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Supporting information

Sticky Tubes and Magnetic Hydrogels Co-Assembled by a Short Peptide and Melanin-like Nanoparticles

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Experimental Section

Materials

Dopamine hydrochloride and 1,1,1,3,3,3-hexafluoro-2-propanol (HFP) were purchased from Sigma-Aldrich. The diphenylalanine was purchased from Bachmen (Bubendorf, Switzerland). The peptides were synthesized in solution phase and do not contain TFA salt. To avoid any pre-aggregation, fresh stock solutions of the peptides were prepared for each experiment. Ferrous chloride and Ferric chloride hydrated salts were purchased from BDH Chemicals LTD (Poole, England).

Substrate and surface modification

Silicon wafers (100) with a diameter of 2 inches were coated with 50 nm titanium (as measured by quartz crystal) by electron beam evaporation (TFDS-141E, VST) at a rate of 1 Å/sec. 1cm X 1cm titanium surfaces were sonicated five min in ethanol, washed with Triple Distilled Water (TDW) and dried under nitrogen. The clean surfaces were used for the HR-SEM analysis.

Synthesis of PDA spheres

PDA spheres were synthesized by self-oxidation in a Tris HCl buffer/isopropyl alcohol. Briefly, the Tris-buffer solution (10 mM, pH =8.5) was mixed with isopropyl alcohol (IPA) in a ratio of 5:2. Then, dopamine hydrochloride (1.5 mg/mL) was added to the mixed solution. The solution was stirred three days. After the reaction, the product was centrifuged and washed with deionized (DI) water three times. The washed powder was then lyophilized.

Synthesis of PDA/Fe_3O_4 core/shell spheres

PDA spheres (10.0 mg) were dispersed in DI water (30 mL) by sonication for 15 minutes. $FeCl_2$ (0.15 mmol) and $FeCl_3$ (0.3 mmol) mixed solution (20 mL) was added to the PDA spheres suspension under stirring. KOH solution (0.85 mmol) was then added to the PDA spheres suspension. After the reaction, the product solution was centrifuged and washed with TDW three times. The washed powders were dried at 60°C.

Self- assembly of diphenylalanine in the presence of PDA or PDA/Fe_3O_4 core/shell spheres

Fresh stock solution of the diphenylalanine peptide was prepared by dissolving the lyophilized forms of the peptide in HFP to a concentration of 100 mg/mL. Spherical PDA nanoparticles or PDA/Fe_3O_4 core/shell spheres were weighted and dispersed in TDW by sonication for 15 minutes to a concentration of 1 mg/mL. This process yielded a black solution. The self-assembly of DPA in the presence of either PDA spheres or PDA/Fe_3O_4 core/shell spheres was performed at several different concentration ratios. In the co-assembly experiments the peptide stock solutions in HFP and PDA or PDA/Fe_3O_4 core/shell MNPs based dispersion were blended together to the desired final concentration. In the case of condition 1 and 2 (co-assembly in the presence of PDA spheres) the final effective concentration of the dipeptide were 2 mg/mL or 5mg/mL respectively. The concentration of the PDA spheres was 1mg/mL. In the case of condition 3 and 4 (Co-assembly in the presence of PDA/Fe_3O_4 core/shell spheres) the final effective concentration of the dipeptide was 2 mg/mL or 5 mg/mL respectively. The concentration of PDA/Fe_3O_4 core/shell spheres was 1 mg/mL. The blended mixtures were incubated for 18 h at room temperature.

Freeze-drying of the hydrogel

The hydrogel was floated on liquid nitrogen N_2(l) for 20 min until it was frozen. Then, the frozen hydrogel, was lyophilized using a freeze-dryer (LABCONCO Free zone 4.5 plus.) for two days in order to completely dry the samples. The frozen hydrogels were transferred to the HR-SEM apparatus for morphological analysis (HR-SEM).

