Available online at www.sciencedirect.com
ScienceDirect
Bioorganic & Medicinal Chemistry Letters 17 (2007) 1920–1923
Bioorganic & Medicinal Chemistry Letters

Self-aggregation of zinc chlorophylls possessing perfluoroalkyl chains in fluorous solvents: Selective extraction of the self-aggregates with fluorous phase and accelerated formation of the ordered supramolecules in this phase

Hitoshi Tamiaki,* Takashi Nishiyama and Reiko Shibata

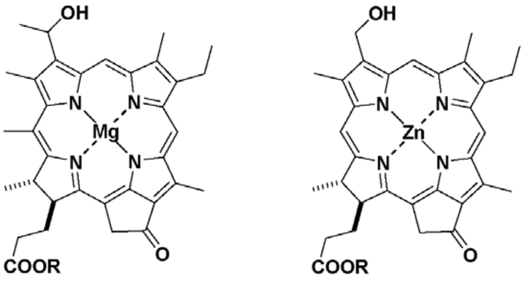
Department of Bioscience and Biotechnology, Faculty of Science and Engineering, Ritsumeikan University, Kusatsu, Shiga 525-8577, Japan

Received 10 November 2006; revised 5 January 2007; accepted 10 January 2007
Available online 24 January 2007

Abstract—Zinc 3^1-hydroxy-13^1-oxo-chlorins possessing various perfluorooctyl groups in the 17-propionate were prepared by modifying naturally occurring chlorophyll-*a*. The synthetic compound having four perfluorooctyl groups readily self-assembled to form large *J*-aggregates, which were dissolved in fluorous phases with more ordered supramolecular structures than in a hydrocarbon solvent.

Self-assemblies of organic molecules make a variety of supramolecular structures and induce different physical properties from those of their monomeric states. Construction of such supramolecules is dependent upon both the composite molecular structure and the assembling conditions. For example, lipid molecules can hardly be dissolved in water as their monomeric states but their bilayer-type self-assemblies are dispersed in water: hydrophobic interaction among the long hydrocarbon chains and hydrophilic interaction of the polar head group with external water molecules. Instead of water, fluorinated solvents have also been used as a lipophobic environment.[1] Kunitake and his colleagues reported that synthetic lipids possessing a fluorophilic head group self-aggregated in a fluorocarbon medium to make vesicles.[2] Specific interaction including coordination[3] and hydrogen bonding[4] was further useful for an increase of fluorophilicity through formation of complexes. Here, we report fluorophilic self-aggregates of a synthetic zinc chlorophyll derivative whose supramolecules consisted of a fluorophobic core (*J*-aggregates of its chlorin π-systems) and a fluorophilic surface (fluorocarbon chains). While the monomeric chlorophyll could not be dissolved in any perfluorinated solvents, its self-aggregates were soluble in a fluorous phase and the resulting supramolecules were in more order than those in a hydrocarbon solvent.

In major light-harvesting apparatus of photosynthetic green bacteria, bacteriochlorophyll (BChl)-*c* (see Fig. 1, left) self-aggregates to form a large oligomer.[5–9] We earlier reported that self-aggregates of synthetic zinc



BChl-*c* (**R = stearyl, etc**) 1 (**R = alkyl chain, etc**)

Figure 1. Naturally occurring light-harvesting chlorophylls in a green nonsulfur bacterium, *Chloroflexus aurantiacus* (left) and their synthetic model **1** (right).

Keywords: Bacteriochlorophyll; Chlorophyll; Chlorosome; Fluorophilicity; Light-harvesting antenna; Self-assembly; Photosynthetic green bacteria.

