Design of a functional test of MexB, a pump from *Pseudomonas aeruginosa*

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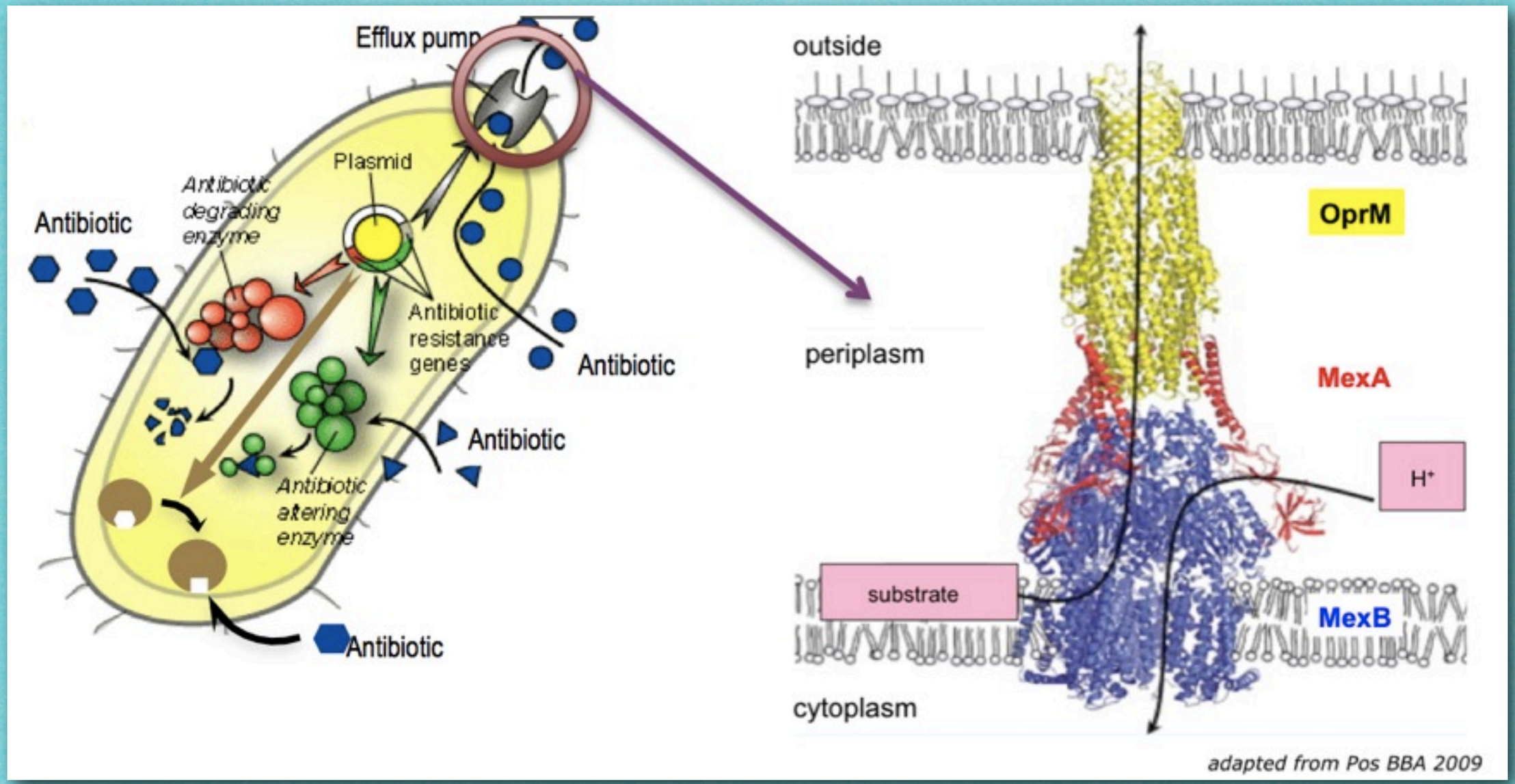
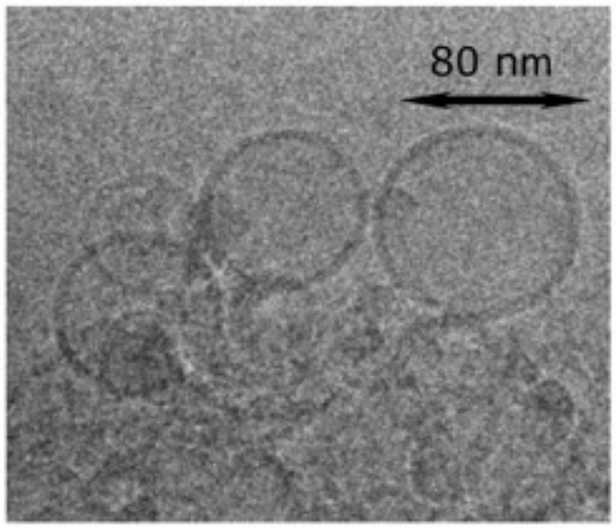
10th ABC meeting - October 20th 2012

