

**Bunty Sharma<sup>a,b</sup>, Gurpreet Kaur<sup>a</sup>, Ganga Ram Chaudhary<sup>a</sup>, Lluisa Perez-Garcia<sup>b</sup>**

a. Department of Chemistry and Center for Advance Study in Chemistry, Panjab University, Chandigarh, 160014, India  
b. Advanced Materials and Healthcare Technologies Group, School of Pharmacy, University of Nottingham, NG7 2RD, Nottingham, UK

## Introduction

### Metallosurfactant

The metallosurfactant (MS) are the new class of materials in which surfactant molecule contains metal ions as integral structural component<sup>1</sup>. These MSs shown surface active and delivering properties of both metal and surfactant, respectively

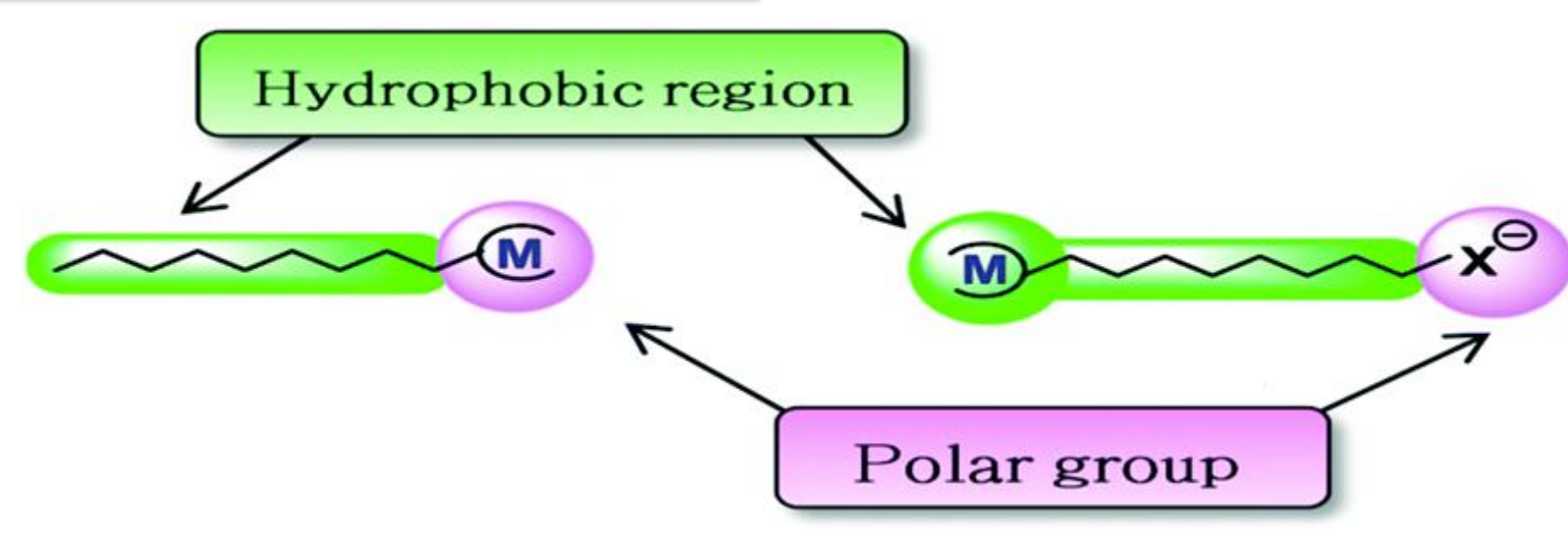


Figure 1. Pictorial representation of the metallosurfactant

### Catanionic Vesicles

Catanionic vesicles synthesized by mixing the non-stoichiometric ratios of cationic and anionic surfactants (single or double chain) in aqueous medium. These vesicles does not require any high energy method<sup>2</sup>

Unique properties of catanionic vesicle –

- Entrapment of both hydrophilic and hydrophobic drugs and control release of drug
- Temperature, light and pH responsive.
- Surface charge, size and permeability variations by changing cationic/anionic ratios