* Corresponding author. Tel.: +81 77 566 1111; fax: +81 77 561 2659; e-mail: tamiaki@se.ritsumei.ac.jp

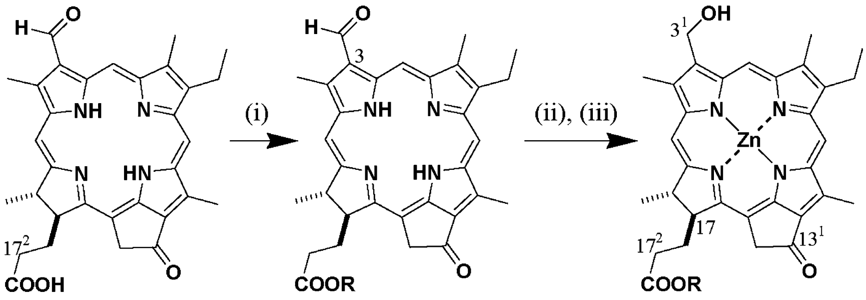
0960-894X/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2007.01.044

chlorophylls **1** (see Fig. 1, right) in hydrophobic environments were good structural and functional models.[10,11] As an alternative hydrophobic condition, a fluorous phase was examined here. For introduction of fluorophilicity in a molecule, zinc 3^1-hydroxy-13^1-oxo-chlorins **1a–c** possessing perfluorooctyl chains at the 17^2-ester group were prepared as shown in Scheme 1 (see also Electronic supporting information (ESI)). The 17^2-carboxy group of pyropheophorbide-*d* was esterified[12,13] with a mixture of primary alcohol ROH possessing perfluorooctyl groups. The resulting esters **2** were separated by gel permeation chromatography to give three compounds having two, three, and four perfluorooctyl groups, which were further purified by flash silica gel column chromatography. Each separated aldehyde **2** was selectively reduced and zinc-metallated[10,13,14] to afford the desired complex **1**.

In 0.4% (v/v) THF and hexane, compounds **1a–c** afforded species absorbing longer wavelength light compared to monomeric absorption bands in THF: $\lambda_{\max}$ (Soret)/(Q_y) = 424/646 → 442/730 nm. The red-shifted spectra are characteristic of their *J*-aggregates based on special connection of Zn···O(3^1)—H···O=C13^1 and π–π interaction of chlorin systems,[10,15] and similar to those of previous models with a single alkyl chain[10,13,14] and natural chlorosomes,[16,17] indicating that such multi-perfluoroalkyl chains did not disturb the formation of their self-aggregates and all the supramolecular structures resemble each other. All the synthetic compounds **1a–c** in the solid state were insoluble in perfluorinated solvents due to the lower content (41–50%) of their fluoro atoms than approximate 60% which was reported to be at least requisite for dissolution in such solvents.[1]

When the above self-aggregated solution of **1c** was placed on perfluoro-2-butyltetrahydrofuran (FC-75) and the biphase mixture was shaken, the upper green colored hexane phase was almost colorless and the lower fluorous phase became green (see Fig. 2a → b, top). The self-aggregates moved from hexane to FC-75 phase and such self-aggregation increased the fluorophilicity in a supramolecule. The visible spectrum of the hexane phase after shaking gave small peaks of monomeric **1c** which had been detected before the shaking (see Fig. 2a → b, middle). Such a selective extraction of self-aggregates with fluorous phase was apparently observed by use of 7%(v/v) THF–hexane solution containing ca. a 2:3 mixture of monomeric and oligomeric **1c**, which afforded a hexane solution of monomeric **1c** and a FC-75 solution of oligomeric **1c** after shaking with FC-75 (see Fig. S1 of ESI). The separated FC-75 solution of self-aggregated **1c** was shaken with THF and the self-aggregates were disconnected by coordination of a THF molecule to zinc complex to give the corresponding monomeric state, then all the monomers moved to THF phase (Fig. 2b → c). These observations also clearly show that less fluorophilic monomeric zinc complex **1c** could not be dissolved in the fluorous phase and selective movement of more fluorophilic self-aggregates $(\mathbf{1c})_n$ did occur from the hexane to FC-75 phase.