Rheology measurements

Rheology measurements were performed at 25°C using a Thermo-Haake Rheoscope 1 rheometer (Thermo Electron, Karlsruhe, Germany). A cone-plate sensor was used with a diameter of 60 mm, cone angle of 1° and a gap of 0.022 mm. Shear rate was between 0 and 120 s^{-1}. The rheometer is fitted with a solvent trap and a peltier device

that controls temperature within 298±0.1 K. Oscillatory stress sweeps from 10 to 10000 Pa were measured at a constant frequency of 1Hz at 25°C to obtain storage modulus ($G^{/}$) and loss modulus ($G^{//}$). The frequency sweep measurements were performed in linear viscoelastic range at a constant stress of 100 Pa.

High Resolution Scanning Electron Microscopy (HR-SEM)

A 10 µL drop of the dipeptide, PDA/Fe_3O_4 core/shell spheres and the co-assembled mixtures incubated at RT (for several time intervals) were placed on a glass cover slip or clean titanium surfaces and allowed to dry at RT. The substrates were then coated with gold using a Polaron SC7640 Sputter Coater. SEM analysis was performed using a high resolution scanning electron microscope (HR-SEM, Serion equipped with X-MAX20 SDD Inca 450 EDS LN^2 free detector) operating at 1 kV.

Transmission Electron Microscopy (TEM)

A 10 µL drop of the dipeptide, PDA/Fe_3O_4 core/shell spheres and the co-assembled mixtures incubated at RT for 18h was placed on 200-mesh copper grid, covered by carbon-stabilized Formvar® film (Electron Microscopy Science, PA, USA). After 1 min, excess fluid was removed from the grid. The samples were analyzed using a Tecnai T12 G^2 Spirit (Cryo-TEM) operating at 120 kV.

Fourier Transform Infrared Spectroscopy (FT-IR)

Infrared spectra were recorded using a Nicolet 6700 FT-IR spectrometer with a deuterated triglycine sulfate (DTGS) detector (Thermo Fisher Scientific, MA, USA). The peptide solutions and the assembled samples were deposited on a CaF_2 plate and dried under vacuum. The peptide deposits were re-suspended with D_2O and subsequently dried to form thin films. The re-suspension procedure was repeated twice to ensure maximal hydrogen-to-deuterium exchange. The measurements were taken using a 4 cm^{-1} resolution and averaging 2000 scans. The transmittance minimal values were determined by the OMNIC analysis program (Nicolet). FT-IR analysis of the PDA/Fe_3O_4 core/shell magnetic nanoparticles and the magnetic hydrogels were performed using a KBr Plate.

X-Ray Photoelectron Spectroscopy (XPS)

X-ray Photoelectron Spectroscopy (XPS) measurements were performed using a Kratos Axis Ultra X-ray photoelectron spectrometer (Karatos Analytical Ltd., Manchester, UK) using an Al Kα monochromatic radiation source (1,486.7 eV) with 90° takeoff angle (normal to analyzer). The high-resolution XPS spectra were collected for O 1s peaks with pass energy 20 eV and step size 0.1 eV. Data analysis was performed using Casa XPS (Casa Software Ltd.) and Vision data processing program (Kratos Analytical Ltd.).

Adhesion test

A 10 μL drop of the DPA, PDA and the co-assembled mixtures (condition 2) incubated at RT for 18 h were placed on a clean titanium surface and dried properly. Then, the surfaces were washed with water using a pipette and dried for morphological analysis (HR-SEM).

UV-Vis spectroscopy

UV-Vis absorption spectra of DPA, PDA and the co-assembled peptides (condition 2) recorded using a UV/Vis spectrophotometer (SHIMADZU, UV-1650PC).

