipyrrins.[34] Aside from such reactions that require amelioration to achieve high yields throughout, three possibly precarious aspects of the synthesis bear on the integrity of the *trans*-dialkyl configuration in ring D:

(i) Epimerization of the dihydrodipyrrin 2-position (equivalent to the bacteriochlorin 17-position) could occur by site-localized inversion and also by imine–enamine tautomerization ($\Delta^{1,10} \rightarrow \Delta^{1,2}$). See Schemes 2 and 5 for position numbering. Such processes can proceed in all structures containing a dihydrodipyrrin unit: **12**, **13**, and **14**.

(ii) Imine–enamine tautomerization ($\Delta^{1,10} \rightarrow \Delta^{1,2}$) followed by alkene migration (dihydrodipyrrin $\Delta^{4,5} \rightarrow \Delta^{3,4}$) together would convert the 2,3-dihydrodipyrrin to the dipyrromethane (see Scheme 6 for the conversion). The dipyrromethane is regarded as the thermodynamic product. Such isomerization would not only cause loss of the *trans*-dialkyl stereochemistry but would also yield a chlorin rather than a bacteriochlorin.

(iii) Adventitious dehydrogenation of the dihydrodipyrrin 2,3-positions would afford the dipyrrin; similar dehydrogenation of the bacteriochlorin 17,18-positions would afford the chlorin. Dehydrogenation of bacteriochlorins (that lack stabilizing structural features such as gem-dimethyl groups) to form chlorins on routine handling in air is a well-established phenomenon.[10−13]

An explicit search in the crude reaction mixture for the chlorin byproduct, using an authentic sample (**C-1**), revealed the presence of the latter at 1.4% relative to that of bacteriochlorin **BC-1**. The presence of the chlorin could not be observed by the routine method of absorption spectroscopy but was detectable by the sensitive technique of fluorescence spectroscopy. While the quantity of chlorin byproduct is already quite low, the fluorescence assay may prove useful in further studies of reaction conditions that further decrease the yield of chlorin.

The reaction conditions for conversion of **14-*E*** to **BC-1** have been developed by extension of methods reported for the Nazarov reaction of pyrrole-substituted enones[43] and the macrocyclization of dihydrodipyrrin–acetals leading to bacteriochlorins.[17,62] While Nazarov cyclization is an essential step in the overall reaction process, the conditions here are 1000-fold more dilute (~0.2 mM) and use a greater relative amount of acid (10 equiv) versus the typical Nazarov cyclization of divinyl ketones (0.1–0.3 M substrate, catalytic quantities of acid).[43,44,63] The Nazarov cyclization entails (conrotatory) electrocyclization of a pentadienyl cationic intermediate. While E–Z isomerization is facile for divinyl ketones and heteroaryl vinyl ketones upon exposure to the acidic conditions of the Nazarov reaction,[41−45] we employed exclusively the major isomer (**14-*E***) for studies herein. The electrocyclization yields two stereocenters, here at the 15- and 13^2-positions, of which the former is lost upon subsequent aromatization. The origin of the 7:1 ratio of epimeric 13^2-CO_2Me substituents is

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attributed[50] to the stereochemistry and size of the adjacent 17-substituent, which is an ethyl group in **BC-1** versus a phytyl propionate in **BChl *a***. Further studies of diverse conditions (perhaps including the effects of chiral catalysts) are required to understand the reaction of Knoevenagel enones (**14-*E*** and analogues) as well as the kinetic stability of the observed epimeric ratio at the 13^2-position.

At present, the only means to access native bacteriochlorophylls and analogues is via semisynthesis[12,64,65] of biosynthesized native pigments. The biosynthesis of bacteriochlorophylls relies on first forming a porphyrin (protoporphyrin IX, the ligand of heme), which is then subjected to two stereoselective reduction processes to form the *trans*-dialkyl configuration in rings D and B.[66] The enzymes for the two processes are protochlorophyllide reductase and chlorin reductase, respectively. The availability of (nonenzymic) asymmetric catalysts for stereoselective reduction of a porphyrin in rings B and D would provide an attractive complementary route, as the synthetic chemistry of porphyrins is more advanced than that of bacteriochlorins. In the absence of such catalysts, the introduction of stereochemically defined groups in precursors to the pyrroline rings seems the most suitable approach. The bacteriochlorophyll model compound (**BC-1**) prepared herein is unusual, given the presence of one gem-dimethyl group and one *trans*-dialkyl group in pyrroline rings B and D, respectively, yet has the virtue of enabling focus on the integrity of only one site of vulnerability, the *trans*-dialkyl-substituted ring D. The sharp focus validates a synthetic approach that may prove viable for gaining access to native bacteriochlorophylls and analogues, which to date have fallen outside the scope of chemical synthesis.

4. EXPERIMENTAL SECTION

4.1. General Methods. All chemicals from commercial sources were used as received. Silica gel for column chromatography (230−400 mesh, 60 Å) was obtained from Silicycle Inc. Preparative TLC plates (1000 μm thickness) were purchased from Analtech. THF for use in inert−atmosphere reactions was freshly distilled from sodium/benzophenone ketyl. DMF and acetonitrile used as reaction media were stirred overnight in the presence of 3 Å molecular sieves, followed by distillation. Dichloromethane employed as a solvent for reactions was dried overnight over 3 Å molecular sieves. All other solvents were reagent grade from commercial suppliers.

Compounds **1**,[17] **2**,[18] and **7**[22] have been prepared and isolated in 5.12, 2.56, and 2.85 g quantities, respectively, and fully characterized. Compound **5**[21] has been prepared in 2.0 g quantity according to a synthetic route that is different from the more streamlined route reported herein. Compound **3** is listed in ACS Scifinder but without any reference. Dihydrodipyrrin **II**[7] is known.

4.1.1. Ethyl 1H-Pyrrole-3-carboxylate (1). Following a literature procedure[17] at 1.5-fold larger scale, a sample of NaH (60% in mineral oil, 6.68 g, 167 mmol) was washed with hexanes (3 × 25 mL), suspended in anhydrous Et_2O (270 mL), and stirred at room temperature. A solution of ethyl acrylate (10.0 g, 100 mmol) and TosMIC (28.2 g, 144 mmol) in the anhydrous co-solvent of Et_2O (400 mL) and DMSO (200 mL) was added slowly to the ethereal suspension of NaH. The reaction mixture was stirred overnight at room temperature. Water (400 mL) was added slowly, followed by extraction with ethyl acetate (4 × 200 mL). The combined organic extract was dried (Na_2SO_4), concentrated, and chromatographed [silica, hexanes/ethyl acetate (2:1), 6.5 cm × 40 cm] to afford a yellow oil (9.15, 66%): silica TLC R_f 0.33 in hexanes/ethyl acetate (2:1); ^{1}H NMR (500 MHz, $CDCl_3$): δ 1.34 (t, J = 7.1 Hz, 3H), 4.28 (q, J = 7.1 Hz, 2H), 6.66−6.67 (m, 1H), 6.75−6.76 (m, 1H), 7.43−7.44 (m, 1H), 8.51 (br, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 14.5, 59.8, 109.7, 116.6, 118.8, 123.5, 165.3; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_7H_{10}NO_2$, 140.0706; found, 141.0708.

