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# Total Syntheses of Sterically Locked Phycocyanobilin Derivatives Bearing a 15*Z-anti* or a 15*E-anti* CD-Ring Component

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Total syntheses of sterically locked phycocyanobilin derivatives with a 15*Z-anti* or a 15*E-anti* CD-ring component were performed toward elucidation of the stereochemistry and function of the chromophore in phytochromes. In the course of the construction of a sterically locked 15*E-anti* CD-ring component employing 5-tosylpyrrolin-2-one derivatives as the D-ring, the Ts group was found to be rearranged under acidic conditions to give a mixture of regioisomers, both of which could be transformed into the same CD-ring precursor via detosylation with a base followed by Wittig-like coupling reaction. In addition, a sterically locked 15*E-anti* biliverdin derivative was also synthesized.

Phytochromes, one of the best-characterized photoreceptors in plants, are a widespread family of red/far-red light responsive photoreceptors first discovered in plants[1] and have been recently also discovered in bacteria,[2] fungi,[3] and slime molds.[4] They play critical roles in various light-regulated processes, ranging from phototaxis and pigmentation in bacteria to seed germination, chloroplast development, shade avoidance, and flowering in higher plants. All phytochromes have a covalently attached linear tetrapyrrole (bilin) chromophore that absorbs light in the red and far-red region.[5] Three different bilins are used as chromophores: land plants use phytochromobilin (PΦB),[6] and cyanobacteria use phycocyanobilin (PCB),[7] which is also a chromophore of the light-harvesting pigment, phycocyanin, and differs from PΦB only by substitution of the vinyl group at the C18 position with an ethyl group.[6,7b,8] The PΦB and PCB chromophores bind covalently to the apoprotein by a thioether bond through the A-ring ethylidene side chain. Some bacterial phytochromes carry biliverdin (BV) as a natural chromophore (Figure 1).[3,9] We found that the BV covalently binds to the apoprotein of *Agrobacterium* phytochrome Agp1 via its A-ring vinyl side chain,[10] which was recently confirmed by X-ray crystallographic analysis of other bacteriophytochromes.[11]

The interchange between the physiologically inactive red light absorbing Pr-form and the active far-red light absorbing Pfr-form is the most essential for the light absorbing and biological processes in the phytochrome chromophore function (Figure 1). It is commonly accepted that the first step in the photoconversion from Pr to Pfr is a *Z* to *E* isomerization around the C15=C16 double bond between the C- and D-rings of the bilin chromophores.[12]

In order to analyze the structure and function of the chromophores in the reconstituted phytochromes, we have synthesized PΦB, PCB, the modified PCBs, and BVs, in free acid forms by developing efficient methods for the preparation of each pyrrole ring, a new coupling reaction between them, and palladium-catalyzed deprotection of allyl propanoate side chains of the B- and C-rings under mild conditions.[13] Furthermore, we have succeeded for the first time in synthesizing the sterically locked 15*Z-syn*, 15*Z-anti*, 15*E-syn*, and 15*E-anti* BV derivatives,[14,15] which made it possible to directly confirm the stereochemistry around the C15 position of the chromophores in *Agrobacterium* phytochromes Agp1 and Agp2.[16] Doubly locked BV derivatives were also recently synthesized to assemble with Agp1 and Agp2.[17] Assembly experiments of the synthesized chromophores with phytochrome apoproteins in vitro and in vivo have provided us insights into the structure and function of phytochromes, in particular the stereochemistry at the C15 position of the BV chromophore in Pr- and Pfr-forms of Agp1 and Agp2 was determined to be 15*Z-anti* and 15*E-anti*, respectively.[13]

Recently it was reported that light-induced rotation of the A-ring but not the D-ring is the primary motion of the chromophore during photoconversion from Pr to Pfr in the cyanobacterial phytochrome from *Synechococcus* OSB′, which has PCB as a chromophore.[18] In order to reveal the exact mechanism of photoconversion in the cyanobacterial phytochrome, the syntheses of sterically locked PCB derivatives are crucial. In this paper, we first describe a total synthesis of 15*Z-anti* PCB derivative, in which the stereochemistry between the

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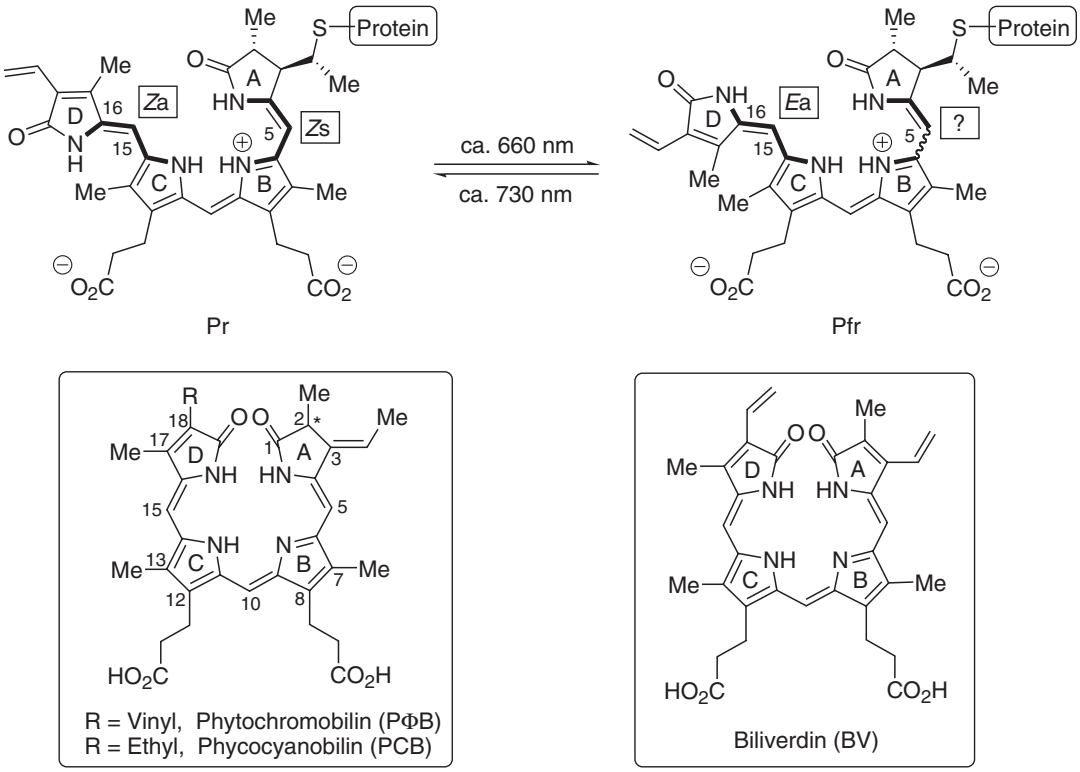



**Figure 1.**

C- and D-rings is locked in *Z*-configuration and *anti*-conformation, respectively, corresponding to the Pr-form of Agp1 and Agp2. Next, a total synthesis 15*E-anti* BV derivative, which has been reported briefly in a preliminary communication,[15] and finally, 15*E-anti* PCB derivative corresponding to the Pfr-form will be described.

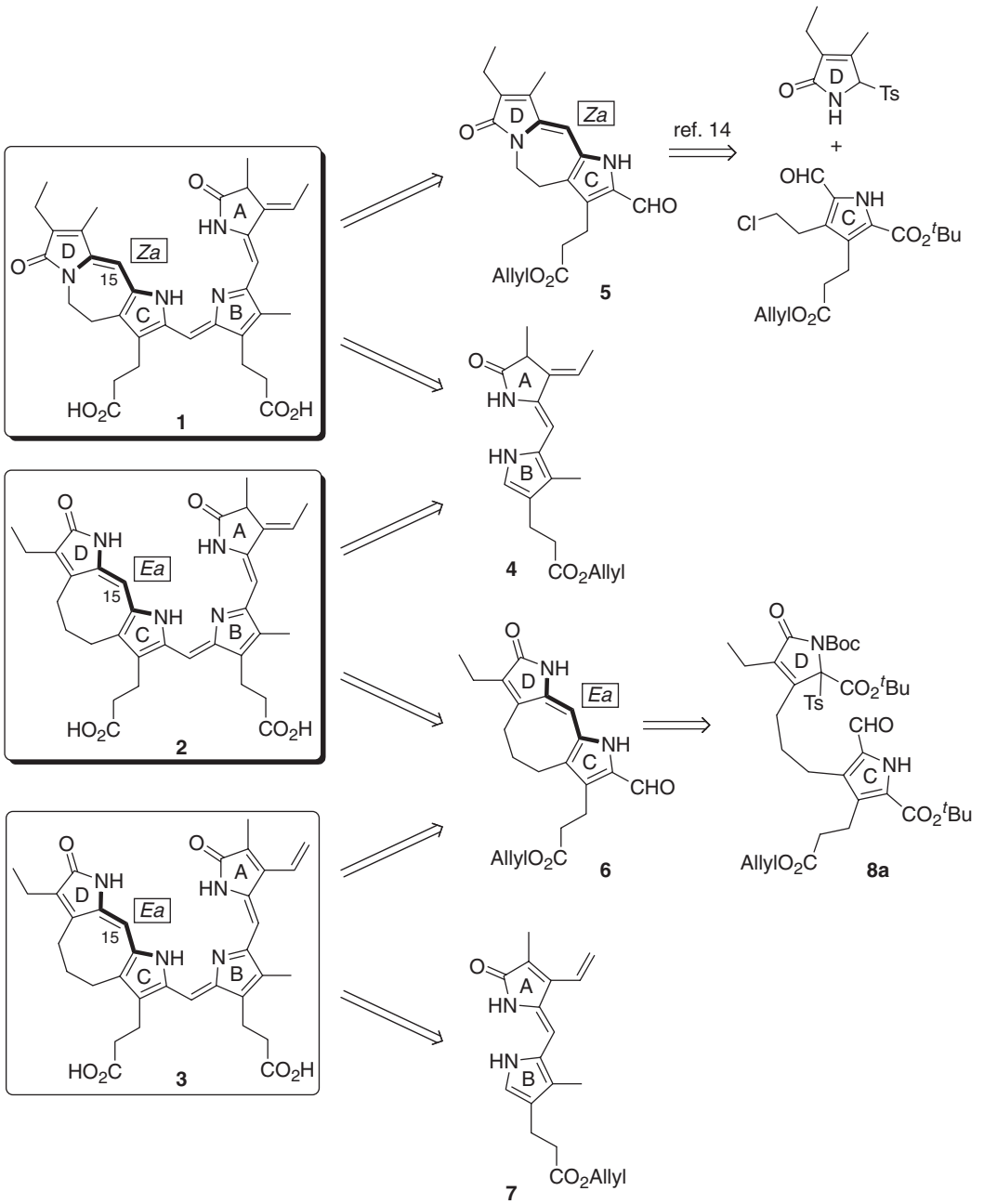
The retro-synthetic analyses are shown in Scheme 1. In the synthesis of sterically locked 15*Z-anti* PCB derivative **1**, ring closure was carried out by the intramolecular $S_N2$ reaction of a nitrogen atom of the D-ring toward an alkyl halide moiety of the C-ring to form **5** as previously described.[14] On the contrary, the intramolecular Wittig-like reaction between the tethered C- and D-rings in **8a** was designed for the synthesis of a sterically locked *E-anti* CD-ring component **6**, which allowed production of 15*E-anti* PCB derivative **2** and 15*E-anti* BV derivative **3** corresponding to Pfr-form.

## Results and Discussion

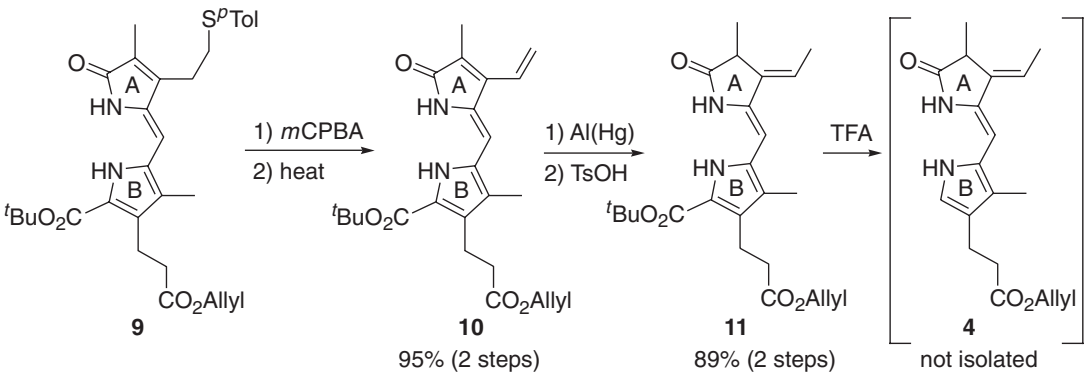
The AB-ring component **4** of PCB derivative was prepared from **9**, whose preparation was previously reported during the course of the synthesis of the 15*Z-anti* locked BV derivative.[14] Oxidation of sulfide **9** by *m*CPBA and subsequent heating in xylene gave a vinylic compound **10** (Scheme 2). The resulting compound **10** was then reduced with aluminum amalgam[19] followed by acidic isomerization to give **11** in good chemical yield. Compound **4** generated in situ from **11** by treating with trifluoroacetic acid (TFA) was coupled with **5**[14] under acidic conditions to afford the sterically locked 15*Z-anti* PCB diallyl ester **12** in 54% yield (Scheme 3).[20] The allyl ester groups of **12** were deprotected via a Pd(0)-catalyzed reaction using sodium *p*-toluenesulfinate (TsNa) as a nucleophile in THF/MeOH to give the desired locked chromophore **1** in 76% yield in a free acid form.

