Chirally self-assembled porphyrin nanowires assisted by L-glutamide-derived lipid for excitation energy transfer†

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Received 20th October 2008, Accepted 3rd March 2009
First published as an Advance Article on the web 2nd April 2009
DOI: 10.1039/b818358a

An L-glutamide-functionalized tetraphenylporphyrin (**1**) has been newly synthesized and its self-assembling behavior in organic solvents is reported. **1** forms nanofibrous assemblies in a dilute solution to show specific induced circular dichroism at the absorption band around the porphyrin moiety. It enables us to observe the meso-stable and stepwise H- and J-like hierarchical aggregations. Energy transfer studies in a mixed system with a pyrene-functionalized L-glutamide reveal the promotion of energy transfer from the pyrene excimer to the porphyrin moiety.

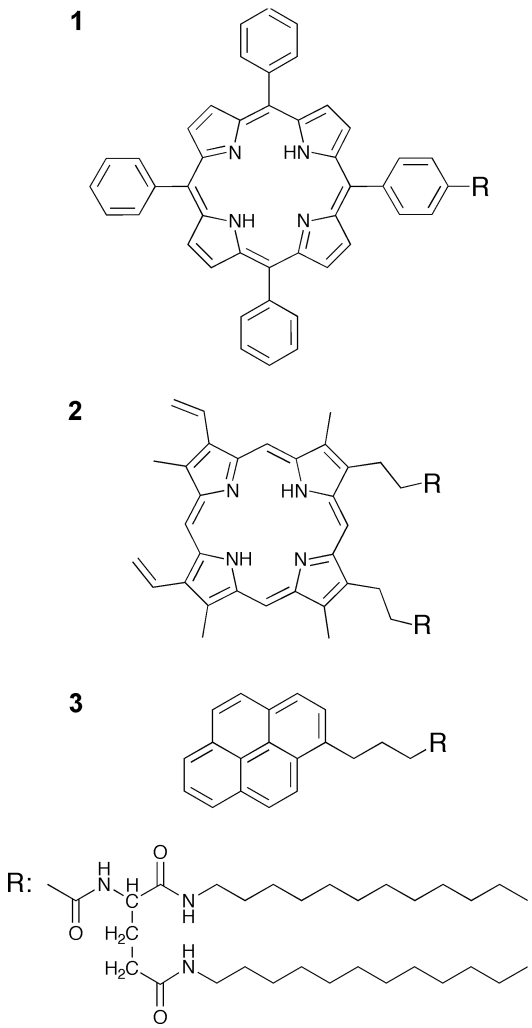
Introduction

Molecular assemblies of porphyrins and their derivatives are attractive as molecular electronic devices for mimicking natural photosynthesis[1] in terms of light-harvesting and energy migration.[2] In this context, we have previously reported on a mixed system composed of L-glutamide derivatives functionalized with protoporphyrin IX (**2**) and pyrene (**3**) for energy transfer as shown in Scheme 1.[3] Spectroscopic studies suggested that **3** could form highly-ordered molecular gels in both benzene and cyclohexane to promote the formation of co-facial chiral aggregates with **2** through their amide hydrogen bonding. Energy transfer studies in the mixed assemblies provided evidence for steady-state singlet–singlet energy transfer from the pyrene excimer to the free base porphyrin moiety. However, the protoporphyrin IX with four long chain alkyl groups (**2**) failed to gelate any of the solvents tried because of low solubility. For the sake of making a highly oriented molecular gel system with porphyrin as a light-harvesting group, we report herein a novel fabrication of a porphyrin combined with an L-glutamide-derived lipid with two long chain alkyl groups as a lipophilic part and tetraphenylporphyrin as a functional head group (**1**).

Results and discussion

Solubility and gelation properties

Various kinds of organic solvents such as acetonitrile, methanol, ethanol, DMF, THF, chloroform and benzene were good solvents to dissolve **1** as shown in Table 1, while **2** was less soluble in any solvent. However, it was observed that addition of a poor solvent such as cyclohexane converted a solution to a gel state. In this study, we mainly examined the association behaviors of **1** in a cyclohexane–THF (20 : 1) mixed system.



Scheme 1 L-Glutamide derivatives with a porphyrin (**1** or **2**) or pyrene (**3**) head group as self-assembling dyes.

