Quatsomes: stable and versatile vesicular scaffolds for nanostructuring organic dyes in water

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Fluorescence imaging is a powerful tool for research and diagnostics. The potential of this technique is strongly affected by the optical and physicochemical properties of the fluorescent probe¹. Organic dyes, whose properties can be tuned via structural modification, are actively investigated in this direction, although their low solubility in aqueous and biological media strongly limits their use for *in vivo* and *in vitro* applications. Fluorescent organic nanoparticles² represent a promising route to bring organic dyes in aqueous media, with the possibility to add other functionalities, such as drug-delivery or site specific targeting. Indeed multifunctional fluorescent nanoparticles are attracting a large interest for theranostic applications. Here we present Quatsomes (QSs), a class of small unilamellar vesicles (SUVs), as a versatile and stable scaffold for nanostructuring organic dyes in water, using different dyes with different optical and physicochemical properties.

QSs are formed by the self-assembly of quaternary ammonium surfactants (e.g. CTAB) and sterols (e.g. cholesterol) in water^{3, 4}. QS are good protein delivery nanocarriers and their surface can be easily functionalized with targeting peptides.⁴ Several strategies for the preparation of QSs-dyes are presented, including: i) the decoration of the positively charged surface of QSs with anionic water-soluble dyes, ii) the partial substitution of cholesterol with a dye-functionalized cholesterol and iii) the encapsulation of hydrophobic dyes inside the bilayer via a *membrane anchoring* mechanism. By this last strategy, it was possible to disperse in water commercially available as well as newly synthesized hydrophobic dyes with long alkyl chains. When prepared by using compressed fluids (DELOS-SUSP method⁵), the decorated QSs are strongly fluorescent and are stable over months. Moreover, a study on photostability of QSs-dyes showed that in some cases dyes can be more photostable when loaded on QSs membrane than in solution.

QSs constitute therefore a versatile platform for nanostructuring organic dyes in water, preserving their optical properties and providing the possibility of specific targeting and/or drug-delivery, then opening new possibilities in theranostic applications.

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- [1] Resch-Genger, U., Grabolle, M., Cavaliere-Jaricot, S., Nitschke, R. & Nann, T. Quantum dots versus organic dyes as fluorescent labels. *Nat. Methods* **5**, 763–775 (2008).
- [2] Zhang, J. *et al.* Highly Stable Near-Infrared Fluorescent Organic Nanoparticles with a Large Stokes Shift for Noninvasive Long-Term Cellular Imaging. *ACS Appl. Mater. Interfaces* acsami.5b08539 (2015). doi:10.1021/acsami.5b08539
- [3] Ferrer-Tasies, L. *et al.* Quatsomes: Vesicles formed by self-assembly of sterols and quaternary ammonium surfactants. *Langmuir* **29**, 6519–6528 (2013).
- [4] Cabrera, I. *et al.* Multifunctional nanovesicle-bioactive conjugates prepared by a one-step scalable method using CO2-expanded solvents. *Nano Lett.* **13**, 3766–3774 (2013).
- [5] Ventosa et al. Patent EP1843836B1.