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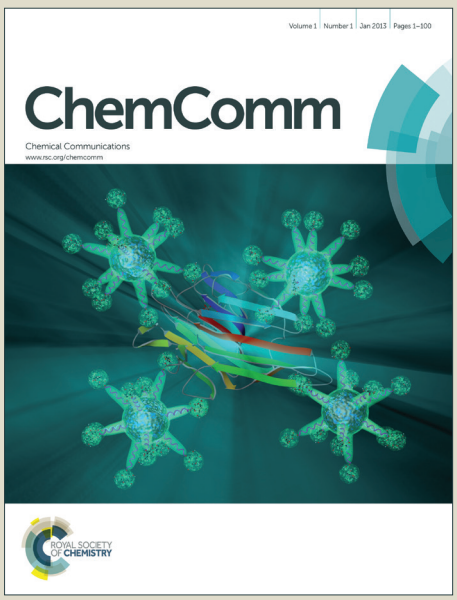

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Three-Component Vesicle Aggregation Driven by Adhesion Interactions between Au Nanoparticles and Polydopamine-Coated Nanotubes

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COMMUNICATION

Three-Component Vesicle Aggregation Driven by Adhesion Interactions between Au Nanoparticles and Polydopamine-Coated Nanotubes

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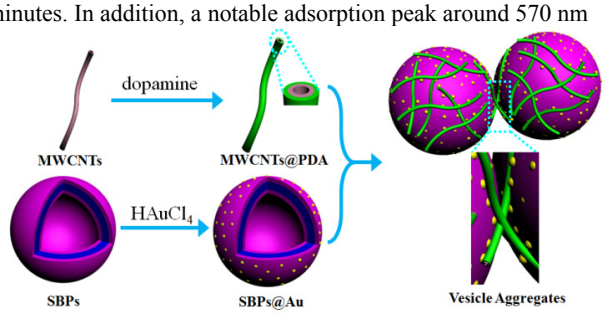
Large-scale and robust vesicle aggregates were obtained through molecular recognitions among cell-sized polymer vesicles, carbon nanotubes and AuNPs driven by adhesion interactions between Au and polydopamine, and vesicle fusion were avoided effectively in such a three-component vesicle aggregation process.

Cell-cell aggregation (CCA) is one of the most important biological processes in the living systems, playing crucial roles in hemostasis, inflammation, embryogenesis, immune response, and development of neuronal tissue.[1] However, despite of the great significance; it is very difficult to directly investigate CCA due to the great complexity of the biomembranes and proteins involved. Alternatively, people have used the synthetic vesicles as model systems to mimic CCA by studying vesicle-vesicle aggregation (VVA) due to the similarities between vesicles and cell membranes in bilayer structure and the properties of fluidity, semipermeability, and deformability, etc.[2-4] Hitherto, the VVA studies have progressed from the simple adhesion of several vesicles to the complicated behaviour of constructing multivesicular arrays as well as to the artificial tissue-like entities. Generally, these reported VVA events are performed among vesicles driven by intervesicular molecular recognition interactions including hydrogen bonding, electrostatic interactions and host-guest interactions or by intervesicular chemical bondings like click chemistry.[5-18] Two-component VVA between nanoparticles and vesicles has also been reported induced by the metal ion coordinations.[19] However, in nature, some CCA events happen among multi-components like cell membranes, adhesion proteins and cytoskeletal filaments. Inspired from them, herein, we reported for the first time on a three-component VVA process through the molecular recognitions among vesicles, nanoparticles and nanotubes driven by the adhesion interactions between metal and polydopamine (PDA), which also represents a new driving force for VVA.

The whole VVA process is illustrated in Scheme 1. Micro-sized hyperbranched polymer vesicles with thiol groups on the surface (SBPs) were selected as the model system, which were coated by Au nanoparticles (AuNPs) on the surface through the reduction of $HAuCl_4$ to form SBPs@Au composites. Meanwhile, PDA coated multiwall carbon nanotubes (MWCNTs@PDA) were prepared by inserting MWCNTs into dopamine solution. Subsequently, SBPs@Au were linked by MWCNTs@PDA to get large vesicle aggregate through the adhesion interaction between AuNPs and PDA layers.[20]

