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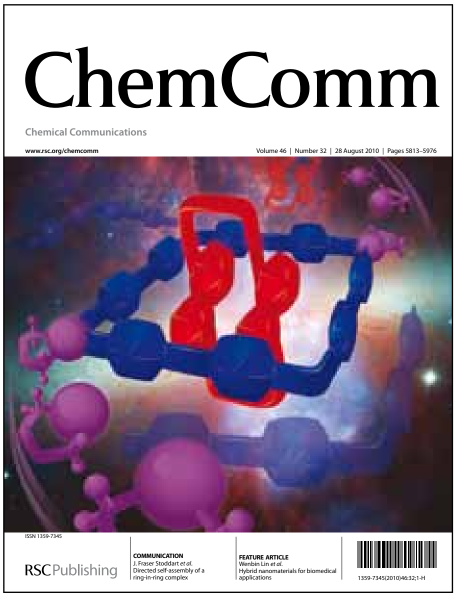

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ARTICLE TYPE

Spontaneous Transformation from Micelle to Vesicle Associated with Sequential Conversions of Comprising Amphiphiles within Assemblies

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A morphological transformation from hybrid micelles to giant vesicles was observed in aqueous dispersion associated with formation of a double-chained amphiphile as a result of the migration of dodecylamine from the amphiphilic imine to the amphiphilic aldehyde within the hydrophobic environment of amphiphilic aggregates.

Self-assemblies composed of amphiphiles, e.g. micelles or vesicles, in water have drawn considerable attention from various aspects, e.g. morphological changes of self-assemblies induced by external stimuli,[1] construction of micro-reactors,[2] design of phase-transfer catalysts, and a drug delivery system.[3] Such self-assemblies enable to from a "local hydrophobic region" at the water/ lipid interface, and the hydrophobic region within a self-assemblies or a membrane serves as a minimal reaction environment for chemical reactions, sometimes associated with uptake and release of water molecules.[4] Hence if a comprising amphiphile itself is converted in this reaction field to a different amphiphile with a smaller spontaneous curvature, the morphology of the self-assembly itself must change.[5] For example, amphiphilic imine which is in an equilibrium with its components, aldehyde and amine, is an appropriate target to investigate this plausibility because its equilibrium constant is largely affected by hydrophobic or hydrophilic environment.[6]

Recent success in the construction of two types of self-reproductive giant vesicle (GV), which is a closed-shaped bimolecular membrane composed of amphiphiles, was brought about by the inclination of the equilibrium between an imine derivative and its components depending on hydrophobic and hydrophilic environments, respectively.[7-11] This strategy turned out to be also valid for converting micelles, consisting of an amphiphilic imine, to larger aggregates, such as tubular GVs and spherical GVs.[12] Here we found that the migration of octylamine, which is a hydrolyzed product of a single-chained amphiphilic imine $\mathbf{N_1}$, to a single-chained amphiphilic aldehyde $\mathbf{N_2}$ took place within the self-assembly, resulting in the formation of a new double-chained amphiphile **V** with a smaller spontaneous curvature as an assembly. The migration of octylamine is accompanied by the morphological transformation of the self-assembly from a hybrid micelle, through a tubular vesicle, to a giant vesicle. This chemical conversion, consisting of two reversible processes of the hydrolysis (Eq. 1) and the imine formation (Eq. 2) as described in Fig. 1, proceeded in a sequential manner by utilizing hydrophilic or hydrophobic environment, respectively, depending on the molecular structures of two kinds of amphiphilic imines; one bears a single-chain, while the other bears double-chains.

Single-chained amphiphilic imine $\mathbf{N_1}$, amphiphilic aldehyde $\mathbf{N_2}$ and double-chained amphiphilic imine **V** were prepared according to the procedure reported by Kunitake *et al.*[6b] Dynamic light scattering (DLS) data revealed that $\mathbf{N_1}$ and $\mathbf{N_2}$ formed only micelles with the average diameters of 5.8 nm (standard deviation = 2 nm) and 4.6 nm (standard deviation = 1 nm) in aqueous solutions (10 mM), respectively (ESI Fig. S11).