XRD analysis

The phase of the product was identified by X-ray powder diffraction (***X***-Ray Diffractometer - D8 Advance), using Cu K α (λ = *0.15406 nm) and* Solid state NaI dynamic scintillation detector. Full Diffrac^{plus} package softwarewas used for data acquisition, phase analysis, crystallography and thin film characterization

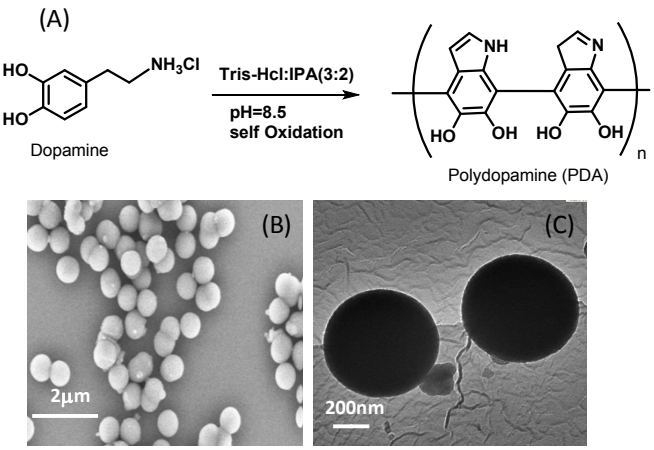

Figure S1. (A) The synthetic route adopted for the synthesis of spherical polydopamine particles, PDA. (B) Representative SEM and (C) TEM micrographs of the spherical PDA nanoparticles.

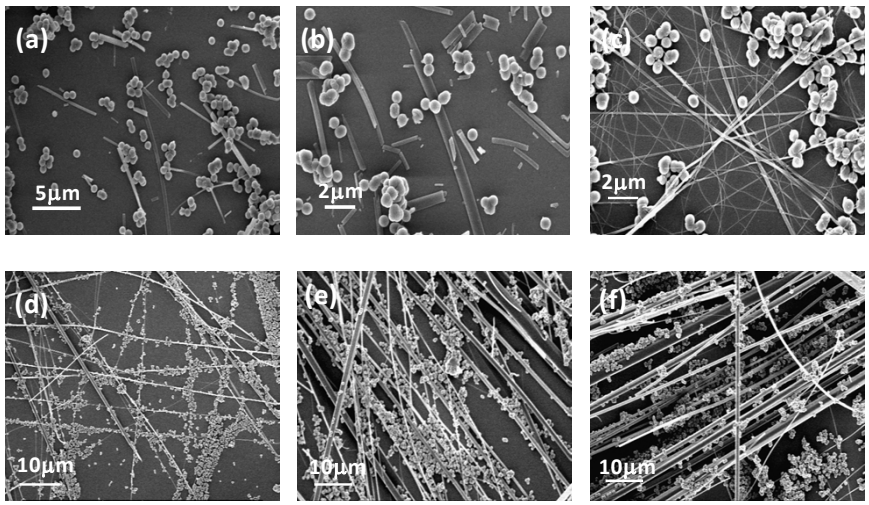

Figure S2. Representative SEM micrographs of different morphological states at different time interval of the co-assembly process (condition 2) after (A) 0 min, (B) 30 mins (C) 2h (D) 4h, (E) 6h and (F) 8h.

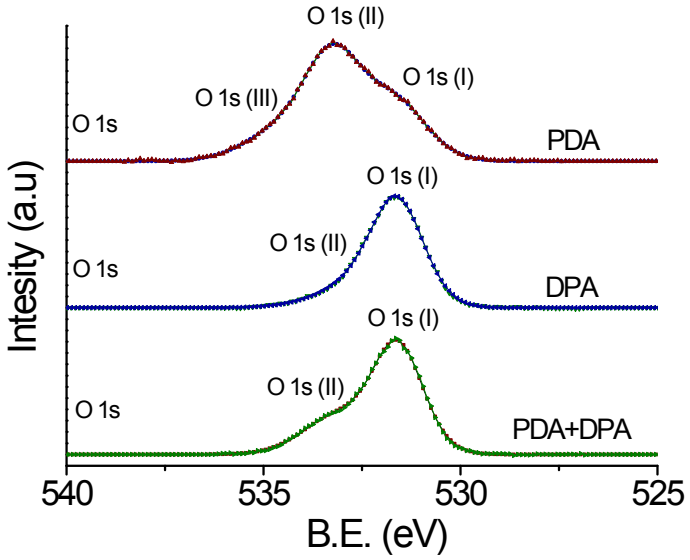

Figure S3. High resolution XPS spectra of O 1s regions for PDA, DPA and the co-assembled structures formed by PDA and DPA (condition 2).