Previously, we prepared a polymer vesicle named as branched-polymersome (BP) by the aqueous self-assembly of an amphiphilic hyperbranched multiarm copolymer of HBPO-star-PEO with a hydrophobic hyperbranched poly(3-ethyl-3-oxetanemethanol) (HBPO) core and many hydrophilic poly(ethylene oxide) (PEO) arms.[21] Cell-sized bilayer vesicles around 1–10 μm were generated from the self-assembly of HBPO-star-PEO with the molar fraction of PEO segments (f_{EO}) of 67% (Fig. S1, ESI†).[18] Thiol-modified BPs (SBPs) were prepared through the direct hydration of thiol-functionalized HBPO-star-PEOs (HBPO-star-PEO-SHs) obtained by modifying the terminal hydroxyl groups of HBPO-star-PEOs into thiol groups through the chemistry of Bunte salts (Scheme S1, ESI†).[24] The average conversion ratio from the hydroxyl into thiol groups is about 36% according to the ^{1}H NMR result (Fig. S2, ESI†), and the final polymer concentration is 5 mg•mL^{-1}. There are no evident changes in size and morphology when comparing SBPs with BPs, and the average size of SBPs is about 1-10 μm, too. The hollow lumens of SBPs were confirmed by dye encapsulation experiments according to the colour phase-contrast microscopy, and the red rhodamine dyes inside the lumens could be clearly discerned from the skins (Fig. S3a, ESI†). In addition, the fluorescent image of the pyrene-labelled SBPs (inset of Fig. S3a, ESI†) shows a significant decrease in fluorescence intensity toward the centre of the particle, a typical evidence to support the vesicular structure. The SEM image (Fig. S3b, ESI†) also support it by showing the collapsed microsized spherical particles.

Subsequently, SBPs were modified with AuNPs by adding chloroauric acid into SBP aqueous solution, followed with the reduction by $NaBH_4$. After adding the reductive agent, the solution turned gradually from pale yellow to purple within 10 minutes. In addition, a notable adsorption peak around 570 nm



Scheme 1 Three-component vesicle aggregation triggered by adhesion interactions between AuNPs and PDA-coated MWCNTs

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attributed to the formation of AuNPs was observed in the UV-Vis spectrum of the solution, while, as a control, no adsorption was observed in the pure SBP solution (Fig. 1a). These results clearly prove the successful reduction from $HAuCl_4$ into AuNPs. The optical microscope (OM) image shows that the spherical and micro-sized vesicles are kept after the formation of AuNPs (Fig. S4, ESI†). Both the SEM (Fig. 1b) and transmission electron microscopy (TEM, Fig. 1c) images indicate AuNPs are coated onto the membranes of SBPs to form SBPs@Au composites. According to the magnified TEM image, the average size of AuNPs is about 9.3 nm. As a further evidence, energy-dispersive X-ray analysis (EDX) measurement (Fig. 1d) shows the SBPs@Au composites are composed of gold, sulfur, carbon and oxygen elements. It should be noted that AuNPs-coated vesicles like SBPs@Au had also been observed by Arms despite of the different preparation methods.[23]

PDA layer coated MWCNTs (MWCNTs@PDA) were prepared by mixing MWCNTs and dopamine into tris buffer at pH=8.5 according to the method reported by Messersmith etal.[20b] The FTIR spectrum of the obtained MWCNTs@PDA (Fig. 2b) shows some characteristic peaks at 3449 cm^{-1} (catechol -OH, stretching vibration), 2921 cm^{-1} (-CH_2-, stretching vibration) and 1642 cm^{-1} (aromatic rings, stretching vibration) assigned to the grafted PDA polymers when compared to the spectrum of pristine MWCNTs. As the direct evidence to support the successful sidewall modification of MWCNTs by PDA layers, TEM image shows the MWCNTs are covered with almost uniform polymer shells (Fig. 2c) since the polymer shells display a lighter contrast than the MWCNT sidewalls owing to a lower electron density. The thickness of PDA layer of the so-formed MWCNTs@PDA is about 12.5 nm when directly measured from the TEM images. The TGA measurements under O_2 show that there is evident thermo degradation from 200 to 500 °C for MWCNTs@PDA (Fig. 2d), which should be attributed to the degradation of the coated PDA layers. Thus, the content of the grafted PDA in MWCNTs@PDA is about 45% according to the TGA results.

Vesicle aggregation was performed by mixing SBPs@Au and MWCNTs@PDA together at Tris buffer (pH=8.5). Dimmers, trimers, multimers and oligomers appeared gradually with time according to OM (Fig. 3a). After 24 hours, vesicle aggregates over 100 μm on average in diameter (Fig. 3b) were observed. Under the phase-contrast model, the vesicles were looked in purple to some degree probably due to the colour of the coated AuNPs on the vesicle surface (Fig. 3c).