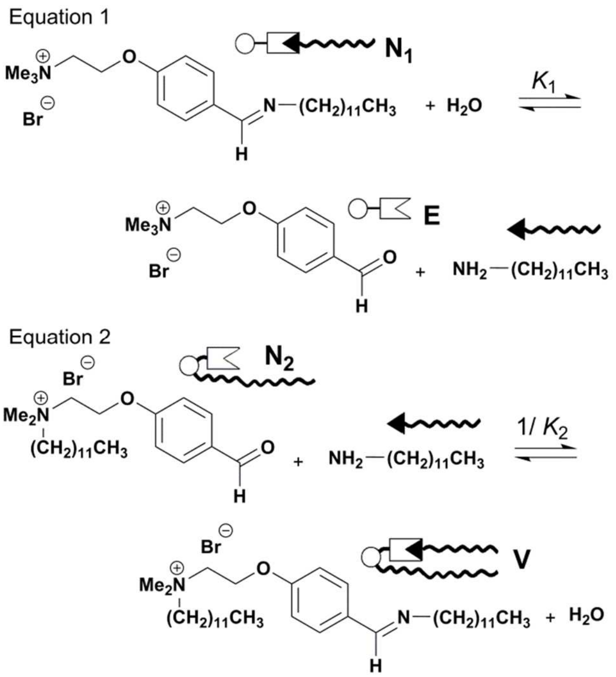

Fig. 1 Equilibriums between single- or double-chained amphiphilic imines ($\mathbf{N_1}$ or **V**) and their components: Eq. 1, top; Eq. 2, bottom. K_1 and K_2 represents the equilibrium constants of the two reaction formulas respectively.

The formation of double-chained amphiphile **V** by migration of dodecylamine from $\mathbf{N_1}$ to $\mathbf{N_2}$ in D_2O (10 mM each) was monitored by ^1H NMR spectroscopy at 25 °C. In compensation for the decrease in the intensities of signals assigned to $\mathbf{N_1}$ and

N_2, signals assigned to the product **V** and **E** increased in intensity (ESI Fig. S10). The molar fraction‡ of each component of aggregates was determined as a function of time (Fig. 2). When the aqueous solution of N_1 and N_2 were mixed, the amount of product **V** increased smoothly for the first 3 h after mixing. It was found that the decay rates of compounds N_1 and N_2 were the same as the rise rate of product **V** and that the molar fraction of dodecylamine remained constant at almost zero. This tendency indicates that the migration of dodecylamine from N_1 to N_2 proceeded smoothly without accumulating the steady-state concentration of the amine. The result indicates that the conversion smoothly increased to be saturated at ca. 90% after 600 min (diamond in Fig. 2). This is because the reaction occurs in the hydrophobic region of the hybrid amphiphilic assembly (Eq.2 in Fig. 1), and the released electrolyte **E** does not form aggregates but dissolve into the aqueous solution because of the poor hydrophobic character. This behaviour ought to shift the equilibrium to the product side, (Eq.1 in Fig. 1).[8]

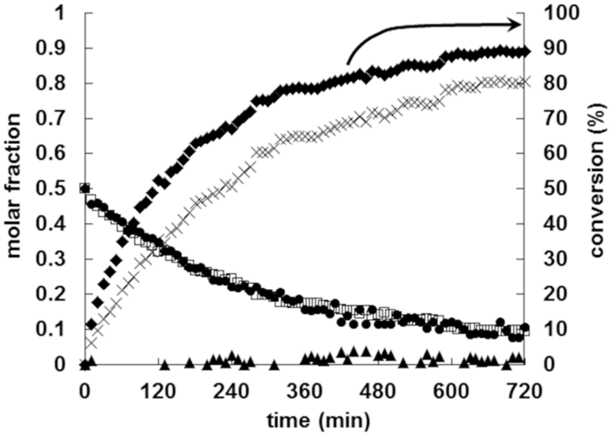

Fig. 2 Time dependence of the molar fractions of the amphiphilic components (filled circle, N_1; open square, N_2; cross, **V**; triangle, dodecylamine) and conversion of the dodecylamine migration ($N_1 + N_2 \rightarrow V + E$; filled diamond). Initial concentrations of N_1 and N_2 are both 10 mM.