Next we intended to synthesize the sterically locked 15*E-anti* BV derivative **3**. The CD-ring component **6** was prepared starting from the commercially available 3,4-dihydro-2*H*-pyran (**13**) via tetrahydropyran-2-ol (**14**) and 5-hydroxypentanal (**15**) as shown in Scheme 4. The Henry reaction with 1-nitropropane followed by acetylation in the presence of 4-(dimethylamino)-pyridine (DMAP) gave a mixture of nitro diacetate compound **17** and the corresponding nitro olefin **18**, which was allowed to react with *t*-butyl isocyanoacetate without separation in the presence of DBU applying Barton's method[21] to give the pyrrole derivative **19** in 26% yield from **13** in four steps. Iodination of the $\alpha$-position of the pyrrole **19** with *N*-iodosuccinimide (NIS) followed by oxidation utilizing $Pb(OAc)_4$ in toluene[22] gave the pyrrolinone derivative **21** in 90% yield from **19**. The $\alpha$-acetoxy group of the pyrrolinone derivative **21** was replaced with a Ts group using anhydrous TsNa in 94% yield, followed by protection of the pyrrolinone-NH using $Boc_2O$ to give **23** in 86% yield.

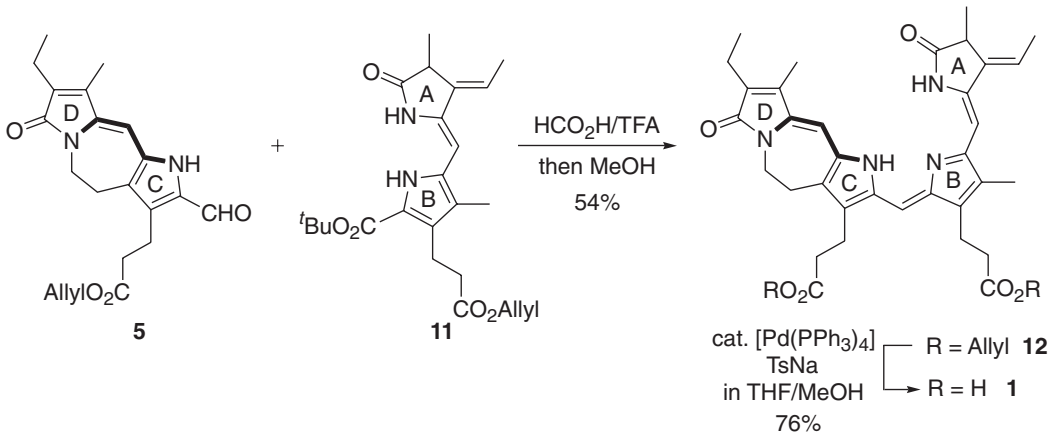
Hydrolysis of the acetate group in 0.5 M methanolic HCl afforded the N-protected tosylpyrrolinone derivative **24** in 97% yield. As described in the preliminary communication,[15] the iodination of the resulting alcohol using iodine and triphenylphosphine in the presence of imidazole followed by nitration using sodium nitrite in the presence of phloroglucinol[23] gave a tosylpyrrolinone derivative bearing a nitro group in its side chain, which was initially considered to be **26a**. However, the spectral data of the product were not identical with the indicated structure of **26a**. The hydrolysis product first assigned as **24a** was confirmed to be a ca. 1:2 mixture of the two



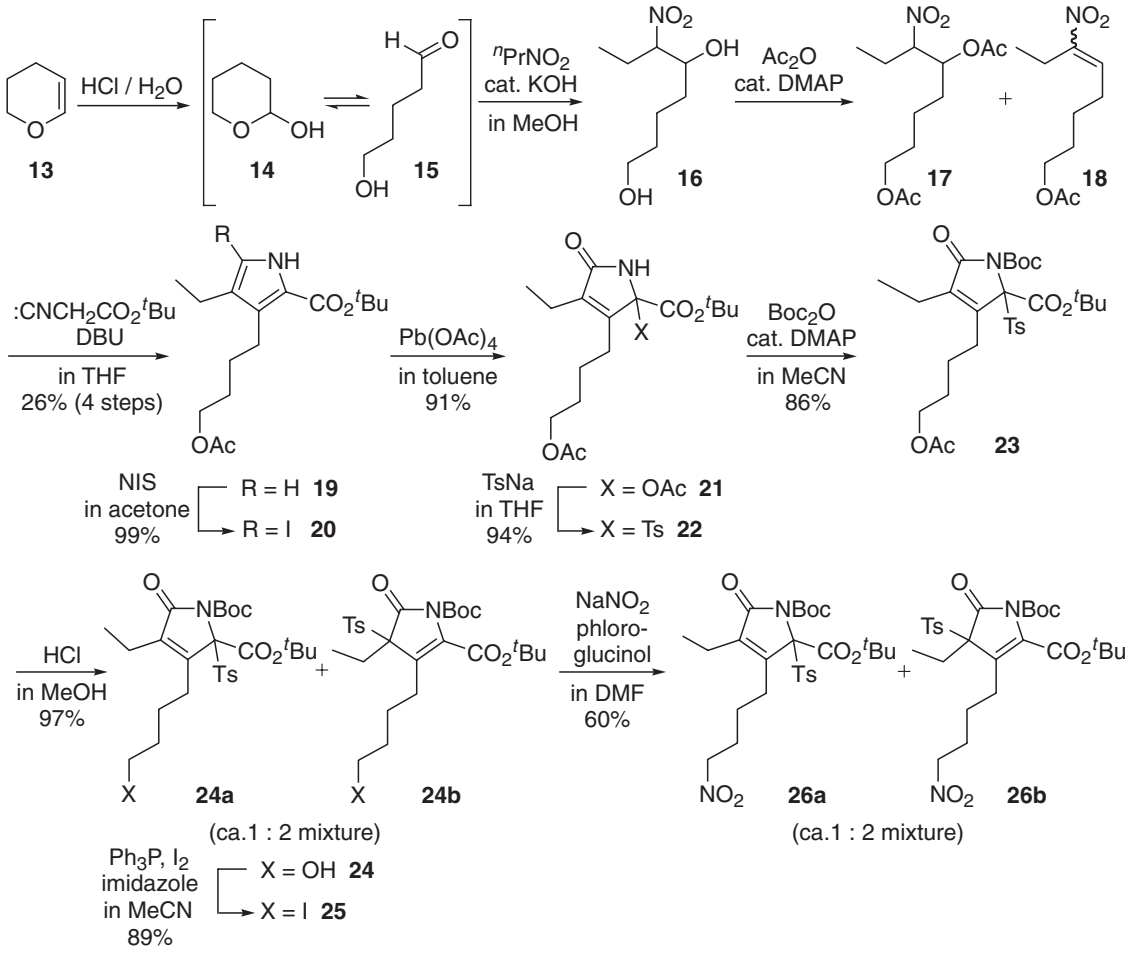

**Scheme 1.**




**Scheme 2.**




**Scheme 3.**




**Scheme 4.**

isomeric alcohols **24a** and **24b** as follows. After conversion of the mixture of alcohols **24** to nitro compounds **26** via iodination, the resulting nitro compounds could be separated into two isomers, and they gave single crystals suitable for X-ray crystallographic analysis. The X-ray analyses revealed that they are 5-tosylpyrrolin-2-one derivative **26a** and its 3-tosyl isomer **26b**, respectively, as shown in Figure 2. This suggests that an isomerization proceeded during the acidic hydrolysis of the acetate **23** due to steric demand.

The separated 5-tosyl isomer of the nitro compound **26a** was allowed to react with allyl 4-oxobutanoate (**27**)[17c] to construct the nitro alcohol side chain of the 5-tosylpyrrolin-2-one derivative **28a** according to the Henry reaction. Acetylation of the resulting alcohol **28a** followed by reaction with *t*-butyl isocyanoacetate gave the dipyrrole derivative **29a** in 27% yield in three steps as shown in Scheme 5. Compound **29a** was subjected to the Vilsmeier reaction[24] to afford the formylated dipyrrole derivative **8a** in 71% yield. Compound **8a** was then

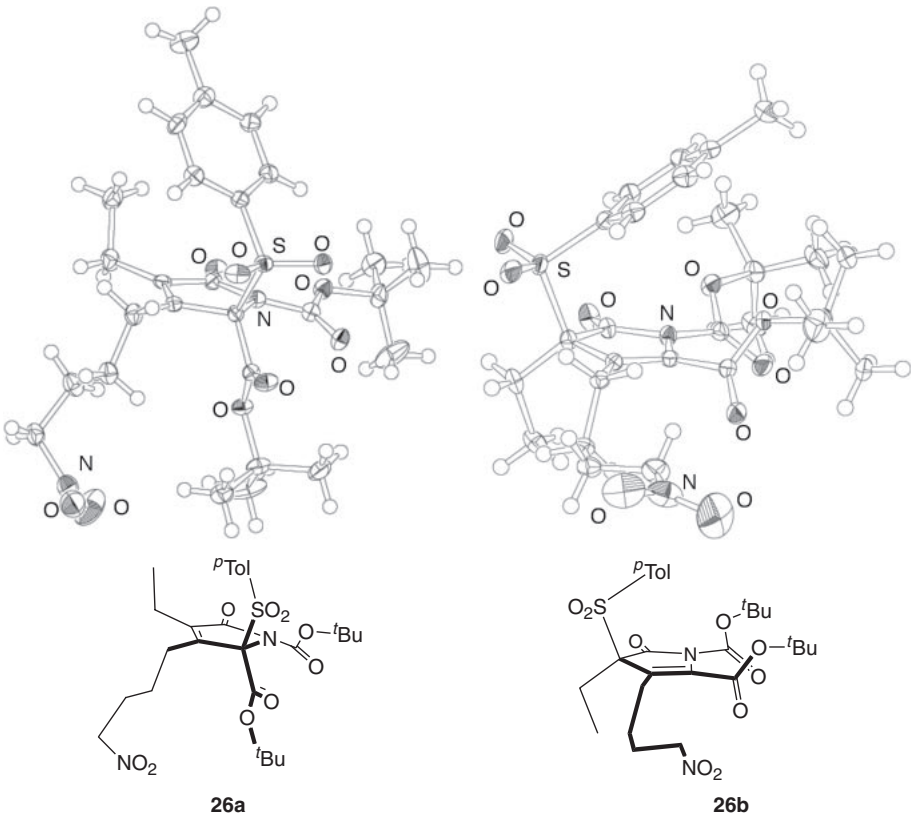



**Figure 2.**

treated with 99% formic acid to cleave the Boc and *t*-butyl ester giving a dicarboxylic acid derivative **30a**, which was subjected to our original Wittig-like coupling reaction using tri(*n*-butyl)-phosphine in the presence of DBU[13,25] to afford the pyrromethenone derivative **31** in 46% yield from **8a** in two steps. In a similar manner, the 3-tosyl isomer **26b** was converted to the formylated dipyrrole derivative **8b**. The treatment of the dipyrrole **8b** with formic acid followed by tri(*n*-butyl)phosphine and DBU gave the same pyrromethenone derivative **31** in 60% yield from **8b**.

Furthermore, the CD-ring **31** could be synthesized without the separation of the isomeric nitro compounds **26a** and **26b** in similar chemical yields (Scheme 6).

Finally, the acid **31** was converted to the corresponding aldehyde **6** via decarboxylation and formylation by treating with trimethyl orthoformate in TFA in 63% yield.