Introduction



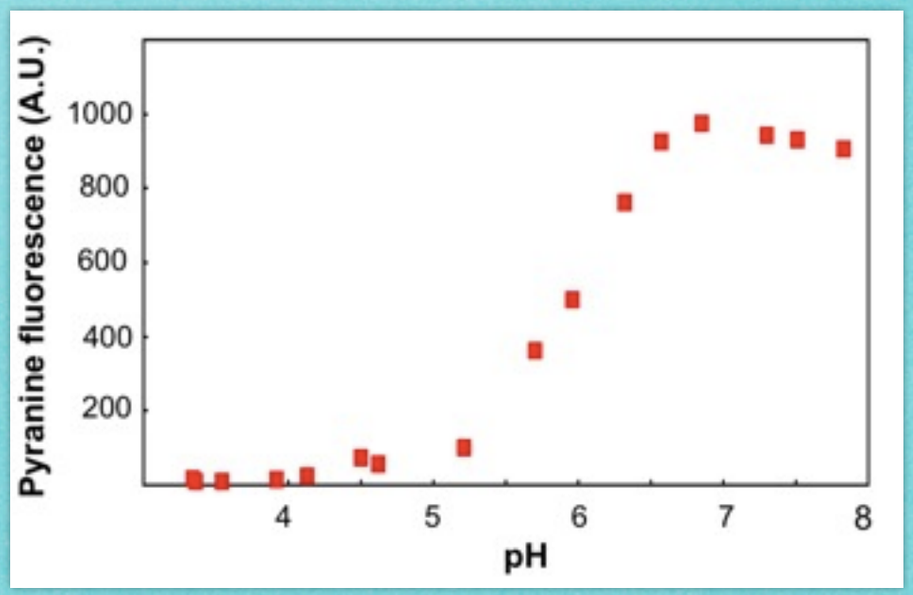
Prerequisites for a functional test of MexB

- Membrane and compartimentation-> Liposomes



Cryo TEM (coll. O.Lambert, UMR 5248)