The same vesicle aggregate was further dried and then characterized by the OM, SEM and TEM. The structure and morphology of the aggregate were mostly kept after dried (Figs. S5a-5b, ESI†), which indicates the vesicle aggregate is robust. The SEM images (Figs. 3d-3e, Fig. S5c in ESI†) show the vesicles in the aggregate are covered by carbon nanotubes either on the vesicle surface or on the boundary of interconnected vesicles. The same result could be found in the TEM images in spite that the detailed structure of the vesicles could not be discerned due to the limited penetrating power of electrons to the samples (Fig. 3f, Figs. S6-S7, ESI†). Moreover, the magnified TEM images clearly show that AuNPs are coated onto the PDA layers of nanotubes. Meanwhile, the UV/Vis absorption peak also shifted from 570 nm for the isolated SBPs@Au to 528 nm (Fig. S8, ESI†) for the vesicle aggregate due to the decrease of inter-nanoparticle coupling between adjacent AuNPs induced by the adhesions from PDA-coated nanotubes. In other words, as shown in Scheme 1, the vesicles are connected together by the carbon nanotubes through the adhesion between the AuNPs on SBPs@Au and the PDA layers of MWCNTs@PDA.

Two control experiments were conducted to support the abovementioned mechanism. Firstly, AuNPs and MWCNTs@PDA were mixed together at Tris buffer (pH=8.5), and it was found AuNPs were coated onto the PDA layers of the nanotubes after incubation for 24 hours (Fig. S9, ESI†). Secondly, it was found that no vesicle aggregation was formed by mixing SBPs@Au and pristine MWCNTs (Fig. S10, ESI†). Thus, these control experiments further confirm that it is the adhesion interactions between PDA and AuNPs that drive the vesicle aggregation. It is understandable since PDA has proved to be adhesive to many kinds of material surfaces including noble metals, oxides, polymers, semiconductors and ceramics.[20]

Generally, vesicle fusion widely occurs during the vesicle aggregation process since the vesicles are tightly connected in the vesicle aggregate.[24] On the contrary, fusion is generally inhibited

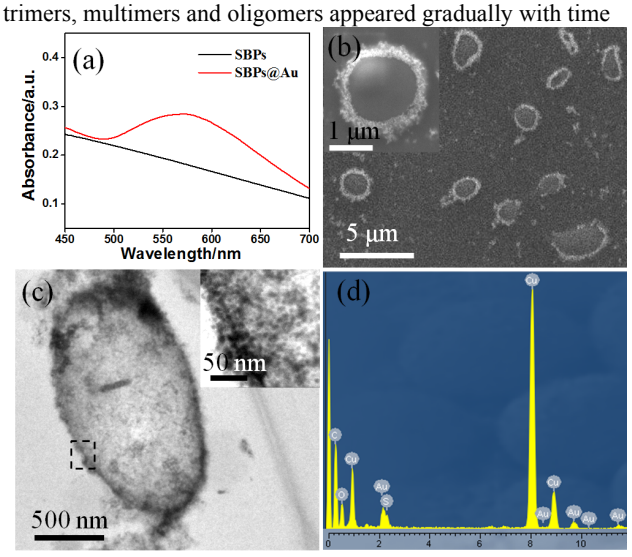

Fig. 1 Characterizations of SBPs@Au composites. (a) UV/Vis spectra; (b) SEM image; (c) TEM image; (d) EDX analysis. The inset in image (c) shows the magnified image indicated by the black dashed framework.

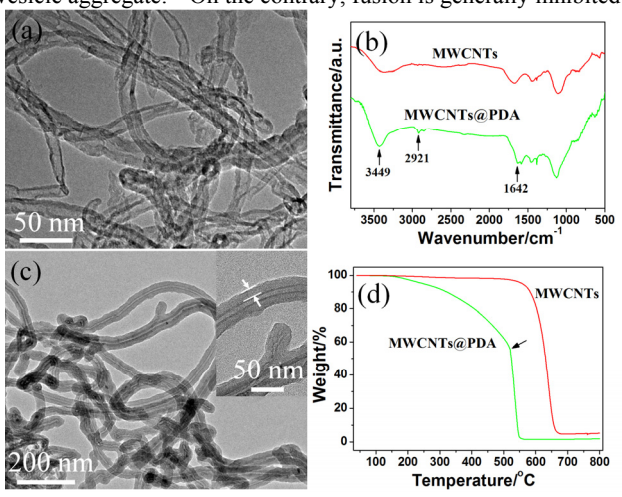

Fig. 2 Characterizations of MWCNTs@PDA. (a) TEM image of pristine MWCNTs; (b) FTIR spectra; (c) TEM image of MWCNTs@PDA; (d) TGA curves under O_2 atmosphere.