Sample Number	Functional group	Binding energy (eV)	%Area O1s(I)	O1s (II)
PDA	O1s	531.0	29.2	58.3
DPA	O1s	529.0	75.6	24.4
PDA+DPA	O1s	528.4	70.0	30.2

Table T1. Binding energy and % area assignments of the XPS peaks for O1s region for PDA, DPA and the co-assembled structures (condition 2).

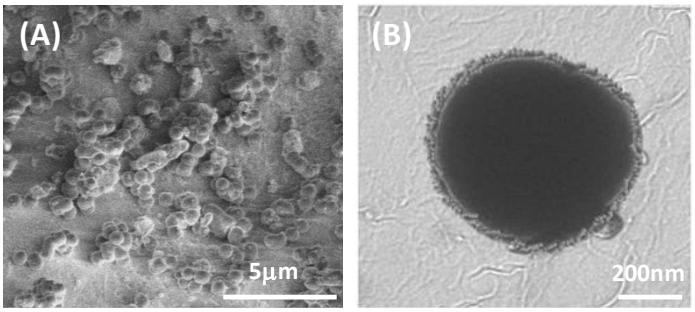

Figure S4. Representative (A) SEM and (B) TEM micrographs of the spherical PDA/Fe_3O_4 core/shell spherical nanoparticles**.**

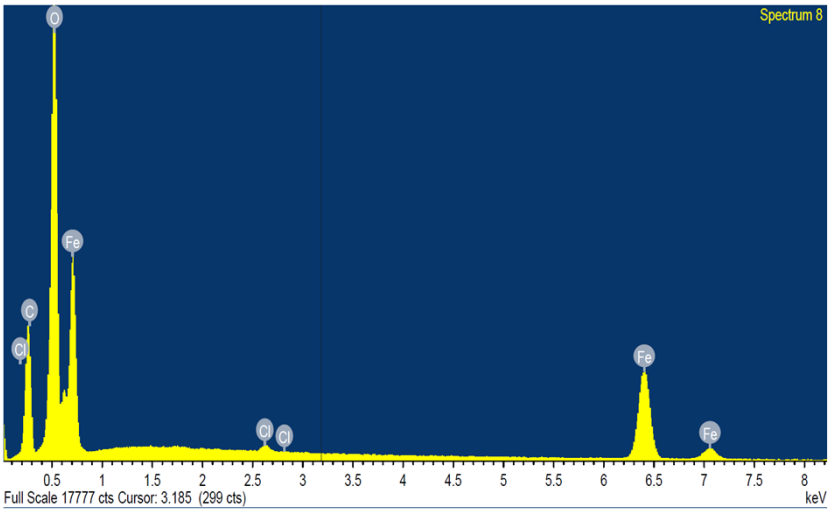

Figure S5. EDS analysis of PDA/Fe_3O_4 core/shell spheres nanoparticles.