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† Electronic supplementary information (ESI) available: Fig. S1: Resolution of CD spectrum; Fig. S2: Polarized optical micrograph observation; Fig. S3: Temperature dependent fluorescence spectral change of **3**; Fig. S4: Concentration dependent fluorescence spectral change at 60 °C; Fig. S5: Concentration dependency of emission intensities at 450 nm. See DOI: 10.1039/b818358a

Table 1 Solubility of **1**[a]

solvent	0 °C	20 °C	60 °C
water	–	I	I
dimethyl sulfoxide (DMSO)	–	S	S
N,*N*-dimethyl formamide (DMF)	S	S	S
acetonitrile	I	S	S
pyridine	S	S	S
methanol	S	S	S
1,4-dioxane	–	S	S
ethanol	S	S	S
tetrahydrofuran (THF)	S	S	S
ethyl acetate	S	S	S
chloroform	S	S	S
ether	I	I	–
benzene	–	S	S
toluene	S	S	S
n-hexane	I	I	I
cyclohexane	–	I	I

[a] [**1**]: 1.0 mM, S: soluble, I: insoluble.

TEM observation of **1** aggregates in the cast film from a cyclohexane–THF (20 : 1) mixture showed well-developed fibrous networks (Fig. 1). The minimum diameters of these fibers are around 4 nanometers although some parts are twisted and bundled. Their lengths are at least in the order of hundreds of micrometers. The geometric dimensions of **1** are 3.75 × 1.75 × 0.44 nm^3 estimated with HyperChem-MM2 calculations. Therefore, we estimate that the minimum sizes of the fibrous aggregates of **1** are based on a molecular-layered structure.

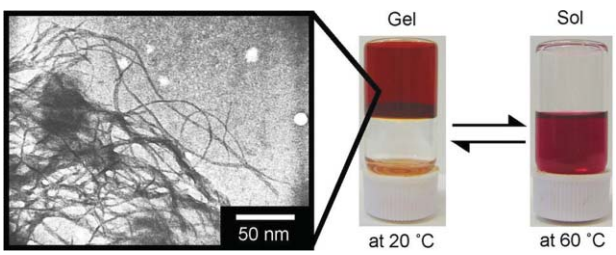

Fig. 1 Temperature-dependent sol-to-gel transition of **1** (0.5 mM) in a cyclohexane–THF (20 : 1) mixture and TEM image of **1** aggregates in the cast film prepared by 0.2 mM. Stained by 2.0 wt% ammonium molybdate.

Photophysical properties

The Soret band (417 nm) of **1** showed a characteristic split by about 30 nm (398 nm and 428 nm) when the concentration was increased from 0.10 mM to 0.75 mM as shown in Fig. 2a. The plots of the molecular coefficient at 417 nm (ε_{417}) *versus* log [**1**] (Fig. 2b) showed that a drastic spectral change was observed by a slight change of the concentration of **1**. Therefore, the Soret band at 417 nm in Fig. 2a may correspond to the monomeric form or weakly aggregated species like micelles of **1**. A similar split pattern was reported in a protoporphyrin IX derivative with bis(glycosamides) in an aqueous solution[4] and a bis(imidazolyl)-porphyrinato-cobalt(III) complex in chloroform[5] although their detailed mechanisms are different from our observation. Also, further important information was obtained by CD spectroscopy. A strong Cotton effect was observed at 375–460 nm on increasing the concentration of **1** as shown in Fig. 2c. The plot of $[\theta]_{417}$