The Q_y absorption band of self-aggregated **1c** in a selectively extracted FC-75 solution was slightly red-shifted and sharpened, compared with that in hexane solution before shaking: $\lambda_{\max}$ = 730 → 734 nm and half-height band width = 990 → 740 cm^{-1} (see Fig. 2a (middle) → b (bottom)) These changes might be partially ascribable to a solvent effect and are mainly due to the composition of self-aggregates. During the extraction, the absorbance on the blue side of the Q_y band decreased to give the sharper band. The lower wavelength absorbing species on the blue side of the Q_y band in hexane are smaller and/or less ordered self-aggregates. Such small fragmented and relatively disordered self-aggregated components in hexane would be altered to the larger and more ordered self-aggregates ($\lambda_{\max}$ = 734 nm) in FC-75, although the basic motif in self-aggregates is unchanged as mentioned above (Zn···O—H···O=C/π–π stacking). The former species hardly dissolved in FC-75 and changed to the more fluorophilic latter species in the biphasic interface. A similar observation was made in other perfluorinated solvents, perfluorooctane and tris(perfluorobutyl)amine (see Figs. S2 and S3 of ESI). Therefore, fluorous solvents accelerated self-aggregation of zinc complexes in a much larger and ordered fashion. The change in supramolecules was supported by circular dichroism (CD) spectra. CD intensities at the red-shifted Q_y region strengthened during the extraction of self-aggregates from hexane to FC-75 phase (Fig. 3),



Pyropheophorbide-*d*

2a: R = CHX_2
2b: R = $CHXCH_2OCHX_2$
2c: R = $CHXCH_2OCHX$–CH_2OCHX_2

1a: R = CHX_2
1b: R = $CHXCH_2OCHX_2$
1c: R = $CHXCH_2OCHX$–CH_2OCHX_2

Scheme 1. Synthesis of self-aggregative zinc 3^1-oxo-13^1-oxo-chlorins **1** possessing perfluorooctyl groups [X = $\mathbf{CH_2O(CH_2)_2(CF_2)_8F}$]. Reagents: (i) **ROH**, EDC–DMAP/CH_2Cl_2; (ii) OsO_4–$NaIO_4$/aq AcOH–THF; (iii) $Zn(OAc)_2$·$2H_2O$/MeOH–CH_2Cl_2.