### Photodynamic Therapy

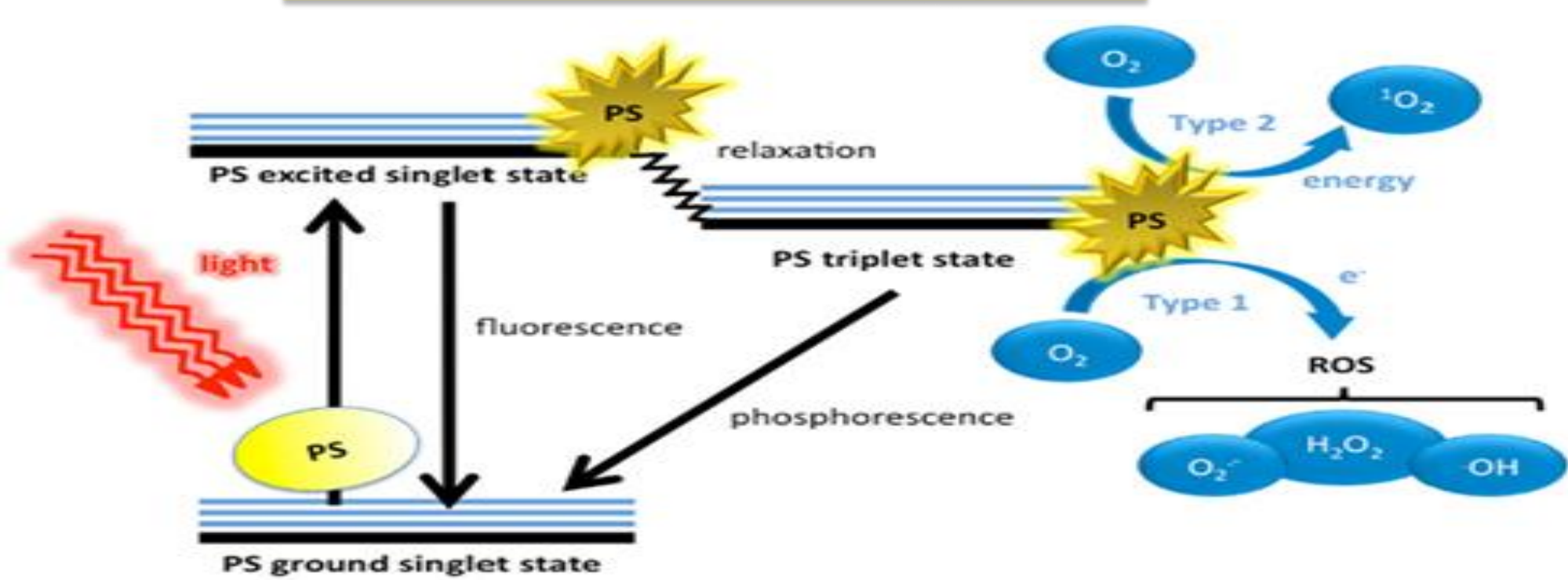


Figure 2. Schematic illustration of photodynamic action<sup>3</sup>

## Methods

### Metallosurfactant Synthesis

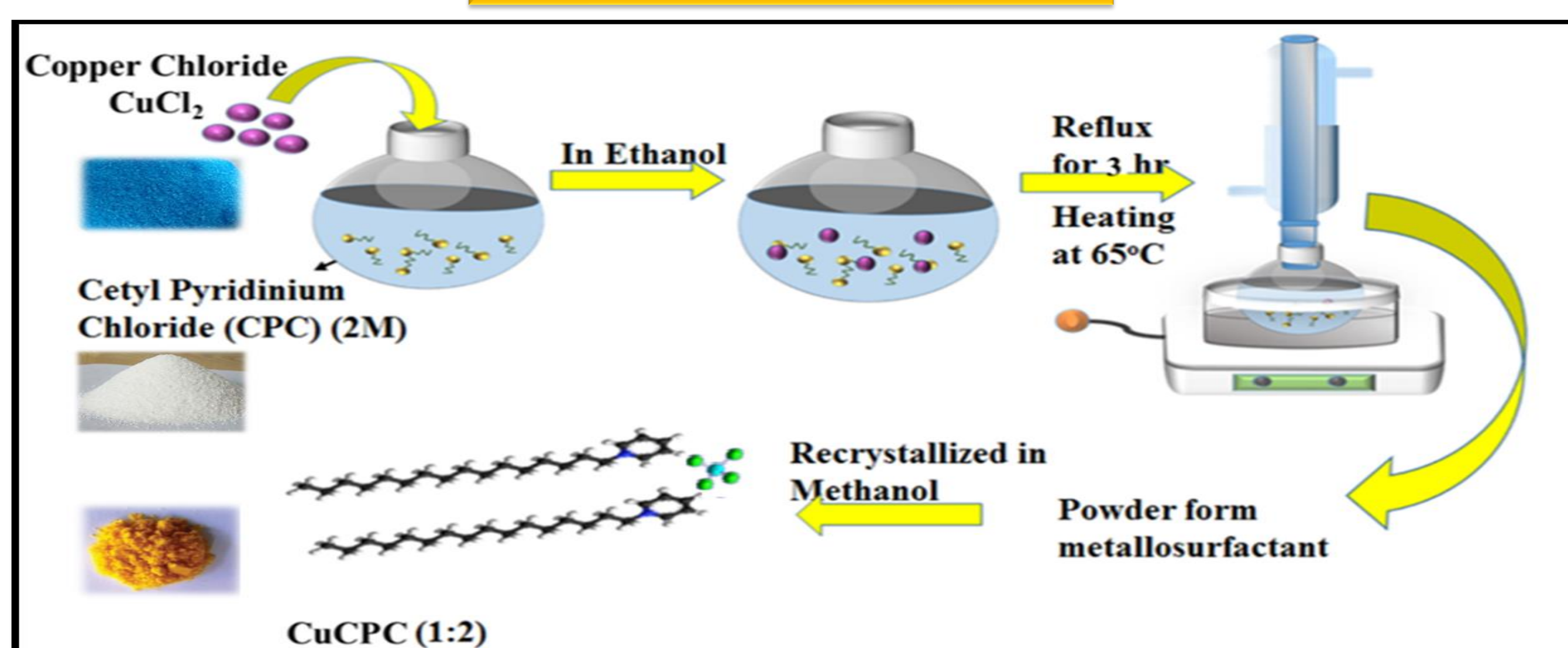


Figure 3. Pictorial representation of Synthesis of CuCPC (1:2) i.e. CuCPC II metallosurfactant<sup>4</sup>

### Catanionic Vesicles Synthesis

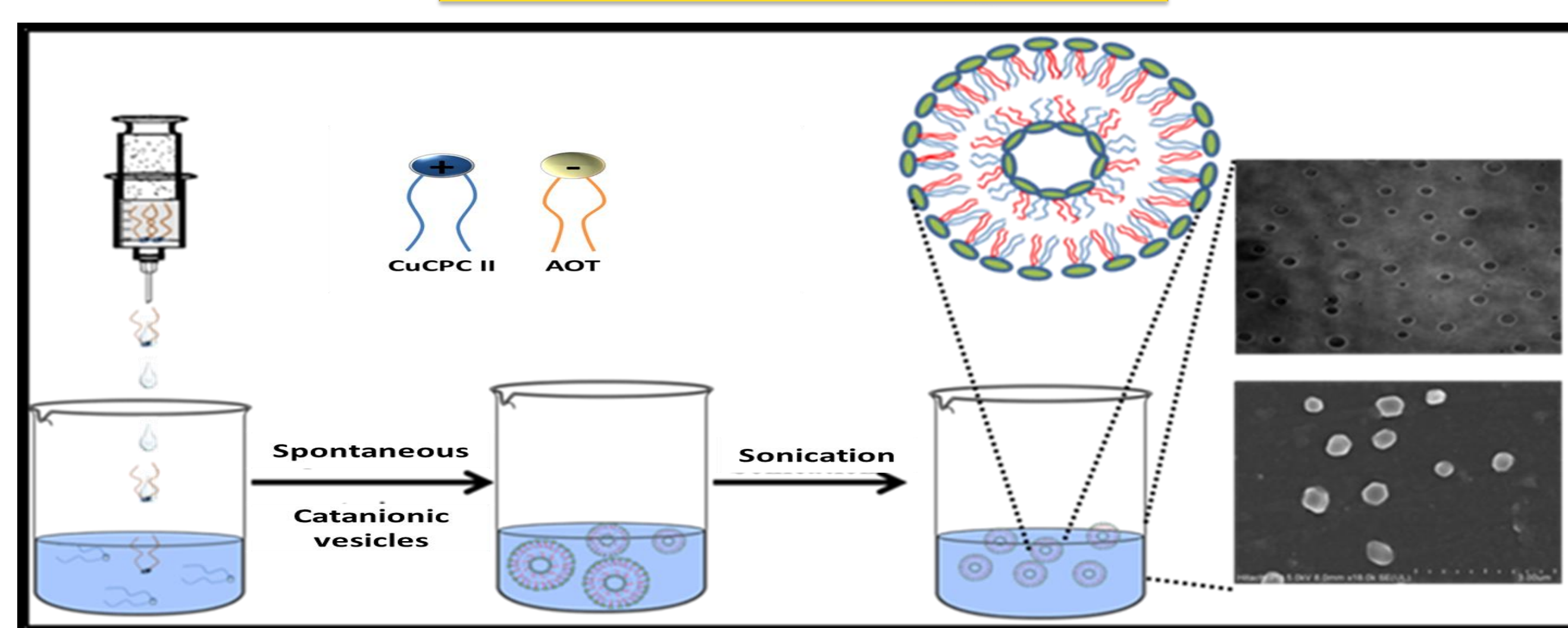


Figure 4. Pictorial representation of synthesis of CuCPC II:AOT (or C A) vesicular mixture<sup>5</sup>

## Results

### Catanionic Vesicles Characterization

Table 1. Summary of CuCPC II:AOT fractions CAC, hydrodynamic diameter ( $d_h$ , nm), PDI, Zeta-potential and RB singlet oxygen quantum yield ( $\Phi_{\Delta}^1O_2$ ) at 25°C

Catanionic vesicles (CuCPCII:AOT)	CAC ( $\mu$ M)	Z-avg. ( $d_h$ , nm)	PDI	Zeta Potential (mV)	RB $\Phi_{\Delta}^1O_2$ (Pure RB= 0.59)
10:90	1.62	174.32	0.124	- 47.70	0.49
20:80	2.47	259.42	0.173	- 25.81	0.35
30:70	2.00	272.53	0.448	6.22	0.58
40:60	2.30	156.11	0.073	74.90	-
60:40	3.75	114.26	0.388	73.75	0.66
70:30	5.08	214.87	0.544	52.70	0.81
80:20	7.50	226.94	0.425	64.20	0.69
90:10	21.87	101.59	0.314	66.44	0.77