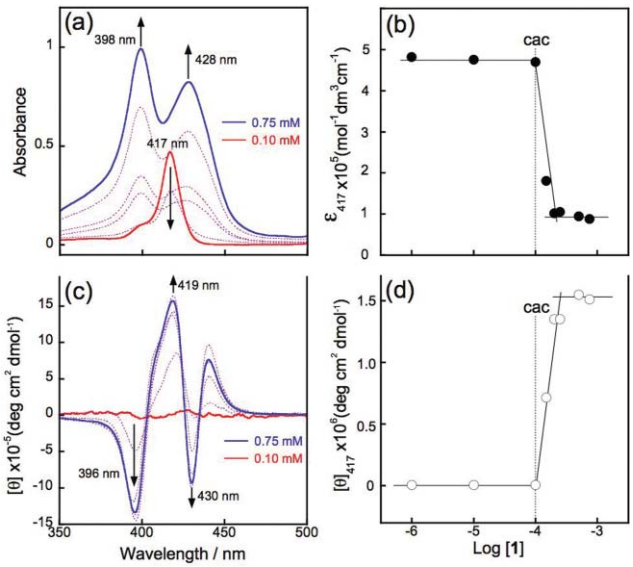

Fig. 2 UV–visible spectra (a) and molecular coefficient (ε) at 417 nm (b), CD spectra (c) and molecular ellipticity (θ) at 417 nm (d) of **1** in a cyclohexane–THF (20 : 1) mixture at 10 °C. The arrows indicate the concentration increase from 0.1 mM (red line) to 0.75 mM (blue line).

versus log [**1**] (Fig. 2d) shows that the $[\theta]_{417}$ value is very small in concentrations below 0.1 mM and almost constant in the concentrations above 0.2 mM. Therefore, Fig. 2b and 2d indicate that the critical aggregation concentration (cac) of **1** is around 0.1 mM in a cyclohexane–THF (20 : 1) mixed system at 10 °C.

After warming a cyclohexane–THF (20:1) mixture containing 0.2 mM of **1** at over 60 °C for 20 min, a UV–vis spectral change of **1** at 10 °C was recorded at every 2 min interval for 20 min as shown in Fig. 3a. Two isosbestic points were observed at 405 and 426 nm and the split Soret band appeared. This result is ascribed to the equilibrium between the solution state of **1** (at 417 nm) and the gel state of aggregated **1** (at 398 nm and 428 nm). A time-course of CD spectral change of **1** at 10 °C revealed complex Cotton effects (Fig. 3b). CD spectra showed hierarchical spectral changes. Positive and negative Cotton bands were observed at around 395 nm and 425 nm at the initial stage (from 0 min to 10 min in Fig. 3b), and a new negative Cotton band was observed at 430 nm at the later stage (from 10 min to 20 min in Fig. 3b). Detailed investigation of the CD spectrum as shown in Fig. 3 confirms that the CD pattern centered at 398 nm can be assigned to hypsochromically shifted H-like aggregates and this is also

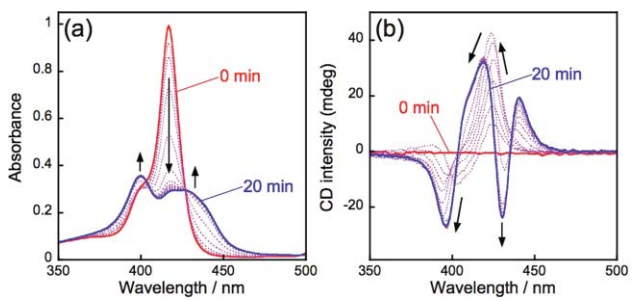

Fig. 3 Time-course of UV–visible (a) and CD (b) spectral change of **1** (0.25 mM) in a cyclohexane–THF (20 : 1) mixture at 10 °C. The arrows indicate a time increase from 0 min (red line) to 20 min (blue line).

Table 2 Absorption frequencies of **1** under various conditions

	frequencies / cm^{-1}	
	amide I	amide II
a (solid)	1639	1542
b (cyclohexane/THF (20 : 1))	1676, 1637	1551
c (chloroform)	1652	1558, 1523

assigned to right-handed (*R*-) chirality, while another centered at 428 nm is assigned to the bathochromically shifted J-like aggregates and left-handed (*S*-) chirality† according to Simonyi's explanation.[6] These results indicate that the dispersed sol state of **1** turned gradually into H-like aggregates with *R*-chirality, then the H-like aggregates started to form J-like aggregates with *S*-chiral orientation and completely developed to keep a steady state. An FT-IR spectrum of the **1** aggregates in a cyclohexane–THF (20 : 1) mixture showed typical amide I absorptions corresponding to parallel and antiparallel hydrogen bonding[7,8] at 1637 and 1678 cm^{-1}, respectively (Table 2). This spectrum is similar to that in a solid state with an absorption peak at 1639 cm^{-1} but different from that in a chloroform solution state with an absorption peak at 1652 cm^{-1} (Fig. 4).