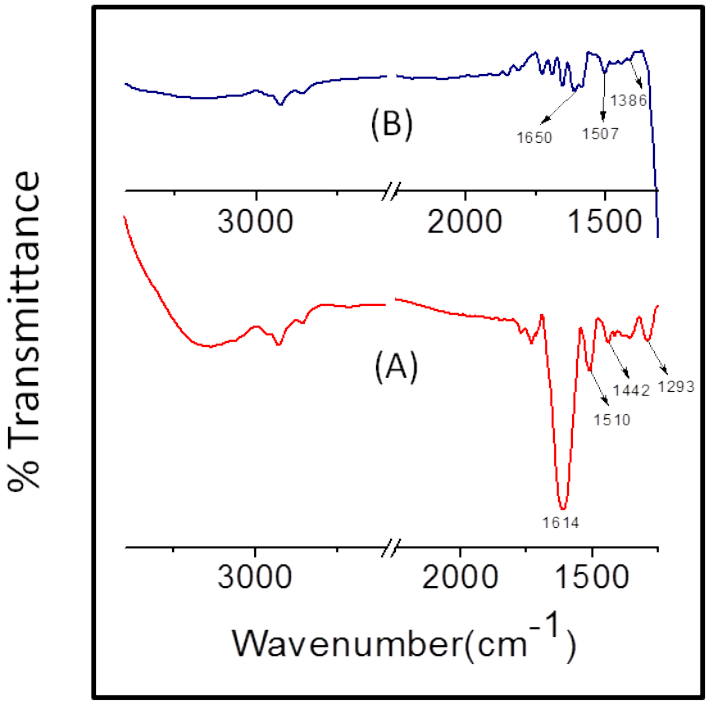

Figure S6. FT-IR transmittance spectra of as synthesized (A) PDA (B) PDA/Fe_3O_4 core/shell spheres.

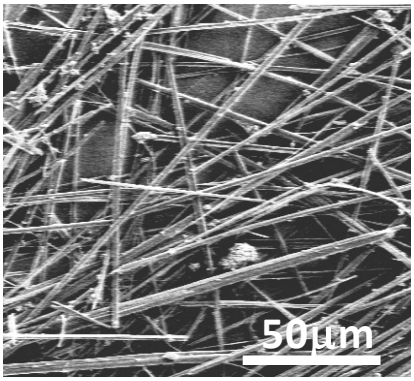

Figure S7. Representative HR-SEM micrograph of the mixture formed by already formed DPA tubes with PDA/Fe_3O_4 core/shell MNPs.

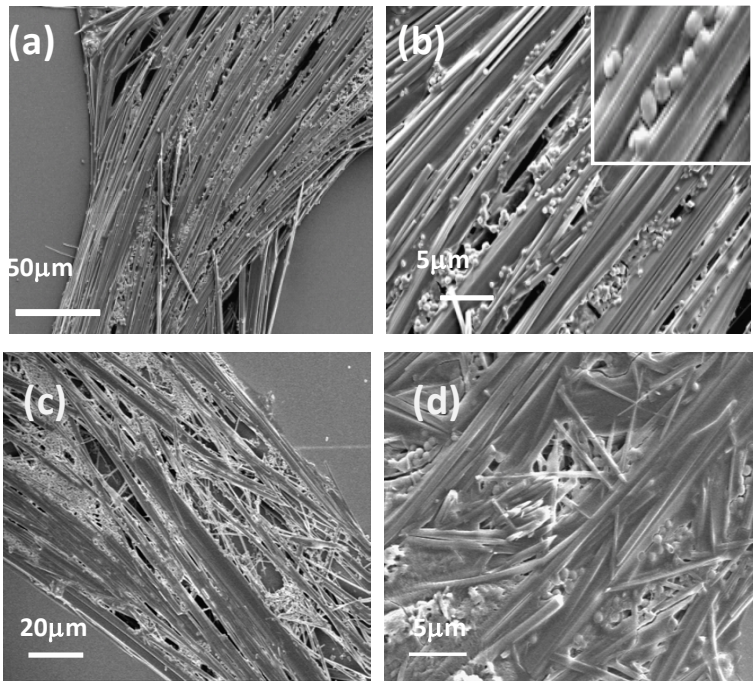

Figure S8. Representative HR-SEM micrographs of the air-dried hydrogel formed by co-assembly under condition 4 (a, b) and condition 3 (c, d).

