Supporting Information

Hybrid-cell membrane coating nanocomplexes loading chikusetsusaponin IVa methyl ester for combinational therapy against breast cancer assisted with Ce6

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1. Experiment and methods

1.1 PLSCR NPs synthesis

PEGylated liposome is conventional material for drug delivering. Hence, in this study we synthesized PEGylated liposome to embrace CSME and Ce6. CSME-Lipo composed of Soybean Lecithin/ cholesterol/ CSME/ = 2:1:0.2:0.15 (mass ratio) were prepared using a film hydration method. At first, At first, the phospholipids were dissolved in a solution of chloroform/ ethanol = 10:1 (v/v) and dried at 37 ℃ under vacuum to obtain a thin lipid film. The film was hydrated with PBS (pH 7.4) at 45 ℃ for 1 h. Then the mixture was probe sonicated for 5 min (80 w, 0.5 s on, 0.5 s off) under ice water bath to obtain PLS. By stirring PLS with Ce6 at 800 rpm for 1 h at 37 °C, the mixture was dialyzed in PBS through a dialysis zone (MCW of dialysis zone is 3 kD) for 12 h to obtain PLSC (CSME 0.4 mg/ml, Ce6 0.08 mg/ml). Then, the DSPE-PEG2000-RGD was added in the LSCM NPs solution and incubated at 37 °C for 1 h. Then, the PLSCR NPs was dialyzed in PBS for 12 h to remove free DSPE-PEG2000-RGD. Finally, the as-prepared PLSCR NPs were harvested by and stored at 4 °C.

1.2 Cell uptake of LSCMR NPs and PLSCR

4T1 cells were seeded in 12-well plates and incubated for 24 h at 37°C (5 to 10^4 cells/well). The new cultivated media containing LSCMR NPs and PLSCR were introduced and incubated for a different period after washing with PBS 3 times (Ce6 final concentration of 5 μg/mL) (2 h). Hoechst 33342 (Beyotime Biotech Co., Ltd., China) stained cell nuclei for 20 min and washed with PBS 3 times. Then the cells were stained. Tumor cell fluorescence pictures have been taken under a confocal laser microscope (CLSM) (FV1200, Olympus, Japan).

2. **Figure caption**

Figure S1. information of compound CSME. (a) Chemical structure of CSME; (b)High resolution mass spectrum; (c)^{1}H NMR spectrum and ^{13}C NMR spectrum; (d)HPLC of CSME method：Column: Agilent 5 TC-C18: 250*4.6 mm*5 μm, Con:10 mM, Inj Vol:10 μl, Wavelength: 210 nm, Temp:30 ℃, Flow Rate:1 ml/min; Mobile Phase: A: purified water, D: ACN, A/D=55:45, 10 min, t=5.573 min.

Figure S2. Chikusetsusapoin IVa methyl ester (CSME) can efficiently affect cancer cells viability and migration. (a) Chikusetsusapoin IVa methyl ester (CSME) showed good activity against breast cancer cells (IC_{50}=17.5 μM (MDA-MB-231); IC_{50}=20.1 μM (4T1) Wound-healing assay (b) and inhibition of migration (c) of 4T1 cells with different concentration of CSME. (d) Different cell shape MDA-MB-231 cell line and 4T1 after treated with Chikusetsusapoin IVa methyl ester (CSME).

Figure S3. CLSM image of the subcellular localization of LSCMR NPs in 4T1 cell line, DAPI (Blue), Ce6 (red), lysosome (green).