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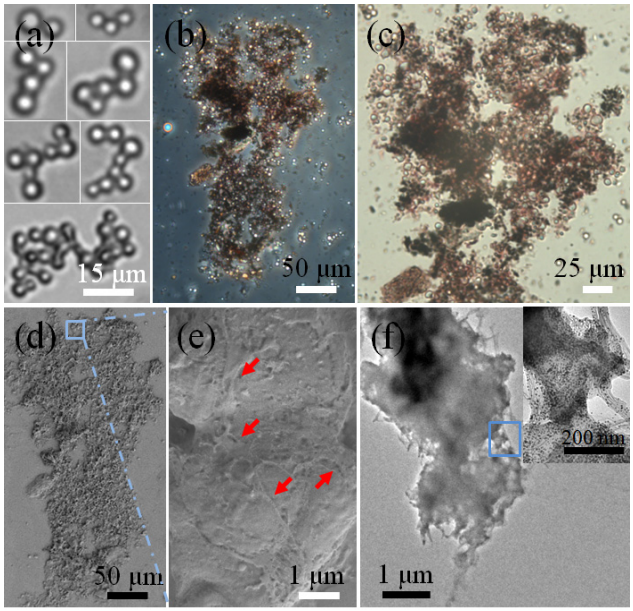

Fig. 3 Characterizations of vesicle aggregates generated by inter-connecting SBPs@Au with MWCNTs@PDA. (a) Vesicle multimers; (b) Optical phase-contrast microscope image; (c) A magnified view of Figure b; (d) SEM image of the same vesicle aggregate after drying; (e) A magnified view; (f) TEM images of the vesicle multimers; inset shows a magnified view of the boundary of two interconnected vesicles.

in the natural CCA process. In the present work, it is expected that the fusion events should be prevented since there are gaps consisting of nanotubes and nanoparticles between the two adhered vesicles (Scheme 1). To prove this point, the vesicle size before and after aggregation (Fig. S11, ESI†) was measured and statistically analysed according to the OM images. SBPs@Au have an average diameter of 4.4 μm based on the statistics of 200 vesicles. The vesicle size is almost kept constant at around 4.3 μm in the vesicle aggregare after the incubation of SBPs@Au and MWCNTs@PDA for 24 and 72 hours. These data do indicate the prohibition of fusion events in the aggregation process.

In conclusion, herein, we have realized a three-component vesicle aggregation event by connecting vesicles with a bridge of nanoparticle/nanotube complexes driven by adhesion interactions between golden nanoparticles and polydopamines. It also represents a multi-component hierarchical self-assembly process through the sequential recognitions of the building blocks of vesicles, nanoparticles and nanotubes. These building blocks are beyond molecules and can be looked as the modules with recognition sites, and thus the spontaneous aggregation process is denoted as "modular self-assembly". Like other molecular recognition interactions to drive self-assembly,[25] we believe the adhesion ability of polydopamine is potential to be a new versatile driving force to trigger self-assembly, especially the "modular self-assembly" and hybrid self-assembly.

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Notes and references

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† Electronic Supplementary Information (ESI) available: [details of experiment and characterization, and supporting figures composed of OM, FM, SEM and TEM results]. See DOI: 10.1039/b000000x/

1 G. De Saint Basile, G. Ménasché and A. Fischer, Nat. Rev. Immunol., 2010, **10**, 568.

2 (a) F. M. Menger and M. I. Angelova, Acc. Chem. Res., 1998, **31**, 789; (b) A.-S. Smith and E. Sackmann, ChemPhysChem, 2009, **10**, 66; (c) H. B. Jin, W. Huang, X. Y. Zhu, Y. F. Zhou and D. Y. Yan, Chem. Soc. Rev., 2012, **41**, 5986.

3 D. E. Discher and A. Eisenberg, Science, 2002, **297**, 967.

4 J. Voskuhl and B. J. Ravoo, Chem. Soc. Rev., 2009, **38**, 495.

5 S. Chiruvolu, S. Walker, J. Israelachvili, F. J. Schmitt, D. Leckband and J. A. Zasadzinski, Science, 1994, **264**, 1753.