4.1.2. Ethyl 5-Iodo-1H-pyrrole-3-carboxylate (2). Following a literature procedure[18] at a 5-fold larger scale, a solution of **1** (7.24 g, 52 mmol) in DMF (130 mL) at 0 °C was treated with NIS (11.70 g, 52 mmol) in portions over 15 min. The reaction mixture was continuously stirred at 0 °C for 1 h, followed by dilution with water (100 mL). The resulting solution was extracted with diethyl ether (4 × 150 mL). The combined organic extract was dried (Na_2SO_4), concentrated, and chromatographed [silica, hexanes/ethyl acetate (2:1), 6.5 cm × 40 cm] to afford a slightly yellow solid (8.98 g, 65%): silica TLC R_f 0.38 in hexanes/ethyl acetate (2:1); mp 64−66 °C; ^{1}H NMR (500 MHz, $CDCl_3$): δ 1.34 (t, J = 7.1 Hz, 3H), 4.28 (q, J = 7.1 Hz, 2H), 6.78 (dd, J = 2.5 and 1.7 Hz, 1H), 7.42 (dd, J = 2.9 and 1.6 Hz, 1H), 8.93 (br, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 14.4, 60.2, 63.5, 119.1, 119.2, 127.3, 163.9; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_7H_9INO_2$, 265.9673; found, 265.9666.

4.1.3. (2S,3S)-2-Ethyl-3-methylpent-4-ynoic Acid (3). Following a general procedure,[24] a solution of **9** (15.41 g, 47.6 mmol) in THF/water (3:1, 600 mL) at 0 °C was treated with aqueous 0.5 M LiOH (286 mL, 143 mmol) and 30% H_2O_2 (44 mL, 380 mmol). The resulting solution was first stirred at 0 °C for 1 h and subsequently at room temperature for 3 h. The completion of the transformation was checked by TLC, whereupon further stirring with addition of a further quantity of 30% H_2O_2 was done if needed. The reaction mixture was then cooled to 0 °C, quenched by the slow addition of 1.5 N Na_2SO_3 until a negative test with KI-starch paper was obtained, stirred at room temperature for 30 min, and then concentrated under reduced pressure to remove THF. The resulting solution was treated with saturated aqueous $NaHCO_3$ (300 mL) and extracted with CH_2Cl_2 (3 × 200 mL). The organic extract was concentrated under reduced pressure to recover (*R*)-4-isopropyloxazolidin-2-one (**6**). The aqueous layer was cooled in ice followed by the dropwise addition of conc. HCl until pH = 2. The acidic solution was extracted with ethyl acetate (5 × 150 mL), and the combined organic extract was concentrated under reduced pressure to afford a clear, colorless oil (6.18 g, 93%): ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.98 (t, J = 7.4 Hz, 3H), 1.25 (d, J = 7.1 Hz, 3H), 1.69−1.74 (m, 2H), 2.11 (d, J = 2.4 Hz, 1H), 2.40 (q, J = 7.9 Hz, 1H), 2.79−2.85 (m, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 11.9, 17.9, 22.2, 27.7, 52.0, 69.8, 85.9, 179.8; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_8H_{13}O_2$, 141.0910; found, 141.0909.

4.1.4. (±) Hexacarbonyldicobalt Complex (5). A solution of **4** (14.67 mL, 100 mmol) in anhydrous THF (200 mL) was cooled to −78 °C under argon and treated with 1.6 M MeLi (1.6 M in Et_2O, 62.5 mL, 100 mmol). The reaction mixture was stirred for 15 min at −78 °C, followed by addition of dimethyl sulfate (9.48 mL, 100 mmol), warming to room temperature, and stirring at room temperature for 28 h. Then, octacarbonyldicobalt 90% (37.99 g, 100 mmol) was added batchwise to the reaction mixture under vigorous stirring. A brisk evolution of (presumed CO) gas was noticed over the course of addition of $Co_2(CO)_8$. The mixture was stirred at room temperature for 12 h. The resulting dark brown solution was concentrated and chromatographed [silica, hexanes/ethyl acetate (50:1), 6.5 cm × 28 cm] to afford a dark red paste (35.11 g, 79%): silica TLC R_f 0.30 in hexanes/ethyl acetate (50:1); ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.31 (s, 9H), 1.48 (d, J = 6.2 Hz, 3H), 3.48 (s, 3H), 4.47 (q, J = 6.2 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 0.8, 22.8, 57.2, 77.8, 113.3, 200.4 (br).

4.1.5. (R)-3-Butyryl-4-isopropyloxazolidin-2-one (7). Following a general procedure[22] at a 5-fold larger scale, a solution of (*R*)-4-isopropyloxazolidin-2-one (**6**, 10.00 g, 77 mmol) in anhydrous THF (200 mL) at −78 °C under argon was treated slowly with *n*-BuLi (1.6 M in hexanes, 48 mL, 77 mmol). The resulting reaction mixture was vigorously stirred at −78 °C for 15 min. The reaction mixture was removed from the cooling bath and treated dropwise with butyryl chloride (8.00 mL, 77 mmol) with vigorous stirring. The resulting clear solution was stirred for 24 h at room temperature before treatment with saturated aqueous NH_4Cl (200 mL). The ethereal layer was collected. The aqueous layer was extracted with CH_2Cl_2 (3 × 100 mL). The combined organic extract was washed with water,

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dried (Na_2SO_4), concentrated, and chromatographed [silica, hexanes/ethyl acetate (2:1), 6.5 cm × 40 cm] to give a clear, colorless oil (13.90 g, 91%): silica TLC R_f 0.45 in hexanes/ethyl acetate (2:1); ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.88 (d, J = 6.9 Hz, 3H), 0.92 (d, J = 7.0 Hz, 3H), 0.99 (t, J = 7.4 Hz, 3H), 1.64−1.75 (m, 2H), 2.33−2.42 (m, 1H), 2.81−2.87 (m, 1H), 2.94−3.00 (m, 1H), 4.20 (dd, J = 9.0 and 3.1 Hz, 1H), 4.27 (t, J = 8.7 Hz, 1H), 4.44 (dt, J = 8.3 and 3.2 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 13.7, 14.7, 17.9, 18.0, 28.4, 37.4, 58.4, 63.3, 154.1, 173.2; HRMS (ESI-TOF) m/z: [M + H]$^+$ calcd for $C_{10}H_{18}NO_3$, 200.1281; found, 200.1274.

*4.1.6. Nicholas Adduct (**8**) and Its Decomplexed Derivative (**9**).* Following a general procedure[23] with modification, a solution of **7** (12.75 g, 64 mmol) in anhydrous CH_2Cl_2 (200 mL) at 0 °C with vigorous stirring under argon was treated dropwise with Bu_2BOTf (1 M in CH_2Cl_2, 100 mL, 100 mmol) and then dropwise with diisopropylethylamine (11.15 mL, 64 mmol). After stirring at 0 °C for 15 min, the mixture was cooled to −78 °C. A solution of hexacarbonyldicobalt complex **5** (28.30 g, 64 mmol) in anhydrous CH_2Cl_2 (170 mL) was added dropwise via a syringe to the *in situ* generated boronate ester. After the addition, the reaction mixture was allowed to warm to 0 °C, then left at 0 °C for 20 min, then allowed to warm to room temperature, and then left at room temperature for 20 min. The reaction was quenched by the addition of a phosphate buffer solution at pH 7 (0.1 M, made from K_2HPO_4 and KH_2PO_4, 400 mL). The aqueous layer was extracted with CH_2Cl_2 (3 × 150 mL). The combined organic extract was washed with water, dried (Na_2SO_4), concentrated, and chromatographed [silica, hexanes/ethyl acetate (5:1), 6.5 cm × 40 cm] to yield the Nicholas adduct **8** as a dark red paste (29.95 g, 77%): silica TLC R_f 0.35 in hexanes/ethyl acetate (5:1); ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.34 (s, 9H), 0.85−0.89 (m, 6H), 0.94 (d, J = 7.0 Hz, 3H), 1.21 (d, J = 7.1 Hz, 3H), 1.54−1.60 (m, 1H), 1.95−2.05 (m, 1H), 2.29−2.36 (m, 1H), 3.43 (qd, J = 7.1 and 1.9 Hz, 1H), 4.00 (d, J = 11.9 Hz, 1H), 4.21 (dd, J = 9.1 and 3.0 Hz, 1H), 4.30 (t, J = 8.8 Hz, 1H), 4.55 (dt, J = 8.5 and 4.2 Hz, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 1.0, 11.9, 14.4, 17.2, 17.8, 18.2, 28.5, 41.5, 50.8, 58.1, 63.0, 79.3, 115.0, 153.2, 173.7, 200.2 (br), 200.6 (br); HRMS (ESI-TOF) m/z: [M + H]$^+$ calcd for $C_{23}H_{30}Co_2NO_9Si$, 610.0348; found, 610.0345.