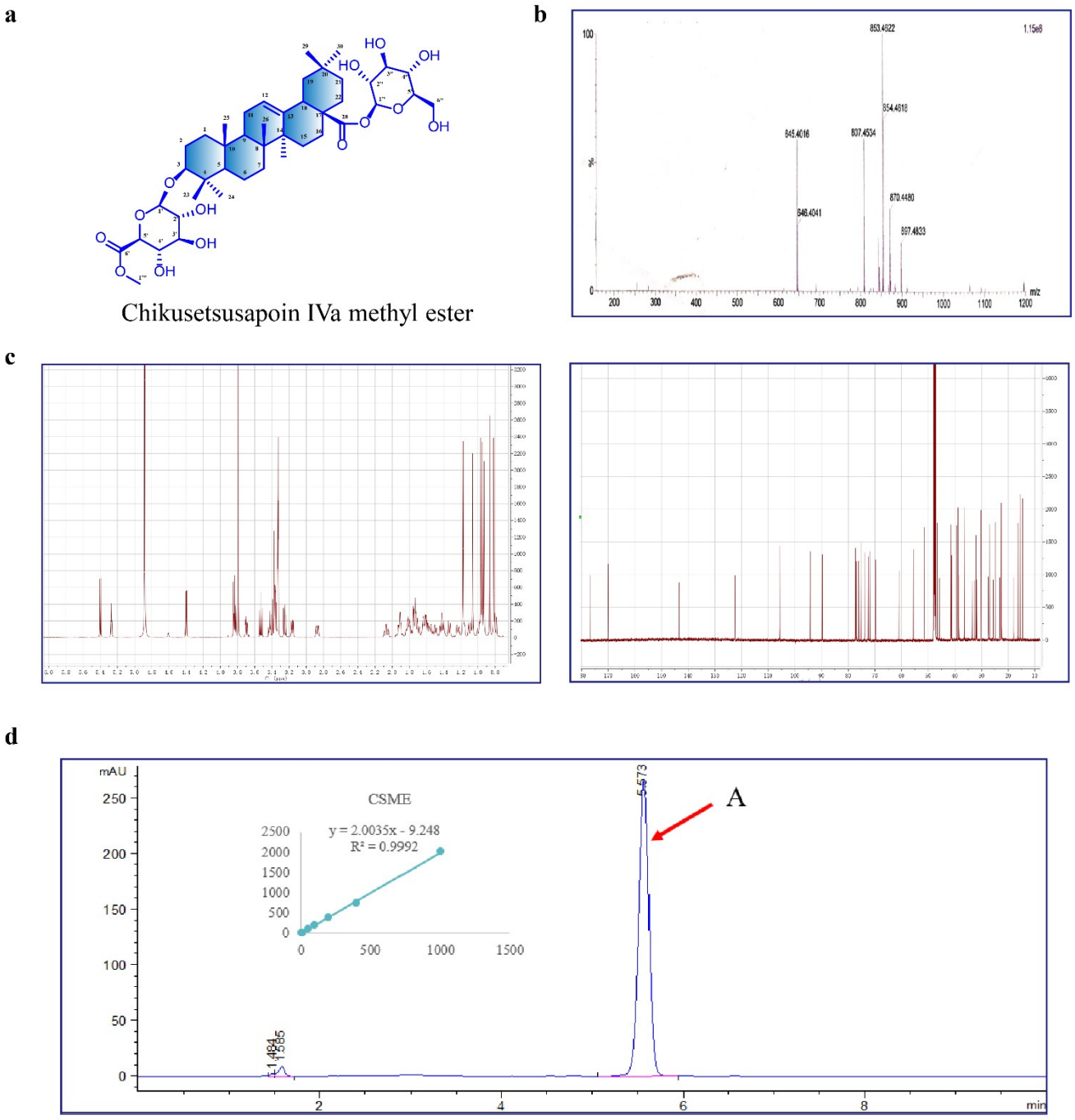
Figure S4. Cell uptake of LSCMR, PLSCR. CLSM images of 4T1 cell after incubation of LSCMR, PLSCR (CSME 15 uM, Ce6 5 ug/ml) for 2h.

Figure S5. Western blotting and quantitative analysis of cyclin D1, p21, MMP-9, Vimentin, and C-casp3 expression in 4T1 cells with various treatments.

Figure S6. *In vivo* toxicity evaluation. (a) Complete blood panel analysis. WBC: white blood cellS, RBC: red blood cells, HGB: hemoglobin, PLT: platelets；Blood biochemistry data including liver function markers: ALT and AST；kidney function markers: CRE and BUN; (b) H&E assay, stained slices of major organs.

Figure S7. Image of different materials.