The morphological transformation of the assembly was monitored by phase contrast microscopy at room temperature. No observable aggregates were found after the addition of an equimolar mixture of N_1 and N_2 (10 mM for each) within 60 min after preparation. DLS measurement showed the formation of hybrid micelles consisting of N_1 and N_2 appeared after mixing (ESI Fig. S11 and S12). About 90 min after mixing (conversion of ca. 45%), small granular aggregates began to appear (Fig. 3a) as observed using the phase contrast microscope, and then the tubular vesicles grew (Fig. 3b, conversion of 65%), followed by the conversion to spherical GV (Fig. 3c, conversion of 70%). The diameters of the observed GVs were in the range of 2.5–20 µm. The wide view of the differential interference contrast microscopy image shown in Fig. 3d indicates that the lamellarity of GV formed by the morphological transformation from hybrid micelles is low. This thin lamellarity is different from that of GVs prepared by the film swelling method using N_1, N_2 and **V** in the presence of **E**. One of the reasons for the difference in the lamellarity would be the release of **E** into a growing GV, accompanied by the hydrolysis of N_1 as discussed above. The local concentration of **E** in a water pool of the restricted volume inside GV becomes much higher than that in the exterior water phase. As a result, the flow-in of water through the bimolecular membrane resulting in swelling the closed membrane may be similar to the GV formation by the film swelling method using an aqueous solution of electrolytes.[13]

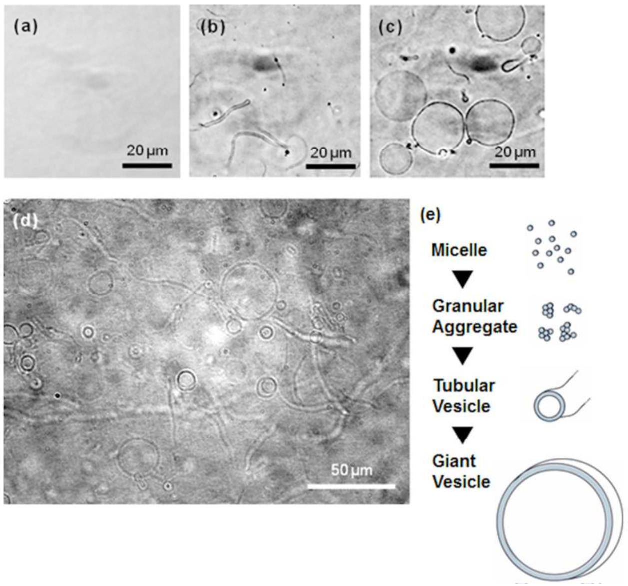

Fig. 3 Optical microscopy images of morphological transformation of aggregates composed of an equimolar mixture of N_1 and N_2 (10 mM for each): (a-c) Phase contrast microscopy images obtained at 90, 180, and 240 min after preparation, respectively. (d) Wide view of differential interference contrast microscopy image of tubular and spherical GVs 240 min after preparation. A number of GVs with the thin vesicular membrane were observed. (e) Schematic illustration of the morphological transformation of the aggregate.

The plausible driving force to promote the dodecylamine-migration is as follows. There are two equilibriums in reference to amphiphilic imines, N_1 and **V**, as expressed in Fig. 1. As for the equilibrium of hydrolysis of N_1, the reversible reaction proceeds in the hydrolysis direction predominantly because protrusive motions of N_1 with a single chain can occur in a hybrid molecular aggregate, which provides a chance for N_1 to place the amine unit near the aqueous layer to be hydrolyzed. Besides, the released electrolyte **E** dissolves into the exterior water phase, which shifts the equilibrium to the amine and aldehyde side ($K_1 >> 1$). A temporally produced dodecylamine immediately reacts with N_2 nearby to form the imine linkage of **V** in the hydrophobic region. Thus the second equilibrium equation is inclined to the imine-formation side ($K_2 << 1$) because the resulted **V** bears double-chained and is firmly buried in the hydrophobic region. Moreover, formation of bilayer membrane accompanied by the increase of **V** in the aggregate would assist the dehydrocondensation of N_2 and dodecylamine to form **V** itself because interior of bilayer membrane is more hydrophobic than that of micelle. As a result, the imine migration from N_1 to N_2 proceeds efficiently, the conversion of the amine migration being up to 90% with neither catalyst nor pH control.