As 15*E-anti* locked CD-ring component **6** was in hand, the synthesis of sterically locked 15*E-anti* BV derivative **3** was then examined by coupling with the AB-ring component. The AB-ring component **7** of BV derivative was obtained from **9** as previously reported.[14] The coupling reaction between the 15*E-anti* CD- and AB-ring components (**6** and **7**) was carried out under acidic conditions to afford the sterically locked BV diallyl ester derivative **32** in 71% yield. The deprotection of the allyl esters was achieved by Pd(0)-catalyzed reaction in the presence of TsNa as a nucleophile to give the desired BV derivative **3** bearing a 15*E-anti* CD-ring component in 70% yield (Scheme 7).

Finally, the synthesis of sterically locked 15*E-anti* PCB **2** was examined by coupling with AB-ring component of PCB. The coupling of **6** with **4**, generated in situ from **11**, was accomplished under acidic conditions to afford the 15*E-anti* PCB derivative **33**. Final deprotection of allyl ester furnished the free acid **2** quantitatively (Scheme 8).

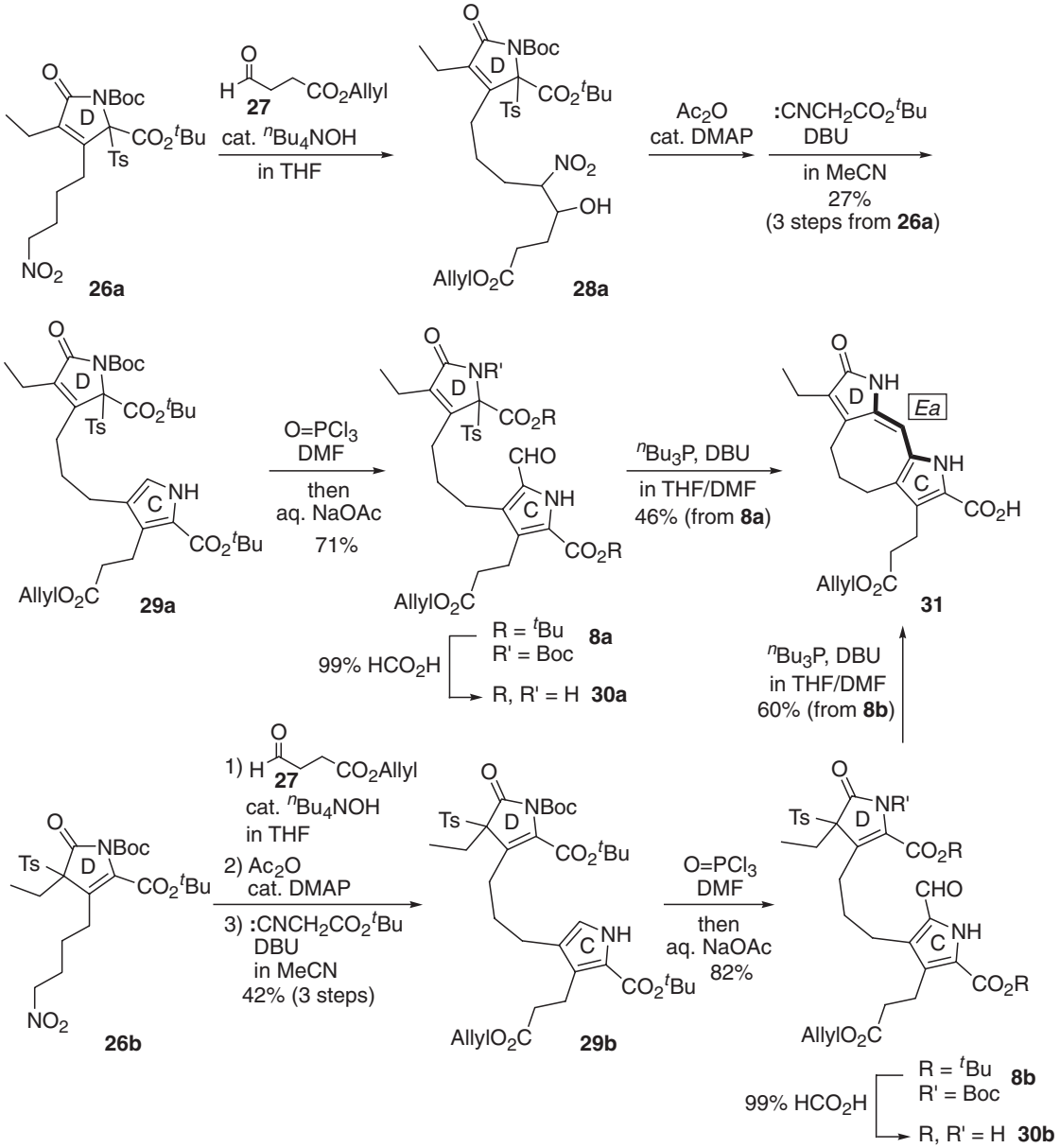
## Conclusion

As described above, total syntheses of the sterically locked 15*Z-anti* and 15*E-anti* PCB derivatives **1**, **2**, and 15*E-anti* BV derivative **3** were achieved. These sterically locked chromophores will make it possible to investigate the stereochemistry and function of phytochrome chromophores both in vitro and in vivo.

## Experimental

The $^{1}$H and $^{13}$C NMR spectra were recorded on JEOL Lambda 400, Lambda 300, and ECS 400 NMR spectrometers. The chemical shifts were determined in the $\delta$-scale relative to TMS ($\delta$ 0) as an internal standard. The IR spectra were measured on a JASCO FT/IR-230 spectrometer. The MS spectra were recorded with a JEOL SX-102A mass spectrometer. THF and $Et_2O$ were freshly distilled from sodium diphenylketyl. All other solvents were distilled and stored over drying agents. Merck silica gel 60 $PF_{254}$ (Art. 7749) was used for thin-layer chromatography (TLC) and Cica-Merck silica gel 60 (No. 9385-5B) and Cica silica gel 60N (No. 37563–84) for flash column chromatography.

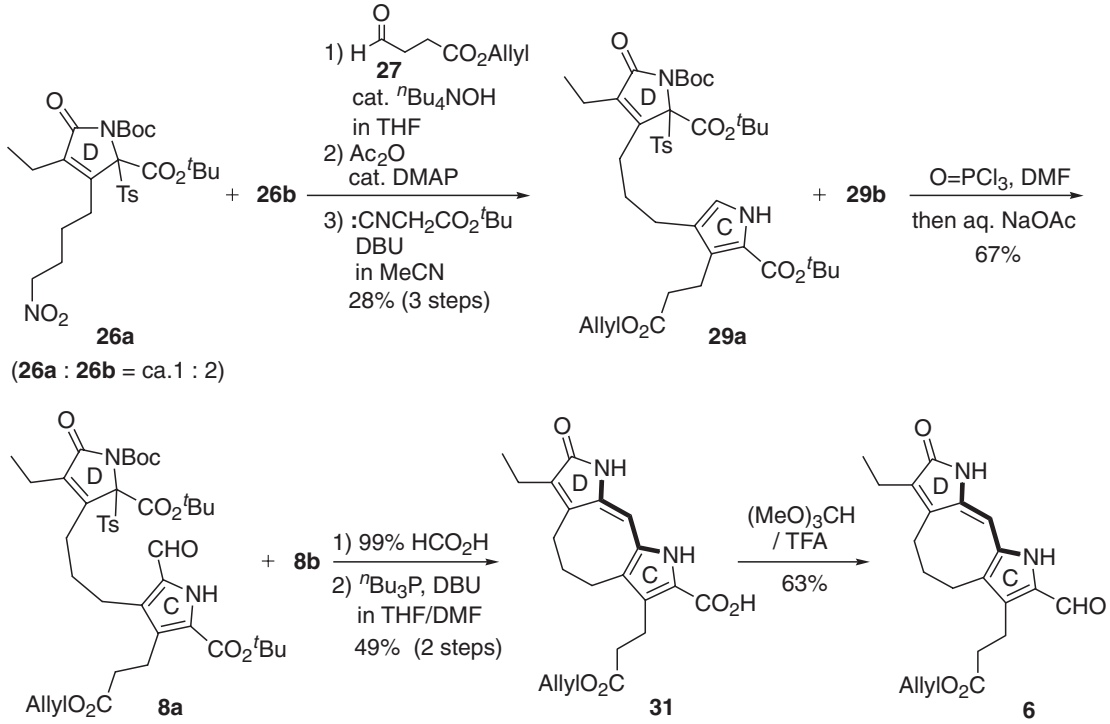
***t*-Butyl 3-(2-Allyloxycarbonylethyl)-4-methyl-5-[(4-methyl-5-oxo-3-vinyl-1*H*-pyrrol-2(5*H*)-ylidene)methyl]-1*H*-pyrrole-2-carboxylate (10).** To a solution of *t*-butyl 3-(2-allyloxycarbonylethyl)-4-methyl-5-({4-methyl-5-oxo-3-[2-(*p*-



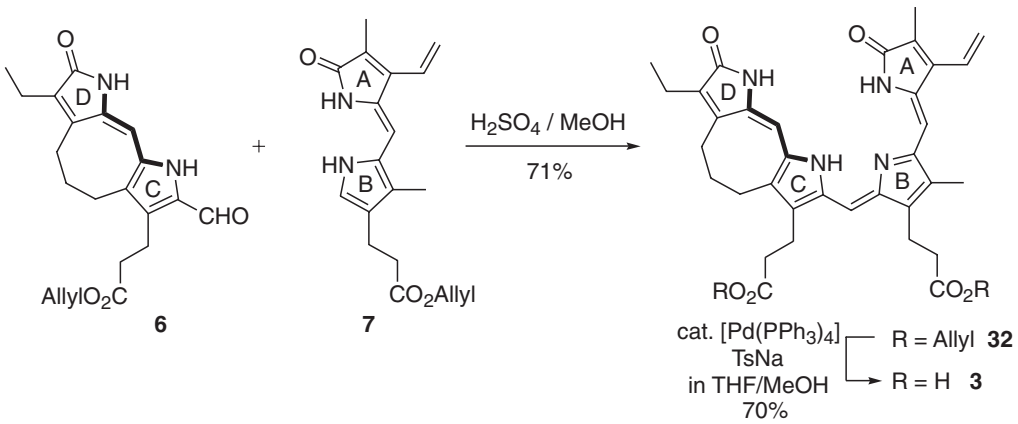

**Scheme 5.**

tolylthio)ethyl]-1*H*-pyrrol-2(5*H*)-ylidene}methyl)-1*H*-pyrrole-2-carboxylate (**9**)[14] (627 mg, 1.14 mmol) in $CH_2Cl_2$ (15 mL), a solution of *m*CPBA (70% purity, 281 mg, 1.14 mmol) in $CH_2Cl_2$ (5 mL) was added dropwise at 0 °C under a nitrogen atmosphere, and the mixture was allowed to stir for 5 min at 0 °C. The organic solvent was removed under reduced pressure, and the residue was partitioned between AcOEt and water. The organic extract was washed successively with saturated aqueous solutions of $NaHSO_3$, $NaHCO_3$, brine, and dried over $MgSO_4$. After evaporation of the solvent, the residue was dissolved in xylene (10 mL) and the solution was refluxed with stirring for 1 h. After removal of the solvent under reduced pressure, the obtained solid product was recrystallized from AcOEt/hexane to give **10**. The residue obtained by evaporation of the mother liquid was purified by flash column chromatography (hexane/AcOEt = 3/1, v/v) to give additional **10**. The total amount of **10** was 463 mg (95% yield) as a yellow solid. Mp 192.5–193.5 °C (from AcOEt). IR (KBr): 3350, 3124, 2979, 1730, 1686, 1656, 1449, 1366, 1275, 1159, 1130, 1056, 992, 915, 849, 765, 733, 688 $cm^{-1}$. $^1H$ NMR (400 MHz, $CDCl_3$): $\delta$ 1.57 (s, 9H), 2.09 (s, 6H), 2.55 (t, 2H, $J = 8.1$ Hz), 3.02 (t, 2H, $J = 8.1$ Hz), 4.59 (d, 2H, $J = 5.6$ Hz), 5.21 (dd, 1H, $J = 10.5$, 1.3 Hz), 5.30 (dd, 1H, $J = 17.1$, 1.3 Hz), 5.65 (dd, 1H, $J = 17.5$, 1.3 Hz), 5.67 (dd, 1H, $J = 11.5$, 1.3 Hz), 5.91 (ddt, 1H, $J = 17.1$, 10.5, 5.6 Hz), 6.10 (s, 1H), 6.63 (dd, 1H, $J = 17.5$, 11.5 Hz). Two N*H* protons were not observed clearly. Found: C, 67.68; H, 7.17; N, 6.46%. Calcd for $C_{24}H_{30}N_2O_5$: C, 67.58; H, 7.09; N, 6.57%.