- pH monitoring -> Pyranine



Prerequisites for a functional test of MexB

- Proton gradient -> Bacteriorhodopsin

Communication

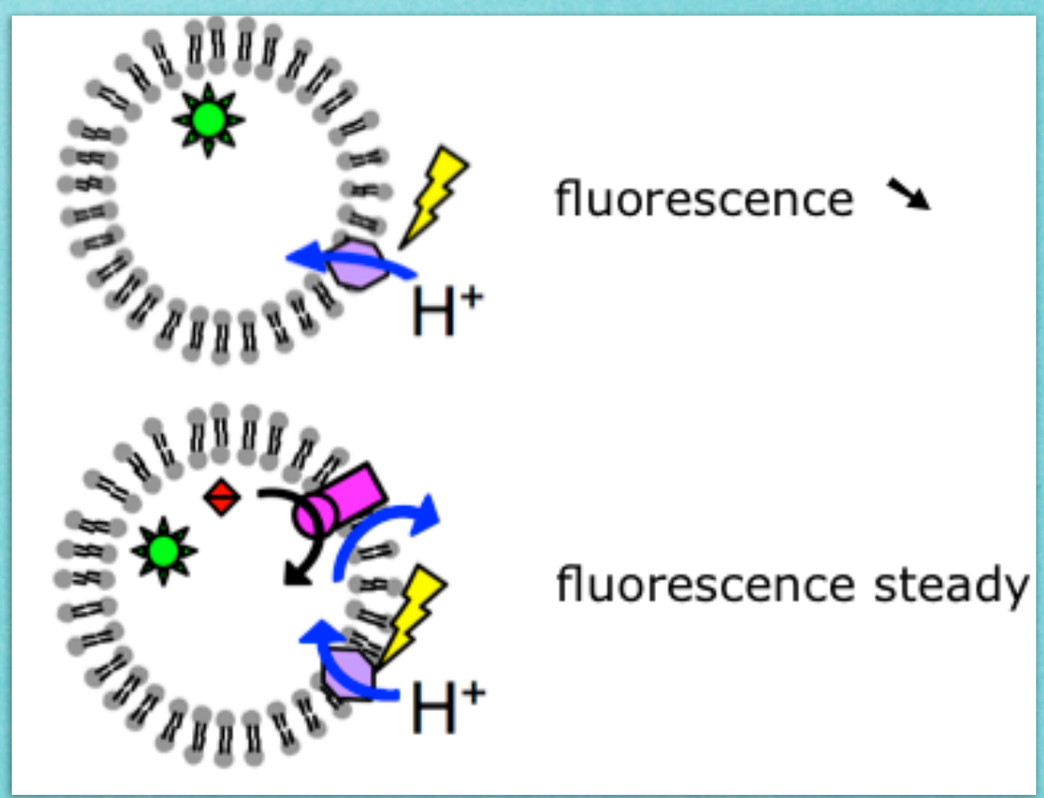
THE JOURNAL OF BIOLOGICAL CHEMISTRY
Vol. 249, No. 2, Issue of January 25, pp. 662–663, 1974
Printed in U.S.A.

Reconstitution of Purple Membrane Vesicles Catalyzing Light-driven Proton Uptake and Adenosine Triphosphate Formation*