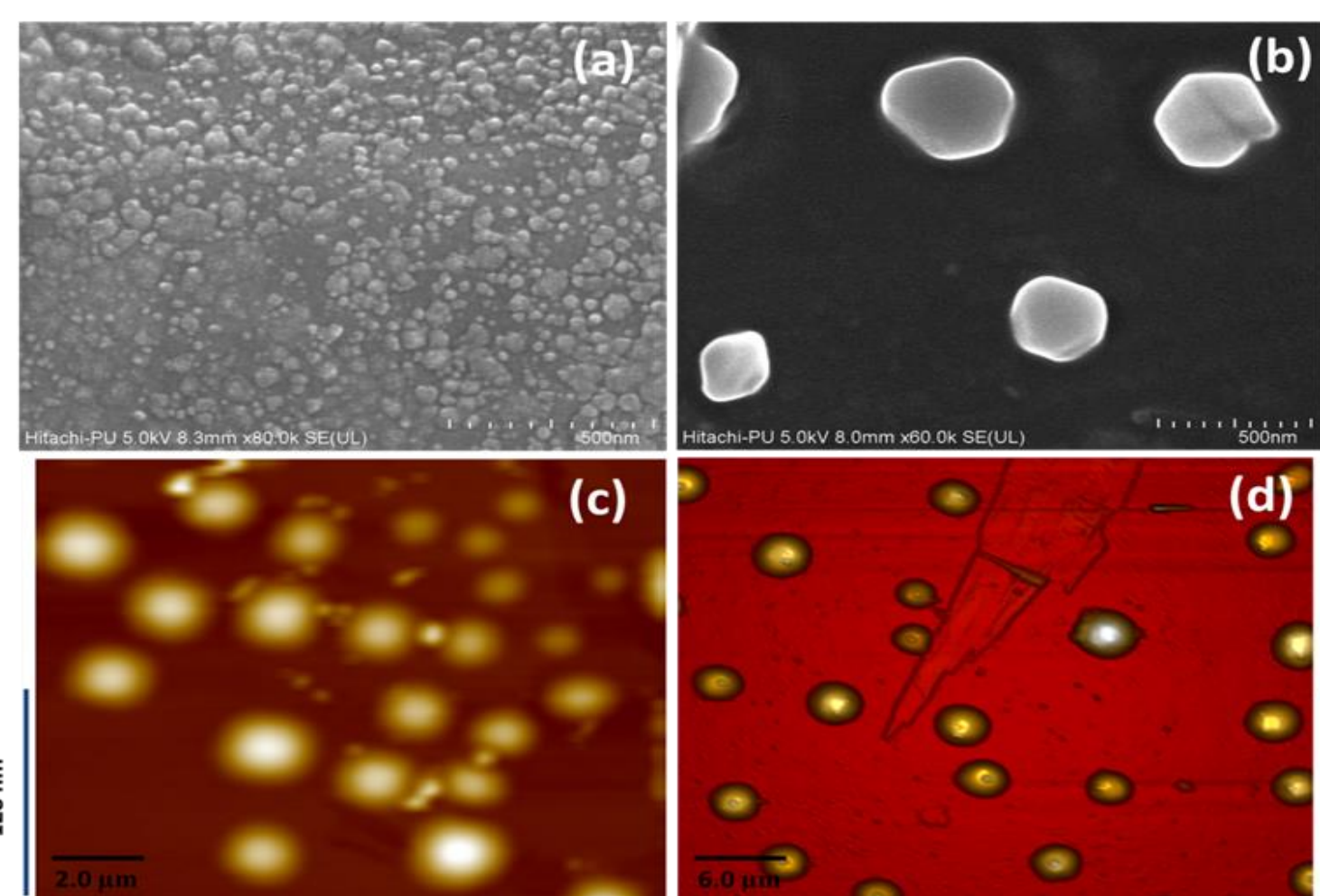


Figure 5. FESEM images of catanionic vesicular molar ratios of CuCPC II:AOT (a) 70:30 (b) 30:70 and morphological two-dimensional images of Atomic force microscope of (c) 70:30 (d) 30:70