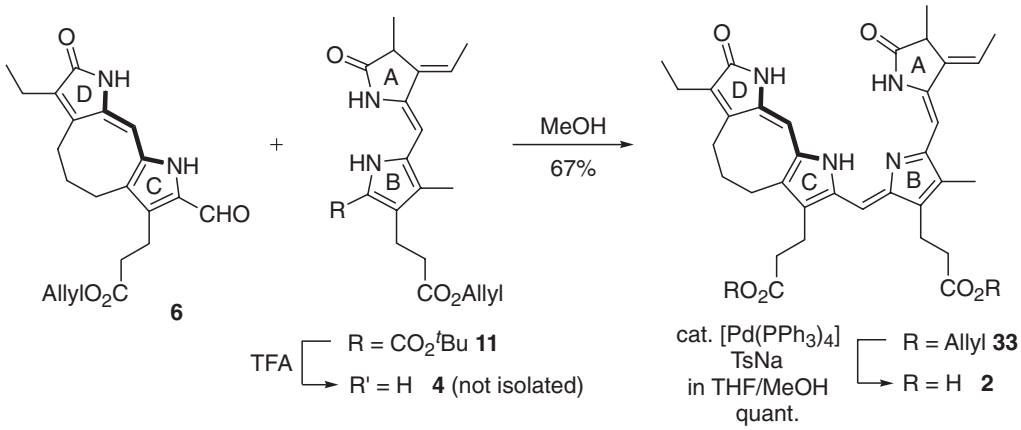
***t*-Butyl 3-(2-Allyloxycarbonylethyl)-5-{(*Z*)-[(*E*)-3-ethylidene-4-methyl-5-oxopyrrolidin-2-ylidene]methyl}-4-methyl-1*H*-pyrrole-2-carboxylate (11).** To **10** (200 mg, 0.468 mmol) in THF/$H_2O$ (2.6 mL/0.26 mL) was added aluminum amalgam,[19] which was prepared from aluminum foil (64 mg, 2.37 mmol) by the successive treatment with 1 M NaOH aq,




**Scheme 6.**




**Scheme 7.**




**Scheme 8.**

$H_2O$, THF, 0.5% $HgCl_2$ aq, $H_2O$, THF, and the resulting suspension was stirred for 2 h at room temperature. The reaction mixture was filtered through a pad of Celite. The filtrate was condensed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was washed with brine and then dried over $MgSO_4$. After evaporation of the solvent, the residue was dissolved in $CH_2Cl_2$ (2.6 mL), and *p*-TsOH·$H_2O$ (89 mg, 0.468 mmol) was added under a nitrogen atmosphere. After stirring for 5 min at room temperature, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was washed with brine and dried over $MgSO_4$, followed by evaporation of the solvent. The residue was separated by TLC on $SiO_2$ (hexane/AcOEt = 2/1, v/v) to give **11** (178 mg, 0.416 mmol) in 89% yield as a yellow solid. The spectral data were identical with those previously reported.[22,26]

**Allyl 3-{2-[1-(2-Allyloxycarbonylethyl)-6-ethyl-5-methyl-7-oxo-3,7,8,9-tetrahydro-3,7a-diazacyclopenta[*f*]azulen-2-ylmethylene]-5-(3-ethylidene-4-methyl-5-oxopyrrolidin-2-ylidenemethyl)-4-methyl-2*H*-pyrrol-3-yl}propanoate (12).** To **11** (13 mg, 0.030 mmol) and **5**[14] (8 mg, 0.022 mmol) was added $HCO_2H$ (0.14 mL) and TFA (0.07 mL) at room temperature under a nitrogen atmosphere. After stirring for 10 min, the resulting brown solution was condensed under reduced pressure. To the residue was added MeOH (5.0 mL) and the reaction mixture was stirred for 2 h. The blue reaction mixture was quenched with a phosphate buffer solution (pH 7.0) and extracted with AcOEt. The organic layer was washed with brine, and then dried over $Na_2SO_4$. The solvent was evaporated and the residue was separated by TLC on $SiO_2$ ($CHCl_3$/AcOEt/EtOH = 40/8/1, v/v/v) to afford **12** (12 mg, 0.012 mmol) in 54% yield as a blue solid. Mp 113–115 °C (from $CH_2Cl_2$/hexane). IR(KBr): 3456, 2935, 1730, 1674, 1615, 1586, 1455, 1412, 1338, 1314, 1280, 1233, 1212, 1160, 1100, 1040, 991, 953, 824, 771, 662 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 1.11 (t, 3H, $J = 7.6$ Hz), 1.45 (d, 3H, $J = 7.3$ Hz), 1.94 (d, 3H, $J = 7.1$ Hz), 2.06 (s, 3H), 2.20 (s, 3H), 2.39 (q, 2H, $J = 7.6$ Hz), 2.56 (t, 2H, $J = 7.3$ Hz), 2.57 (t, 2H, $J = 7.6$ Hz), 2.87–2.89 (m, 2H), 2.91 (t, 2H, $J = 7.6$ Hz), 2.95 (t, 2H, $J = 7.3$ Hz), 3.28 (q, 1H, $J = 7.3$ Hz), 3.96–4.02 (m, 2H), 4.56 (d, 2H, $J = 5.9$ Hz), 4.57 (d, 2H, $J = 5.9$ Hz), 5.20 (d, 1H, $J = 10.1$ Hz), 5.21 (d, 1H, $J = 10.1$ Hz), 5.27 (d, 1H, $J = 17.1$ Hz), 5.28 (d, 1H, $J = 17.1$ Hz), 5.85 (s, 1H), 5.88 (ddt, 2H, $J = 17.1$, 10.1, 5.9 Hz), 6.35 (s, 1H), 6.42 (q, 1H, $J = 7.1$ Hz), 6.64 (s, 1H). Two N*H* protons were not observed clearly. HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 679.3485. Calcd for $C_{40}H_{47}N_4O_6$: 679.3496.

**3-{2-[1-(2-Carboxyethyl)-6-ethyl-5-methyl-7-oxo-3,7,8,9-tetrahydro-3,7a-diazacyclopenta[*f*]azulen-2-ylmethylene]-5-(3-ethylidene-4-methyl-5-oxopyrrolidin-2-ylidenemethyl)-4-methyl-2*H*-pyrrol-3-yl}propanoic Acid (1).** To a mixed solution of **12** (27 mg, 0.045 mmol) and $[Pd(PPh_3)_4]$ (10 mg, 0.009 mmol) in THF (1.0 mL), a solution of TsNa (16 mg, 0.090 mmol) in MeOH (1.0 mL) was added under a nitrogen atmosphere at room temperature. After stirring for 10 min, the reaction mixture was quenched by the addition of thiourea (3 mg, 0.036 mmol). The solution was directly separated by flash column chromatography ($SiO_2$, $CHCl_3$/MeOH/AcOH = 250/15/1, v/v/v). The blue fraction was evaporated and the resulting solid residue was recrystallized from $CHCl_3$/hexane. Free acid **1**: a blue solid, 19 mg (0.034 mmol), in 76% yield. Mp >280 °C (from $CHCl_3$/hexane). IR (KBr): 3213, 2926, 1778, 1694, 1599, 1454, 1395, 1277, 1159, 1064, 963, 895, 824 $cm^{-1}$. $^1H$ NMR ($C_5D_5N$): $\delta$ 1.10 (t, 3H, $J = 7.6$ Hz), 1.38 (d, 3H, $J = 7.6$ Hz), 1.70 (d, 3H, $J = 7.3$ Hz), 2.12 (s, 3H), 2.13 (s, 3H), 2.40 (q, 2H, $J = 7.6$ Hz), 2.84 (t, 2H, $J = 7.1$ Hz), 2.87–2.92 (m, 4H), 3.14–3.20 (m, 4H), 3.17 (q, 1H, $J = 7.6$ Hz), 4.06–4.10 (m, 2H), 6.06 (s, 1H), 6.32 (q, 1H, $J = 7.3$ Hz), 6.78 (s, 1H), 7.41 (s, 1H). Two N*H* protons and two COO*H* protons were not observed clearly. UV–vis (MeOH) $\lambda_{max}$ 365 ($\varepsilon = 21100$), 608 ($\varepsilon = 72000$) nm. HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 599.2881. Calcd for $C_{34}H_{39}N_4O_6$: 599.2870.

***t*-Butyl 3-(4-Acetoxybutyl)-4-ethyl-1*H*-pyrrole-2-carboxylate (19).** To a mixture of $H_2O$ (30 mL) and concd HCl (2.5 mL), 3,4-dihydro-2*H*-pyran (**13**) (10.046 g, 0.120 mol) was added at room temperature. After stirring for 40 min, the mixture was neutralized with a saturated aqueous solution of $NaHCO_3$ and then extracted with $Et_2O$. The organic extract was dried over $MgSO_4$ and the solvent was evaporated to give a colorless oil of a mixture of tetrahydropyran-2-ol (**14**) and 5-hydroxypentanal (**15**) (5.570 g). To the mixture of **14** and **15** (5.570 g, 0.055 mol) and 1-nitropropane (9.730 g, 0.109 mol) was added KOH (0.616 g, 0.011 mol) in MeOH (11 mL) dropwise at 0 °C. After stirring overnight at room temperature, the reaction mixture was neutralized by adding 1 M HCl and the organic solvent was removed under reduced pressure. The residue was partitioned between AcOEt and water, and the organic extract was successively washed with a saturated aqueous solution of $NaHCO_3$, brine, and dried over $MgSO_4$. The solvent was evaporated to give the nitro alcohol **16** (9.033 g) as a yellow oil. To a mixture of **16** (9.033 g, 0.047 mol) and DMAP (1.160 g, 9.5 mmol) in THF (10 mL), $Ac_2O$ (9.8 mL, 0.104 mol) was added dropwise at 0 °C. After stirring at room temperature for 3 h, the reaction mixture was quenched by adding MeOH and the solvent was removed under reduced pressure. The residue was partitioned between AcOEt and water and the organic extract was successively washed with a saturated aqueous solution of $NaHCO_3$, brine, and dried over $Na_2SO_4$. The solvent was evaporated to give an oil of a mixture of 6-nitrooctane-1,5-diyl diacetate (**17**) and 6-nitrooct-5-enyl acetate (**18**) (12.285 g). To a solution of *t*-butyl isocyanoacetate (3.133 g, 22.22 mmol) in THF (10 mL), DBU (7.435 g, 48.914 mmol) was added at 0 °C under a nitrogen atmosphere, followed by dropwise addition of the mixture of **17** and **18** (12.285 g) in THF (5 mL).[21] After stirring overnight at room temperature, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was successively washed with 1 M HCl, a saturated aqueous solution of $NaHCO_3$, brine, and then dried over $Na_2SO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 6/1, v/v) to give **19** (4.764 g, 26% yield in 4 steps) as an oil. IR (neat): 3320, 2968, 2934, 2868, 2360, 2341, 1739, 1684, 1566, 1552, 1505, 1457, 1406, 1367, 1317, 1246, 1140, 1113, 1083, 1059, 941, 907, 840, 785, 745 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 1.18 (t, 3H, $J = 7.5$ Hz), 1.54–1.74 (m, 4H), 1.57 (s,

9H), 2.04 (s, 3H), 2.43 (q, 2H, $J = 7.5$ Hz), 2.72 (t, 2H, $J = 7.9$ Hz), 4.08 (t, 2H, $J = 6.4$ Hz), 6.65 (d, 1H, $J = 2.7$ Hz), 9.32 (s, 1H). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 310.2018. Calcd for $C_{17}H_{28}NO_4$: 310.2038.

***t*-Butyl 3-(4-Acetoxybutyl)-4-ethyl-5-iodo-1*H*-pyrrole-2-carboxylate (20).** To a solution of compound **19** (3.40 g, 11 mmol) in acetone (50 mL), *N*-iodosuccinimide (2.97 g, 13.2 mmol) was added at room temperature under a nitrogen atmosphere.[22] After stirring for 1 h, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was successively washed with saturated aqueous solutions of $NaHSO_3$, $NaHCO_3$, brine, and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 7/1, v/v) to give **20** (4.740 g, 99% yield) as an oil. IR (neat): 3457, 3296, 2965, 2869, 1738, 1665, 1557, 1476, 1457, 1402, 1367, 1318, 1242, 1169, 1139, 1109, 1058, 948, 860, 846, 779 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 1.07 (t, 3H, $J = 7.5$ Hz), 1.56–1.75 (m, 4H), 1.57 (s, 9H), 2.06 (s, 3H), 2.40 (q, 2H, $J = 7.5$ Hz), 2.75 (t, 2H, $J = 7.8$ Hz), 4.09 (t, 2H, $J = 6.2$ Hz), 9.20 (s, 1H). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 436.0985. Calcd for $C_{17}H_{27}INO_4$: 436.0998.

