Supporting information

Multifunctional Integrated Compartment Systems for Incompatible Pickering Interfacial Catalysis Cascade Reactions Based on Responsive Core–Shell Nanoparticles

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Experimental Procedures

Materials

NaCas was purchased from Zhengzhou Taber Trading Co. Ltd. Chloroauric acid trihydrate ($HAuCl_4 \cdot 3H_2O$, 47% Au base), Cy5 and ethyl acetate were obtained from Aladdin Chemical Reagent Company (Shanghai, China). Sodium borohydride ($NaBH_4$) was purchased from Lanzhou Institute of Chemical Physics (Lanzhou, China). SBA-15 (pore size: 6-11 nm, SSA: 550-600 m^2/g) and metal organic framework materials (100-400 nm) were obtained from Nanjing Xianfeng nanomaterials technology Co., Ltd. Methyl phenyl sulfide and fluorescent dyes, i.e., FITCs, Nile Blue A, and Nile Red, were bought from Sigma-Aldrich, Inc. (St. Louis, MO, USA). All other chemicals were analytically pure.

NaCas synergistic mesoporous nanoparticle to construct CO_2/N_2-response emulsion.

In order to make emulsions, NaCas-mesoporous nanoparticle dispersions and oil phases of equal volume (10 mL) were typically combined and sheared in 25 mL glass containers using a T10 homogenizer (IKA, Germany) at a speed of 15,000 rpm for 1 minute.

Adding aliquots of CO_2 (67 mL/min) to break up the emulsion and then aerating N_2 to remove the CO_2 allowed us to test the emulsions' responsive performance. Moreover, we used cryo-SEM, TEM, CSLM, and an optical microscope to analyze the emulsion's microscopic properties. The mesoporous particles used included mesoporous silica (MSNP), metal organic framework material (MOF) and SBA-15, of which MOF and SBA-15 were purchased from commercial companies and MSNP was obtained by previous report. The synthesis method of MSNP is as follows: triethylamine (0.5 g) and cetyltrimethylammonium chloride aqueous solution (2.5 mL, 25 wt%) are added into the mixture of water (15 mL) and ethanol (2.5 mL) under stirring condition. After stirring at 60 ℃ for 1 h, aliquots of tetraethyl silicate (1.5 mL) were added. After 2 hours of heating, the mixture was centrifuged to separate the product. The white powder was generated by freeze-drying after being washed with water and ethanol six times.

Characterization of emulsifier interfacial activity

The three-phase contact angle was also measured using ODG-20. First, the samples were pressed into disks approximately 1.5 cm in diameter and 0.2 cm thick. Place the disc into an optical glass cuvette, then add oil slowly, and allowed to stand for 500s. Finally a drop of distilled water was added and the contact angle was calculated after 180s. All samples were repeated 3 times.

Synthesis of NaCas-Au NCs (Au-loaded NaCas).

The growth of Au NCs on NaCas were performed by reducing $HAuCl_4$ with $NaBH_4$ using previous procedure, with slight modifications. Briefly, $HAuCl_4 \cdot 3H_2O$ (10 mM, 4 mL) was mixed with 50 mL of NaCas dispersion (0.5 wt%, pH 7.0). Afterwards, aliquots of freshly prepared ice-cold $NaBH_4$ (10 mM, 0.5 mL) were added gradually to the as-prepared suspension until a stable brown-yellow colloid appeared (2.5 mL of $NaBH_4$). The suspension was incubated at room

temperature to evaluate phycial stabilization for 24 h. Au NP-labelled NaCas was purified using acidic precipation procedure by adjusting the pH to 4.7 and centrifuging at 5000 rpm, followed by repeatedly washing the sediment with deionized water until the clear supernatant occured. Finally, the powered NaCas-Au NCs was obtained by freeze-drying the as-prepared purided samples. AFM images were captured using a Bruker Multimode instrument controlled by a Quadrexed Nanoscope 3D. For the preparation of the samples, a single drop of 0.02 wt% protein solution was placed on a clean silica wafer and dried under vacuum for one day. The Kratos Axis Ultra DLD instrument was used to acquire the XPS spectra, with the C1S line at 284.6 e V serving as a standard. We collected the samples' FT-IR spectra at room temperature using a Perkin Elmer Spectrum RXIFT-IR Spectrometer (USA). Atomic emission spectroscopy with an inductively coupled plasma was used to determine the amount of gold present (ICP-AES, Atom Scan16, TJA Co.). A sample aliquot (10 mg) was combined with nitric acid (5 mL) and hydrogen peroxide (1 mL) before being digested by heating at 300 °C on a hot plate.

HRP-Mimicking Property.

To evaluate the HRP-Mimicking catalytic performances of NaCas-Au NC, TMB was applied as a typical substrate since the oxidation of TMB with H_2O_2 generated colored product (TMB*+) by the peroxidase or its mimics (H_2O_2+TMB→H_2O+TMB*+). Typically, NaCas-Au NC (1 mL), TMB (2 mM, 1 mL), and H_2O_2 (100 mM, 0.5 mL) were put successively into a 5-mL tube. After gentle mixing, the absorption profiles were recorded. HRP-mimicking property of NaCas-Au NC at different reation times and pHs was also evaluated using the above-mentiuoned procedures.

Physicochemical Characterization of NaCas-Au NCs.

TEM observations were performed using a JEM-2100 Plus (JEOL Ltd, Japan) at an acceleration voltage of 200 kV. The sample (0.05 wt%) was dispersed in deionized water and homogenized at 25 °C for 5 min. A drop of the suspension was spread onto copper grids coated with a carbon support film. After 1 min, excess liquid was blotted with filter paper, and the remaining film was allowed to dry for observation.