Figure S1



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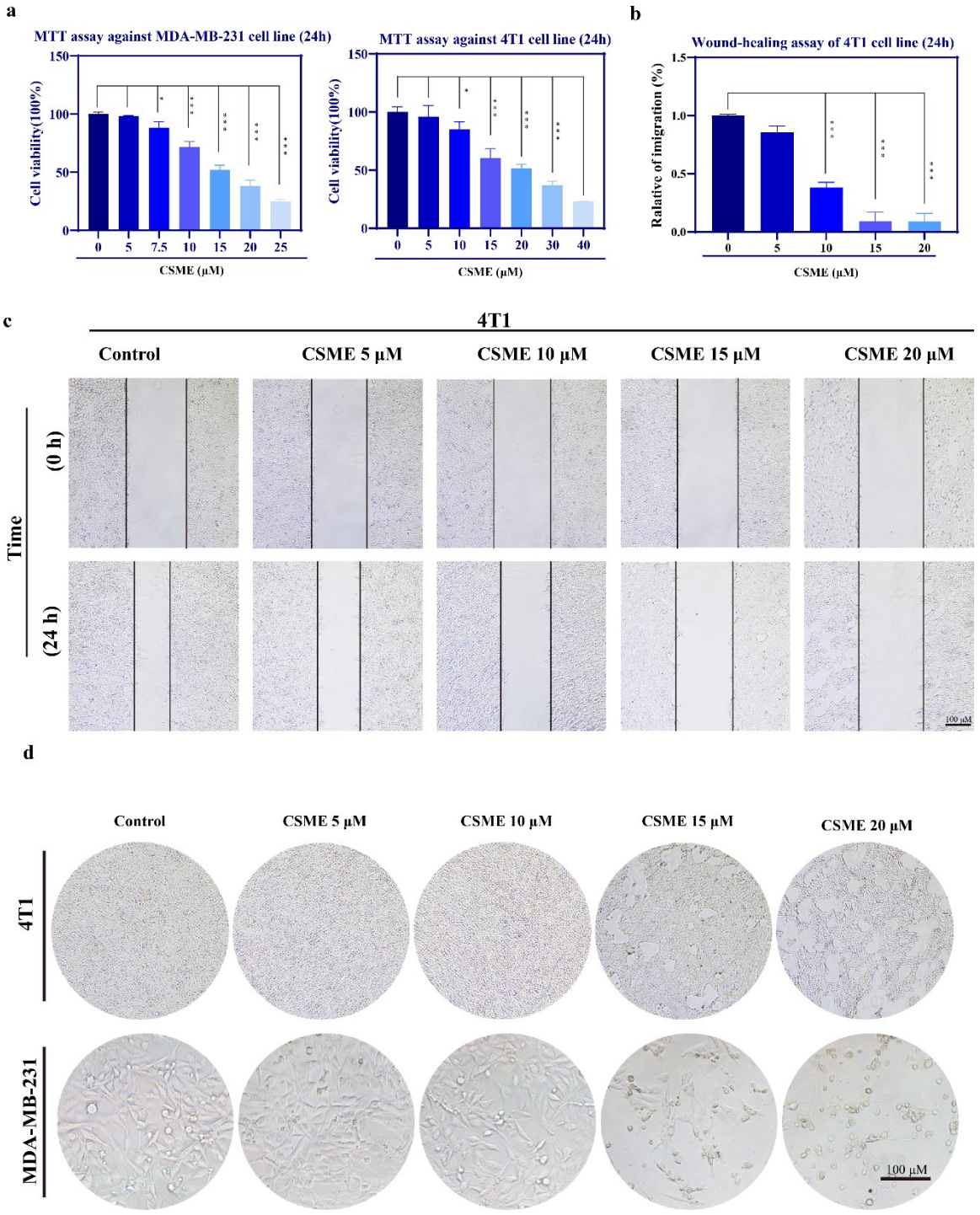