A schematic representation of the morphological transformation from micron-sized granular aggregates to GV is shown in Fig. 4. When the molar fraction of **V** amounted to *ca.* 50% as determined from ^{1}H NMR measurements, the morphological transformation proceeded from micelles to granular aggregates. The growth of these self-assembled structures would be achieved not only by conversion from $\mathbf{N_1}$ to double-chained amphiphile **V** but also by incorporating hybrid micelles containing the already converted **V**, which makes a spontaneous curvature of the surface of aggregates. Electrolyte **E**, which is produced by the hydrolysis of $\mathbf{N_1}$ within the aggregate, may also contribute to enlarging the aggregate by filling water of the exterior phase. The morphological transformation from the micron-sized granular aggregates to the spherical GVs is, thus, achieved by the stabilization of the membrane by the accumulated double-chained amphiphile **V** and by the minimization of the surface area of the resulting vesicular structure.[5, 14]

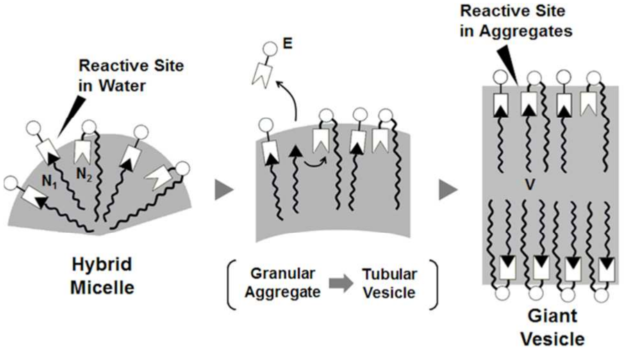

Fig. 4 Schematic representation of the morphological transformation from micelles to giant vesicles accompanied with the accumulation of **V** in the hybrid aggregates ($\mathbf{N_1}$+ $\mathbf{N_2}$).

Finally, the emergence of kinetically stable aggregates of submicron-size as minor products will be mentioned here (ESI Fig. S12). DLS measurements revealed that these aggregates remained unchanged even 120 h after the preparation. The high stability of the submicron-size aggregate may be interpreted by means of a tight packing of amphiphilic imines that are not hydrolyzed (ESI Fig. S13). However, the high conversion of **V** (90%) strongly suggests that the formation of spherical GVs is the major process.

Conclusions

We found that the morphological transformation of amphiphilic aggregates from micelles to GVs proceeded, induced by the migration of amine with a long alkyl chain from imine-type amphiphile $\mathbf{N_1}$ to aldehyde-type amphiphile $\mathbf{N_2}$ to yield double-chained amphiphile **V** and electrolyte **E**. Note that the efficient migration of an alkylamine with a long alkyl chain from amphiphilic imine to amphiphilic aldehyde within the aggregate was attributable to the utilization of a hydrophilic or hydrophobic environment depending on the molecular structure of amphiphilic imines. The current system is a good example of the material conversion of amphiphiles in a locally lipophilic environment in water, which can be utilized as a conventional method for advanced organic synthesis.[2]

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Notes and references

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† Electronic Supplementary Information (ESI) available: Full experimental details, spectral charts of synthetic amphiphiles, introduction of the equations expressing molar fractions of each component consisting of aggregates from ^{1}H NMR signal areas, and dynamic light scattering measurement of morphological transformation of aggregates consisting of $\mathbf{N_1}$ and $\mathbf{N_2}$. See DOI: 10.1039/b000000x/

‡ The molar fraction of each component of aggregates was determined as a function of time by applying the corresponding equations (equations (6)-(9) described in Electronic Supplementary Information) to ^{1}H NMR spectroscopic data.

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