Atomic force microscopy (AFM) images were acquired on a Bruker Multimode instrument with a Quadrexed Nanoscope 3D controller. Samples were prepared by adding one drop of proteinosome solution (0.02 wt%) onto a clean silica wafer and drying under vacuum for one day.

XPS spectra were recorded on a Kratos Axis Ultra DLD instrument, and the C_{1S} line at 284.6 e V was used as a reference.

The FT-IR spectra of the samples were recorded using a Perkin Elmer Spectrum RXIFT-IR Spectrometer (USA) at room temperature. The sample powder was blended with KBr powder and pressed into tablets before spectra were obtained over the range of 4000 to 500 cm^{-1} at a resolution of 8 cm^{-1}.

Au content were analyzed with an inductively coupled plasma-atomic emission spectrometry (ICP-AES, Atom Scan16, TJA Co.). An aliquot of sample (10 mg) was taken, and 5 mL of nitric acid and 1 mL of hydrogen peroxide were added, and the mixture was heated on a hot plate at 300 °C for digestion.

MSNP loaded GOx

The MSNP powder (125 mg) was suspended in 10 ml phosphate buffer (0.1 M, pH 7.40). Then GOx (30 mg) was added. It was slightly stirred and adsorbed for 24 h at 0-4 ℃. Finally, the free GOx was removed by freeze centrifugation and freeze-dried for standby.

Cascade Catalysis.

Aqueous phase was prepared by dispersing glucose (3.47mmoL) in deinozed water (containing 0.2 wt%NaCas-Au NCs-MSNP-GOx), and aqueous phase and ethyl acetate in the presence of 100mM methyl phenyl sulfide) were mixed at aqual volume fraction (2.5 mL). The stable emulsion cascade system was gernerated by shearing the mixture, and the cascade catalysis reaction proceeded under oxygen atmosphere. The yield was determined by gas chromatography and the products were identified by gas chromatography-mass spectrometry. Averages were plotted for each time period, and error bars were calculated using the standard deviation.

General methods

Experiments in cryo-SEM were performed on a Hitachi S-4800 apparatus set to 2.0 kV. Emulsion samples were frozen with liquid nitrogen before observation, and then sublimated (-90 °C, 30 min) to remove ice crystals and show the surface of the droplet. Moreover, observation of the interfacial structure was also facilitated by scraping the surface of the sample away with a blade before taking immediate measurements. To capture CLSM pictures, a Leica TCS SP5 CLSM was used (Heidelberg, Germany). In brief, Nile blue (0.1%) and Nile red (0.1%) were used to dye the emulsions, which were then mounted on concave confocal microscope slides. Then, the dyed emulsions were excited at 488 nm and 633 nm, repectively, using a argon/krypton laser and a helium/neon (He-Ne) laser. FITC labeled protein: 1 mL sodium bicarbonate-sodium carbonate (0.15 mol / L, pH 9.0) buffer was added to 9 mL NaCl solution (0.15 mol / L). Then 10 mg / mL protein solution was prepared by adding protein into the above solution. Fluorescein (protein: fluorescein mass ratio = 50:1) was added and mixed at 4 ℃ for 12 h before dialysis. Cy5 labeling enzyme: Solution 1: Cy5 (1 mg) added to brown glass bottle. Added 400 μL DMSO to the above solution and stirred for 30 min. Solution 2: 1 mg of enzyme was dissolved in 400 mg μL DMSO. Mix solution 1 and solution 2, then add 15 μL triethylamine, room temperature and stirring reaction for 12 h before dialysis. The adsorption kinetics of NaCas or NaCas-mesoporous particles solutions (0.5 mg mL^{-1}) were determined at pHs 7.0 and 4.7 using an OCA-20 optical contact angle meter (Dataphysics Instruments GmbH, Germany). Interfacial adsorption experiment was performed by placing a drop of aqueous solutions using an inverted syringe tip into an optical glass cell with puried oil and letting the mixture incubate for 500s. High-speed video was used to capture pictures of the drop, and the interfacial tension (γ) was then determined using the drop's geometry using the Young-Laplace equation. The experiments were performed at room temperature in triplicate.

Results and Discussion

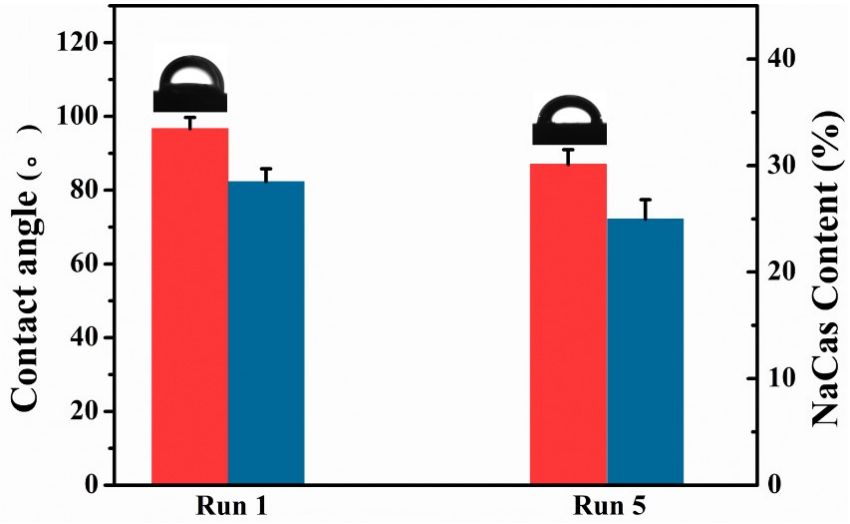

Figure S1 The three-phase contact angle of MSNP-NaCas at 1st and 5th cycle. To clarify the stability of MSNP-NaCas, the three-phase contact angle and NaCas absorption content of particle was examined after treating using CO_2/N_2. We surprisingly showed that those two parameters have the almost same results between the 1 cycle and 5th cycle. Our findings suggest that the Composite structure (MSNP-NaCas) have the excellent stability.