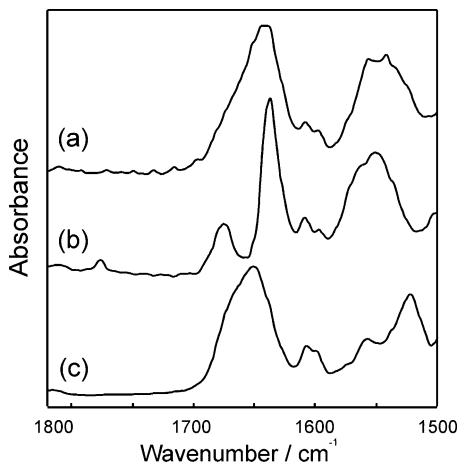

Fig. 4 FT-IR spectra of **1** under various conditions. Solid state (a), cyclohexane–THF (20 : 1) solution (b), and chloroform solution (c) at 25 °C.

The schematic illustration shown in Fig. 5 satisfies this assumption. Fig. 5 shows that fibrous networks can consist of structures chirally ordered to become nanofibrillar aggregates. One-dimensional morphology can be derived from a long-range face-to-face stacking (H-like aggregation) and an edge-to-edge interaction (J-like aggregation) can be almost simultaneously formed to lead to a ribbon-like morphology. Polarized micrograph observation of the cast film of **1** also revealed that the quasi-solid state of **1** was a highly oriented liquid crystalline phase.†

Energy transfer system

We have reported that the pyrene-functionalized L-glutamide (**3**) undergoes a temperature-induced phase transition between monomeric and aggregated states in benzene and that the excimeric emission of a pyrene moiety can be observed only in the aggregated states.[3,9] Therefore, in this study, **3** was selected as an

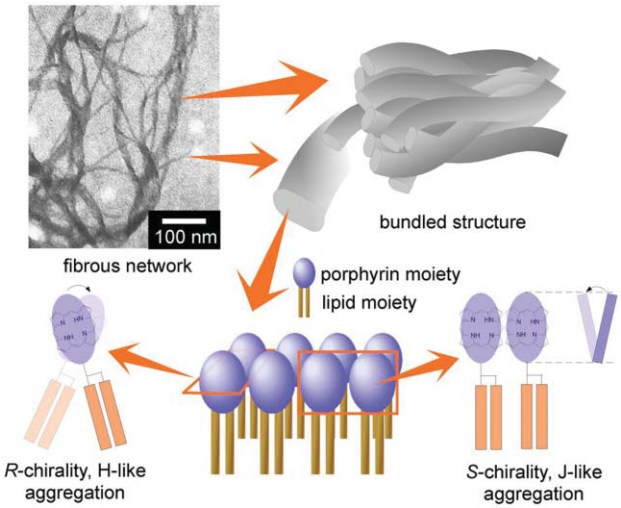

Fig. 5 Schematic proposal for the ordered structure in the **1** aggregate.

energy-donating compound for energy transfer to **1** because the emission band of a pyrene excimer can overlap partially with an absorption band of **1**.

A fluorescence spectrum of **3** was examined in a mixed system of cyclohexane and THF (20 : 1). The excimeric emission band of **3** was observed at 450 nm as a peak top (10 °C), while a monomeric emission band could be detected with peak tops at 378, 398 and 418 nm (60 °C).† According to our previous work, these results indicate that a pyrene moiety can form an excimeric state induced by highly oriented assembling at 10 °C.

As shown in a UV–vis spectrum of **1** (0.01 mM) and a fluorescence spectrum of **3** (0.05 mM), partial spectral overlapping can be realized between excimeric emission and UV–vis spectral bands at 10 °C (Fig. 6). This result indicates the possibility of energy transfer from the pyrene excimer to the porphyrin moiety.

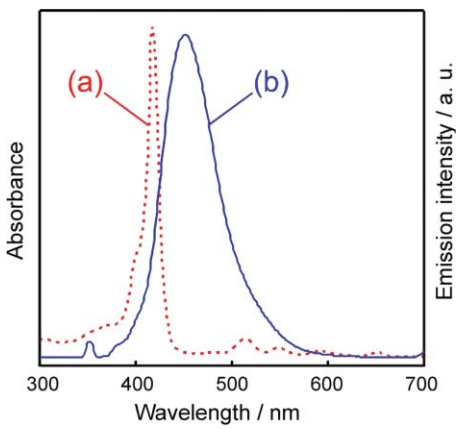

Fig. 6 UV–vis spectrum of monomeric **1** (a) and excimeric emission spectrum of **3** (b) in a cyclohexane–THF (20 : 1) mixture at 10 °C. Excitation wavelength was 350 nm.