A solution of the entire sample of **8** (29.95 g, 49 mmol) in acetone (400 mL) was treated in portions with CAN (~115 g total) under vigorous stirring until the brisk evolution of (presumed CO) gas as well as the dark brown color of solution was no longer detected. The resulting solution was diluted with water, concentrated under reduced pressure, and then extracted with CH_2Cl_2 (5 × 150 mL). The combined organic extract was washed with water, dried (Na_2SO_4), and concentrated to afford a clear, colorless oil (15.41 g, 97%): ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.11 (s, 9H), 0.85−0.94 (m, 9H), 1.18 (d, J = 7.2 Hz, 3H), 1.71 (quint, J = 7.2 Hz, 2H), 2.35−2.44 (m, 1H), 2.92 (quint, J = 7.2 Hz, 1H), 3.94 (q, J = 6.9 Hz, 1H), 4.20 (d, J = 9.0 Hz, 1H), 4.26 (t, J = 8.7 Hz, 1H), 4.49−4.50 (m, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 0.1, 11.0, 15.0, 17.5, 18.0, 21.8, 28.4, 28.6, 48.4, 58.6, 63.1, 85.2, 109.4, 153.6, 174.3; HRMS (ESI-TOF) m/z: [M + H]$^+$ calcd for $C_{17}H_{30}NO_3Si$, 324.1990; found, 324.1983.

*4.1.7. Ethyl 5-(((3S,4S)-4-Ethyl-3-methyl-5-oxodihydrofuran-2(3H)-ylidene)methyl)-1H-pyrrole-3-carboxylate (**10-E/Z** and **10-E**).* Following a general procedure[18] with modification, a 500 mL Schlenk flask was charged with **2** (11.66 g, 44 mmol), **3** (6.17 g, 44 mmol), $BnNEt_3Cl$ (12.22 g, 44 mmol), and Et_3N (52.5 mL) in 250 mL of acetonitrile. The mixture was degassed by three cycles of freeze−pump−thaw prior to addition of $Pd(PPh_3)_4$ (2.54 g, 2.20 mmol). The resulting mixture was subjected to one more degassing cycle and then stirred at 80 °C in an oil bath for 18 h. The mixture was allowed to cool to room temperature, diluted by addition of water (300 mL), and extracted with CH_2Cl_2 (4 × 200 mL). The combined organic extract was dried (Na_2SO_4), concentrated, and chromatographed [silica, hexanes/ethyl acetate (2:1), 6.5 cm × 40 cm, R_f 0.28 in hexanes/ethyl acetate (2:1)] to deliver a brown paste (9.68 g), which was found to comprise a mixture of two (E, Z) isomers in 9:1 ratio along with deiodinated compound **1**. This mixture, referred to as the postcolumn chromatographed sample of **10-E/Z**, was used directly in the next synthetic transformation. Analysis of the 9.68 g of isolated sample by ^{1}H NMR spectroscopy showed the presence of **10-E/Z** at 87% purity (with **1** accounting for the remaining 13%), implying a 69% yield in total for both E and Z isomeric forms. Further purification of a small sample by preparative TLC [20 cm × 20 cm, 1000 μm thickness, silica, hexanes/ethyl acetate (5:1)] gave the E-isomer of the title compound: ^{1}H NMR (500 MHz, $CDCl_3$): δ 1.03 (t, J = 7.4 Hz, 3H), 1.33−1.37 (m, 6H), 1.64−1.72 (m, 1H), 1.75−1.83 (m, 1H), 2.35−2.39 (m, 1H), 3.17 (q, J = 7.0 Hz, 1H), 4.30 (q, J = 7.1 Hz, 2H), 6.05 (s, 1H), 6.52 (s, 1H), 7.38 (s, 1H), 8.60 (br, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$) 11.2, 14.5, 18.6, 25.0, 37.8, 50.2, 60.0, 96.9, 107.0, 118.0, 123.1, 126.6, 155.4, 164.8, 176.3; HRMS (ESI-TOF) m/z: [M + H]$^+$ calcd for $C_{15}H_{20}NO_4$, 278.1387; found, 218.1385.

*4.1.8. Ethyl 5-(((3S,4S)-4-Ethyl-3-methyl-5-methylenedihydrofuran-2(3H)-ylidene)methyl)-1H-pyrrole-3-carboxylate (**11-E/Z**).* Preparation of the Petasis reagent and application in the olefination of **10-E/Z** were conducted according to a standard procedure.[18] A solution of Cp_2TiCl_2 (33.12 g, 133 mmol) in anhydrous toluene (350 mL) at 0 °C under argon was treated dropwise with MeLi (1.6 M in Et_2O, 182 mL, 291 mmol). The reaction mixture was stirred at 0 °C for 1 h, and then, saturated aqueous NH_4Cl (400 mL) was added. The organic layer was washed with water and brine, dried (Na_2SO_4), and filtered. The filtrate (containing the Petasis reagent) was treated with the above postcolumn chromatographed sample of **10-E/Z** (8.93 g, 87% pure, 28 mmol) and additional Cp_2TiCl_2 (418 mg). The reaction mixture was heated at 80 °C in an oil bath for 5 h in the dark under argon. Then, the resulting solution was allowed to cool to room temperature, followed by the addition of MeOH (33 mL), $NaHCO_3$ (1.39 g), and water (333 μL). The resulting mixture was stirred at 40 °C in an oil bath for 12 h, diluted with CH_2Cl_2, and then filtered through Celite. The filtrate (clear yellow) was concentrated and chromatographed [silica, hexanes/ethyl acetate (2:1) in the presence of 1% Et_3N, 6.5 cm × 40 cm, R_f 0.38 in hexanes/ethyl acetate (2:1)] to yield an orange solid. Characterization by ^{1}H NMR spectroscopy and LC-HRMS indicated the presence of the E and Z isomers in 7:1 ratio (4.95 g, 64%): mp 70−72 °C; the following NMR listing is for the *E* isomer only: ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.95 (t, J = 7.4 Hz, 3H), 1.24 (d, J = 7.1 Hz, 3H), 1.34 (t, J = 7.1 Hz, 3H), 1.48 (quintd, J = 7.3 and 0.9 Hz, 2H), 2.36 (t, J = 7.1 Hz, 1H), 2.96 (q, J = 7.1 Hz, 1H), 4.08 (d, J = 1.7 Hz, 1H), 4.29 (q, J = 7.1 Hz, 2H), 4.51 (d, J = 1.5 Hz, 1H), 5.73 (s, 1H), 6.43 (s, 1H), 7.32−7.33 (m, 1H), 8.18 (br, 1H); ^{13}C{^{1}H} NMR (125 MHz, $CDCl_3$): δ 11.3, 14.5, 18.2, 27.8, 39.8, 50.6, 59.8, 84.4, 91.7, 105.3, 117.8, 122.2, 128.7, 161.4, 162.9, 165.0; HRMS (ESI-TOF) m/z: [M + H]$^+$ calcd for $C_{16}H_{22}NO_3$, 276.1594; found, 276.1589.