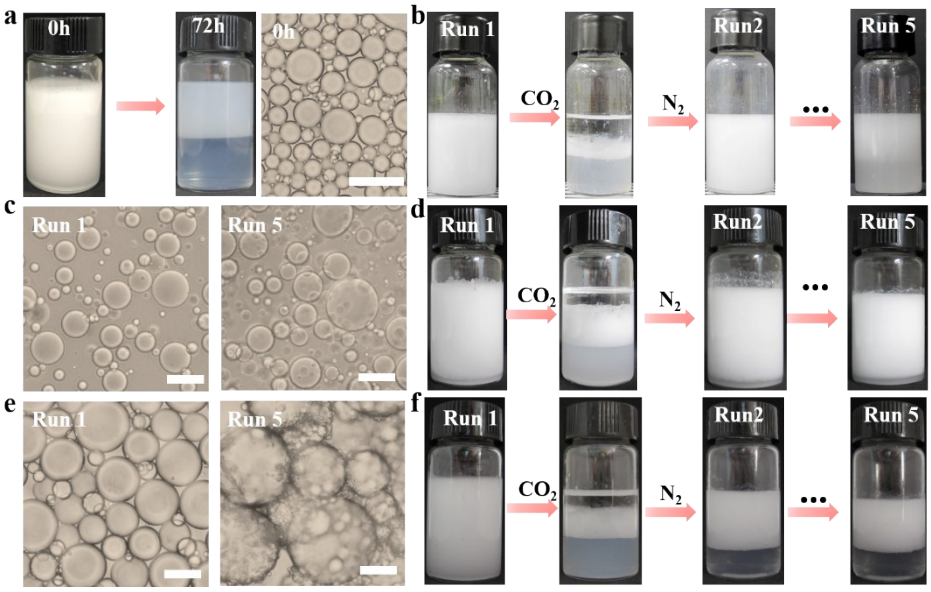

Figure S2 CO_2/N_2-responsive emulsions stabilized by NaCas-MSNP (a) Appearance and optical micrographs of NaCas-MSNP-stabilized emulsion using ethyl acetate as the oil phase after storage for 0h and 72h. The appearance images for the successive CO_2/N_2-triggered NaCas-MSNP stabilized emulsion inversion using ethyl acetate (b), toluene (c-d) and N-heptane (e-f) as oil phase, respectively. After addition of an equal volume of oil phase (ethyl acetate) into water containing 0.2 wt % of the sample (with respect to water), and subsequent homogenizing at a rate of 15 000 rpm for 1 min, different phenomena were observed for these samples. The as-prepared emulsion stabilized by NaCas-MSNP was stable at neutral condition. Interestingly, bubbling CO_2 resulted in complete macroscopic phase separation of the emulsions stabilized. This system, however, rapidly restored the o/w emulsion by shearing the oil-water mixture after CO_2 was removed from the mixture by bubbling N_2. Impressively, this CO_2 switchable emulsion could be reversibly switched on and off over 5 cycles. The emulsifying and responsive performance of NaCas-MSNP remained unaffected throughout the cycles because the type and droplet size of the regenerated emulsions were similar to the equivalents of the original emulsions. In addition to the ethyl acetate/water system, we found that NaCas-MSNP was also a good emulsifier for stabilizing other oils, including toluene and N-heptane. In these systems, all emulsions could be switched on or off through the bubbling of the CO_2/N_2. The type and droplet size of the regenerated emulsions remained almost the same during and after 5 cycles. Scale bar, 50μm.

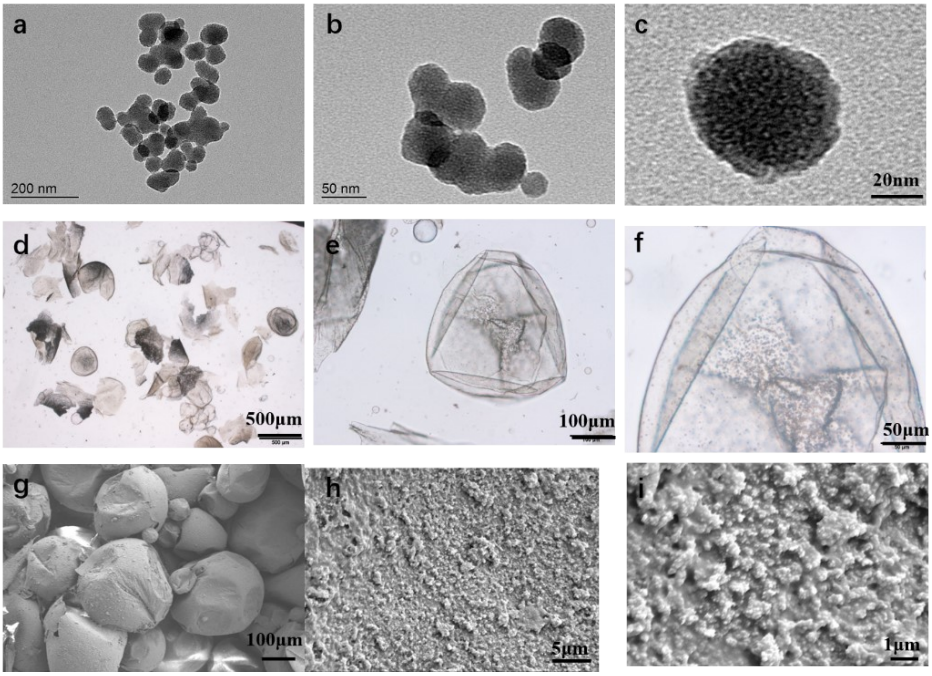

Figure S3 (a-c) TEM image of MSNP; optical micrographs (d-f) and SEM (g-i) of NaCas-MSNP-stabilized emulsion. SEM image reveal that the surface of emulsion droplets consisted of closely packed NaCas-MSNP. Figure S3a-c shows a typical magnification transmission electron microscopy (TEM) image of the synthesized MSNP, in which nanometers of porous structure are evenly distributed inside. Interestingly, in the presence of NaCas, those MSNP are uniformly anchored on the oil-water interface forming oil-in water emulsion, as shown in Figure S3 g-i, an enlarged SEM image.