As shown in Fig. 7a, an increase in the emission intensity at 653 nm, which was assigned to the emission band of **1**, was observed by addition of **3** at 10 °C. No similar increase was detected at 60 °C.† It was also confirmed that 5-(4-carboxyphenyl)-10,15,20-triphenyl-21*H*,23*H*-porphine (MCTPP) having no

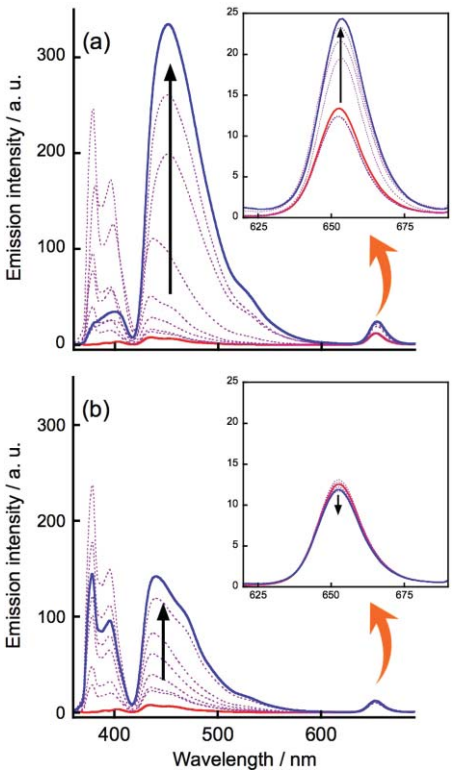

Fig. 7 Fluorescence spectra of mixtures containing 0.01 mM of **1** and 0–0.05 mM of **3** (a), and 0.01 mM of MCTPP and 0–0.05 mM of **3** (b) in a cyclohexane–THF (20 : 1) mixture at 10 °C. The arrows indicate increasing concentrations of **3** from 0 mM (red line) to 0.05 mM (blue line). Excitation wavelength was 350 nm.

L-glutamide moiety showed no significant effect under the same conditions (Fig. 7b).

The concentration dependence of **3**'s emission (Fig. 8) shows that a distinct increase in the emission intensity at 653 nm (I_{653}) can be seen at concentrations between 0.025 mM and 0.05 mM. On the other hand, the I_{653} of MCTPP was not enhanced by addition of **3** but rather decreased at concentrations above the cac of **3**.† In the absence of **3**, the I_{653} per molecule was almost constant in 0–0.1 mM of **1** but decreased at concentrations above 0.1 mM. This is attributed to quenching due to the aggregation of **1** (the cac was estimated to be around 0.1 mM by both the UV–vis and CD methods). These results indicate that the enhancement of I_{653} in

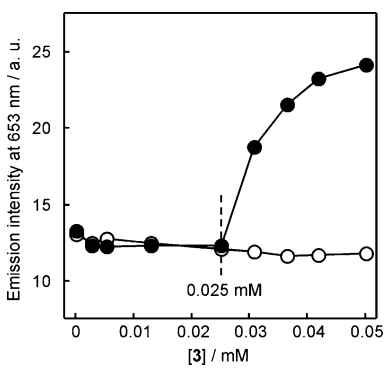

Fig. 8 Emission intensities at 653 nm of a mixture of **3** and 0.01 mM of **1** (●) or **3** and 0.01 mM of MCTPP (○).

the **1** and **3** mixed system is brought about through (1) aggregation of **3**, (2) incorporation of **1** into the aggregates of **3** and (3) energy transfer from the pyrene excimer to the porphyrin moiety.

Sol-gel matrices have been sometimes utilized to retard structural changes in the macromolecules of proteins such as the cooperative behavior of the rearrangement of the packing of the subunits, quaternary conformational changes, and so on.[10] Such a matrix-assisted assembly in sol-gel states allows the molecules to diffuse slowly and enables us to measure the meso-stable structure of the molecular assemblies even by the steady-state observation.[11] In this regard, our new system of porphyrin **1** and pyrene **3** in combination with an L-glutamide part might enable the observation of effective energy transfer from **3** to **1**.