6 F. M. Menger and H. L. Zhang, J. Am. Chem. Soc., 2006, **128**, 1414.

7 A. Richard, V. Marchi-Artzner, M. N. Lalloz, M. J. Brienne, F. Artzner, T. Gulik-Krzywicki, M. A. Guedeau-Boudeville and J. M. Lehn, Proc. Natl. Acad. Sci. U.S.A., 2004, **101**, 15279.

8 I. Tsogas, Z. Sideratou, D. Tsiourvas, T. A. Theodossiou and C. M. Paleos, ChemBioChem, 2007, **8**, 1865.

9 E. C. Constable, S. Mundwiler, W. Meier and C. Nardin, Chem. Commun., 1999, 1483.

10 M. Ma, Y. Gong and D. Bong, J. Am. Chem. Soc., 2009, **131**, 16919.

11 G. Stengel, R. Zahn and F. Höök, J. Am. Chem. Soc., 2007, **129**, 9584.

12 R. J. Mart, K. P. Liem, X. Wang and S. J. Webb, J. Am. Chem. Soc., 2006, **128**, 14462.

13 (a) S. K. Mohan Nalluri, J. B. Bultema, E. J. Boekema and B. J. Ravoo, Chem. Sci., 2011, **2**, 2383; (b) A. Samanta, M. C. A. Stuart and B. J. Ravoo, J. Am. Chem. Soc., 2012, **134**, 19909.

14 P. J. Photos, L. Bacakova, B. Discher, F. S. Bates and D. E. Discher, J. Controlled Release, 2003, **90**, 323.

15 G. P. Robbins, D. Lee, J. S. Katz, P. R. Frail, M. J. Therien, J. C. Crocker and D. A. Hammer, Soft Matter, 2011, **7**, 769.

16 T. P. Smart, C. Fernyhough, A. J. Ryan and G. Battaglia, Macromol. Rapid Commun., 2008, **29**, 1855.

17 F. Loosli, D. A. Doval, D. Grassi, P.-L. Zaffalon, F. Favarger and A. Zumbuehl, Chem. Commun., 2012, **48**, 1604.

18 (a) H. B. Jin, Y. L. Zheng, Y. Liu, H. X. Cheng, Y. F. Zhou and D. Y. Yan, Angew. Chem. Int. Ed., 2011, **50**, 10352; (b) H. B. Jin, Y. Liu, Y. L. Zheng, W. Huang, Y. F. Zhou and D. Y. Yan, Langmuir 2012, **28**, 2066; (c) H. B. Jin, W. Huang, Y. L. Zheng, Y. F. Zhou and D. Y. Yan, Chem.–Eur. J., 2012, **18**, 8641.

19 P. L. Kwan, R. J. Mart and S. J. Webb, J. Am. Chem. Soc., 2007, **129**, 12080.

20 (a) H. Lee, S. M. Dellatore, W. M. Miller and P. B. Messersmith, Science, 2007, **318**, 426; (b) B. Fei, B. Qian, Z. Yang, R. Wang, W. C. Liu, C. L. Mak and J. H. Xin, Carbon, 2008, **46**, 1795; (c) Q. Ye, F. Zhou and W. Liu, Chem. Soc. Rev., 2011, **40**, 4244.

21 (a) Y. F. Zhou and D. Y. Yan, Angew. Chem. Int. Ed., 2004, **43**, 4896; (b) Y. F. Zhou, D. Y. Yan, W. Y. Dong and Y. Tian, J. Phys. Chem. B, 2007, **111**, 1262.

22 H. Wolf, H. Ringsdorf, E. Delamarche, T. Takami, H. Kang, B. Michel, C. Gerber, M. Jaschke, H. J. Butt and E. Bamberg, J. Phys. Chem., 1995, **99**, 7102.

23 J. Z. Du, Y. P. Tang, A. L. Lewis and S. P. Armes, J. Am. Chem. Soc., 2005, **127**, 17982.

24 R. J. Mart, K. P. Liem and S. J. Webb, Pharm. Res., 2009, **26**, 1701.

25 (a) K. Wang, C. Y. Wang, Y. Wang, H. Li, C. Y. Bao, J. Y. Liu, S. X. A. Zhang and Y. W. Yang, Chem. Commun., 2013, **49**, 10528; (b) D. D. Zheng, D. Y. Fu, Y. Wu, Y. L. Sun, L. L. Tan, T. Zhou, S. Q. Ma, X. Zha and Y. W. Yang, Chem. Commun., 2014, **50**, 3201; (c) H. Li, D. X. Chen, Y. L. Sun, Y. B. Zheng, L. L. Tan, P. S. Weiss and Y. W. Yang, J. Am. Chem. Soc. 2013, **135**, 1570.

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