***t*-Butyl 2-Acetoxy-3-(4-acetoxybutyl)-4-ethyl-5-oxo-2,5-dihydro-1*H*-pyrrole-2-carboxylate (21).** A mixed solution **20** (4.25 g, 9.76 mmol) and lead tetraacetate [$Pb(OAc)_4$, 6.49 g, 14.6 mmol] in toluene (30 mL) was stirred at room temperature for 2 d.[22] The reaction mixture was filtered through a pad of Celite and the filtrate was partitioned between AcOEt and water. The organic extract was successively washed with saturated aqueous solutions of $NaHSO_3$, $NaHCO_3$, brine, and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 4/1, v/v) to give **21** (3.405 g, 91% yield) as an oil. IR (neat): 3382, 2977, 2938, 2876, 2360, 2342, 1727, 1459, 1370, 1241, 1156, 1117, 1048, 841, 819, 782 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 1.11 (t, 3H, $J = 7.5$ Hz), 1.45 (s, 9H), 1.50–1.73 (m, 4H), 2.05 (s, 3H), 2.14 (s, 3H), 2.30 (q, 2H, $J = 7.5$ Hz), 2.34–2.49 (m, 2H), 4.10 (t, 2H, $J = 6.2$ Hz), 6.77 (s, 1H). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 384.2022. Calcd for $C_{19}H_{30}NO_7$: 384.2038.

***t*-Butyl 3-(4-Acetoxybutyl)-4-ethyl-5-oxo-2-tosyl-2,5-dihydro-1*H*-pyrrole-2-carboxylate (22).** A mixture of compound **21** (1.99 g, 5.19 mmol) and anhydrous TsNa (1.938 g, 10.89 mmol) in THF (50 mL) was refluxed for 1.5 h under a nitrogen atmosphere. The solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was washed with brine and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 4/1, v/v) to give **22** (2.345 g, 94% yield) as an oil. IR (neat): 3580–3200, 2978, 2940, 2880, 2360, 2340, 1728, 1710, 1578, 1459, 1367, 1321, 1278, 1243, 1154, 1082, 1065, 840, 816, 710 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.72 (t, 3H, $J = 7.5$ Hz), 1.59 (s, 9H), 1.67–1.82 (m, 2H), 1.94–2.35 (m, 4H), 2.05 (s, 3H), 2.41 (s, 3H), 2.49 (dt, 1H, $J = 13.4$, 4.4 Hz), 3.02 (dt, 1H, $J = 13.4$, 4.4 Hz), 4.09–4.16 (m, 2H), 6.62 (s, 1H), 7.29 (d, 2H, $J = 8.6$ Hz), 7.66 (d, 2H, $J = 8.6$ Hz). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 480.2056. Calcd for $C_{24}H_{34}NO_7S$: 480.2074.

**Di-*t*-butyl 3-(4-Acetoxybutyl)-4-ethyl-5-oxo-2-tosyl-1*H*-pyrrole-1,2(2*H*,5*H*)-dicarboxylate (23).** To a mixture of **22** (17.64 g, 42 mmol) and di-*t*-butyl dicarbonate ($Boc_2O$, 13.75 g, 63 mol) in MeCN (100 mL), DMAP (1.03 g, 8.4 mmol) in MeCN (10 mL) was added dropwise at −40 °C under a nitrogen atmosphere. After stirring for 30 min at room temperature, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was washed with brine and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 1/1, v/v) to give **23** (18.78 g, 86% yield) as a white solid. Mp 102–104 °C (from $Et_2O$/hexane). IR (KBr): 2979, 2940, 2880, 1766, 1739, 1647, 1597, 1464, 1369, 1314, 1285, 1254, 1232, 1214, 1154, 1086, 1043, 1006, 976, 950, 843, 819, 796, 778, 742, 706, 661 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.81 (t, 3H, $J = 7.5$ Hz), 1.48 (s, 18H), 1.64–1.78 (m, 4H), 2.05 (s, 3H), 2.21–2.33 (m, 1H), 2.39–2.66 (m, 3H), 2.43 (s, 3H), 4.10 (t, 2H, $J = 6.1$ Hz), 7.29 (d, 2H, $J = 8.3$ Hz), 7.65 (d, 2H, $J = 8.3$ Hz). Found: C, 60.03; H, 6.98; N, 2.50%. Calcd for $C_{29}H_{41}NO_9S$: C, 60.08; H, 7.13; N, 2.42%.

**Di-*t*-butyl 4-Ethyl-3-(4-hydroxybutyl)-5-oxo-2-tosyl-1*H*-pyrrole-1,2(2*H*,5*H*)-dicarboxylate (24a) and Di-*t*-butyl 4-Ethyl-3-(4-hydroxybutyl)-5-oxo-4-tosyl-1*H*-pyrrole-1,2-(4*H*,5*H*)-dicarboxylate (24b).** To compound **23** (18.78 g, 36 mmol) was added 0.5 M methanolic HCl (229 mL) and the solution was stirred overnight at room temperature. The mixture was neutralized with a saturated aqueous solution of $NaHCO_3$ and the solvent was removed under reduced pressure. The residue was partitioned between AcOEt and water and the organic extract was washed with brine and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 4/1, 3/1, 2/1, 1/1, v/v) to give a ca. 1:2 mixture of **24a** and **24b** (16.08 g, 97% yield) as an oil. Analytical samples of **24a** and **24b** were obtained by further separation of a part of the mixture by flash column chromatography.

**24a**: a white soild. Mp 87 °C (from AcOEt). IR (KBr): 3555, 2980, 2942, 2868, 1783, 1729, 1623, 1595, 1460, 1395, 1371, 1321, 1281, 1254, 1226, 1153, 1083, 1013, 960, 847, 815, 795, 764, 706, 658 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.91 (t, 3H, $J = 7.4$ Hz), 1.50 (s, 9H), 1.52 (s, 9H), 1.55–1.70 (m, 5H), 2.12–2.29 (m, 2H), 2.40 (s, 3H), 2.55–2.68 (m, 1H), 2.75–2.89 (m, 1H), 3.66–3.76 (m, 2H), 7.26 (d, 2H, $J = 8.3$ Hz), 7.70 (d, 2H, $J = 8.3$ Hz). Found: C, 60.39; H, 7.22; N, 2.66%. Calcd for $C_{27}H_{39}NO_8S$: C, 60.31; H, 7.31; N, 2.61%.

**24b**: an oil. IR (neat): 3559, 2980, 2936, 2877, 1791, 1730, 1628, 1597, 1458, 1393, 1369, 1322, 1254, 1211, 1152, 1084, 1006, 944, 846, 817, 795, 759, 706, 661 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.81 (t, 3H, $J = 7.8$ Hz), 1.47 (s, 9H), 1.48 (s, 9H), 1.61–1.86 (m, 5H), 2.24–2.34 (m, 1H), 2.43 (s, 3H), 2.46–2.52 (m, 3H), 3.68–3.75 (m, 2H), 7.30 (d, 2H, $J = 8.2$ Hz), 7.66 (d, 2H, $J = 8.2$ Hz). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 538.2469. Calcd for $C_{27}H_{40}NO_8S$: 538.2474.

**Di-*t*-butyl 4-Ethyl-3-(4-iodobutyl)-5-oxo-2-tosyl-1*H*-pyrrole-1,2(2*H*,5*H*)-dicarboxylate (25a) and Di-*t*-butyl 4-Ethyl-3-(4-iodobutyl)-5-oxo-4-tosyl-1*H*-pyrrole-1,2(4*H*,5*H*)-dicarboxylate (25b).** To a ca. 1:2 mixture of **24a** and **24b** (9.240 g, 20.0 mmol) was added $Ph_3P$ (6.295 g, 24.0 mmol), $I_2$ (6.096 g,

24.0 mmol), and imidazole (3.404 g, 50.0 mmol) in MeCN (50 mL) at room temperature under a nitrogen atmosphere. After stirring for 1 h, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was successively washed with a saturated aqueous solution of $NaHCO_3$ and brine, and then dried over $Na_2SO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 4/1, 3/1, 2/1, 1/1, v/v) to give a ca. 1:2 mixture of **25a** and **25b** (10.195 g, 89% yield) as an oil. Analytical samples of the iodides were obtained by further separation of a part of the mixture by flash column chromatography.

**25a**: an oil. IR (neat): 2979, 2935, 2877, 2257, 1782, 1748, 1658, 1596, 1458, 1395, 1370, 1305, 1255, 1151, 1083, 1045, 983, 912, 874, 817, 779, 733 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.94 (t, 3H, $J = 7.6$ Hz), 1.50 (s, 9H), 1.53 (s, 9H), 1.60–1.96 (m, 4H), 2.13–2.84 (m, 4H), 2.40 (s, 3H), 3.22 (t, 2H, $J = 6.6$ Hz), 7.29 (d, 2H, $J = 8.3$ Hz), 7.70 (d, 2H, $J = 8.3$ Hz). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 648.1495. Calcd for $C_{27}H_{39}NO_7SI$: 648.1492.

**25b**: an oil. IR (neat): 2981, 2938, 2879, 1760, 1742, 1721, 1641, 1596, 1460, 1392, 1369, 1353, 1327, 1304, 1257, 1221, 1201, 1151, 1080, 1003, 940, 871, 842, 811, 779, 732, 675 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.83 (t, 3H, $J = 7.6$ Hz), 1.47 (s, 9H), 1.48 (s, 9H), 1.64–1.95 (m, 4H), 2.24–2.67 (m, 4H), 2.43 (s, 3H), 3.22 (t, 2H, $J = 7.1$ Hz), 7.30 (d, 2H, $J = 8.2$ Hz), 7.65 (d, 2H, $J = 8.2$ Hz). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 648.1495. Calcd for $C_{27}H_{39}NO_7SI$: 648.1492.

**Di-*t*-butyl 4-Ethyl-3-(4-nitrobutyl)-5-oxo-2-tosyl-1*H*-pyrrole-1,2(2*H*,5*H*)-dicarboxylate (26a) and Di-*t*-butyl 4-Ethyl-3-(4-nitrobutyl)-5-oxo-4-tosyl-1*H*-pyrrole-1,2(4*H*,5*H*)-dicarboxylate (26b).** To ca. 1:2 mixture of the iodides **25a** and **25b** (8.64 g, 15 mmol), $NaNO_2$ (2.14 g, 31 mmol), and phloroglucinol (2.76 g, 17 mmol), DMF (100 mL) was added under a nitrogen atmosphere.[23] After stirring overnight at room temperature, the mixture was partitioned between AcOEt and water. The organic extract was washed with brine and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 4/1, v/v) to give a ca. 1:2 mixture of **26a** and **26b** (4.64 g, 60% yield) as an oil. Analytical samples of **26a** and **26b** were obtained by further separation of a part of the mixture by flash column chromatography. Their structures were confirmed by X-ray crystallographic analysis.

**26a**: a white solid. Mp 117–120 °C (from $Et_2O$/hexane). IR (KBr): 2980, 2937, 2878, 1785, 1748, 1658, 1596, 1552, 1460, 1372, 1332, 1304, 1255, 1149, 1083, 1046, 1028, 941, 873, 820, 779, 713, 655 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.93 (t, 3H, $J = 7.6$ Hz), 1.47 (s, 9H), 1.48 (s, 9H), 1.58–2.29 (m, 6H), 2.59–2.88 (m, 2H), 2.41 (s, 3H), 4.44 (t, 2H, $J = 6.9$ Hz), 7.27 (d, 2H, $J = 8.2$ Hz), 7.70 (d, 2H, $J = 8.2$ Hz). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 567.2368. Calcd for $C_{27}H_{39}N_2O_9S$: 567.2376. Crystal data: $C_{27}H_{38}N_2O_9S$, FW 566.67, orthorhombic, $Pca2_1$, $a = 9.567(2)$ Å, $b = 15.836(3)$ Å, $c = 19.335(3)$ Å, $V = 2929.3(9)$ Å$^3$, $Z = 4$, $D_{calcd} = 1.285$ g cm$^{-3}$, $R = 0.044$ ($R_w = 0.081$) for 6470 reflections with $I > 3.00\sigma(I)$ and 352 variable parameters.