*4.1.9. Ethyl 5-((Z)-((3S,4S)-4-Ethyl-3,5-dimethyl-3,4-dihydro-2H-pyrrol-2-ylidene)methyl)-1H-pyrrole-3-carboxylate (**12**).* Following a general procedure[18] with some modifications, aqueous 1 M HCl (3.4 mL) was added to a solution of **11-E/Z** (1.58 g, 5.75 mmol) in DMF (67 mL). The reaction mixture was stirred at room temperature for 30 min. Afterward, NH_4OAc (8.86 g, 115 mmol) and Et_3N (16.0 mL, 115 mmol) were added, and the resulting solution was stirred at 55 °C in an oil bath for 15 min. The reaction was rapidly cooled to 0 °C in an ice bath before being quenched and diluted by sequential addition of cold saturated aqueous KH_2PO_4 (150 mL) solution and ethyl acetate (150 mL). The organic layer was washed with water, dried (Na_2SO_4), concentrated, and chromatographed [deactivated silica prepared by pretreating with Et_3N in hexanes and then removing the solvent under reduced pressure, hexanes/ethyl acetate (2:1), 4.5 cm × 40 cm] to afford the title dihydrodipyrrin (0.88 g, 56%, R_f 0.48 in hexanes/ethyl acetate (2:1)) and dipyrromethane byproduct **12-DPM** (0.27 g, 17%, R_f 0.35 in hexanes/ethyl acetate (2:1)) as a brown and yellow solid, respectively.

12: Characterization by ^{1}H NMR spectroscopy and LC-HRMS indicated the presence of a single isomer (Z only): mp 57−59 °C; ^{1}H NMR (500 MHz, $CDCl_3$): δ 0.94 (t, J = 7.4 Hz, 3H), 1.19 (d, J = 7.1 Hz, 3H), 1.34 (t, J = 7.1 Hz, 3H), 1.38−1.45 (m, 1H), 1.74−1.80 (m, 1H), 2.18 (s, 3H), 2.31−2.34 (m, 1H), 2.60−2.63 (m, 1H), 4.27 (q, J = 7.1 Hz, 2H), 5.75 (s, 1H), 6.45 (s, 1H), 7.41−7.42 (m, 1H), 11.21

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(br, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 11.1, 14.5, 18.9, 20.9, 24.3, 40.5, 59.3, 59.5, 104.8, 108.2, 116.3, 124.0, 132.1, 157.6, 165.4, 182.1; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_{16}H_{23}N_2O_2$, 275.1754; found, 275.1759.

*4.1.10. 8-Carboethoxy-2-ethyl-1,3-dimethyldipyrromethane (**12-DPM**).* mp 102−104 °C; ^1H NMR (500 MHz, $CDCl_3$): δ 1.07 (t, J = 7.5 Hz, 3H), 1.33 (t, J = 7.1 Hz, 3H), 1.97 (s, 3H), 2.10 (s, 3H), 2.37 (q, J = 7.5 Hz, 2H), 3.84 (s, 2H), 4.27 (q, J = 7.1 Hz, 2H), 6.42 (s, 1H), 7.28 (br, 2H), 8.20 (br, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 9.0, 10.9, 14.5, 15.7, 17.7, 24.2, 59.7, 107.0, 114.2, 116.7, 120.8, 120.9, 121.9, 122.8, 130.9, 165.1; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_{16}H_{23}N_2O_2$, 275.1754; found, 275.1744.

*4.1.11. (2S,3S)-8-Carboethoxy-2-ethyl-1-formyl-3-methyl-2,3-dihydrodipyrrin (**13**).* Following a general procedure,[18] SeO_2 (1.33 g, 12.0 mmol) was added in one portion to a solution of compound **12** (1.10 g, 4.00 mmol) in distilled 1,4-dioxane (120 mL) in the presence of added deionized water (200 μL). The reaction mixture was stirred at room temperature for 15 min. Upon completion, ethyl acetate (150 mL) and saturated aqueous $NaHCO_3$ (150 mL) were added. The organic layer was washed with water, dried (Na_2SO_4), concentrated, and chromatographed [deactivated silica prepared by pretreating with Et_3N in hexanes and then removing the solvent under reduced pressure, hexanes/ethyl acetate (2:1), 4.5 cm × 30 cm, R_f 0.47 in hexanes/ethyl acetate (2:1)] to give a yellow paste (247 mg, 21%): ^1H NMR (500 MHz, $CDCl_3$): δ 0.90 (t, J = 7.4 Hz, 3H), 1.21 (d, J = 7.0 Hz, 3H), 1.35 (t, J = 7.1 Hz, 3H), 1.40−1.51 (m, 1H), 1.84−1.92 (m, 1H), 2.72−2.78 (m, 2H), 4.29 (q, J = 7.1 Hz, 2H), 6.21 (s, 1H), 6.69 (s, 1H), 7.54 (dd, J = 3.1 and 1.5 Hz, 1H), 9.98 (s, 1H), 10.84 (br, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 11.0, 14.5, 21.5, 24.5, 41.2, 53.8, 59.8, 112.8, 114.9, 117.4, 126.5, 131.3, 157.2, 164.7, 173.9, 190.1; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_{16}H_{21}N_2O_3$, 289.1547; found, 289.1543.

*4.1.12. 2-Carbomethoxy-3-[(2S,3S)-8-carboethoxy-2-ethyl-1-formyl-3-methyl-2,3-dihydrodipyrrin-1-yl]-1-[1-(1,1-dimethoxymethyl)-3,3-dimethyl-2,3-dihydrodipyrrin-8-yl]prop-2-en-1-one (**14**).* Following a literature procedure[7] with modification, samples of **13** (38.0 mg, 132 μmol), **II** (50.0 mg, 143 μmol), and dried molecular sieves powder (3 Å, 45 mg) were added to a solution of piperidine/acetic acid in acetonitrile (15 mM/15 mM, 3.50 mL, 52.8 μmol/52.8 μmol). The reaction mixture was stirred at room temperature for 20 h, whereupon an additional amount of **II** (9.2 mg, 26 μmol) was added. Then, the resulting reaction mixture was stirred at room temperature for another 20 h, followed by filtration on a Celite pad. The filtrate was concentrated and purified by preparative TLC [20 cm × 20 cm, 1000 μm thickness, silica, hexanes/ethyl acetate (1:1)] to afford two orange bands, which were characterized as two isomeric forms of the title compound. Band 2 was subsequently found by single-crystal X-ray crystallography to be the E isomer (see the Supporting Information).

Band 1, implied to be the Z isomer (R_f 0.55 in hexanes/ethyl acetate (1:1), 2.3 mg, 3%): ^1H NMR (500 MHz, $CDCl_3$): δ 0.94 (t, J = 7.4 Hz, 3H), 1.21−1.23 (m, 9H), 1.34 (t, J = 7.1 Hz, 3H), 1.41−1.50 (m, 1H), 1.72−1.80 (m, 1H), 2.54−2.57 (m, 1H), 2.66 (s, 2H), 2.68−2.71 (m, 1H), 3.46 (s, 6H), 3.78 (s, 3H), 4.28 (q, J = 7.1 Hz, 2H), 5.04 (s, 1H), 5.86 (s, 1H), 5.99 (s, 1H), 6.57 (s, 1H), 6.59 (s, 1H), 7.11 (s, 1H), 7.52 (dd, J = 3.1 and 1.6 Hz, 1H), 7.57 (dd, J = 3.1 and 1.5 Hz, 1H), 10.79 (br, 1H), 11.26 (br, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 10.9, 14.5, 21.2, 25.0, 28.9, 40.2, 40.7, 48.4, 52.7, 54.5, 57.8, 59.6, 102.3, 106.2, 109.1, 110.2, 110.7, 116.6, 124.1, 125.9, 126.5, 130.0, 131.5, 132.7, 140.0, 157.2, 162.3, 165.0, 168.6, 172.8, 176.3, 183.5; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_{34}H_{43}N_4O_7$, 619.3126; found, 619.3123.