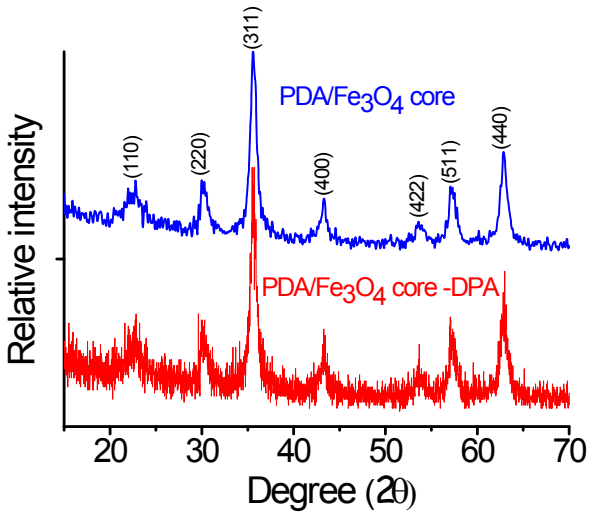

Figure S9. XRD analysis of PDA/Fe_3O_4 core/shell spheres and the hydrogel formed by PDA/Fe_3O_4 core/shell spheres and DPA.

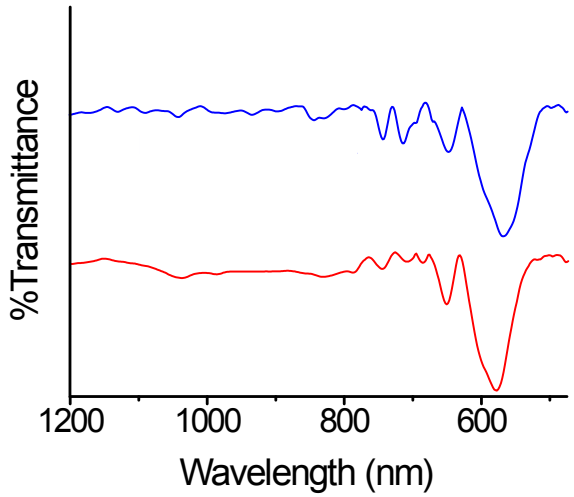

Figure S10. FT-IR transmittance spectra of PDA/Fe_3O_4 core/shell spheres (red) and the hydrogel formed by PDA/Fe_3O_4 core/shell spheres and DPA (blue).

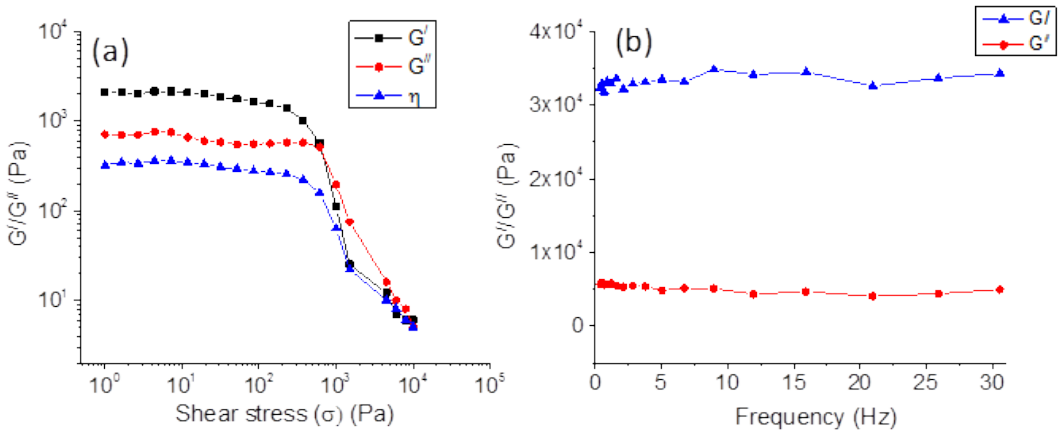

Figure S11. (a) Variation of storage modulus ($G^{/}$) and loss modulus ($G^{//}$) with shear stress (σ) of the hydrogel. (b) Variation of storage modulus ($G^{/}$) and loss modulus ($G^{//}$) with frequency of the hydrogel.