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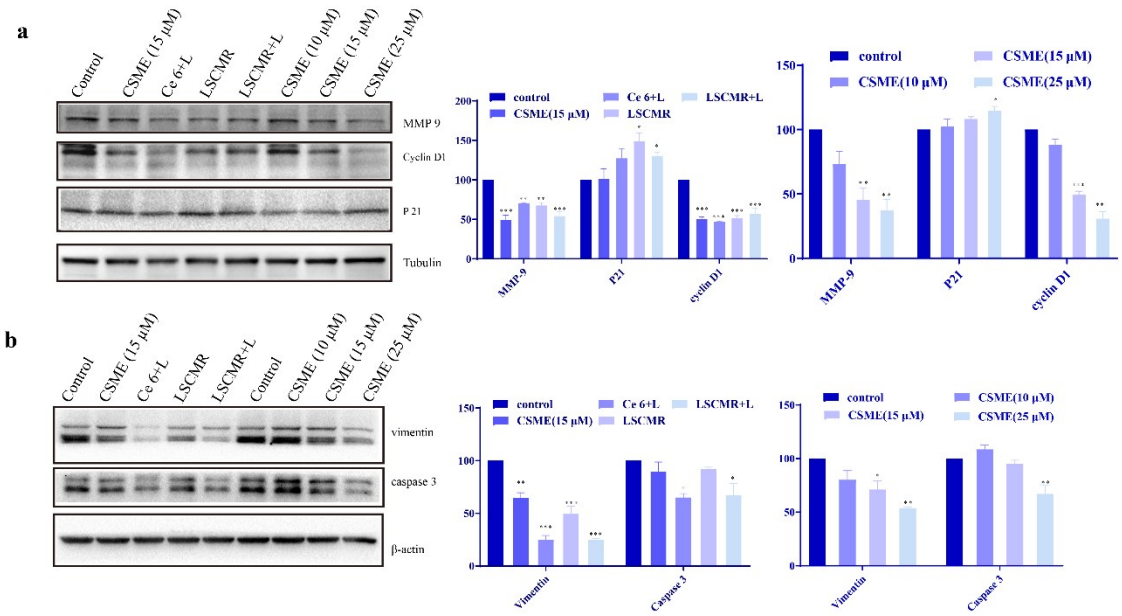

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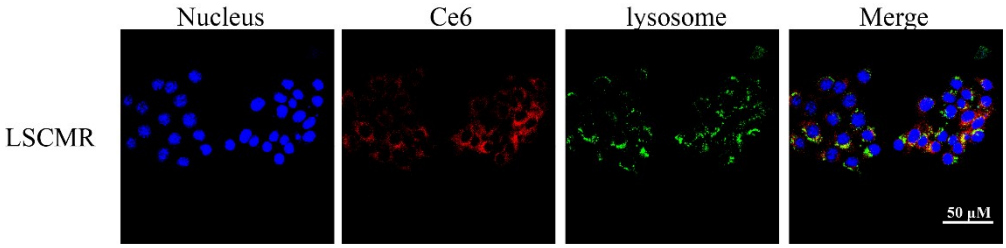

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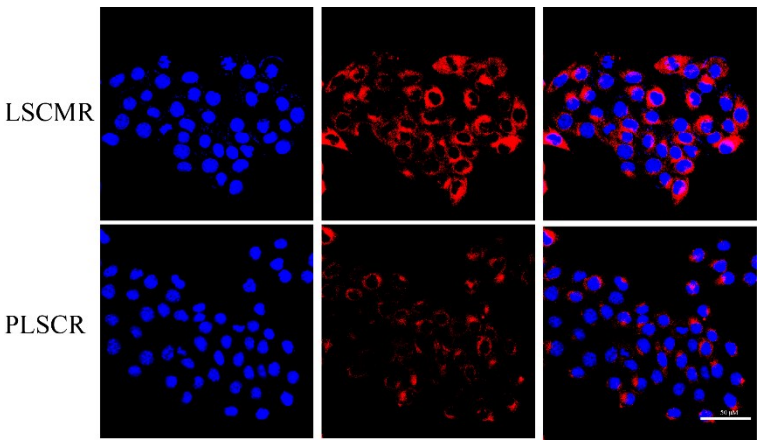