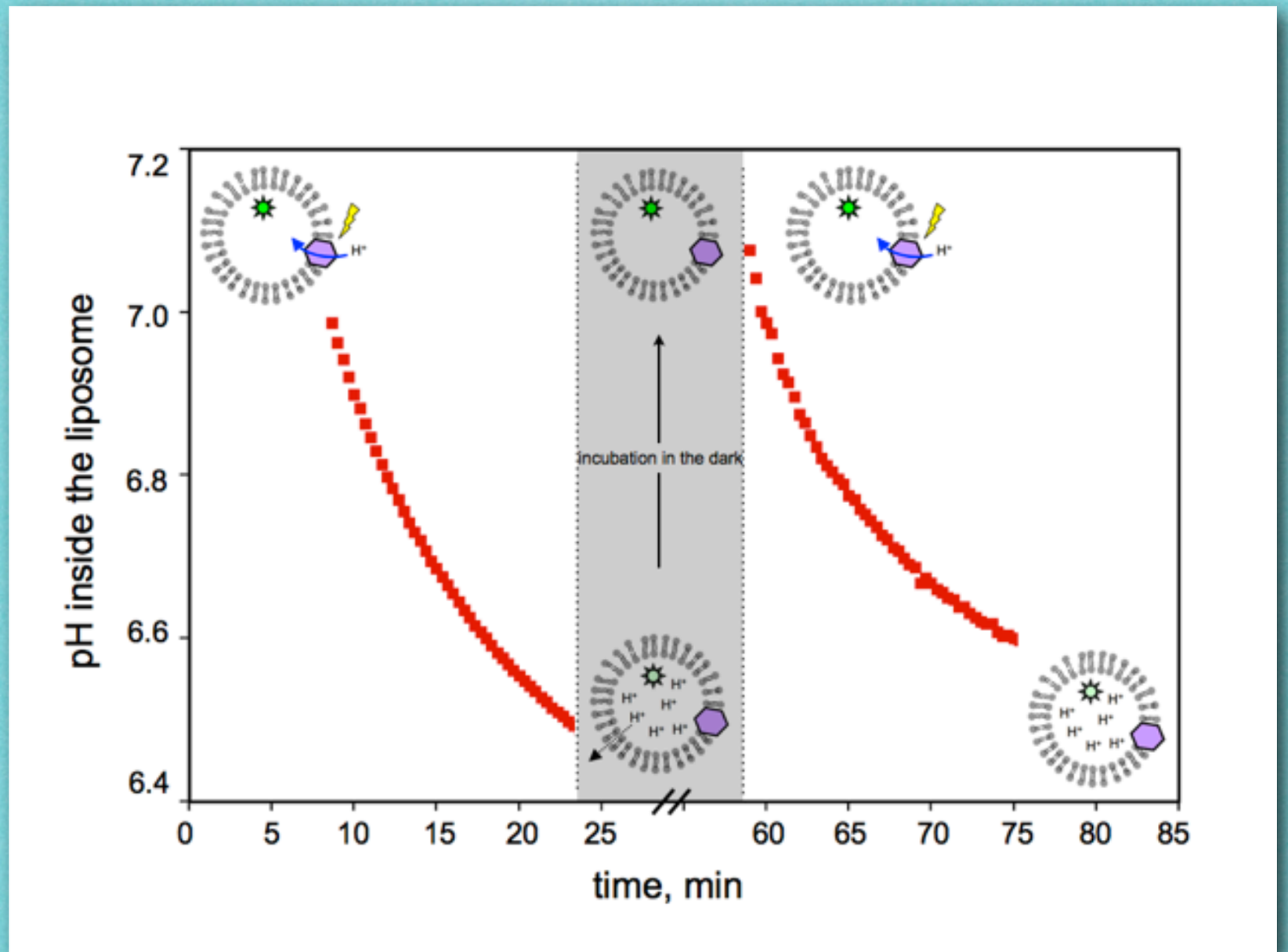
(Received for publication, August 20, 1973)