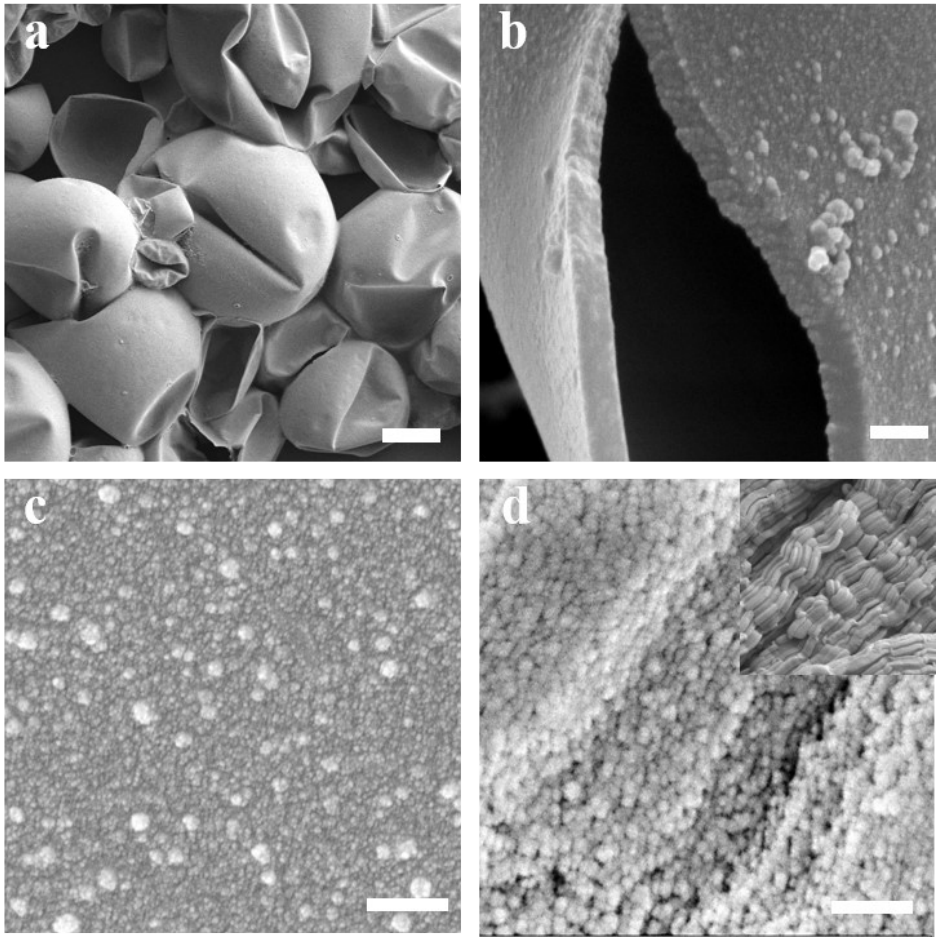

Figure S4 (a-c) SEM image of emulsions stabilized by NaCas-SBA-15. (d) SEM image of SBA-15. The scanning electron microscopy (SEM) images of emulsions showed essentially rough surface morphology composing of closely packed particles, indicating that each particle was an SBA-15 (SBA-15-NaCas) since this particle size was similar to the SEM results of single SBA-15. Scale bar, 10μm (a), 200nm (b) and 100nm (c-d).

CO_2/N_2-responsive emulsions stabilized by NaCas-MOF

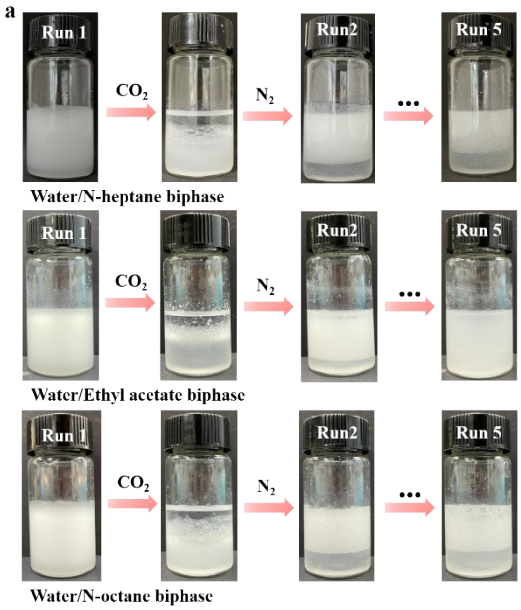

Figure S5 CO_2/N_2-responsive emulsions stabilized by NaCas-MOF. (a) Appearance image of successive CO_2/N_2-responsive NaCas-MOF stabilized emulsion conversion cycles using different oil phase. After addition of an equal volume of oil phase (benzene) into water containing 0.2 wt % of the sample (with respect to water), and subsequent homogenizing at a rate of 15 000 rpm for 1 min, different phenomena were observed for these samples. The as-prepared emulsion stabilized by NaCas-MOF was stable at neutral condition. Interestingly, bubbling CO_2 resulted in complete macroscopic phase separation of the emulsions stabilized. This system, however, rapidly restored the o/w emulsion by shearing the oil-water mixture after CO_2 was removed from the mixture by bubbling N_2. Impressively, this CO_2 switchable emulsion could be reversibly switched on and off over 5 cycles. The emulsifying and responsive performance of NaCas-MOF remained unaffected throughout the cycles because the type and droplet size of the regenerated emulsions were similar to the equivalents of the original emulsions. In addition to the benzene/water system, we found that NaCas-MOF was also a good emulsifier for stabilizing other oils, including toluene and N-octane. In these systems, all emulsions could be switched on or off through the bubbling of the CO_2/N_2. The type and droplet size of the regenerated emulsions remained almost the same during and after 5 cycles.