Conclusions

It has been described that the porphyrin-functionalized L-glutamide derivative **1** can form a molecular gel in organic media through three-dimensional network formation with self-assembled nanofibrillar aggregates. Particularly, **1** formed a unique chiral assembly with both *R*-chiral H-like and *S*-chiral J-like aggregations in a hydrophobic environment. These phenomena are reversible with temperature, solvent-combination, concentration, and so on. In this paper, a mixed system of **1** with the pyrene-functionalized L-glutamide derivative **3** has been also investigated as an energy transfer model.

Experimental

^{1}H NMR (400 MHz) spectra were recorded in $CDCl_3$, $SiMe_4$ as an internal standard with JNM-EX400 (JEOL). IR spectra were measured in a KBr method with FT/IR-4100 (JASCO). TEM images were observed with JEM-2000X (JEOL). The gel was cast in a carbon-coated copper grid and stained by 2.0 wt% ammonium molybdate after casting and dried by a vacuum pump under reduced pressure. The accelerating voltage of the TEM was 80 kV and the beam current was 40 A. MALDI TOF-MS spectra were recorded on a Voyager RP (PerSeptiv Biosystem). UV–visible, CD and fluorescence spectra were measured with V-560 (JASCO), J725 (JASCO) and FP-6500 (JASCO) respectively.

A solution of 1 N NaOH 3.75 mL was added dropwise to 5-(4-methoxycarbonylphenyl)-10,15,20-triphenyl-21*H*,23*H*-porphine (0.25 g, 0.4 mmol) in DMF (100 mL) at room temperature for 30 min. After the reaction mixture was stirred at room temperature for 3 h, it was acidified with aqueous 2.0 wt% HCl to pH 2.0. The precipitates were filtered and dried *in vacuo*, to give 5-(4-carboxyphenyl)-10,15,20-triphenyl-21*H*,23*H*-porphine (0.23 g, 92%) as a purple solid. 0.40 g (0.6 mmol) of 5-(4-carboxyphenyl)-10,15,20-triphenyl-21*H*,23*H*-porphine and 0.35 g (0.7 mmol) of *N*,*N*′-didodecyl-L-glutamide, which was obtained by the previously reported procedure[12] with slight modification, were dissolved in THF (150 mL) in the presence of triethylamine (1.2 mL, 0.9 mmol) and diethyl cyanophosphate (1.5 mL, 0.9 mmol). After being stirred for 1 day at room temperature, the solution was concentrated *in vacuo.* The residue was redissolved in chloroform, and the organic layer was washed three times with 0.2 N HCl and 0.2 N NaOH, then washed with water, and dried with sodium sulfate. The solution was concentrated *in vacuo*, and the residue was reprecipitated from chloroform/n-hexane and

then dried *in vacuo* to give **1** (0.50 g, 80%) as a purple solid. mp 229.5–233.0 °C. (Found: C, 78.88; H, 8.09; N, 8.61. $C_{74}H_{87}N_7O_3$ requires C, 79.01; H, 7.81; N, 8.72%) ν_{max}(KBr)/cm^{-1} 3316, 2925, 2853, 1639, 1542; δ_H(400 MHz; $CDCl_3$; Me_4Si) –2.78 (2H, s, -N*H*), 0.80–0.87 (6H, m, -C*H*$_3$), 1.17–1.35 (40H, br, -(C*H*$_2$)$_{10}$-), 2.32 (2H, quintet, J = 5.2 Hz, -C*H*$_2$-), 2.45–2.75 (2H, br, -C*H*$_2$CO-), 3.30–3.38 (4H, m, -NC*H*$_2$-), 4.74 (1H, quintet, J = 6.0 Hz, -C*H*-), 6.03–6.06 (1H, br, N*H*), 7.04–7.07 (1H, br, N*H*), 7.73–7.79 (9H, m, Ar*H*), 8.20–8.33 (10H, m, Ar*H*), 8.42–8.44 (1H, br, N*H*), 8.79–8.87 (8H, m, Ar*H*). MALDI TOF MS (DHB matrix) (Mw = 1122.51) m/z = 1122.59 (M^+).

Acknowledgements

This work was supported in part by a Grant-in-Aid for Scientific Research from the ministry of Education, Science, Sports, and Culture of Japan. This work was financially supported by the Sasakawa Scientific Research Grant from The Japan Science Society.

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