EFRAIM RACKER AND WALTHER STOECKENIUS

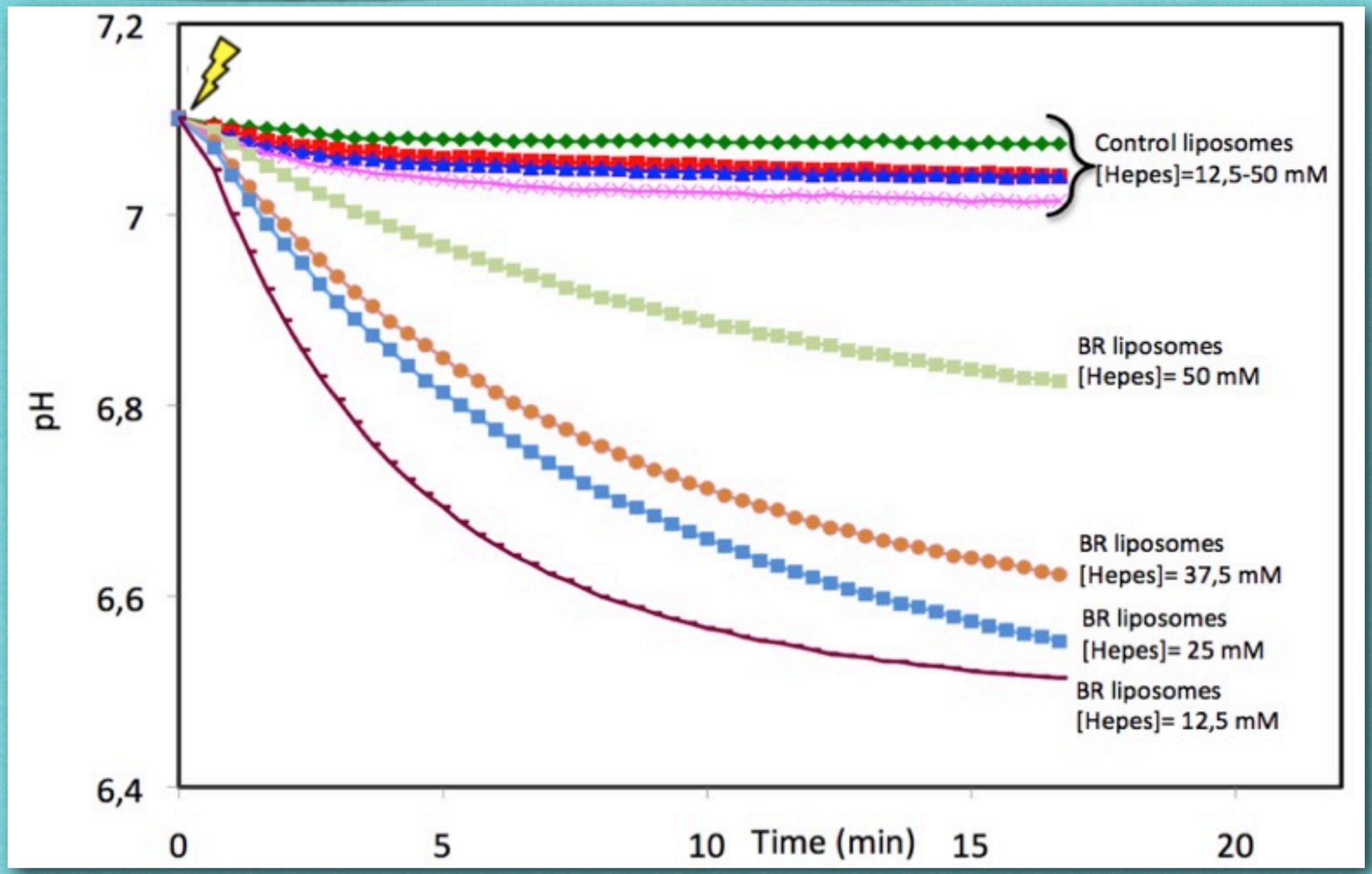
From the Section of Biochemistry, Molecular and Cell Biology, Cornell University, Ithaca, New York 14850, and Cardiovascular Research Institute and Department of Biochemistry and Biophysics, University of California, San Francisco, California 94143