**26b**: a white solid. Mp 98–100 °C (from $Et_2O$/hexane). IR (KBr): 2980, 2938, 2875, 1792, 1760, 1728, 1596, 1551, 1457, 1368, 1321, 1252, 1210, 1151, 1085, 1005, 943, 843, 817, 775, 705, 661 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 0.78 (t, 3H, $J = 7.6$ Hz), 1.47 (s, 9H), 1.49 (s, 9H), 1.68–2.16 (m, 4H), 2.21–2.69 (m, 4H), 2.42 (s, 3H), 4.44 (t, 2H, $J = 6.9$ Hz), 7.31 (d, 2H, $J = 8.2$ Hz), 7.64 (d, 2H, $J = 8.2$ Hz). Found: C, 57.24; H, 6.80; N, 4.92%. Calcd for $C_{27}H_{38}N_2O_9S$: C, 57.23; H, 6.76; N, 4.94%. Crystal data: $C_{27}H_{38}N_2O_9S$, FW 566.67, triclinic, $P\bar{1}$, $a = 16.601(2)$ Å, $b = 18.798(3)$ Å, $c = 21.8398(3)$ Å, $\alpha = 66.44(3)°$, $\beta = 71.18(2)°$, $\gamma = 84.41(3)°$, $V = 5909(1)$ Å$^3$, $Z = 8$, $D_{calcd} = 1.274$ g cm$^{-3}$, $R = 0.046$ ($R_w = 0.066$) for 19815 reflections with $I > 3.00\sigma(I)$ and 1404 variable parameters.

Crystallographic data have been deposited with Cambridge Crystallographic Data Centre: Deposition numbers CCDC-789497 and -789498 for compounds **26a** and **26b**, respectively. Copies of the data can be obtained free of charge via http://www.ccdc.cam.ac.uk/conts/retrieving.html (or from the Cambridge Crystallographic Data Centre, 12, Union Road, Cambridge, CB2 1EZ, U.K.; Fax: +44 1223 336033; e-mail: deposit@ccdc.cam.ac.uk).

**Di-*t*-butyl 3-{3-[4-(2-Allyloxycarbonylethyl)-5-(*t*-butoxycarbonyl)-1*H*-pyrrol-3-yl]propyl}-4-ethyl-5-oxo-2-tosyl-1*H*-pyrrole-1,2(2*H*,5*H*)-dicarboxylate (29a) and Di-*t*-butyl 3-{3-[4-(2-Allyloxycarbonylethyl)-5-(*t*-butoxycarbonyl)-1*H*-pyrrol-3-yl]propyl}-4-ethyl-5-oxo-4-tosyl-1*H*-pyrrole-1,2(4*H*,5*H*)-dicarboxylate (29b). Step 1**: To a ca. 1:2 mixture of **26a** and **26b** (625 mg, 1.245 mmol) and allyl 4-oxobutanoate (**27**)[17c] (353 mg, 2.490 mol), 0.1 M $^nBu_4NOH$ in THF (3.7 mL, 0.37 mmol) was added dropwise at 0 °C. After stirring for 4 h at room temperature, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic layer was successively washed with 1 M HCl, a saturated aqueous solution of $NaHCO_3$, brine, and dried over $MgSO_4$. The solvent was evaporated and the residue was separated by TLC on $SiO_2$ (hexane/AcOEt = 3/1, v/v) to give a mixture of crude nitro alcohols **28a** and **28b** (600 mg) as an oil.

**Step 2**: To the mixture of **28a** and **28b** (total 600 mg, ca. 0.811 mmol) and DMAP (20 mg, 0.162 mmol) was added $Ac_2O$ (91 mg, 0.892 mmol) dropwise at 0 °C under a nitrogen atmosphere. After stirring for 2 h at room temperature, the reaction mixture was quenched by adding MeOH and the solvent was removed under reduced pressure. The residue was partitioned between AcOEt and water, and the organic extract was successively washed with a saturated aqueous solution of $NaHCO_3$, brine, and dried over $MgSO_4$. The solvent was evaporated and the product was separated by TLC on $SiO_2$ (hexane/AcOEt = 2/1, v/v) to give a mixture of acetates of **28a** and its isomer **28b** and the corresponding nitro olefins (570 mg) as an oil.

**Step 3**: To a solution of *t*-butyl isocyanoacetate (321 mg, 2.280 mmol) in MeCN (10 mL) at −40 °C under a nitrogen atmosphere, DBU (578 mg, 3.8 mmol) was added, followed by dropwise addition of the mixture of nitro acetates and the corresponding nitro olefins obtained above (total 570 mg) in THF (2 mL).[21] After stirring overnight at room temperature, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water. The organic extract was successively washed with saturated aqueous solutions of

$NaHSO_3$, $NaHCO_3$, brine, and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by TLC on $SiO_2$ (hexane/AcOEt = 2/1, v/v) to give a ca. 1:2 mixture of **29a** and **29b** (268 mg) as an oil. Total yield over these 3 steps was 28%.

In a similar manner, 2-tosyl isomer **29a** was prepared from the isolated **26a**: The nitro compound **26a** (200 mg, 0.398 mmol), allyl 4-oxobutanoate (**27**) (113 mg, 0.796 mmol), and 0.1 M $^n$Bu$_4$NOH in THF (1.2 mL, 0.12 mmol) gave nitro alcohol **28a** (195 mg, 0.264 mmol) in Step 1. The nitro alcohol **28a** (50 mg, 0.068 mmol), DMAP (2 mg, 0.014 mmol), and $Ac_2O$ (91 mg, 0.892 mmol) afforded a mixture of the acetate of **28a** and the corresponding nitro olefin (total 27 mg) in Step 2. The mixture of the nitro acetate and the corresponding nitro olefin (total 27 mg), *t*-butyl isocyanoacetate (15 mg, 0.108 mmol), and DBU (27 mg, 0.036 mmol) afforded **29a** (23 mg, 0.028 mmol) in Step 3. Total yield over these 3 steps was 27%.

Furthermore, the isomer **29b** was prepared from the isolated **26b**: The nitro compound **26b** (200 mg, 0.398 mmol), allyl 4-oxobutanoate (**27**) (113 mg, 0.796 mmol), and 0.1 M $^n$Bu$_4$NOH in THF (1.2 mL, 0.12 mmol) gave **28b** (187 mg, 0.253 mmol) in Step 1. The nitro alcohol **28b** (76 mg, 0.103 mmol), DMAP (2 mg, 0.014 mmol), and $Ac_2O$ (12 mg, 0.113 mmol) afforded a mixture of the acetate of **28b** and the corresponding nitro olefin (total 62 mg) in Step 2. The mixture of the nitro acetate and the corresponding nitro olefin (total 62 mg), *t*-butyl isocyanoacetate (35 mg, 0.250 mmol), and DBU (63 mg, 0.420 mmol) produced **29b** (51 mg, 0.063 mmol) in Step 3. Total yield over these 3 steps was 42%.

**29a**: an oil. IR (neat): 3378, 3318, 2979, 2936, 1791, 1733, 1689, 1597, 1565, 1456, 1402, 1369, 1324, 1281, 1254, 1213, 1153, 1086, 1058, 1001, 941, 843, 816, 757, 706, 662 cm$^{-1}$. $^1$H NMR ($CDCl_3$): $\delta$ 0.87 (t, 3H, $J$ = 7.6 Hz), 1.45 (s, 9H), 1.51 (s, 9H), 1.54 (s, 9H), 1.70–1.81 (m, 2H), 2.02–2.20 (m, 2H), 2.34–2.66 (m, 5H), 2.40 (s, 3H), 2.79–2.86 (m, 1H), 3.00 (d, 2H, $J$ = 8.2 Hz), 4.56 (d, 2H, $J$ = 5.5 Hz), 5.20 (d, 1H, $J$ = 10.8 Hz), 5.28 (d, 1H, $J$ = 17.2 Hz), 5.91 (ddt, 1H, $J$ = 17.2, 10.8, 5.5 Hz), 6.67 (d, 1H, $J$ = 2.8 Hz), 7.25 (d, 2H, $J$ = 8.3 Hz), 7.68 (d, 2H, $J$ = 8.3 Hz), 8.92 (brs, 1H). HRMS (FAB$^+$) ($M^+ + 1$), Found: $m/z$ 785.3684. Calcd for $C_{41}H_{57}N_2O_{11}S$: 785.3684.

**29b**: an oil. IR (neat): 3378, 3318, 2979, 2936, 1791, 1733, 1689, 1597, 1565, 1456, 1402, 1369, 1324, 1281, 1254, 1213, 1153, 1086, 1058, 1001, 941, 843, 816, 757, 706, 662 cm$^{-1}$. $^1$H NMR ($CDCl_3$): $\delta$ 0.80 (t, 3H, $J$ = 7.6 Hz), 1.42 (s, 9H), 1.45 (s, 9H), 1.54 (s, 9H), 1.78–1.98 (m, 2H), 2.18–2.28 (m, 1H), 2.36–2.45 (m, 1H), 2.43 (s, 3H), 2.47–2.66 (m, 6H), 3.02 (t, 2H, $J$ = 8.0 Hz), 4.59 (d, 2H, $J$ = 5.5 Hz), 5.22 (d, 1H, $J$ = 10.1 Hz), 5.30 (d, 1H, $J$ = 17.4 Hz), 5.89 (ddt, 1H, $J$ = 17.4, 10.1, 5.5 Hz), 6.73 (d, 1H, $J$ = 2.7 Hz), 7.29 (d, 2H, $J$ = 8.3 Hz), 7.65 (d, 2H, $J$ = 8.3 Hz), 8.89 (brs, 1H). (FAB$^+$) ($M^+ + 1$), Found: $m/z$ 785.3684. Calcd for $C_{41}H_{57}N_2O_{11}S$: 785.3684.

**Di-*t*-butyl 3-{3-[4-(2-Allyloxycarbonylethyl)-5-(*t*-butoxycarbonyl)-2-formyl-1*H*-pyrrol-3-yl]propyl}-4-ethyl-5-oxo-2-tosyl-1*H*-pyrrole-1,2(2*H*,5*H*)-dicarboxylate (8a) and Di-*t*-butyl 3-{3-[4-(2-Allyloxycarbonylethyl)-5-(*t*-butoxycarbonyl)-2-formyl-1*H*-pyrrol-3-yl]propyl}-4-ethyl-5-oxo-4-tosyl-1*H*-pyrrole-1,2(4*H*,5*H*)-dicarboxylate (8b).** To DMF (30 mL) was added $POCl_3$ (781 mg, 5.094 mmol) dropwise at room temperature and the mixture was stirred for 30 min under a nitrogen atmosphere.[24] A solution of a ca. 1:2 mixture of **29a** and **29b** (2.560 g, 3.208 mmol) in DMF (2 mL) was then added dropwise at 0 °C and the reaction mixture was stirred for 10 min at 0 °C and then for 2 h at 65 °C. The reaction mixture was quenched by addition of a 10% aqueous NaOAc solution (52 mL) with stirring for 2 h at 65 °C. The mixture was partitioned between AcOEt and water, and the organic extract was washed with brine, and then dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 3/1, v/v) to give a ca. 1:2 mixture of aldehydes **8a** and **8b** (1.780 g, 67% yield) as an oil.

In a similar manner, the reaction of **29a** (150 mg, 0.188 mmol) with $POCl_3$ (46 mg, 0.301 mmol) and DMF (total 1.8 mL) gave **8a** (110 mg, 71% yield). Furthermore, the reaction of **29b** (150 mg, 0.188 mmol) with $POCl_3$ (46 mg, 0.301 mmol) and DMF (total 1.8 mL) gave **8b** (127 mg, 82% yield).

**8a**: a white solid. Mp 59 °C (from AcOEt). IR (KBr): 3299, 2979, 2937, 1783, 1746, 1668, 1596, 1550, 1460, 1393, 1370, 1303, 1256, 1153, 1082, 1046, 989, 932, 843, 816, 783, 710, 665 cm$^{-1}$. $^1$H NMR ($CDCl_3$): $\delta$ 0.90 (t, 3H, $J$ = 7.3 Hz), 1.44 (s, 9H), 1.50 (s, 9H), 1.58 (s, 9H), 1.75–1.95 (m, 2H), 2.09–2.23 (m, 2H), 2.40 (s, 3H), 2.60–2.89 (m, 6H), 3.01 (t, 2H, $J$ = 8.2 Hz), 4.55 (d, 2H, $J$ = 6.0 Hz), 5.20 (d, 1H, $J$ = 10.8 Hz), 5.27 (d, 1H, $J$ = 17.6 Hz), 5.87 (ddt, 1H, $J$ = 17.6, 10.8, 6.0 Hz), 7.26 (d, 2H, $J$ = 8.2 Hz), 7.70 (d, 2H, $J$ = 8.2 Hz), 9.51 (brs, 1H), 9.76 (s, 1H). HRMS (FAB$^+$) ($M^+ + 1$), Found: $m/z$ 813.3623. Calcd for $C_{42}H_{57}N_2O_{12}S$: 813.3633.