CO_2/N_2-responsive emulsions stabilized by NaCas-SBA-15

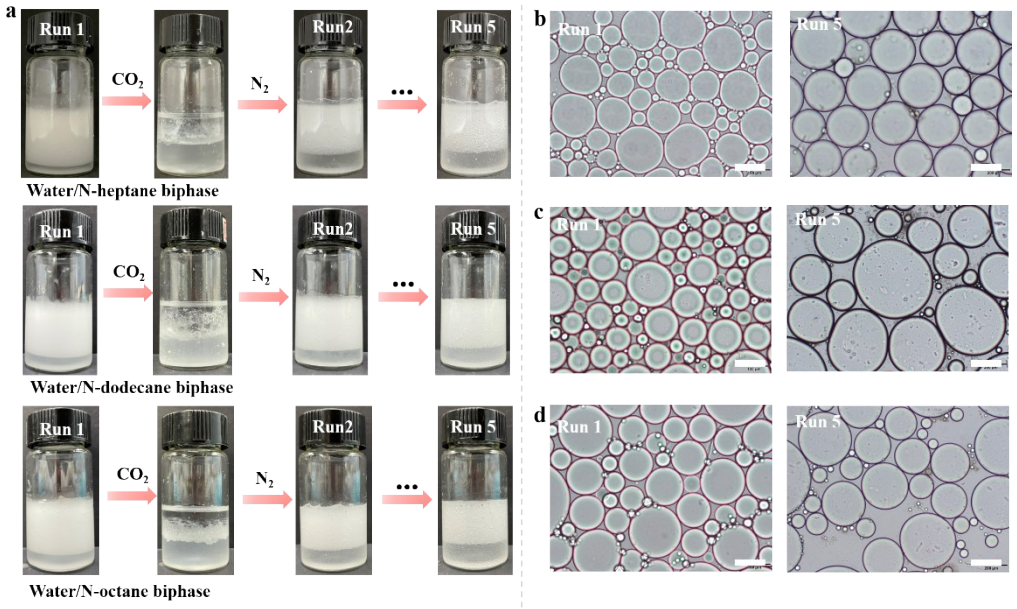

Figure S6 CO_2/N_2-responsive emulsions stabilized by NaCas-SBA-15. (a) Appearance image of successive CO_2/N_2-responsive NaCas-SBA-15 stabilized emulsion conversion cycles using different oil phase. Optical micrographs of NaCas-MOF-stabilized emulsion using N-heptane (b), toluene (c) and N-octane (d) as the oil phase respectively. After addition of an equal volume of oil phase (N-heptane) into water containing 0.2 wt % of the sample (with respect to water), and subsequent homogenizing at a rate of 15 000 rpm for 1 min, different phenomena were observed for these samples. The as-prepared emulsion stabilized by NaCas-SBA-15 was stable at neutral condition. Interestingly, bubbling CO_2 resulted in complete macroscopic phase separation of the emulsions stabilized. This system, however, rapidly restored the o/w emulsion by shearing the oil-water mixture after CO_2 was removed from the mixture by bubbling N_2. Impressively, this CO_2 switchable emulsion could be reversibly switched on and off over 5 cycles. The emulsifying and responsive performance of NaCas-SBA-15 remained unaffected throughout the cycles because the type and droplet size of the regenerated emulsions were similar to the equivalents of the original emulsions. In addition to the benzene/water system, we found that NaCas-SBA-15 was also a good emulsifier for stabilizing other oils, including toluene and N-octane. In these systems, all emulsions could be switched on or off through the bubbling of the CO_2/N_2. The type and droplet size of the regenerated emulsions remained almost the same during and after 5 cycles. Scale bar, 100μm (run 1 of b-d) and 200μm (run 5 of b-d).

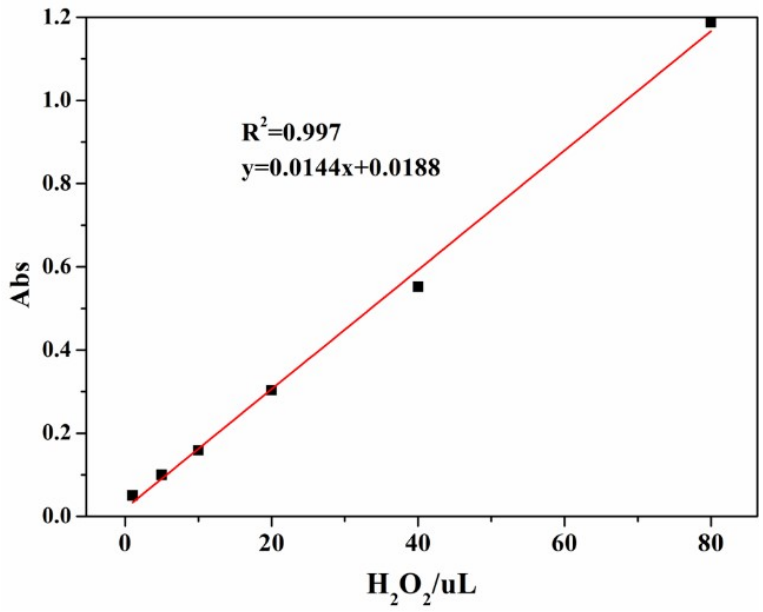

Figure S7 Hydrogen peroxide reaction varies with concentration. According to the amount of hydrogen peroxide produced by msnp GOx, the enzyme activity of GOx after loading is calculated as 95 U / mg.