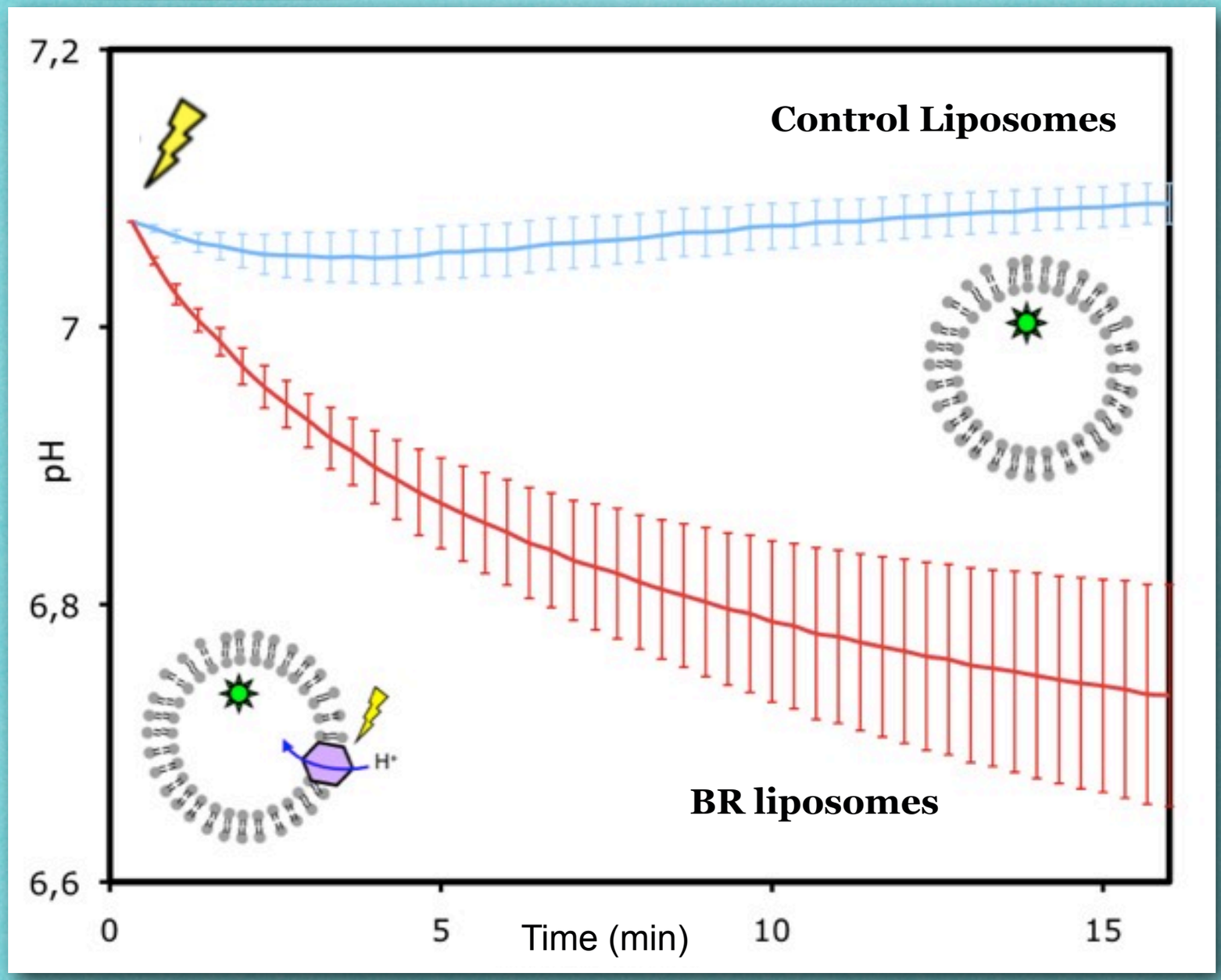
Reversible proton gradient



Tunable proton gradient

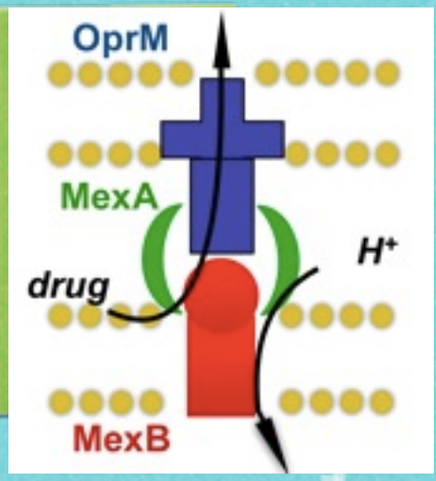


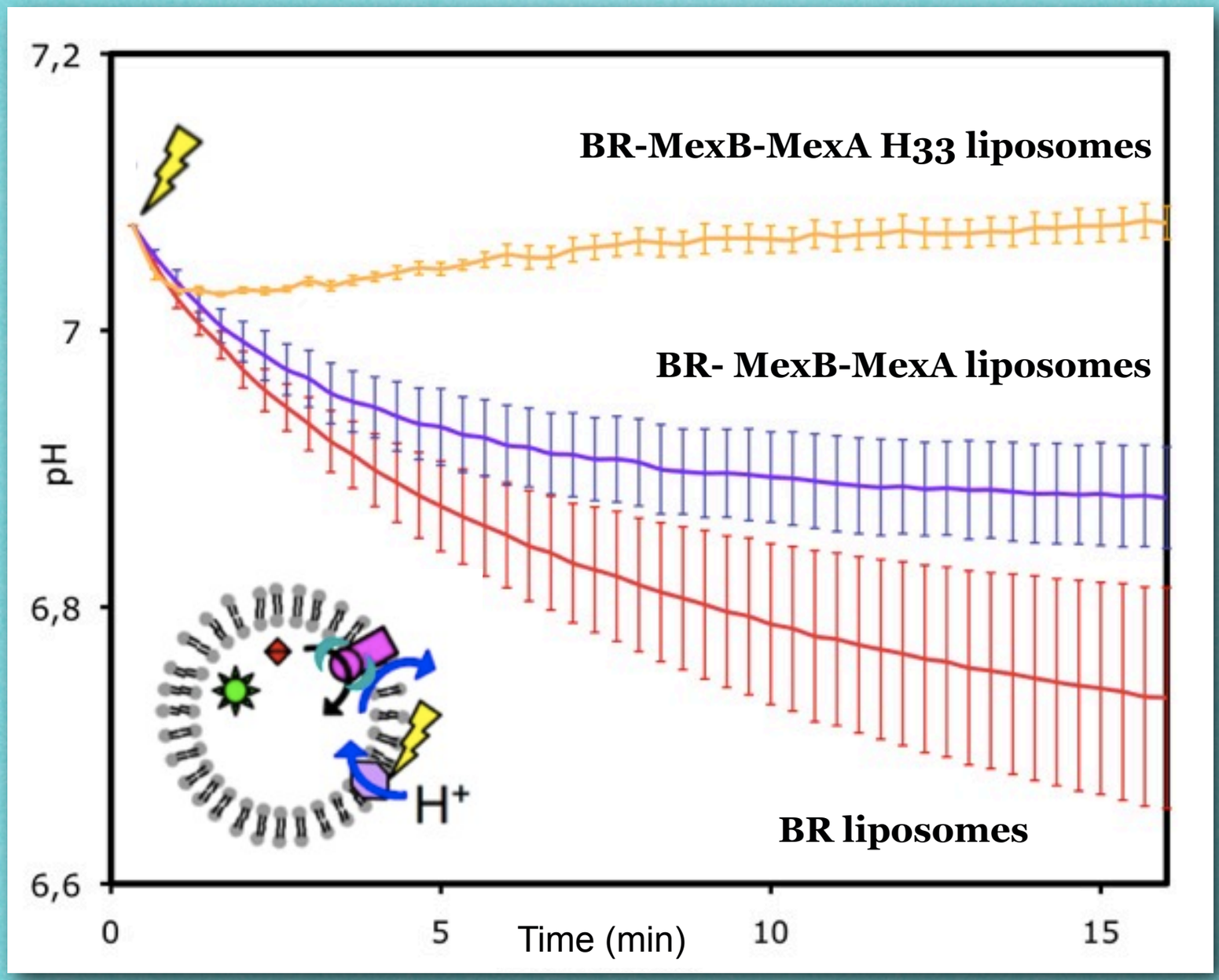
Results



Verchère, Broutin & Picard, *Sci Rep* 2012

Results





Verchère, Broutin & Picard, *Sci Rep* 2012

Functional test for MexB: conclusions and perspective

)-Conclusions:

- Reproductible test, reversible and tunable proton gradient
- Possible adaptation to other pmf-driven membrane proteins.