Band 2, E-isomer (R_f 0.48 in hexanes/ethyl acetate (1:1), 57.3 mg, 70%): mp 155−157 °C; ^1H NMR (500 MHz, $CDCl_3$): δ 0.92 (t, J = 7.4 Hz, 3H), 1.12 (d, J = 7.1 Hz, 3H), 1.20 (d, J = 2.8 Hz, 6H), 1.33 (t, J = 7.1 Hz, 3H), 1.39−1.47 (m, 1H), 1.74−1.81 (m, 1H), 2.47−2.50 (m, 1H), 2.57−2.58 (m, 1H), 2.62 (s, 2H), 3.42 (d, J = 1.6 Hz, 6H), 3.78 (s, 3H), 4.26 (q, J = 7.1 Hz, 2H), 4.99 (s, 1H), 5.83 (s, 1H), 5.85 (s, 1H), 6.46 (s, 1H), 6.51 (s, 1H), 7.37 (s, 1H), 7.43 (s, 1H), 7.45 (s, 1H), 10.36 (br, 1H), 11.20 (br, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 11.0, 14.5, 21.1, 25.1, 28.92, 28.94, 40.2, 40.6, 48.4, 52.9, 54.57, 54.58, 58.0, 59.5, 102.4, 106.3, 108.0, 110.5, 110.8, 116.2, 125.0, 126.2, 126.4, 131.4, 131.5, 133.0, 138.8, 157.1, 162.4, 165.0, 165.1, 171.9, 176.4, 188.9; HRMS (ESI-TOF) m/z $[M + H]^+$ calcd for $C_{34}H_{43}N_4O_7$ 619.3126; found, 619.3119.

*4.1.13. (17S,18S)-3-Carboethoxy-13^2-carbomethoxy-17-ethyl-8,8,18-trimethyl-13^1-oxo-bacteriophorbine (**BC-1**).* Following a literature procedure[7] with modification, a sample of $Yb(OTf)_3$ (241 mg, 388 μmol) was added to a solution of **14-*E*** (24.0 mg, 38.8 μmol) in acetonitrile (200 mL) in a glovebox. The reaction mixture was then stirred at 80 °C in a sand bath for 20 h under the argon atmosphere inside the glovebox. Then, the reaction mixture was allowed to cool to room temperature, withdrawn from the glovebox, and filtered through a Celite pad. The filtrate was concentrated and purified by preparative TLC [20 cm × 20 cm, 1000 μm thickness, silica, hexanes/ethyl acetate (3:1)] to yield a purple solid comprising two epimeric forms in 7:1 ratio (11.5 mg, 53%). The major product determined by NOESY spectroscopy possesses a trans−trans configuration with respect to the three stereocenters in the molecule: ^1H NMR (500 MHz, $CDCl_3$): δ −0.67 (br, 1H), 0.66 (br, 1H), 1.00 (t, J = 7.4 Hz, 3H), 1.66 (t, J = 7.1 Hz, 3H), 1.73 (d, J = 7.4 Hz, 3H), 1.86 (s, 3H), 1.90 (s, 3H), 1.95−2.04 (m, 1H), 2.25−2.33 (m, 1H), 3.85 (s, 3H), 3.89−3.92 (m, 1H), 4.23−4.33 (m, 3H), 4.68−4.77 (m, 2H), 6.09 (s, 1H), 8.40 (s, 1H), 8.44 (s, 1H), 8.50 (s, 1H), 9.15 (d, J = 1.9 Hz, 1H), 9.49 (s, 1H); $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): δ 10.8, 14.7, 22.9, 27.6, 31.05, 31.12, 44.5, 49.5, 52.97, 53.00, 53.3, 61.5, 64.3, 98.9, 100.2, 100.6, 108.4, 109.2, 126.6, 129.8, 131.2, 137.7, 137.8, 141.0, 149.6, 159.5, 164.7, 165.2, 169.6, 171.2, 171.4, 188.9; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_{32}H_{35}N_4O_5$, 555.2602; found, 555.2591. λ_{abs} = 749 nm, λ_{em} = 753 nm, Φ_f = 0.17 (λ_{ex} = 536 nm), in toluene.

*4.1.14. 3-Carboethoxy-13^2-carbomethoxy-17-ethyl-8,8,18-trimethyl-13^1-oxophorbine (**C-1**).* A solution of bacteriochlorin **BC-1** (4.0 mg, 7.2 μmol) and DDQ (3.27 mg, 14.4 μmol) in toluene (7.2 mL) was stirred at room temperature for 17 h in a glovebox filled with argon. The reaction mixture was then quenched by the addition of saturated aqueous $NaHCO_3$ (10 mL). The organic phase was collected. The aqueous phase was extracted with Et_2O (3 × 10 mL). The combined organic extract was dried (Na_2SO_4) and concentrated to give a crude brown solid, which was then chromatographed in a Pasteur pipette (silica, CH_2Cl_2) to yield a greenish deposit (1.4 mg, 35%): ^1H NMR (500 MHz, $CDCl_3$): δ 1.53 (t, J = 7.6 Hz, 3H), 1.70 (t, J = 7.1 Hz, 3H), 1.98 (s, 3H), 2.03 (s, 3H), 3.22 (s, 3H), 3.56 (ddt, J = 18.5, 14.9, and 7.5 Hz, 2H), 3.79 (s, 3H), 4.54 (s, 2H), 4.78 (q, J = 7.2 Hz, 2H), 6.64 (s, 1H), 8.77 (s, 1H), 8.87 (s, 1H), 9.43 (d, J = 3.2 Hz, 1H), 9.58 (d, J = 4.1 Hz, 1H), 9.83 (s, 1H), two exchangeable protons bound to nitrogen atoms were not unambiguously determined; $^{13}C\{^1H\}$ NMR (125 MHz, $CDCl_3$): 11.3, 14.8, 16.3, 20.2, 31.1, 31.3, 45.4, 52.7, 53.4, 61.5, 66.2, 97.5, 98.5, 104.2, 115.0, 143.5, 165.0, 169.6, 188.9, signals for 2 methine carbons and 11 quaternary carbons in the tetrapyrrole macrocycle were not observed; HRMS (ESI-TOF) m/z: $[M + H]^+$ calcd for $C_{32}H_{33}N_4O_5$, 553.2446; found, 553.2435. λ_{abs} = 692 nm, λ_{em} = 695 nm, Φ_f = 0.41 (λ_{ex} = 553 nm), in toluene.

4.2. Crystallization Protocol. Enone **14-*E*** (20 mg) was dissolved in acetonitrile (1 mL) and then divided equally among six 500 μL glass insert tubes. Each insert tube was placed in a 5 mL vial containing 1 mL of one of the following solvents: pentane, hexane, heptane, diethyl ether, toluene, or benzene (Figure S6). The vials were allowed to stand at room temperature for vapor diffusion. The sample in acetonitrile/diethyl ether yielded the crystalline sample used for X-ray crystallography analysis.

4.3. Molar Absorption Coefficient of BC-1. A stock solution of 1.30 mM **BC-1** (7.20 mg, 13.0 μmol) in HPLC-grade toluene (10 mL) was serially diluted to form six solutions with concentrations spanning a range from 0.13 to 6.5 μM. The absorption spectrum of each solution was recorded. The graph of the absorbance at λ = 749 nm versus concentration was plotted, and the molar coefficient was determined to be 72,100 M^{-1} cm^{-1} from the slope of the graph (Figure S12).