**8b**: a white solid. Mp 60 °C (from AcOEt). IR (KBr): 3309, 2979, 2937, 1790, 1737, 1663, 1597, 1550, 1459, 1370, 1324, 1255, 1208, 1152, 1086, 1051, 999, 930, 844, 816, 706, 661 cm$^{-1}$. $^1$H NMR ($CDCl_3$): $\delta$ 0.76 (t, 3H, $J$ = 7.3 Hz), 1.43 (s, 9H), 1.47 (s, 9H), 1.58 (s, 9H), 1.80–2.05 (m, 2H), 2.22–2.66 (m, 6H), 2.43 (s, 3H), 2.85 (t, 2H, $J$ = 7.8 Hz), 3.03 (t, 2H, $J$ = 7.6 Hz), 4.56 (d, 2H, $J$ = 6.0 Hz), 5.19 (d, 1H, $J$ = 10.0 Hz), 5.27 (d, 1H, $J$ = 17.2 Hz), 5.87 (ddt, 1H, $J$ = 17.2, 10.0, 6.0 Hz), 7.30 (d, 2H, $J$ = 8.2 Hz), 7.63 (d, 2H, $J$ = 8.2 Hz), 9.48 (brs, 1H), 9.80 (s, 1H). HRMS (FAB$^+$) ($M^+ + 1$), Found: $m/z$ 813.3627. Calcd for $C_{42}H_{57}N_2O_{12}S$: 813.3633.

**1-(2-Allyloxycarbonylethyl)-7-ethyl-6-oxo-3,5,6,8,9,10-hexahydro-3,5-diazadicyclopenta[*a*,*d*]cyclooctene-2-carboxylic Acid (31).** A ca. 1:2 mixture of **8a** and **8b** (1.780 g, 2.160 mmol) was dissolved in 99% formic acid (10.8 mL) at 10 °C under a nitrogen atmosphere. After stirring for 5 h at room temperature, the solvent was removed under reduced pressure and the residue was partitioned between AcOEt and water and the organic extract was washed with brine, and then dried over $Na_2SO_4$. The evaporation of the solvent gave a crude mixture of dicarboxylic acid **30a** and its isomer **30b** (900 mg), which was dried under vacuum over solid NaOH overnight and used for the next reaction without further purification. To the mixed solution of the carboxylic acids (900 mg) obtained above in THF/DMF (2/1, v/v, 27 mL) was added $^n$Bu$_3$P (693 mg, 3.425 mmol) at −78 °C under a nitrogen atmosphere, followed by dropwise addition of DBU (626 mg, 4.112 mmol).[13,25] After stirring overnight at room temperature, the solvent was

removed under reduced pressure and the residue was dissolved in AcOEt. The organic solution was treated with a saturated aqueous solution of $NaHCO_3$ to convert the resulting carboxylic acid to the corresponding sodium salt and the organic phase was discarded. The aqueous solution was acidified to pH 3 by adding 1 M HCl and the precipitated solid carboxylic acid was filtered off and dried under vacuum to give **31** (410 mg, 49% yield in 2 steps) as a yellow solid. It was used for the next step without further purification.

In a similar manner, **8a** (60 mg, 0.073 mmol) was converted to 2-tosyl isomer of the carboxylic acid **30a** (40 mg). The crude acid **30a** (58 mg) was treated with $^{n}Bu_3P$ (45 mg, 0.222 mmol) and DBU (41 mg, 0.269 mmol) to give **31** (19 mg, 46% yield in 2 steps). Furthermore, **8b** (40 mg, 0.049 mmol) was converted to **30b** (39 mg), which was treated with $^{n}Bu_3P$ (25 mg, 0.125 mmol) and DBU (23 mg, 0.151 mmol) to give **31** (11 mg, 60% yield in 2 steps). Mp 171–175 °C (from THF/hexane). IR (KBr): 3255, 2970, 2935, 2876, 2583, 1677, 1557, 1463, 1371, 1262, 1179, 1154, 1092, 1057, 988, 935, 844, 820, 784 $cm^{-1}$. $^1H$ NMR ($CDCl_3$/THF-$d_8$): $\delta$ 1.10 (t, 3H, $J = 7.3$ Hz), 1.93–1.98 (m, 2H), 2.35 (q, 2H, $J = 7.3$ Hz), 2.36–2.38 (m, 2H), 2.45–2.53 (m, 2H), 2.59 (t, 2H, $J = 8.2$ Hz), 3.05 (t, 2H, $J = 8.2$ Hz), 4.53 (d, 2H, $J = 5.4$ Hz), 5.16 (d, 1H, $J = 9.9$ Hz), 5.26 (d, 1H, $J = 17.4$ Hz), 5.90 (ddt, 1H, $J = 17.4$, 9.9, 5.4 Hz), 6.09 (s, 1H), 7.73 (s, 1H), 9.04 (s, 1H), 10.65 (s, 1H). HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 385.1759. Calcd for $C_{21}H_{25}N_2O_5$: 385.1764.

**Allyl 1-(7-Ethyl-2-formyl-6-oxo-3,5,6,8,9,10-hexahydro-3,5-diazadicyclopenta[*a*,*d*]cycloocten-1-yl)propanoate (6).** To a solution of **31** (110 mg, 0.3 mmol) in TFA (3 mL) was added $(MeO)_3CH$ (1.5 mL) at 0 °C under a nitrogen atmosphere and allowed to stir for 25 min at 0 °C and for 20 min at room temperature. The reaction mixture was quenched with water and extracted with AcOEt. The organic layer was successively washed with a saturated aqueous solution of $NaHCO_3$, brine, and dried over $MgSO_4$. The solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, hexane/AcOEt = 2/1, v/v) to give **6** (67 mg) in 63% yield as a yellow solid. Mp 128–130 °C (from AcOEt/hexane). IR (KBr): 3605, 3565, 3005, 2967, 2925, 1714, 1645, 1421, 1362, 1223, 1092, 903, 785 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 1.13 (t, 3H, $J = 7.6$ Hz), 1.94 (quint, 2H, $J = 6.4$ Hz), 2.36–2.42 (m, 4H), 2.51 (t, 2H, $J = 6.4$ Hz), 2.64 (t, 2H, $J = 7.7$ Hz), 3.08 (t, 2H, $J = 7.7$ Hz), 4.57 (d, 2H, $J = 5.9$ Hz), 5.23 (dd, 1H, $J = 10.5$, 1.5 Hz), 5.28 (dd, 1H, $J = 17.3$, 1.5 Hz), 5.88 (ddt, 1H, $J = 17.3$, 10.5, 5.9 Hz), 6.19 (s, 1H), 8.31 (brs, 1H), 9.63 (s, 1H), 9.98 (brs, 1H). Found: C, 68.39; H, 6.52; N, 7.49%. Calcd for $C_{21}H_{24}N_2O_4$: C, 68.46; H, 6.57; N, 7.60%.

**Allyl 3-{2-[1-(2-Allyloxycarbonylethyl)-7-ethyl-6-oxo-3,5,6,8,9,10-hexahydro-3,5-diazadicyclopenta[*a*,*d*]cycloocten-2-ylmethylene]-4-methyl-5-(4-methyl-5-oxo-3-vinyl-2,5-dihydro-1*H*-pyrrol-2-ylidenemethyl)-2*H*-pyrrol-3-yl}propanoate (32).** To a mixed solution of **6** (15 mg, 0.041 mmol) and (*Z*)-allyl 3-{4-methyl-5-[4-methyl-5-oxo-3-vinyl-1*H*-pyrrol-2(5*H*)-ylidenemethyl]-1*H*-pyrrol-3-yl}propanoate (**7**)[14] (13 mg, 0.041 mmol) in MeOH (1.5 mL), a solution of $H_2SO_4$ (8 mg, 0.082 mmol) in MeOH (0.6 mL) was added dropwise at room temperature under a nitrogen atmosphere. After stirring for 1 h, the reaction mixture was quenched with a phosphate buffer solution (pH 7.0) and extracted with AcOEt. The organic layer was washed with brine, and then dried over $Na_2SO_4$. The solvent was evaporated and the blue residue was separated by thin layer chromatography ($SiO_2$, hexane/$CHCl_3$/MeOH = 7/4/0.5, v/v/v) to afford **32** in 71% (20 mg) yield as a blue solid. Mp 173–175 °C (from $CH_2Cl_2$/hexane). IR (KBr): 3345, 2927, 1734, 1685, 1586, 1456, 1257, 1160, 1099, 953, 931, 874, 850 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 1.04 (t, 3H, $J = 7.4$ Hz), 1.92–1.98 (m, 2H), 2.07 (s, 3H), 2.08 (s, 3H), 2.30 (q, 2H, $J = 7.4$ Hz), 2.32–2.37 (m, 2H), 2.48 (brt, 2H, $J = 5.9$ Hz), 2.58 (t, 2H, $J = 7.8$ Hz), 2.59 (t, 2H, $J = 7.8$ Hz), 2.95 (t, 2H, $J = 7.8$ Hz), 2.97 (t, 2H, $J = 7.8$ Hz), 4.55–4.59 (m, 4H), 5.21 (d, 2H, $J = 10.2$ Hz), 5.28 (d, 2H, $J = 17.3$ Hz), 5.67 (d, 1H, $J = 11.7$ Hz), 5.70 (d, 1H, $J = 17.8$ Hz), 5.84–5.94 (m, 2H), 6.11 (s, 1H), 6.38 (s, 1H), 6.64 (dd, 1H, $J = 17.8$, 11.7 Hz), 6.86 (s, 1H), 8.78 (brs, 1H). Two N*H* protons were not observed clearly. Found: C, 70.95; H, 6.49; N, 8.22%. Calcd for $C_{40}H_{44}N_4O_6$: C, 70.99; H, 6.55; N, 8.28%.

**3-{2-[1-(2-Carboxyethyl)-7-ethyl-6-oxo-3,5,6,8,9,10-hexahydro-3,5-diazadicyclopenta[*a*,*d*]cycloocten-2-ylmethylene]-4-methyl-5-(4-methyl-5-oxo-3-vinyl-2,5-dihydro-1*H*-pyrrol-2-ylidenemethyl)-2*H*-pyrrol-3-yl}propanoic Acid (3).** To a mixed solution **32** (20 mg, 0.0296 mmol) and $[Pd(PPh_3)_4]$ (7 mg, 0.0059 mmol) in THF (0.7 mL), a solution of TsNa (10 mg, 0.059 mmol) in MeOH (0.7 mL) was added under a nitrogen atmosphere at room temperature. After stirring for 10 min, the solvent was evaporated and the residue was separated by flash column chromatography ($SiO_2$, $CHCl_3$/MeOH/AcOH = 200/15/1, v/v/v). The blue fraction was evaporated and the resulting solid residue was recrystallized from $CHCl_3$/hexane. Free acid **3**: a blue solid, 12 mg, 70% yield. Mp 260 °C (decomp.). IR (KBr): 3400, 3232, 2968, 2933, 1702, 1601, 1458, 1416, 1292, 1094, 954, 890, 839 $cm^{-1}$. $^1H$ NMR ($C_5D_5N$): $\delta$ 1.17 (t, 3H, $J = 7.5$ Hz), 1.88–1.96 (m, 2H), 2.06 (s, 3H), 2.12 (s, 3H), 2.32 (t, 2H, $J = 6.1$ Hz), 2.44 (q, 2H, $J = 7.5$ Hz), 2.53 (t, 2H, $J = 6.1$ Hz), 2.88 (t, 4H, $J = 7.3$ Hz), 3.20 (t, 2H, $J = 7.3$ Hz), 3.21 (t, 2H, $J = 7.3$ Hz), 5.60 (d, 1H, $J = 11.7$ Hz), 5.74 (d, 1H, $J = 17.8$ Hz), 6.27 (s, 1H), 6.72 (dd, 1H, $J = 17.8$, 11.7 Hz), 6.72 (s, 1H), 7.59 (s, 1H), 8.42 (s, 1H), 11.76 (s, 1H). Two $CO_2H$ protons and one N*H* proton were not observed clearly. UV–vis (MeOH) $\lambda_{max}$ 381 ($\varepsilon = 40533$), 630 ($\varepsilon = 29260$) nm. HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 597.2741. Calcd for $C_{34}H_{37}N_4O_6$: 597.2713.