)-Perspective:

- Scale-up: screening of inhibitors

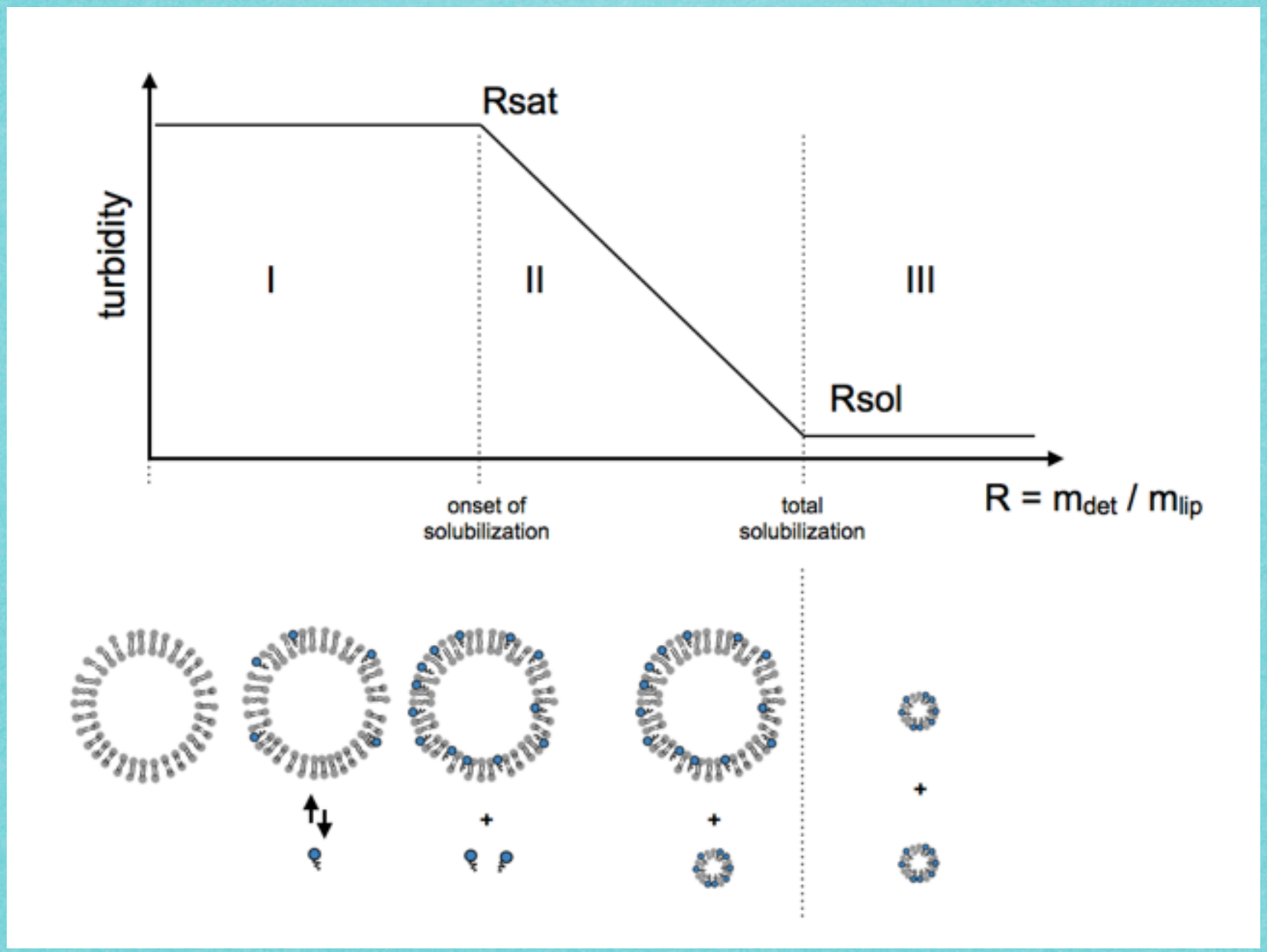
Acknowlegements

- People I work with:
  - Martin Picard
  - Manuela Dezi
  - Isabelle Broutin
  - Vladimir Adrien
  - Laura Monlezun
  - Bili Seijo

Thank you!

îledeFrance

Reconstitution



Adpated from Rigaud et al. *Meth Enzymol* 2003

Passive diffusion of protons