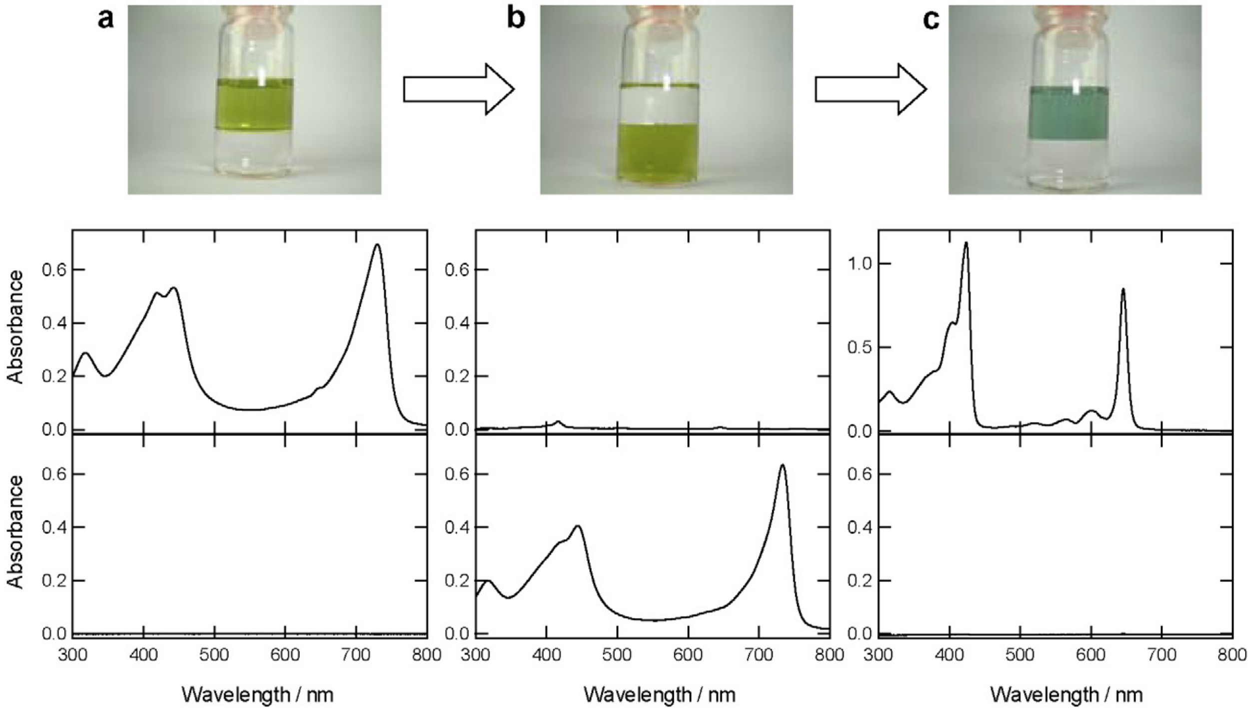


Figure 2. Fluorous biphasic systems of upper hexane and lower FC-75 phases. A hexane solution containing 0.4% (v/v) THF of **1c** (ca. 60 μM) was dropped on FC-75 (a), the resulting biphase was shaken and allowed to stand (b), and the separated FC-75 phase was shaken with THF (c). The middle and bottom electronic absorption spectra came from 4-fold dilution of the upper and lower phases, respectively.

indicating that the supramolecular structure in FC-75 was more ordered.

Under the same conditions as described above, self-aggregates of **1a** and **1b** possessing two and three perfluorooctyl groups, respectively, in hexane were extracted with FC-75 (see Figs. S4 and S5 of ESI). They were less stable in FC-75 than oligomeric **1c** and part of them were insoluble to give solids in the interface. This can be explained as follows. Supramolecules of self-aggregated **1a** and **1b** prepared in hexane were partially surrounded by their perfluorooctyl groups. Such self-aggregates further self-assembled in the extracted fluorous phase to make significant large species. The resulting larger assemblies were less soluble in FC-75 and produced their solid films on the fluorous phase with a high specific gravity ($1.77\ \mathrm{g\,cm^{-3}}$). In contrast, self-aggregates of **1c** in FC-75 were sufficiently surrounded by their perfluorooctyl groups so that their supramolecules were stabilized in the fluorous phase. Such supramolecules would be rod-like structures consisting of self-aggregated chlorin π-systems surrounded by 17^2-ester groups, which were observed in natural chlorosomes by freeze-fracture transmission electron microscopy.[18,19] The supramolecular structures were first proposed from molecular modeling[20] and experimentally confirmed by various techniques including optical spectroscopy,[13,14,21] NMR,[22] and atomic force microscopy.[23]

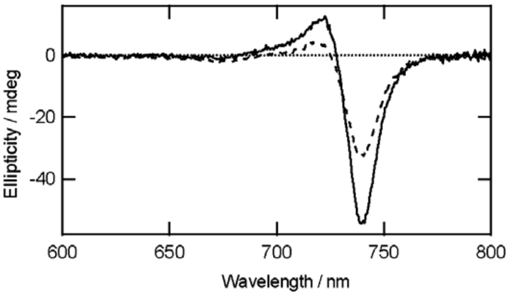


Figure 3. CD spectra of 0.4% (v/v) THF–hexane solution of **1c** (ca. 8 μM) before shaking (broken) and FC-75 solution of **1c** after shaking (solid).

In summary, monomeric forms of synthetic zinc chlorophyll derivatives **1** possessing perfluoroalkyl groups were not soluble in perfluorinated solvents but their self-aggregates in hexane were extracted with fluorous phase due to an increase of their fluorophilicity. In fluorous solvents, self-aggregation of fluorinated zinc chlorophylls **1** accelerated to form large oligomers from the small fragments and the supramolecular structure became more ordered. The fluorous biphasic system[24] is thus demonstrated to be a useful technique for preparation of supramolecularly ordered self-aggregates which would provide photoactive nanodevices with fewer structural defects.

Acknowledgments

This work was partially supported by Grants-in-Aid for Scientific Research (No. 17029065) on Priority Areas (417) from the Ministry of Education, Culture, Sports, Science and Technology (MEXT) of the Japanese Government and for Scientific Research (B) (No. 15350107) from the Japan Society for the Promotion of Science

(JSPS), as well as by the 'Academic Frontier' Project for Private Universities: matching fund subsidy from MEXT, 2003–2007.