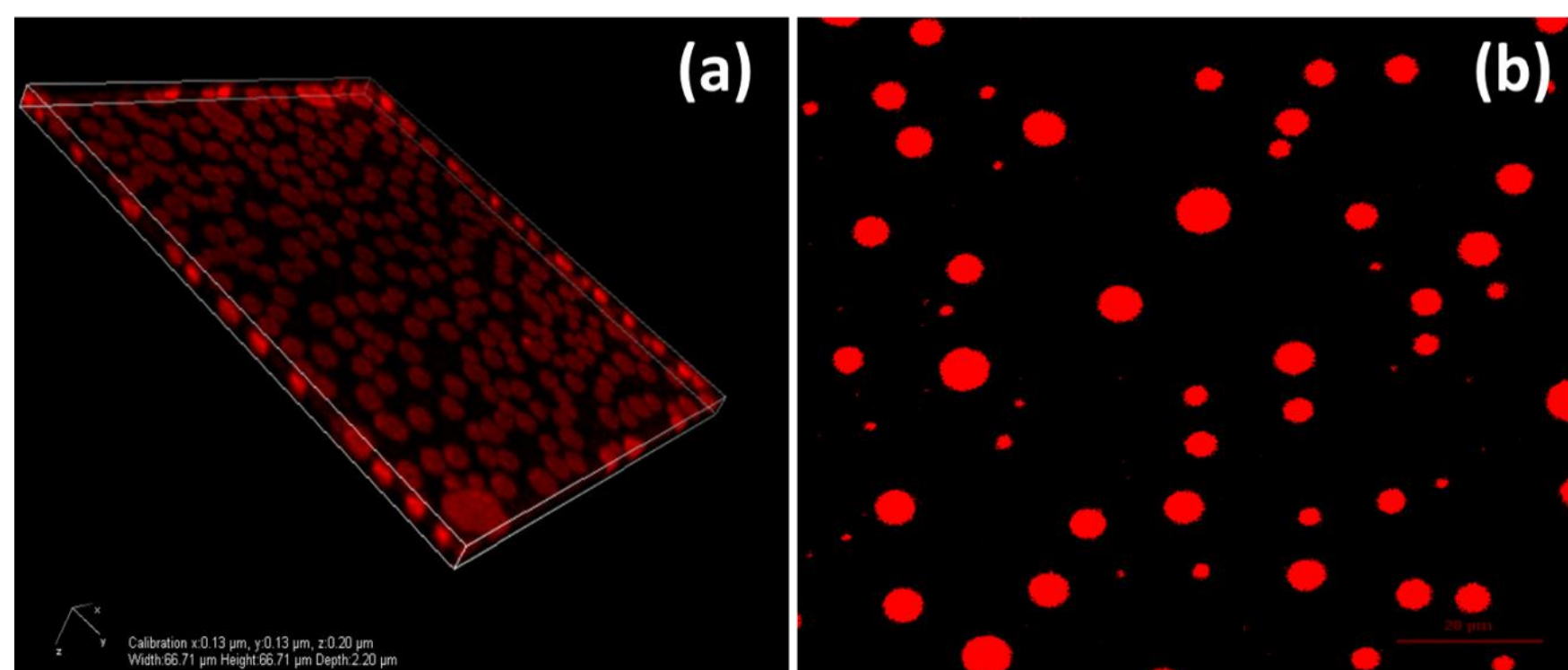


Figure 6. Confocal images of catanionic vesicles encapsulating RB PS (a) 70:30 (b) C A (30:70)