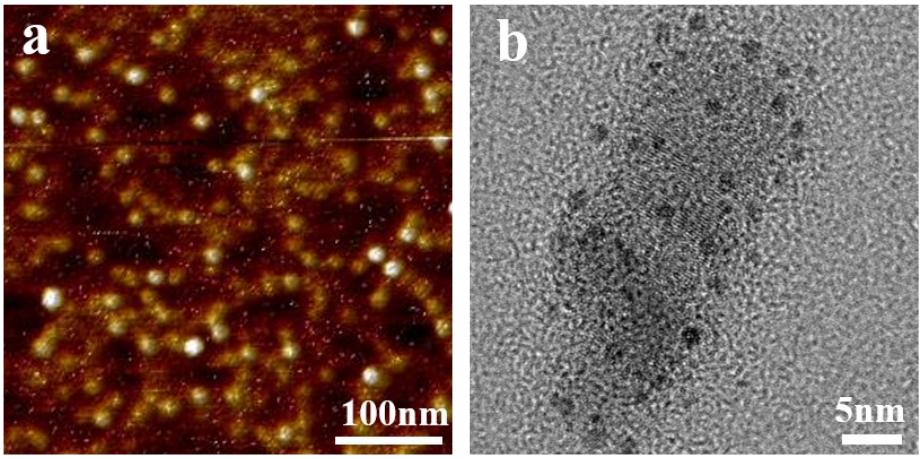

Figure S8 AFM and TEM image of NaCas-Au NCs. Transmission electron microscopy (TEM) analysis found that the Au NCs in the samples are anchored evenly over the NaCas and have a narrow distribution in average size of 1.4 nm. Scale bar, 100nm (a) and 5nm (b).

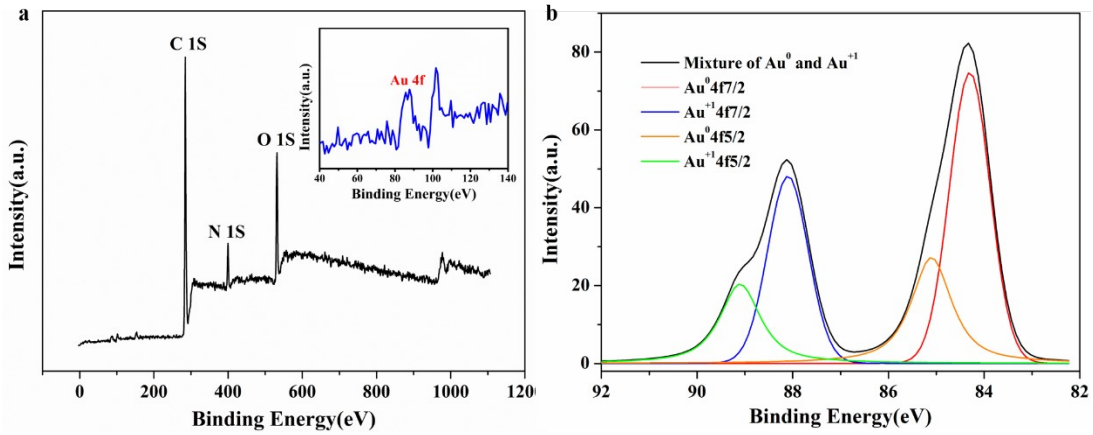

Figure S9 XPS of NaCas-Au NCs. The full range NaCas-Au NCs XPS spectra depicted in Figure which illustrates binding energies of all elements, including Au, O, N, and C, in NaCas-Au NCs. Elemental peaks of C, N, and O were specifically derived from NaCas protein and Au peak was derived from the Au nanocluster. In the full range spectrum, binding energies at 285, 399 and 531 eV were attributed to C1s, N1s, and O1s, respectively. XPS spectrum of Au 4 f is depicted in above Figure. The oxidation state of gold atoms in the NaCas-Au NCs was determined by X-ray photoelectron spectroscopy (XPS).

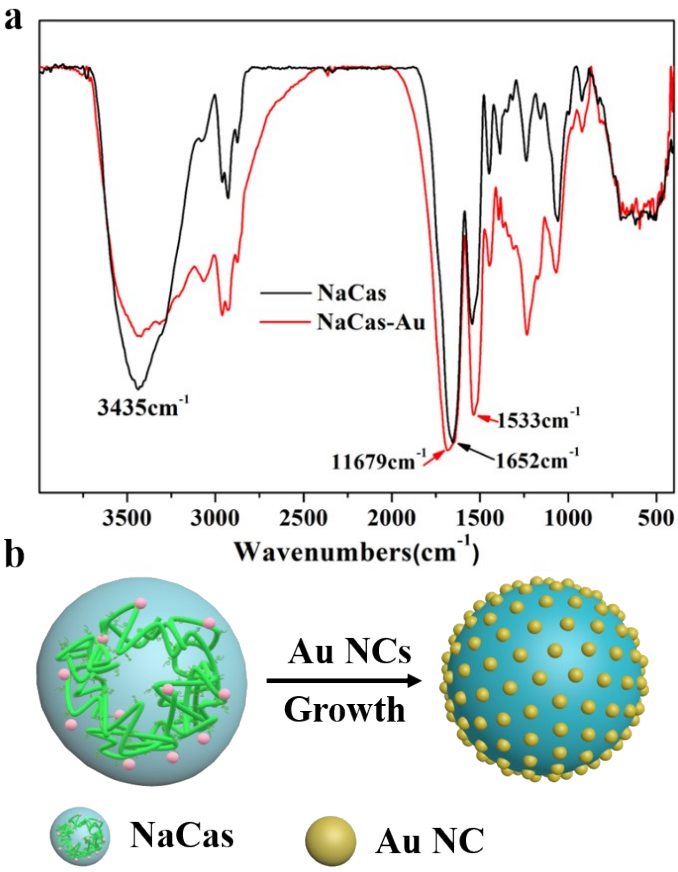

Figure S10 (a) FTIR of NaCas-Au NCs. The shift and enhancement of protein amide I (C=O, 1652cm^{-1}) and amide II (NH_2 in-plane deformation vibration, 1533cm^{-1}) peaks between NaCas and NaCas Au indicate the binding of Au to amide bonds on proteins. (b) Schematic representation of possible mechanisms for the formation of NaCas-Au NCs.