Supplementary data

Supplementary data associated with this article can be found in the online version at doi: Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmcl.2007.01.044.

References and notes

1. Gladysz, J. A.; Emnet, C.; Rábai, J. In *Handbook of Fluorous Chemistry*; Gladysz, J. A., Curran, D. P., Horváth, I. T., Eds.; Wiely: Weinheim, 2004; pp 56–100.
2. Kuwahara, H.; Hamada, M.; Ishikawa, Y.; Kunitake, T. *J. Am. Chem. Soc.* **1993**, *115*, 3002.
3. Richter, B.; Spek, A. L.; van Koten, G.; Deelman, B.-J. *J. Am. Chem. Soc.* **2000**, *122*, 3945.
4. Palomo, C.; Aizpurua, J. M.; Loinaz, I.; Fernandez-Berridi, M. J.; Irusta, L. *Org. Lett.* **2001**, *3*, 2361.
5. Blankenship, R. E.; Matsuura, K. In *Light-Harvesting Antennas in Photosynthesis*; Green, B. R., Parson, W. W., Eds.; Kluwer Academic: Dordrecht, 2003; pp 195–217.
6. Frigaard, N.-U.; Chew, A. G. M.; Li, H.; Maresca, J. A.; Bryant, D. A. *Photosynth. Res.* **2003**, *78*, 93.
7. Balaban, T. S.; Tamiaki, H.; Holzwarth, A. R. *Top. Curr. Chem.* **2005**, *258*, 1.
8. Miyatake, T.; Tamiaki, H. *J. Photochem. Photobiol. C: Photochem. Rev.* **2005**, *6*, 89.
9. Saga, Y.; Wazawa, T.; Ishii, Y.; Yanagida, T.; Tamiaki, H. *J. Nanosci. Nanotechnol.* **2006**, *6*, 1750.
10. Tamiaki, H.; Amakawa, M.; Shimono, Y.; Tanikaga, R.; Holzwarth, A. R.; Schaffner, K. *Photochem. Photobiol.* **1996**, *63*, 92.
11. Saga, Y.; Akai, S.; Miyatake, T.; Tamiaki, H. *Chem. Lett.* **2004**, *33*, 544.
12. Miyatake, T.; Tamiaki, H.; Shinoda, H.; Fujiwara, M.; Matsushita, T. *Tetrahedron* **2002**, *58*, 9989.
13. Shibata, R.; Tamiaki, H. *Bioorg. Med. Chem.* **2006**, *14*, 2235.
14. Tamiaki, H.; Miyata, S.; Kureishi, Y.; Tanikaga, R. *Tetrahedron* **1996**, *52*, 12421.
15. Tamiaki, H. *Coord. Chem. Rev.* **1996**, *148*, 183.
16. Tamiaki, H. *Photochem. Photobiol. Sci.* **2005**, *4*, 675.
17. Saga, Y.; Kim, T.-Y.; Hisai, T.; Tamiaki, H. *Thin Solid Films* **2006**, *500*, 278.
18. Staehelin, L. A.; Golecki, J. R.; Fuller, R. C.; Drews, G. *Arch. Microbiol.* **1978**, *119*, 269.
19. Saga, Y.; Tamiaki, H. *J. Biosci. Bioeng.* **2006**, *102*, 118.
20. Holzwarth, A. R.; Schaffner, K. *Photosynth. Res.* **1994**, *41*, 225.
21. Tamiaki, H.; Shibata, R.; Mizoguchi, T. *Photochem. Photobiol.* **2007**, *83*, in press, doi:10.1562/2006-02-27-1R-819.
22. de Boer, I.; Matysik, J.; Erkelens, K.; Sasaki, S.; Miyatake, T.; Yagai, S.; Tamiaki, H.; Holzwarth, A. R.; de Groot, H. J. M. *J. Phys. Chem. B* **2004**, *108*, 16556.
23. Huber, V.; Katterle, M.; Lysetska, M.; Würthner, F. *Angew. Chem., Int. Ed.* **2005**, *44*, 3147.
24. Horváth, I. T. *Acc. Chem. Res* **1998**, *31*, 641.