### Antibacterial Photodynamic Therapy

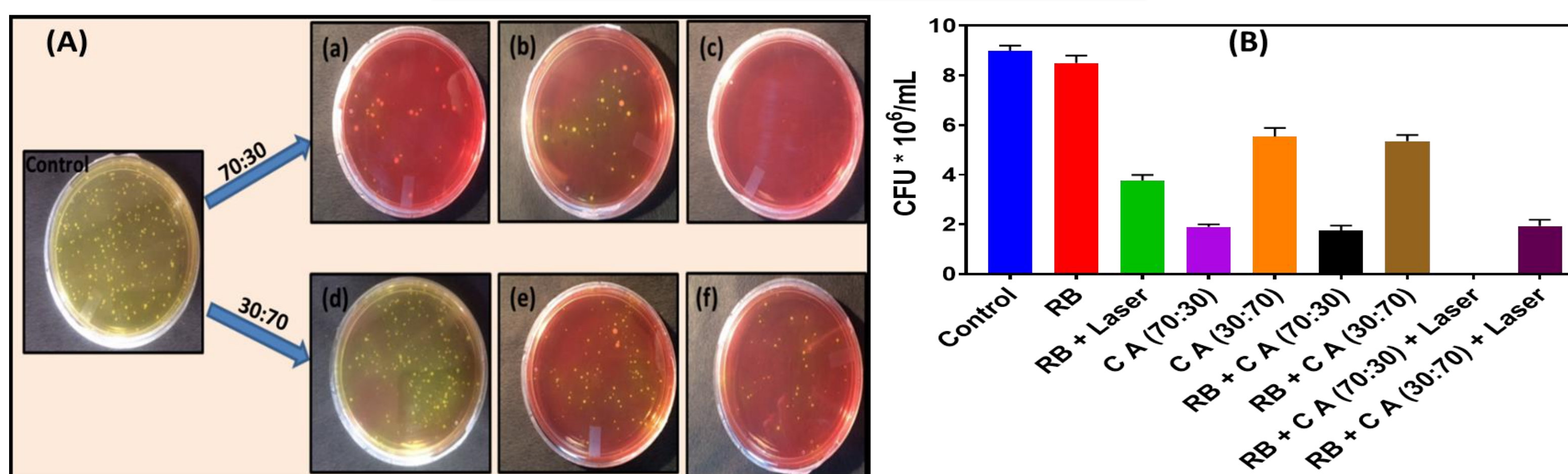


Figure 7. (A) Images of *S. aureus* colonies after different doses (a) C A (70:30) {dark conditions} (b) RB encapsulated C A (70:30) {dark conditions} (c) RB encapsulated C A (70:30) {irradiation 30 minutes} (d) C A (30:70) {dark conditions} (e) RB encapsulated C A (30:70) {dark conditions} (f) RB encapsulated C A (30:70) {irradiation 30 minutes} and (B) their respective CFU value graphs.

Figure 8. FESEM images *S. aureus* bacteria cells (a) Control {dark condition} and (b) with RB loaded C A (70:30) {irradiation 30 minutes} and *E. Coli* bacteria cells (c) Control {dark condition} (d) RB encapsulated C A (70:30) {irradiation 30 minutes}

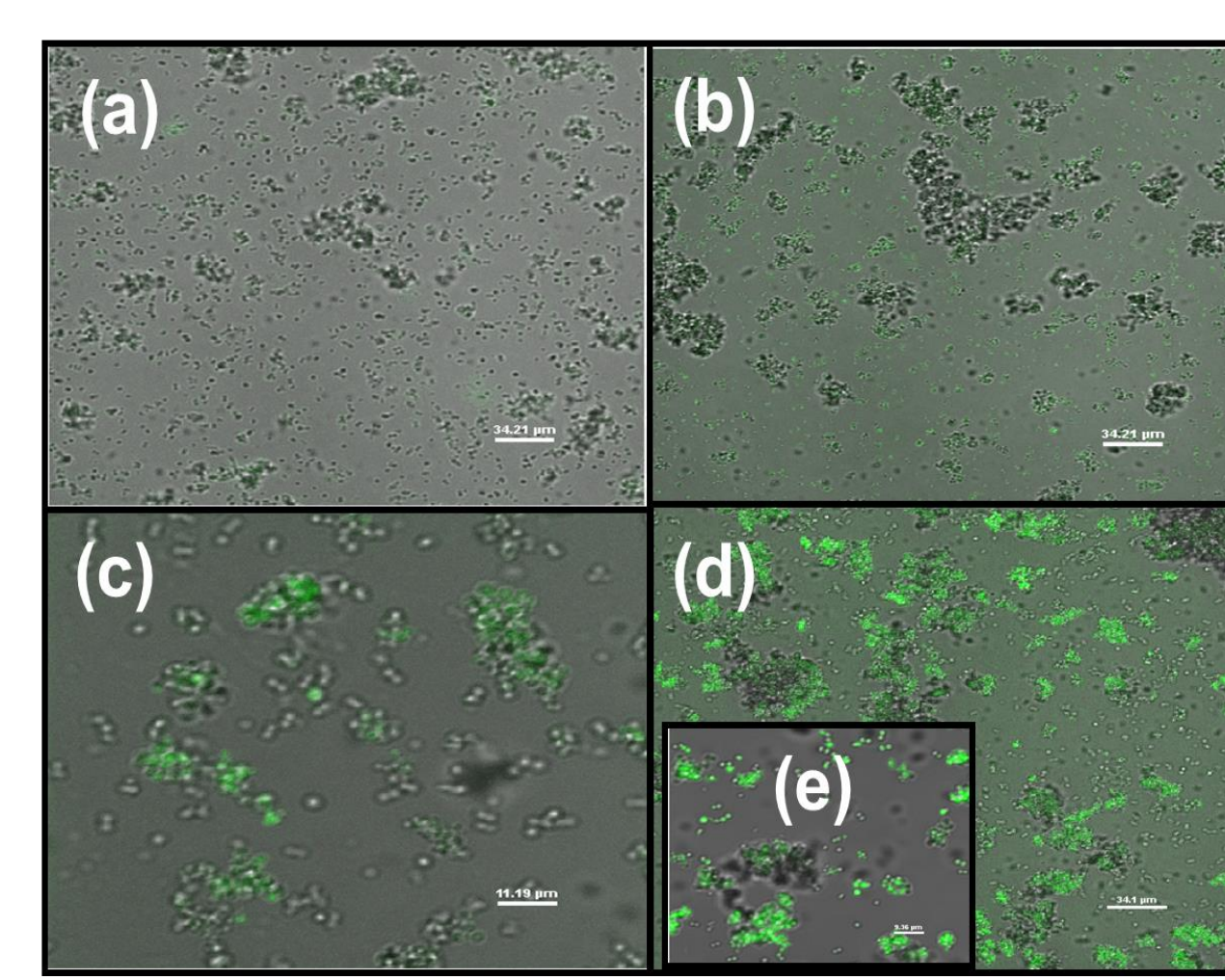
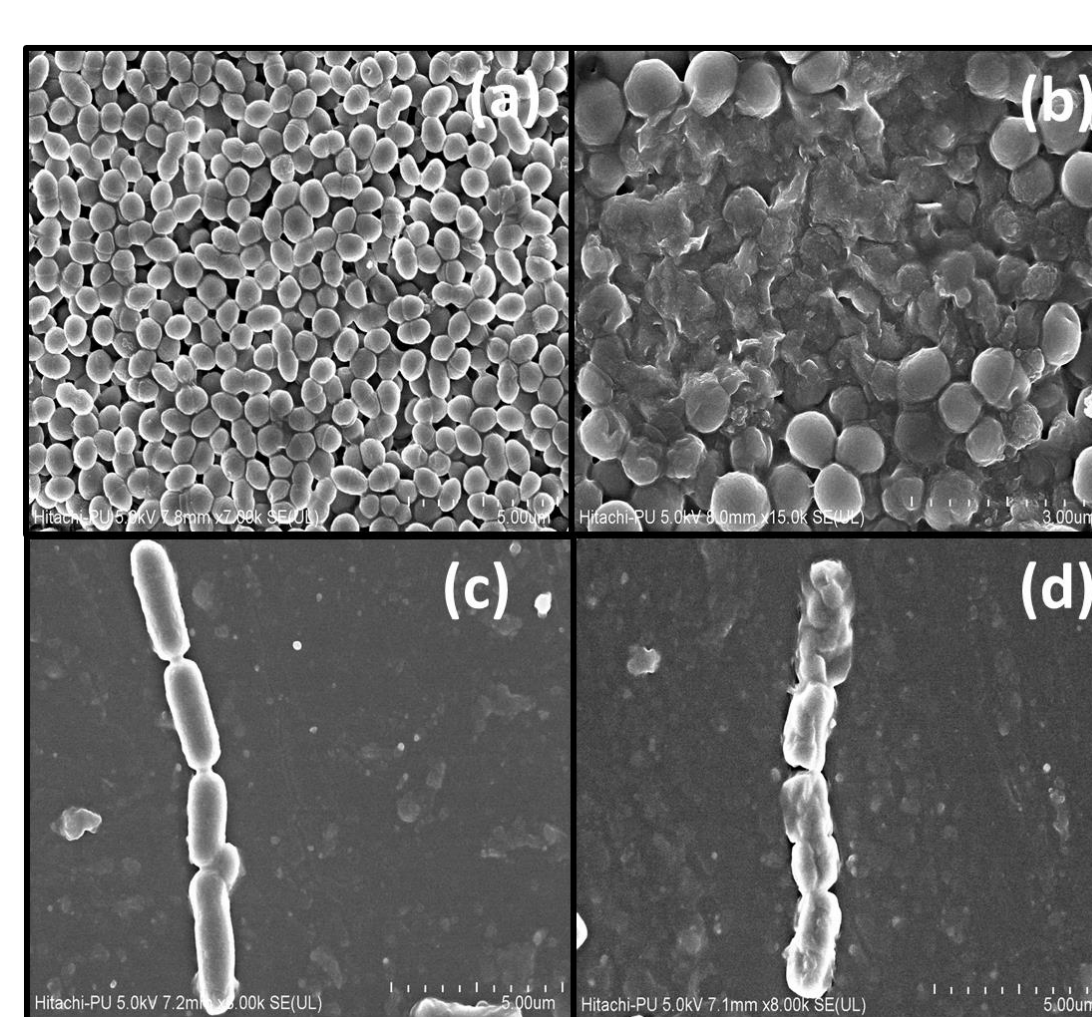