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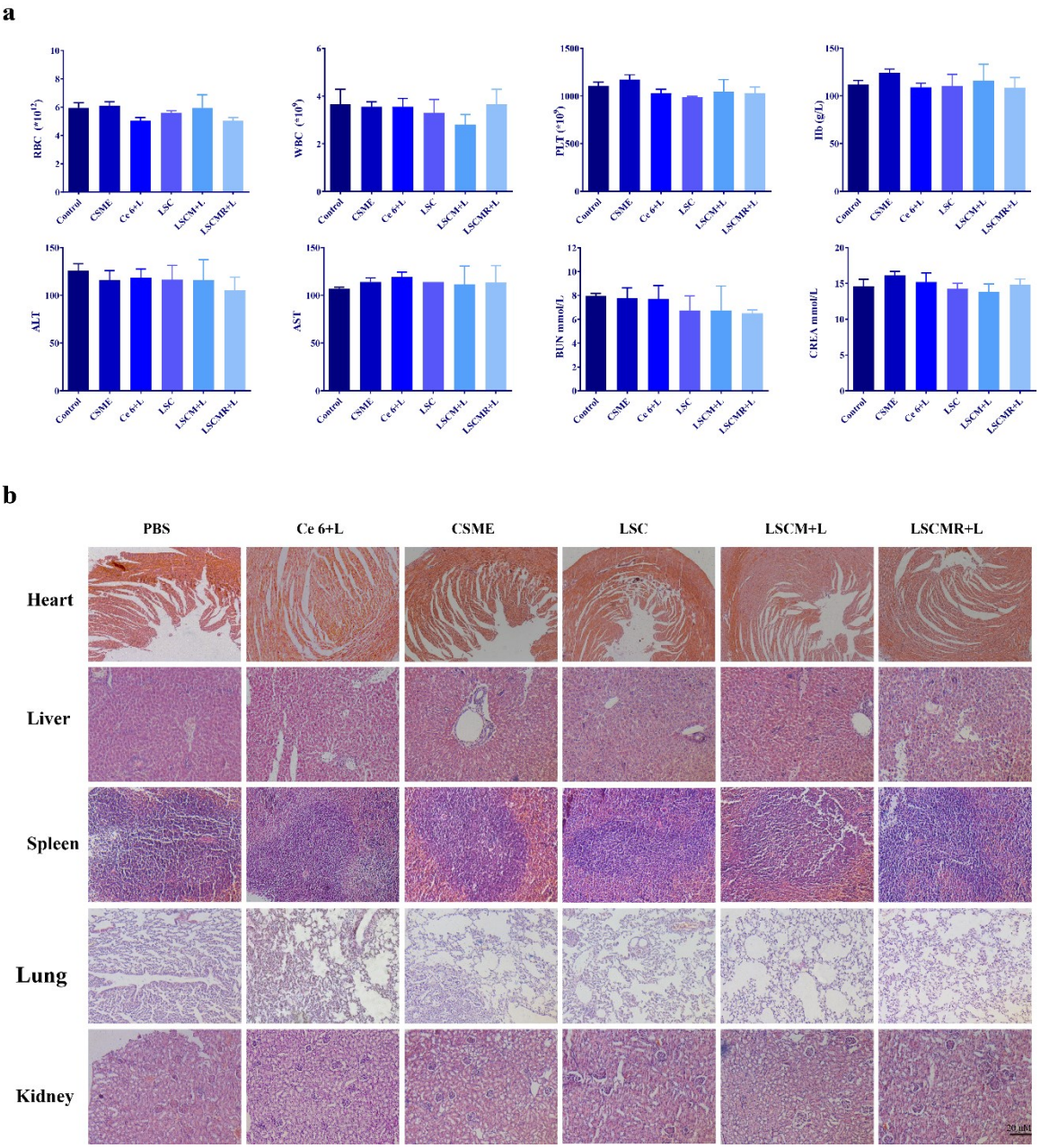

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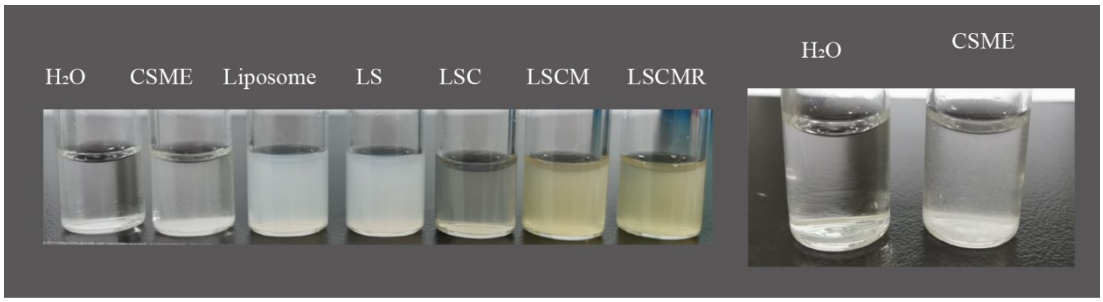

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