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4.4. Fluorescence Spectroscopy. The fluorescence spectrometer instrument parameters were as follows: excitation and emission slit widths = 1.5 nm (0.375 mm); photomultiplier tube (Hamamatsu R928P) voltage = 1000; and integration time = 1 nm/s. The fluorescence spectra were corrected for sample absorption at the wavelength of excitation and for instrument sensitivity as a function of wavelength. Fluorescence quantum yields were determined with use of the standard 2,12-di-*p*-tolyl-8,8,18,18-tetramethylbacteriochlorin (Φ_f = 0.18, toluene).[67] The Φ_f of **C-1** is 0.41, whereas that of **BC-1** is 0.17.

4.5. Spectroscopic Examination of the Chlorin Content in the Bacteriochlorin-Forming Reaction Mixtures. Stock solutions of bacteriochlorin **BC-1** and chlorin **C-1** were prepared in toluene. The concentrations were estimated as 4.0 and 2.0 μM, respectively, from the absorbance of the Q_y band in the absorption spectrum and reported values of the molar absorption coefficient for the analogues bacteriopheophytin *a* (BPheo *a*, λ_{abs} = 748 nm, ε = 67,600 M^{-1} cm^{-1})[3,4] and **C-3** (λ = 682 nm, ε_{682nm} = 3.65 × 10^4 M^{-1} cm^{-1}, CH_2Cl_2/THF).[60] Four mock solutions with a constant concentration of **BC-1** at 2.0 μM and serially diluted concentrations of **C-1** ranging from 0.2 to 0.0002 μM were made from the two stock solutions.

To determine the most appropriate excitation wavelength, the fluorescence spectrum was examined at five excitation wavelengths: 406, 420, 430, 440, and 553 nm (Figure S13). A strong emission of **C-1** (λ_{em} = 695 nm) versus low emission of **BC-1** (λ_{em} = 753 nm) was achieved upon excitation at 406 nm. The graph of integrated fluorescence intensity at λ_{em} = 695 nm versus concentration of **C-1** in mock solutions is plotted in Figure S14. An aliquot from the crude mixture (of **14-*E*** at 80 °C for 21 h) was taken and diluted in toluene to achieve a concentration of bacteriochlorin **BC-1** of ~2.0 μM (determined by absorption spectroscopy). The resulting solution was analyzed by emission spectroscopy, showing that chlorin **C-1** was present at the concentration of 0.028 μM (interpolated from the linear plot in Figure S14), which corresponds to the molar ratio 1:71 with respect to **BC-1** in the same sample (Figure 6). Note that the intensity of fluorescence emission depends on (1) the absorption at the wavelength of excitation (which in turn depends on the concentration and the molar absorption coefficient at that wavelength) and (2) the value of the fluorescence quantum yield.

4.6. Nomenclature. Compound **BC-1** shares a common hydrocarbon skeleton with a free base bacteriochlorophyll (i.e., a bacteriopheophytin) but also differs significantly with regard to peripheral substituents, namely, the lack of the phytyl ester and the lack of the *trans*-dialkyl groups in ring B. For a more exact description, **BC-1** is (1) a bacteriochlorin owing to the presence of pyrroline rings B and D, and (2) a phorbine[68] (with 7,8-saturation) owing to the presence of the isocyclic ring. The hydrocarbon skeleton of **BC-1** is hence a bacteriophorbine.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.joc.0c00608.

^{1}H NMR and $^{13}C\{^1H\}$ NMR data for all new compounds; single-crystal X-ray data for **14-*E***; fluorescence spectra for the chlorin assay; and molar absorption coefficient data for **BC-1** (PDF)

Crystallographic data for compound **14** (CIF)

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Notes

The authors declare the following competing financial interest(s): J.S.L. is a cofounder of NIRvana Sciences, which may license aspects of the technology described herein, for which a patent has been applied.

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■ REFERENCES

(1) Blankenship, R. E. *Molecular Mechanisms of Photosynthesis*, 1st ed.; Blackwell Science: Malden, MA, 2002.

(2) Scheer, H. An Overview of Chlorophylls and Bacteriochlorophylls: Biochemistry, Biophysics, Functions and Applications. In *Chlorophylls and Bacteriochlorophylls. Biochemistry, Biophysics, Functions and Applications*; Grimm, B., Porra, R. J., Rüdiger, W., Scheer, H., Eds.; Springer: Dordrecht, The Netherlands, 2006; Vol. 25, pp 1−26.

(3) Kobayashi, M.; Akiyama, M.; Kano, H.; Kise, H. Spectroscopy and Structure Determination. In *Chlorophylls and Bacteriochlorophylls. Biochemistry, Biophysics, Functions and Applications*; Grimm, B., Porra, R. J., Rüdiger, W., Scheer, H., Eds.; Springer: Dordrecht, The Netherlands, 2006; Vol. 25, pp 79−94.

(4) Kobayashi, M.; Sorimachi, Y.; Fukayama, D.; Komatsu, H.; Kanjoh, T.; Wada, K.; Kawachi, M.; Miyashita, H.; Ohnishi-Kameyama, M.; Ono, H. Physicochemical Properties of Chlorophylls and Bacteriochlorophylls. In *Handbook of Photosynthesis*, 3rd ed.; Pessarakli, M., Ed.; CRC Press: Boca Raton, FL, 2016; pp 95−147.

(5) Liu, Y.; Zhang, S.; Lindsey, J. S. Total Synthesis Campaigns Toward Chlorophylls and Related Natural Hydroporphyrins − Diverse Macrocycles, Unrealized Opportunities. *Nat. Prod. Rep.* **2018**, *35*, 879−901.

(6) Lindsey, J. S. De Novo Synthesis of Gem-Dialkyl Chlorophyll Analogues for Probing and Emulating our Green World. *Chem. Rev.* **2015**, *115*, 6534−6620.

(7) Zhang, S.; Lindsey, J. S. Construction of the Bacteriochlorin Macrocycle with Concomitant Nazarov Cyclization to Form the Annulated Isocyclic Ring: Analogues of Bacteriochlorophyll a. *J. Org. Chem.* **2017**, *82*, 2489−2504.

(8) Wang, P.; Lu, F.; Lindsey, J. S. Use of the Nascent Isocyclic Ring to Anchor Assembly of the Full Skeleton of Model Chlorophylls. *J. Org. Chem.* **2020**, *85*, 702−715.

(9) Wang, P.; Chau Nguyen, K.; Lindsey, J. S. Synthesis of the Ring C Pyrrole of Native Chlorophylls and Bacteriochlorophylls. *J. Org. Chem.* **2019**, *84*, 11286−11293.