Figure 9. Merge (DIC + Confocal) Confocal fluorescence images of *S. aureus* bacterial cells with different mixture irradiation of 532 nm wavelength laser (a) SOSG only, (b) C A (70:30) with SOSG, (c) RB with SOSG, (d) RB loaded C A (70:30) with SOSG and (e) zoom in image of (d)

### Comet Assay

The "Comet Assay" also known as single cell gel electrophoresis is an easy and sensitive technique for the detection of DNA damage at the level of individual cells<sup>6</sup>. We have found that PDT with RB loaded C A 70:30 shown maximum tail length which shown maximum DNA damage.

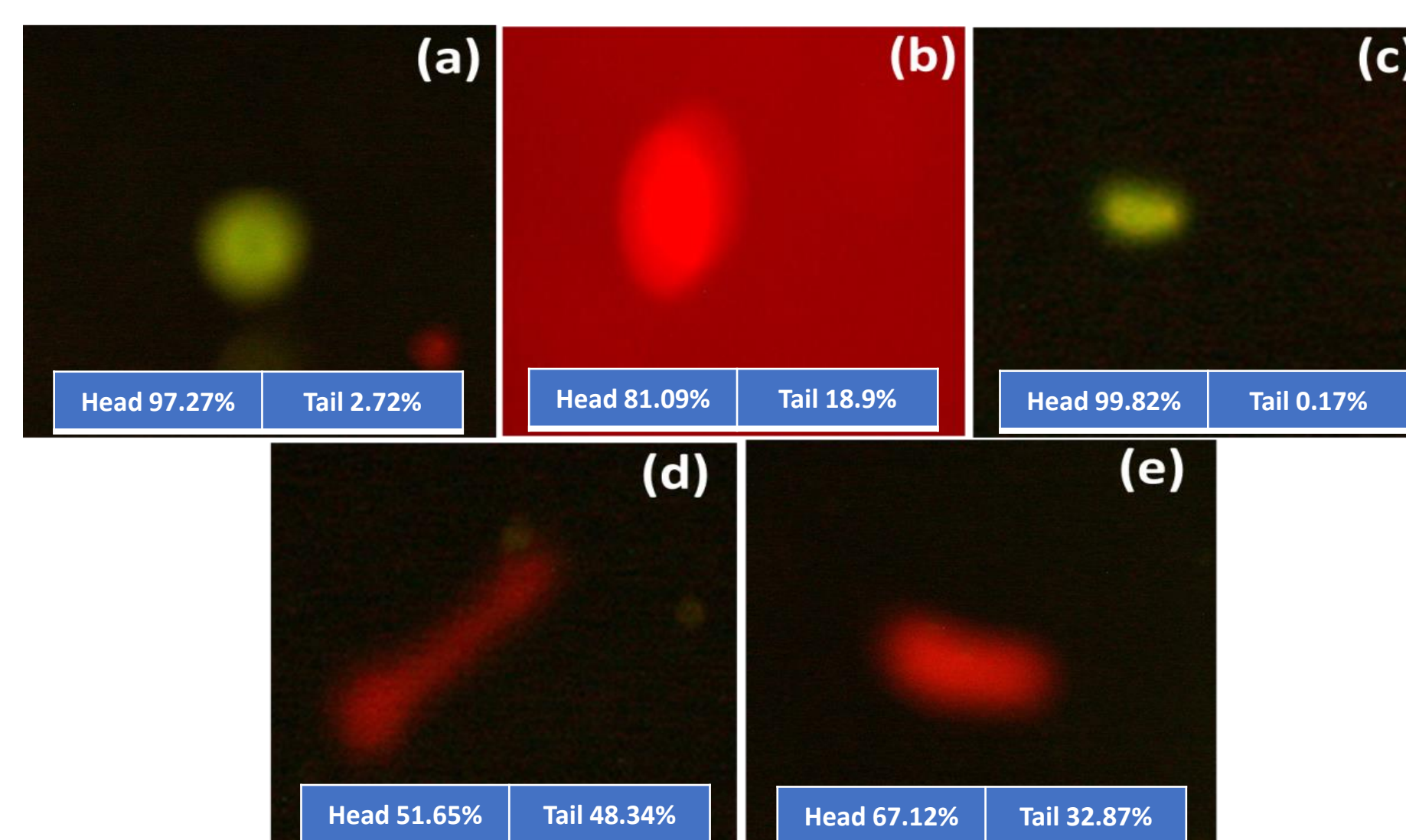


Figure 10. Comet assay images of *S. aureus* cells (a) control undamaged, (b) with C A 70:30 (c) with C A 30:70 (d) RB loaded C A (70:30) after irradiation (30 min) and (e) RB loaded C A (30:70) after irradiations (30 min) and their respective head and tail percentage calculated from CASP (Comet Assay Software Project) lab software

## Conclusion

- First time we have successfully synthesised metallocatanionic vesicles based on CuCPC II metallosurfactant.
- Optimization of the self-assembled structures was carried out by using various analytical techniques like conductivity, Zeta-potential etc.

- Interaction and effect on RB singlet oxygen quantum yield generation were further checked with all fractions using chemical trapping methods using Uric Acid as quencher.
- Antibacterial Photodynamic therapy was performed against bacteria and Comet assay confirmed the DNA damage of the bacterial cells.

### Acronyms

CA = CuCPC II:AOT  
RB = Rose Bengal  
SOSG = Singlet Oxygen Sensor Green  
CAC = Critical aggregation concentrations  
PS = Photosensitizer

### References

- [1] P. C. Griffiths et al., *J. Colloid Interface Sci.*, **2006**, 122, 107-117.
- [2] Kaler et al *Science*, **1989**, 245, 1371-1374.
- [3] Dai et al. *Front. Microbiol.*, **2012**, 3, 1-16.
- [4] G. Kaur et al., *Phys. Chem. Chem. Phys.*, **2016**, 18, 23961-23971.
- [5] Sharma et al., *Biomater. Sci.*, **2020**, 8, 2905-2920.
- [6] F. M. Nair et al., *Br. J. Cancer*, **1997**, 75, 1721-1729

### Acknowledgements