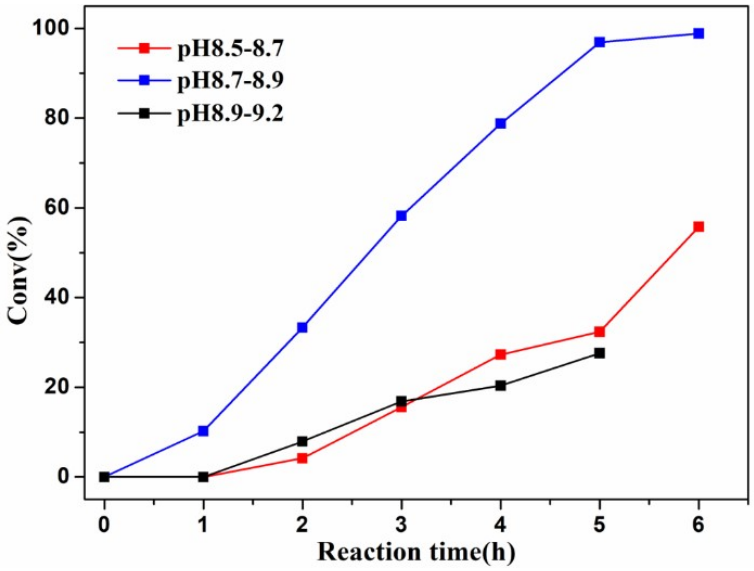

Figure S11 Plot of the conversion of the oxidation of methyl phenyl sulfide catalysed under different pH conditions versus reaction time. As shown in the Figure, too high or too low pH will affect the final conversion. This may be due to the low activity of metal mimetic enzyme under low pH conditions, and higher pH will affect the activity of glucose oxidase.

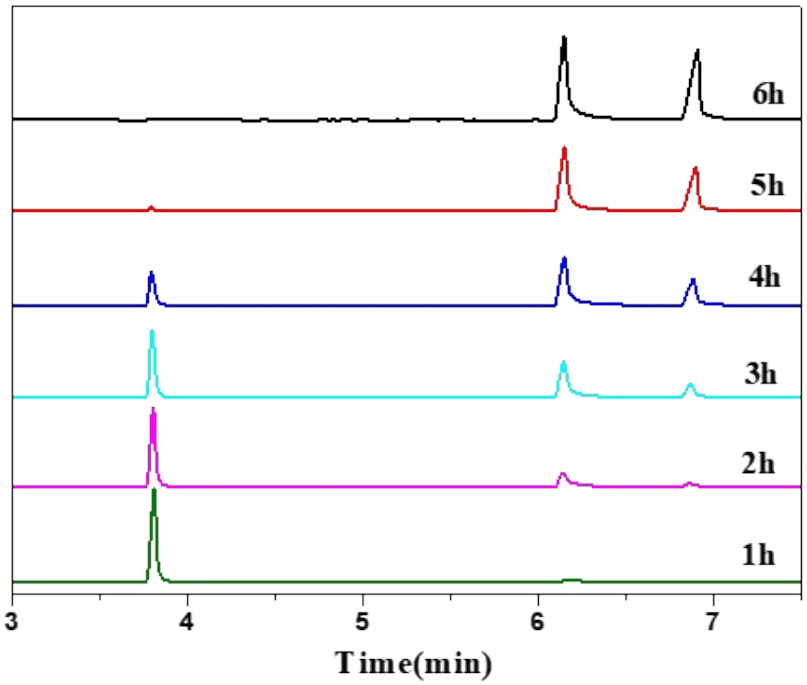

Figure S12 The results of catalyst reaction for NaCas-Au NCs-MSNP-GOx at different times. The substrate is methyl phenyl sulfide (3.79 min). The products of the catalytic reaction were methyl phenyl sulfoxide (6.15 min) and methyl phenyl sulfone (6.91 min). With the extension of time, the substrate peak decreased rapidly, and the transformation was basically complete at 5 hours.

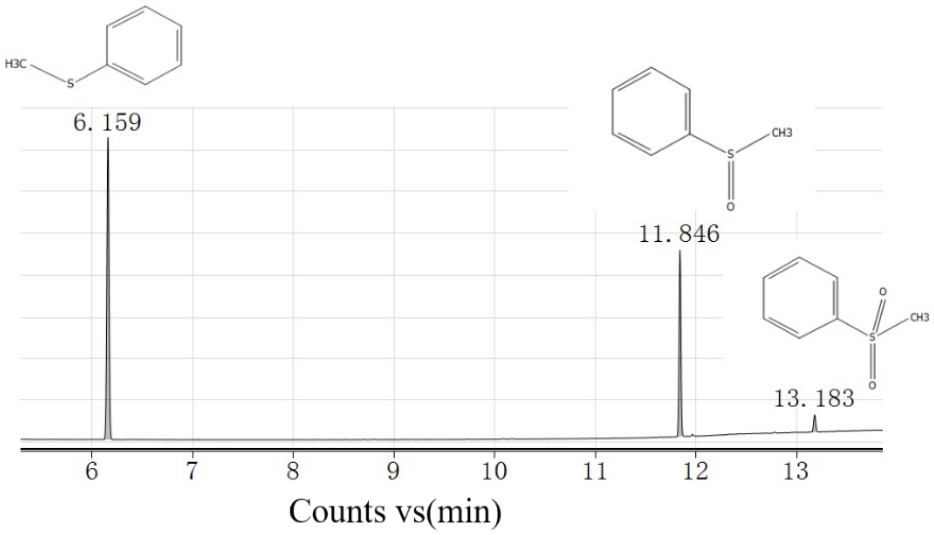

Figure S13 GC-MS of products. The substrate is methyl phenyl sulfide (6.159 min). The products of the catalytic reaction were methyl phenyl sulfoxide (11.846 min) and methyl phenyl sulfone (13.183 min). Due to different instruments and equipment, the peak position is different from the results of GC.