(10) Smith, J. R. L.; Calvin, M. Studies on the Chemical and Photochemical Oxidation of Bacteriochlorophyll. *J. Am. Chem. Soc.* **1966**, *88*, 4500−4506.
(11) Bonnett, R.; Martínez, G. Photobleaching of Sensitisers used in Photodynamic Therapy. *Tetrahedron* **2001**, *57*, 9513−9547.
(12) Chen, Y.; Li, G.; Pandey, R. Synthesis of Bacteriochlorins and Their Potential Utility in Photodynamic Therapy. *Curr. Org. Chem.* **2004**, *8*, 1105−1134.
(13) Limantara, L.; Heriyanto, H. Photostability of Bacteriochlorophyll a and its Derivatives as Potential Sensitizers for Photodynamic Cancer Therapy: The Study on Acetone-Water and Methanol-Water Solvents. *Indones. J. Chem.* **2011**, *11*, 154−162.
(14) O'Neal, W. G.; Roberts, W. P.; Ghosh, I.; Jacobi, P. A. Studies in Chlorin Chemistry. II. A Versatile Synthesis of Dihydrodipyrrins. *J. Org. Chem.* **2005**, *70*, 7243−7251.
(15) van Leusen, A. M.; Siderius, H.; Hoogenboom, B. E.; van Leusen, D. A New and Simple Synthesis of the Pyrrole Ring System from Michael Acceptors and Tosylmethylisocyanides. *Tetrahedron Lett.* **1972**, *13*, 5337−5340.
(16) Ma, Z.; Ma, Z.; Zhang, D. Synthesis of Multi-Substituted Pyrrole Derivatives Through [3+2] Cycloaddition with Tosylmethyl Isocyanides (TosMICs) and Electron-Deficient Compounds. *Molecules* **2018**, *23*, 2666.
(17) Krayer, M.; Ptaszek, M.; Kim, H.-J.; Meneely, K. R.; Fan, D.; Secor, K.; Lindsey, J. S. Expanded Scope of Synthetic Bacteriochlorins via Improved Acid Catalysis Conditions and Diverse Dihydrodipyrrin-Acetals. *J. Org. Chem.* **2010**, *75*, 1016−1039.
(18) Liu, Y.; Lindsey, J. S. Northern−Southern Route to Synthetic Bacteriochlorins. *J. Org. Chem.* **2016**, *81*, 11882−11897.
(19) Nicholas, K. M. Chemistry and Synthetic Utility of Cobalt-Complexed Propargyl Cations. *Acc. Chem. Res.* **1987**, *20*, 207−214.
(20) Evans, D. A.; Bartroli, J.; Shih, T. L. Enantioselective Aldol Condensations. 2. Erythro-Selective Chiral Aldol Condensations via Boron Enolates. *J. Am. Chem. Soc.* **1981**, *103*, 2127−2129.
(21) Schreiber, S. L.; Sammakia, T.; Crowe, W. E. Lewis Acid-Mediated Version of the Nicholas Reaction: Synthesis of Syn-Alkylated Products and Cobalt-Complexed Cycloalkynes. *J. Am. Chem. Soc.* **1986**, *108*, 3128−3130.
(22) Riclea, R.; Aigle, B.; Leblond, P.; Schoenian, I.; Spiteller, D.; Dickschat, J. S. Volatile Lactones from Streptomycetes Arise via the Antimycin Biosynthetic Pathway. *ChemBioChem* **2012**, *13*, 1635−1644.
(23) Jacobi, P. A.; Murphree, S.; Rupprecht, F.; Zheng, W. Formal Total Syntheses of the β-Lactam Antibiotics Thienamycin and PS-5. *J. Org. Chem.* **1996**, *61*, 2413−2427.
(24) Jacobi, P. A.; Buddhu, S. C.; Fry, D.; Rajeswari, S. Studies on the Synthesis of Phytochrome and Related Tetrapyrroles. Dihydropyrromethenones by Photochemical Rearrangement of N-Pyrrolo Enamides. *J. Org. Chem.* **1997**, *62*, 2894−2906.
(25) Evans, D. A.; Sjogren, E. B.; Bartroli, J.; Dow, R. L. Aldol Addition Reactions of Chiral Crotonate Imides. *Tetrahedron Lett.* **1986**, *27*, 4957−4960.
(26) Evans, D. A.; Britton, T. C.; Ellman, J. A. Contrasteric Carboximide Hydrolysis with Lithium Hydroperoxide. *Tetrahedron Lett.* **1987**, *28*, 6141−6144.
(27) Leung, S. H.; Edington, D. G.; Griffith, T. E.; James, J. J. Deiodination of 5-Iodopyrrole-2-carboxylates Using Sodium Formate Catalyzed by Palladium Complexes: Preparation of 5-Unsubstituted Pyrrole-2-carboxylates. *Tetrahedron Lett.* **1999**, *40*, 7189−7191.
(28) Acheson, R. M. *An Introduction to the Chemistry of Heterocyclic Compounds*, 3rd ed.; Wiley-Interscience: New York, 1976; p 110.
(29) Khaghaninejad, S.; Heravi, M. M. Paal-Knorr Reaction in the Synthesis of Heterocyclic Compounds. *Adv. Heterocycl. Chem.* **2014**, *111*, 95−146.
(30) Bean, G. P. The Synthesis of 1H-Pyrroles. In *Pyrroles. Part One. The Synthesis and the Physical and Chemical Aspects of the Pyrrole Ring*; Jones, R. A., Ed.; John Wiley & Sons, Inc.: New York, 1990; pp 105−303.
(31) Mulzer, J.; Innitzer, A. A Tetracarbonyl Paal-Knorr Approach to Semicorrins. *Heterocycles* **2009**, *77*, 873−886.
(32) O'Neal, W. G.; Roberts, W. P.; Ghosh, I.; Wang, H.; Jacobi, P. A. Studies in Chlorin Chemistry. 3. A Practical Synthesis of C,D-Ring Symmetric Chlorins of Potential Utility in Photodynamic Therapy. *J. Org. Chem.* **2006**, *71*, 3472−3480.
(33) Riley, H. L.; Morley, J. F.; Friend, N. A. C. Selenium Dioxide, A New Oxidising Agent. Part I. Its Reaction with Aldehydes and Ketones. *J. Chem. Soc.* **1932**, 1875−1883.
(34) Wang, P.; Lindsey, J. S. Riley Oxidation of Heterocyclic Intermediates on Paths to Hydroporphyrins − A Review. *Molecules* **2020**, *25*, 1858.
(35) Williamson, K. L. Substituent Effects on Nuclear Magnetic Resonance Coupling Constants and Chemical Shifts in a Saturated System: Hexachlorobicyclo [2.2.1]heptenes. *J. Am. Chem. Soc.* **1963**, *85*, 516−519.
(36) Smith, K. M.; Goff, D. A.; Abraham, R. J. The NMR Spectra of Porphyrins. 27−Proton NMR Spectra of Chlorophyll-a and Pheophytin-a. *Org. Magn. Reson.* **1984**, *22*, 779−783.
(37) Lonin, I. S.; Belyaev, E. S.; Tafeenko, V. A.; Chernyshev, V. V.; Savinkina, E. V.; Ponomarev, G. V.; Koifman, O. I.; Tsivadze, A. Y. X-Ray Single-Crystal Structures and NMR Characterization of Three Vinyl Substituted Methylpyropheophorbide a Derivatives. *Macroheterocycles* **2015**, *8*, 366−370.
(38) Khare, R.; Pandey, J.; Smriti, S.; Ruchi, R. The Importance and Applications of Knoevenagel Reaction (Brief Review). *Orient. J. Chem.* **2019**, *35*, 423−429.
(39) List, B. Emil Knoevenagel and the Roots of Aminocatalysis. *Angew. Chem., Int. Ed.* **2010**, *49*, 1730−1734.
(40) Han, Y.-F.; Xia, M. Multicomponent Synthesis of Cyclic Frameworks on Knoevenagel-Initiated Domino Reactions. *Curr. Org. Chem.* **2010**, *14*, 379−413.
(41) Mietke, T.; Cruchter, T.; Larionov, V. A.; Faber, T.; Harms, K.; Meggers, E. Asymmetric Nazarov Cyclizations Catalyzed by Chiral-at-Metal Complexes. *Adv. Synth. Catal.* **2018**, *360*, 2093−2100.
(42) Giese, S.; West, F. G. Ionic Hydrogenation of Oxyallyl Intermediates: The Reductive Nazarov Cyclization. *Tetrahedron* **2000**, *56*, 10221−10228.
(43) Malona, J. A.; Colbourne, J. M.; Frontier, A. J. A General Method for the Catalytic Nazarov Cyclization of Heteroaromatic Compounds. *Org. Lett.* **2006**, *8*, 5661−5664.
(44) He, W.; Herrick, I. R.; Atesin, T. A.; Caruana, P. A.; Kellenberger, C. A.; Frontier, A. J. Polarizing the Nazarov Cyclization: The Impact of Dienone Substitution Pattern on Reactivity and Selectivity. *J. Am. Chem. Soc.* **2008**, *130*, 1003−1011.
(45) Takeda, T.; Harada, S.; Nishida, A. Catalytic Asymmetric Nazarov Cyclization of Heteroaryl Vinyl Ketones through a Crystallographically Defined Chiral Dinuclear Nickel Complex. *Org. Lett.* **2015**, *17*, 5184−5187.
(46) Vaidya, T.; Eisenberg, R.; Frontier, A. J. Catalytic Nazarov Cyclization: The State of the Art. *ChemCatChem* **2011**, *3*, 1531−1548.
(47) Wenz, D. R.; Read de Alaniz, J. The Nazarov Cyclization: A Valuable Method to Synthesize Fully Substituted Carbon Stereocenters. *Eur. J. Org. Chem.* **2015**, 23−37.
(48) Hynninen, P. H. Chemistry of Chlorophylls: Modifications. In *Chlorophylls*; Scheer, H., Ed.; CRC Press, Inc.: Boca Raton, Florida, 1991; pp 145−209.
(49) Mazaki, H.; Watanabe, T.; Takahashi, T.; Struck, A.; Scheer, H. Epimerization of Chlorophyll Derivatives. V. Effects of the Central Magnesium and Ring Substituents on the Epimerization of Chlorophyll Derivatives. *Bull. Chem. Soc. Jpn.* **1992**, *65*, 3080−3087.
(50) Saga, Y.; Nakagawa, S. Structural Effects on Epimerization of Bacteriochlorophyll a and Chlorophyll a Revealed Using 3-Acetyl Chlorophyll a. *J. Porphyrins Phthalocyanines* **2020**, *24*, 499−504.
(51) Storch, K.-F.; Cmiel, E.; Schafer, W.; Scheer, H. Stereoselectivity of Pigment Exchange with 13^2-Hydroxylated Tetrapyrroles in Reaction Centers of Rhodobacter sphaeroides R26. *Eur. J. Biochem.* **1996**, *238*, 280−286.