**Allyl 3-{2-[3-(2-Allyloxycarbonylethyl)-5-(3-ethylidene-4-methyl-5-oxopyrrolidin-2-ylidenemethyl)-4-methylpyrrol-2-ylidenemethyl]-7-ethyl-6-oxo-3,5,6,8,9,10-hexahydro-3,5-diazadicyclopenta[*a*,*d*]cycloocten-1-yl}propanoate (33).** To **11** (13 mg, 0.029 mmol) and **6** (9 mg, 0.024 mmol) was added TFA (0.36 mL) at room temperature under a nitrogen atmosphere. After stirring for 20 min, MeOH (0.72 mL) was added and the reaction mixture was stirred for 1.5 h. The reaction mixture was quenched by adding a phosphate buffer solution (pH 7.0) and extracted with AcOEt. The organic layer was washed with brine, and then dried over $Na_2SO_4$. The solvent was evaporated and the blue residue was separated by TLC on $SiO_2$ ($CHCl_3$/AcOEt/EtOH = 60/8/1, v/v/v) to afford **33** (11 mg, 0.016 mmol) in 67% yield as a blue solid. Mp 73–75 °C (from $CH_2Cl_2$/hexane). IR (KBr): 3243, 2931, 2855,

1733, 1681, 1590, 1453, 1412, 1376, 1338, 1319, 1256, 1212, 1155, 1091, 1052, 987, 957, 934, 895, 835, 700 $cm^{-1}$. $^1H$ NMR ($CDCl_3$): $\delta$ 1.11 (t, 3H, $J = 7.6$ Hz), 1.43 (d, 3H, $J = 7.6$ Hz), 1.92 (d, 3H, $J = 7.3$ Hz), 1.94–2.02 (m, 2H), 2.05 (s, 3H), 2.35–2.40 (m, 2H), 2.38 (q, 2H, $J = 7.6$ Hz), 2.50 (t, 2H, $J = 6.3$ Hz), 2.58 (t, 2H, $J = 7.8$ Hz), 2.59 (t, 2H, $J = 7.8$ Hz), 2.92 (t, 2H, $J = 7.8$ Hz), 2.96 (t, 2H, $J = 7.8$ Hz), 3.25 (q, 1H, $J = 7.6$ Hz), 4.56–4.58 (m, 4H), 5.20 (dd, 1H, $J = 10.5$, 1.5 Hz), 5.21 (dd, 1H, $J = 10.5$, 1.5 Hz), 5.27 (dd, 1H, $J = 17.1$, 1.5 Hz), 5.29 (dd, 1H, $J = 17.1$, 1.5 Hz), 5.84 (s, 1H), 5.89 (ddt, 2H, $J = 17.1$, 10.5, 5.9 Hz), 6.40 (q, 1H, $J = 7.3$ Hz), 6.45 (s, 1H), 6.68 (s, 1H), 9.56 (brs, 1H). Two N*H* protons were not observed clearly. HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 679.3485. Calcd for $C_{40}H_{47}N_4O_6$: 679.3496.

**3-{2-[3-(2-Carboxyethyl)-5-(3-ethylidene-4-methyl-5-oxopyrrolidin-2-ylidenemethyl)-4-methylpyrrol-2-ylidenemethyl]-7-ethyl-6-oxo-3,5,6,8,9,10-hexahydro-3,5-diazadicyclopenta[*a*,*d*]cycloocten-1-yl}propanoic Acid (2).** To a mixed solution of **33** (10 mg, 0.015 mmol) and $[Pd(PPh_3)_4]$ (3 mg, 0.003 mmol) in THF (0.4 mL), a solution of TsNa (5 mg, 0.030 mmol) in MeOH (0.4 mL) was added under a nitrogen atmosphere at room temperature. After stirring for 10 min, thiourea (0.9 mg, 0.012 mmol) was added. The solution was directly separated by flash column chromatography ($SiO_2$, $CHCl_3$/MeOH/AcOH = 250/15/1, v/v/v). The blue fraction was evaporated and the resulting solid residue was recrystallized from $CHCl_3$/hexane. Free acid **2**: a blue solid, 12 mg, in quantitative yield. Mp 175–178 °C (from $CHCl_3$/hexane). IR (KBr): 3434, 2934, 2861, 1668, 1600, 1543, 1455, 1440, 1412, 1375, 1335, 1247, 1222, 1178, 1151, 1104, 1051, 969, 895, 848, 806, 745, 700, 667 $cm^{-1}$. $^1H$ NMR ($C_5D_5N$): $\delta$ 1.16 (t, 3H, $J = 7.6$ Hz), 1.32 (d, 3H, $J = 7.3$ Hz), 1.69 (d, 3H, $J = 7.1$ Hz), 1.93–1.99 (m, 2H), 2.13 (s, 3H), 2.34–2.40 (m, 2H), 2.45 (q, 2H, $J = 7.6$ Hz), 2.55–2.60 (m, 2H), 2.89–2.95 (m, 4H), 3.18–3.28 (m, 5H), 6.04 (s, 1H), 6.29 (q, 1H, $J = 7.1$ Hz), 7.00 (s, 1H), 7.51 (s, 1H), 8.53 (s, 1H). Two N*H* protons and two $CO_2H$ were not observed clearly. UV–vis (MeOH) $\lambda_{max}$ 382 ($\varepsilon$ = 24613), 679 ($\varepsilon = 39057$) nm. HRMS ($FAB^+$) ($M^+ + 1$), Found: $m/z$ 599.2868. Calcd for $C_{34}H_{39}N_4O_6$: 599.2870.

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## References

1 H. A. Borthwick, S. B. Hendricks, M. W. Parker, E. H. Toole, V. K. Toole, *Proc. Natl. Acad. Sci. U.S.A.* **1952**, *38*, 662; W. L. Butler, K. H. Norris, H. W. Siegelman, S. B. Hendricks, *Proc. Natl. Acad. Sci. U.S.A.* **1959**, *45*, 1703.

2 K. C. Yeh, S. H. Wu, J. T. Murphy, J. C. Lagarias, *Science* **1997**, *277*, 1505; T. Lamparter, F. Mittmann, W. Gärtner, T. Börner, E. Hartmann, J. Hughes, *Proc. Natl. Acad. Sci. U.S.A.* **1997**, *94*, 11792; S. J. Davis, A. V. Vener, R. D. Vierstra, *Science* **1999**, *286*, 2517.

3 S. H. Bhoo, S. J. Davis, J. Walker, B. Karniol, R. D. Vierstra, *Nature* **2001**, *414*, 776.

4 C. Starostzik, W. Marwan, *FEBS Lett.* **1995**, *370*, 146.

5 J. C. Lagarias, H. Rapoport, *J. Am. Chem. Soc.* **1980**, *102*, 4821; H. Falk, *The Chemistry of Linear Oligopyrroles and Bile Pigments*, Springer Verlag, New York, **1989**; W. Rüdiger, F. Thümmler, *Angew. Chem., Int. Ed. Engl.* **1991**, *30*, 1216; M. Furuya, P.-S. Song, in *Assembly and Properties of Holophytochrome* in *Photomorphogenesis in Plants*, 2nd ed., ed. by R. E. Kendrick, G. H. M. Kronenberg, Kluwer Academic Publishers, Dordrecht, The Netherlands, **1994**, Chap. 4.3, pp. 105–140; M. Stanek, K. Grubmayr, *Chem.—Eur. J.* **1998**, *4*, 1653; M. Stanek, K. Grubmayr, *Chem.—Eur. J.* **1998**, *4*, 1660; T. Lamparter, *FEBS Lett.* **2004**, *573*, 1; N. C. Rockwell, J. C. Lagarias, *Plant Cell* **2006**, *18*, 4; N. C. Rockwell, Y.-S. Su, J. C. Lagarias, *Annu. Rev. Plant Biol.* **2006**, *57*, 837.

6 W. Rüdiger, F. Thümmler, in *Photomorphogenesis in Plants*, 2nd ed., ed. by R. E. Kendrick, G. H. M. Kronenberg, Kluwer Academic Publishers, Dordrecht, The Netherlands, **1994**.

7 a) N. Frankenberg, K. Mukougawa, T. Kohchi, J. C. Lagarias, *Plant Cell* **2001**, *13*, 965. b) T. Hübschmann, T. Börner, E. Hartmann, T. Lamparter, *Eur. J. Biochem.* **2001**, *268*, 2055.

8 S. H. Wu, M. T. McDowell, J. C. Lagarias, *J. Biol. Chem.* **1997**, *272*, 25700.

9 A. Blumenstein, K. Vienken, R. Tasler, J. Purschwitz, D. Veith, N. Frankenberg-Dinkel, R. Fischer, *Curr. Biol.* **2005**, *15*, 1833; I. Oberpichler, I. Molina, O. Neubauer, T. Lamparter, *FEBS Lett.* **2006**, *580*, 437.

10 T. Lamparter, N. Michael, O. Caspani, T. Miyata, K. Shirai, K. Inomata, *J. Biol. Chem.* **2003**, *278*, 33786.

11 J. R. Wagner, J. S. Brunzelle, K. T. Forest, R. D. Vierstra, *Nature* **2005**, *438*, 325; J. R. Wagner, J. Zhang, J. S. Brunzelle, R. D. Vierstra, K. T. Forest, *J. Biol. Chem.* **2007**, *282*, 12298; X. Yang, E. A. Stojković, J. Kuk, K. Moffat, *Proc. Natl. Acad. Sci. U.S.A.* **2007**, *104*, 12571; X. Yang, J. Kuk, K. Moffat, *Proc. Natl. Acad. Sci. U.S.A.* **2008**, *105*, 14715; X. Yang, J. Kuk, K. Moffat, *Proc. Natl. Acad. Sci. U.S.A.* **2009**, *106*, 15639.

12 W. Rüdiger, F. Thümmler, E. Cmiel, S. Schneider, *Proc. Natl. Acad. Sci. U.S.A.* **1983**, *80*, 6244.

13 K. Inomata, *Bull. Chem. Soc. Jpn.* **2008**, *81*, 25, and references cited therein.

14 M. A. S. Hammam, H. Nakamura, Y. Hirata, H. Khawn, Y. Murata, H. Kinoshita, K. Inomata, *Bull. Chem. Soc. Jpn.* **2006**, *79*, 1561.

15 Preliminary communication of the synthesis of 15*E*-*anti* BV derivative: H. Kinoshita, M. A. S. Hammam, K. Inomata, *Chem. Lett.* **2005**, *34*, 800.

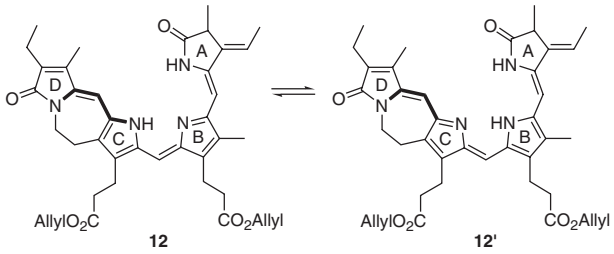
16 K. Inomata, M. A. S. Hammam, H. Kinoshita, Y. Murata, H. Khawn, S. Noack, N. Michael, T. Lamparter, *J. Biol. Chem.* **2005**, *280*, 24491; K. Inomata, S. Noack, M. A. S. Hammam, H. Khawn, H. Kinoshita, Y. Murata, N. Michael, P. Scheerer, N. Krauss, T. Lamparter, *J. Biol. Chem.* **2006**, *281*, 28162.

17 a) H. Khawn, L.-Y. Chen, H. Kinoshita, K. Inomata, *Chem. Lett.* **2008**, *37*, 198. b) K. Inomata, H. Khawn, L.-Y. Chen, H. Kinoshita, B. Zienicke, I. Molina, T. Lamparter, *Biochemistry* **2009**, *48*, 2817. c) L.-Y. Chen, H. Kinoshita, K. Inomata, *Chem. Lett.* **2009**, *38*, 602. d) P. Scheerer, N. Michael, J. H. Park, S. Nagano, H.-W. Choe, K. Inomata, B. Borucki, N. Krauß, T. Lamparter, *ChemPhysChem* **2010**, *11*, 1090.

18 A. T. Ulijasz, G. Cornilescu, C. C. Cornilescu, J. Zhang, M. Rivera, J. L. Markley, R. D. Vierstra, *Nature* **2010**, *463*, 250.

19 E. J. Corey, M. Chaykovsky, *J. Am. Chem. Soc.* **1965**, *87*, 1345.

20 In the B- and C-rings of the obtained PCB derivative, two tautomeric forms such as **12** and **12′** are possible. In this manuscript, one of the tautomeric forms is shown in the schemes including the case of BV derivative.




21 D. H. R. Barton, J. Kervagoret, S. Z. Zard, *Tetrahedron* **1990**, *46*, 7587.

22 K. P. Jayasundera, H. Kinoshita, K. Inomata, *Bull. Chem. Soc. Jpn.* **2000**, *73*, 497.

23 N. Kornblum, J. Powers, *J. Org. Chem.* **1957**, *22*, 455.

24 R. M. Silverstein, E. E. Ryskiewicz, C. Willard, *Org. Synth., Coll. Vol.* **1963**, *IV*, 831.

25 H. Kinoshita, H. Ngwe, K. Kobori, K. Inomata, *Chem. Lett.* **1993**, 1441; K. Kohori, M. Hashimoto, H. Kinoshita, K. Inomata, *Bull. Chem. Soc. Jpn.* **1994**, *67*, 3088.

26 K. P. Jayasundera, H. Kinoshita, K. Inomata, *Chem. Lett.* **1998**, 1227.