

SM1: Evaluation of the encapsulation efficiencies and permeabilities for various solutes.

The solute encapsulations into the vesicles are performed as described in Methods. Aliquots of the vesicles under dialysis are extracted from the cassettes. The residual solute concentration is measured by fluorescence and/or UV-VIS absorption as follows: a known amount of sample is added to a known amount of Triton-X solution to ensure vesicle disruption, and optionally to salt, buffer and CTACl solution to ensure stabilization of the emission yield of the solute against pH and CTA⁺ concentration. The composition of the added solution is given in the table below. The concentration of solute is evaluated after comparison to a calibration curve measured in the same conditions, i.e. known amounts of solute mixed with empty vesicles and additives. Glucose quantitation was performed after enzymatic reaction for production of H₂O₂ (using glucose oxidase), subsequent oxidation of o-Dianisidine (using peroxidase) and finally absorbance measurement at $\lambda = 450$ nm.

Solute to be tested	Method	Additives
Glucose	Absorption ($\lambda = 450$ nm)	1% Triton-X Glucose oxidase/peroxidase enzymatic assay kit (Sigma)
Lucigenin	Absorption ($\lambda = 368$ nm)	1% Triton-X
Rhodamine 6G	Absorption ($\lambda = 526$ nm)	1% Triton-X
Rhodamine 6G	Fluorescence ($\lambda_{\text{exc}} = 526$ nm, $\lambda_{\text{em}} = 552$ nm)	1% Triton-X 1.5 mM CTAC
Rhodamine B	Absorption	1% Triton-X

	($\lambda = 554$ nm)	6.7 mM borate buffer (pH = 9)
Oregon Green 488	Fluorescence ($\lambda_{\text{exc}} = 502$ nm, $\lambda_{\text{em}} = 522$ nm)	1% Triton-X 6.7 mM borate buffer (pH = 9) 1.5 mM CTAC

The resulting concentration-time curves were fitted with an exponential decay:

$$c(t) = c_0 e^{-\frac{P}{3R}t}$$

where c_0 is the concentration of solute extrapolated at $t = 0$, P the permeability of the vesicles to the solute and R the mean radius of the vesicles (2.5 μm).

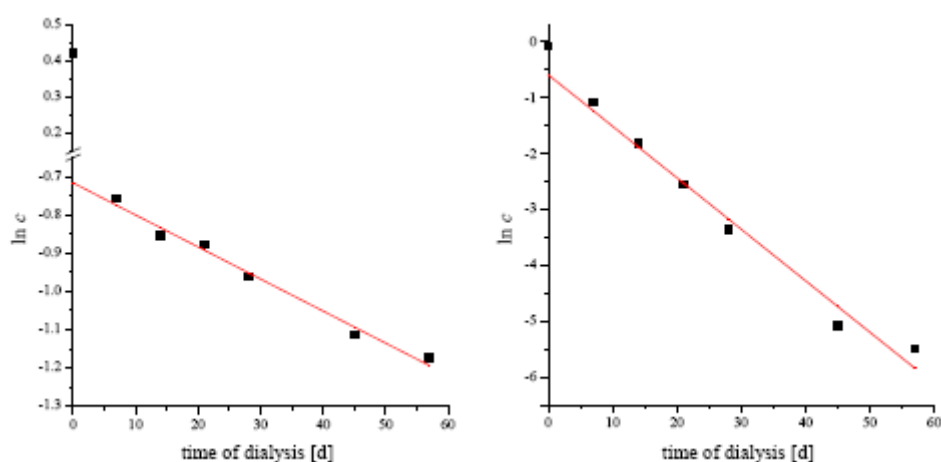


Figure SM1: release profiles of the cationic Rhodamine 6G (left) and anionic Oregon Green (right)

SM2: pH measurement

The pH inside the vesicles has been evaluated from the ratio of the fluorescence of Rhodamine B at 576 nm and 600 nm. This probe is suited as it shows the best sensitivity in the pH = 2 – 5 range. The calibration curve has been recorded with a 10 μ M Rhodamine B solution with variable amounts of added HBr, and shows the typical sigmoidal response:

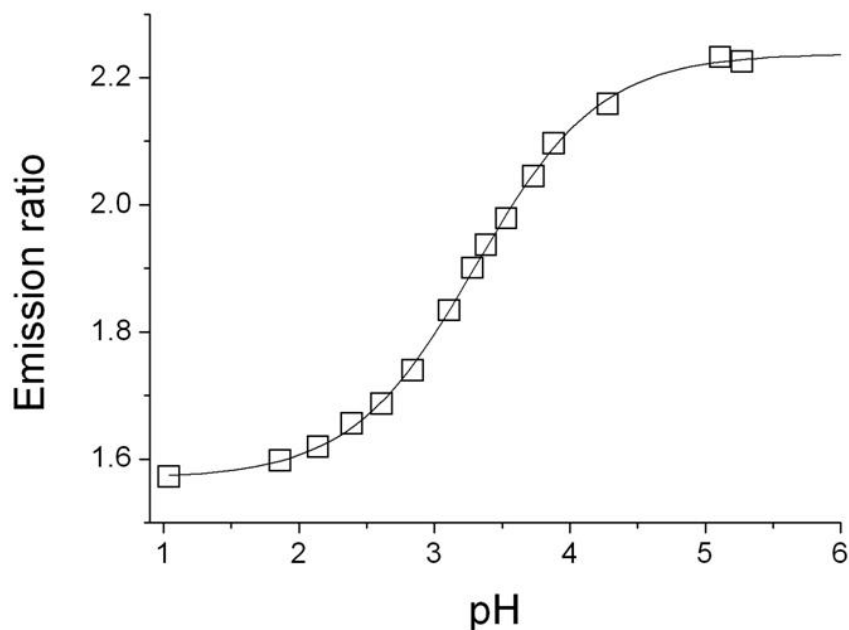


Figure SM2: Emission ratio $F(576\text{nm})/F(600\text{ nm})$ of a 10 μ M Rhodamine B solution excited at 555 nm as a function of pH (adjusted with HBr).

SM3: vesicles with higher internal pH

The vesicles with higher internal pH have been prepared using the same procedure as for the fatty acid/CTAB mixtures, with sodium myristate and N,N dimethyltetradecylamine as amphiphilic molecules in the ratio myristate/(myristate+amine) = 0.35 and a total concentration of 1 wt% in water. After dialysis, the solution consists in a dispersion of spherical vesicles, as demonstrated by confocal microscopy after staining by Rhodamine 6G. The pH of the external solution is in the 5-6 range. After disruption of the vesicles by addition of Triton-X (1 wt%), the pH of the solution is in the 9-10 range.

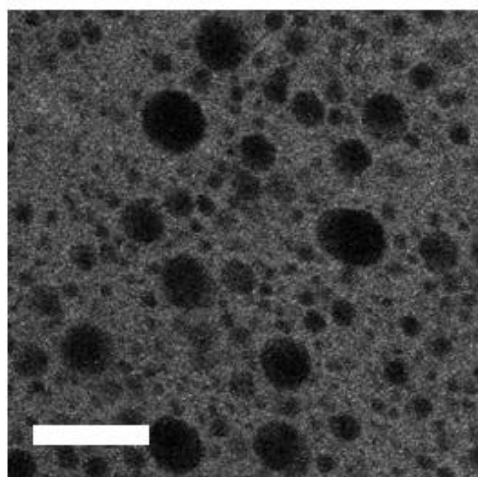


Figure SM3: sodium myristate/N,N tetradecylamine vesicles as observed by confocal microscopy after staining with Rhodamine 6G. The bar is 20 μm .