https://dx.doi.org/10.1021/acs.joc.0c00608
J. Org. Chem. XXXX, XXX, XXX−XXX

(52) Katz, J. J.; Norman, G. D.; Svec, W. A.; Strain, H. H. Chlorophyll Diastereoisomers. The Nature of Chlorophylls a′ and b′ and Evidence for Bacteriochlorophyll Epimers from Proton Magnetic Resonance Studies. *J. Am. Chem. Soc.* **1968**, *90*, 6841−6845.
(53) Connolly, J. S.; Samuel, E. B.; Janzen, A. F. Effects of Solvent on the Fluorescence Properties of Bacteriochlorophyll *α*. *Photochem. Photobiol.* **1982**, *36*, 565−574.
(54) Holten, D.; Gouterman, M.; Parson, W. W.; Windsor, M. W.; Rockley, M. G. Electron Transfer from Photoexcited Singlet and Triplet Bacteriopheophytin. *Photochem. Photobiol.* **1976**, *23*, 415−423.
(55) Gouterman, M.; Holten, D. Electron Transfer from Photo-excited Singlet and Triplet Bacteriopheophytin−II. Theoretical. *Photochem. Photobiol.* **1977**, *25*, 85−92.
(56) Braude, E. A.; Brook, A. G.; Linstead, R. P. Hydrogen Transfer. Part IV. The Use of Quinones of High Potential as Dehydrogenation Reagents. *J. Chem. Soc.* **1954**, 3569−3574.
(57) Eisner, U.; Linstead, R. P. Chlorophyll and Related Substances. Part II. The Dehydrogenation of Chlorin to Porphin and the Number of Extra Hydrogen Atoms in the Chlorins. *J. Chem. Soc.* **1955**, 3749−3754.
(58) Tamiaki, H.; Kouraba, M.; Takeda, K.; Kondo, S.-I.; Tanikaga, R. Asymmetric Synthesis of Methyl Bacteriopheophorbide-d and Analogues by Stereoselective Reduction of the 3-Acetyl to the 3-(1-Hydroxyethyl) Group. *Tetrahedron: Asymmetry* **1998**, *9*, 2101−2111.
(59) Liu, C.; Dobhal, M. P.; Ethirajan, M.; Missert, J. R.; Pandey, R. K.; Balasubramanian, S.; Sukumaran, D. K.; Zhang, M.; Kadish, K. M.; Ohkubo, K.; Fukuzumi, S. Highly Selective Synthesis of the Ring-B Reduced Chlorins by Ferric Chloride-Mediated Oxidation of Bacteriochlorins: Effects of the Fused Imide vs Isocyclic Ring on Photophysical and Electrochemical Properties. *J. Am. Chem. Soc.* **2008**, *130*, 14311−14323.
(60) Joshi, P.; Ethirajan, M.; Goswami, L. N.; Srivatsan, A.; Missert, J. R.; Pandey, R. K. Synthesis, Spectroscopic, and in Vitro Photosensitizing Efficacy of Ketobacteriochlorins Derived from Ring-B and Ring-D Reduced Chlorins via Pinacol−Pinacolone Rearrangement. *J. Org. Chem.* **2011**, *76*, 8629−8640.
(61) Xu, M.; Kinoshita, Y.; Matsubara, S.; Tamiaki, H. Synthesis of Chlorophyll-c Derivatives by Modifying Natural Chlorophyll-a. *Photosynth. Res.* **2016**, *127*, 335−345.
(62) Kim, H.-J.; Lindsey, J. S. De Novo Synthesis of Stable Tetrahydroporphyrinic Macrocycles: Bacteriochlorins and a Tetradehydrocorrin. *J. Org. Chem.* **2005**, *70*, 5475−5486.
(63) Vaidya, T.; Atesin, A. C.; Herrick, I. R.; Frontier, A. J.; Eisenberg, R. A Highly Reactive Dicationic Iridium(III) Catalyst for the Polarized Nazarov Cyclization Reaction. *Angew. Chem., Int. Ed.* **2010**, *49*, 3363−3366.
(64) Grin, M.; Mironov, A.; Shtil, A. Bacteriochlorophyll a and Its Derivatives; Chemistry and Perspectives for Cancer Therapy. *Anti-Cancer Agents Med. Chem.* **2008**, *8*, 683−697.
(65) Grin, M. A.; Mironov, A. F. Chemical Transformations of Bacteriochlorophyll a and Its Medical Applications. *Russ. Chem. Bull., Int. Ed.* **2016**, *65*, 333−349.
(66) Heyes, D. J.; Hunter, C. N. Biosynthesis of Chlorophyll and Bacteriochlorophyll. In *Tetrapyrroles: Birth, Life and Death*; Warren, M. J., Smith, A. G., Eds.; Landes Bioscience: Austin, TX, 2009; pp 235−249.
(67) Yang, E.; Kirmaier, C.; Krayer, M.; Taniguchi, M.; Kim, H.-J.; Diers, J. R.; Bocian, D. F.; Lindsey, J. S.; Holten, D. Photophysical Properties and Electronic Structure of Stable, Tunable Synthetic Bacteriochlorins: Extending the Features of Native Photosynthetic Pigments. *J. Phys. Chem. B* **2011**, *115*, 10801−10816.
(68) Moss, G. P. Nomenclature of Tetrapyrroles. *Pure Appl. Chem.* **1987**, *59*, 779−832.

https://dx.doi.org/10.1021/acs.joc.0c00608
J. Org. Chem. XXXX, XXX, XXX−XXX