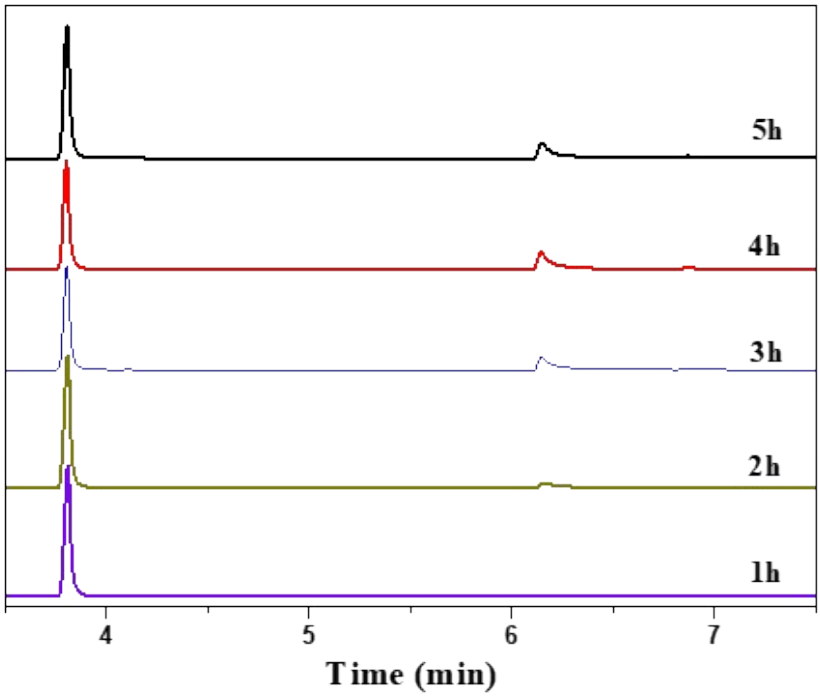

Figure S14. The results of catalyst reaction for NaCas-Au NCs-GOx at different times.

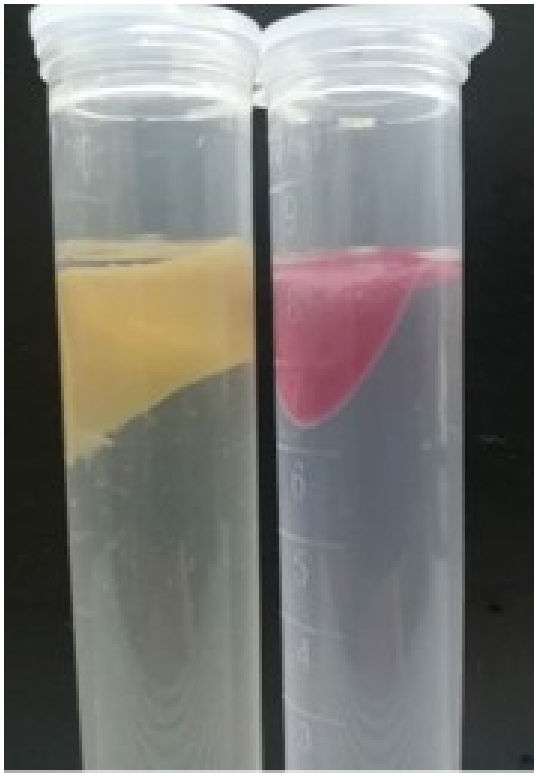

Figure S15. The stability of the catalyst at the gas-water interface. Left (0h), right (24h). The catalyst is easy to oxidize and grow up in the air, which is fatal to the reaction. This system has high advantages because it avoids the oxidation deactivation of catalyst caused by traditional centrifugal drying process.

Table S1 conversion of oxidation of methylphenyl sulphide catalysed under different conditions.

Entry	catalysis	Time	Conversion (%)	Reaction conditions
1	NaCas–Au NCs–MSNP–GOx[a]	5h	96.42	Emulsion
2	NaCas–Au NCs and GOx[b]	5h	48.11	Emulion
3	NaCas–Au NCs[c]	5h	29.62	Emuldion
4	NaCas–Au NCs–MSNP–GOx	5h	0.3	Two phase

[a] an emulsion stabilised by NaCas–Au NCs–MSNP–GOx; [b] an emulsion stabilised by NaCas–Au NCs, and GOx was dispersed in a continuous phase; [c] an emulsion stabilised by NaCas–Au NCs, and H_2O_2 was added in a continuous phase; [d] two phases: NaCas–Au NCs–MSNP–GOx in a biphasic water/ethyl acetate system.

Table S2 The recycling results of NaCas–Au NCs–MSNP–GOx-catalysed oxidation of methylphenyl sulphide in the Pickering emulsion.

Cycle	Conversion (%)[a]	Selectivity (%)	
		Sulfoxide	Sulfone
1	96.42	60	40
2	60.76	80	20
3	42.86	90	10

[a] The reaction time is 5h, the reaction temperature is 40°C.