Select publications (1979 – 2019)

1. Genetic dissection of the secretory pathway in *S. cerevisiae*.

Beginning in 1976, my lab developed a genetic approach to the isolation of secretion mutants in yeast. We predicted that secretory vesicles would convey all proteins to the cell surface thus we expected that these genes would be essential for cell growth. By a variety of approaches, we found temperature sensitive lethal mutations that accumulated proteins at a block defined by mutation in each essential gene in this pathway.

Novick, P. and Schekman, R. (1979) Secretion and cell surface growth are blocked in a temperature sensitive mutant of Saccharomyces cerevisiae. *Proc. Natl. Acad. Sci. USA* 76, 1858-1862. http://www.pnas.org/content/76/4/1858

Novick, P., Field, C., and Schekman, R. (1980) The identification of 23 complementation groups required for post-translational events in the yeast secretory pathway. *Cell 21*, 205-215. PMID: 6996832 DOI: 10.1016/0092-8674(80)90128-2

Novick, P., Ferro, S., and Schekman, R. (1981) Order of events in the yeast secretory pathway. *Cell 25*, 461-469.
PMID: 7026045 DOI: 10.1016/0092-8674(81)90064-7

Deshaies, R., and Schekman, R. (1987) A yeast mutant defective at an early stage in import of secretory protein precursors into the endoplasmic reticulum. *J. Cell Biol. 105*, 633-645. https://www.ncbi.nlm.nih.gov/pubmed/3305520

Payne, G. S., and Schekman, R. (1989) Clathrin: A role in the intracellular retention of a Golgi membrane protein. *Science 245*, 1358-1365. DOI: 10.1126/science.2675311

Kaiser, C.A. and Schekman, R. (1990) Distinct sets of SEC genes govern transport vesicle formation and fusion early in the secretory pathway. *Cell* 61, 723-733. https://www.ncbi.nlm.nih.gov/pubmed/2188733

2. Biochemical dissection of the secretory pathway

The *SEC* genes showed little homology to known genes other than homologs in the genomes of higher eukaryotes thus the function of the gene products remained elusive. For this reason we invested considerable effort to establish a cell-free reaction to reconstitute the function of Sec proteins in the context of a biochemical pathway that could be used to purify functional forms of the proteins. The approach we developed for cell-free reactions with yeast was then reproduced in permeabilized preparations of culture mammalian cells and used to identify the nature of a lesion in one of the COPII subunits to explain the pathology of a human craniofacial disease.

Baker, D., Hicke, L., Rexach, M., Schleyer, M., and Schekman, R. (1988) Reconstitution of *Sec* gene product-dependent intercompartmental protein transport. *Cell 54*, 335-344. PMID: 3293799 DOI: 10.1016/0092-8674(88)90196-1

Deshaies, R. J., Koch, B. D., Werner-Washburne, M., Craig, E. A., and Schekman, R. (1988) A subfamily of stress proteins facilitates translocation of secretory and mitochondrial precursor polypeptides. *Nature 332*, 800-805. PMID: 3282178 DOI: 10.1038/332800a0

Deshaies, R. J., Sanders, S. L., Feldheim, D. A., and Schekman, R. (1991) Assembly of yeast Sec proteins involved in translocation into the endoplasmic reticulum into a membrane-bound multisubunit complex. *Nature 349*, 806-808 PMID: 2000150 DOI: 10.1038/349806a0

Barlowe, C., Orci, L., Yeung, T., Hosobuchi, M., Hamamoto, S., Salama, N., Rexach, M., Ravazzola, M., Amherdt, M., and Schekman, R. (1994) COPII: A membrane coat formed by Sec proteins that drive vesicle budding from the endoplasmic reticulum. *Cell* 77, 895-907. https://www.ncbi.nlm.nih.gov/pubmed/8004676

Fromme, J. C., Ravazzola, M., Hamamoto, S., Al-Balwi, M., Eyaid, W., Boyadjiev, S. A., Cosson, R., Schekman, R., and Orci, L. (2007) The genetic basis of a craniofacial disease provides insight into COPII coat assembly. *Developmental Cell*, 13, 623-634. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2262049/

3. Unconventional secretion and autophagy

Turning our attention to the secretory processes that distinguish mammalian cells from yeast, we probed the mechanism of procollagen traffic from the ER to the Golgi apparatus, the traffic of membrane proteins to the lateral surfaces of polarized epithelial cells, and the biogenesis of the autophagosome membrane and its role in unconventional secretion. We applied morphological approaches including super-resolution fluorescence microscopy, correlative fluorescence and thin section electron microscopy, high resolution cell fractionation and biochemical reconstitution with cultured intact and permeabilized mammalian cells.

Gorur, A., Yuan, L., Kenny, S. J., Baba, S., Xu, Ke, and Schekman, R. (2017) COPII-coated membranes function as transport carriers of intracellular procollagen I. J. Cell Biol., https://doi.org/10.1083/jcb.201702135

Yuan L, Kenny SJ, Hemmati J, Xu K, Schekman R. (2018) TANGO1 and SEC12 are copackaged with procollagen I to facilitate the generation of large COPII carriers. Proc Natl Acad Sci U S A. 2018 Dec 26;115(52):E12255-E12264. doi: 10.1073/pnas.1814810115.

Guo, Y., Zanetti, G., and Schekman, R. (2013) A Novel GTP-binding protein-adaptor protein complex responsible for export of Vangl2 from the *trans* Golgi network. *eLife* DOI: http://dx.doi.org/10.7554/eLife 00160.

Ge, L., Melville, D., Zhang, M. and Schekman, R. (2013) The ER-Golgi intermediate compartment is a key membrane source for the LC3 lipidation step of autophagosome biogenesis. *eLif*e, 2:e00947. DOI: 10.7554.

Zhang M, Kenny SJ, Ge L, Xu K, Schekman R.. (2015) Translocation of interleukin-1β into a vesicle intermediate in autophagy-mediated secretion. Elife. Nov 2;4. pii: e11205

4. Isolation and characterization of extracellular vesicles and the sorting of miRNAs for secretion in exosomes

My research is currently directed to an evaluation of the characteristics and biogenesis of extracellular vesicles (EVs). We have found two distinct species of EV that can be separated on a buoyant density gradient. The denser species, exosomes, has a set of highly enriched and cell type-specific miRNAs but the most abundant RNAs are full-length tRNAs, Y-RNAs and vault RNA. We have devised a biochemical approach to discover molecular basis of miRNA sorting into exosomes and have identified two different RNA binding proteins responsible for the sorting of distinct sets of miRNAs and tRNA.

Shurtleff MJ, Temoche-Diaz MM, Karfilis KV, Ri S, Schekman R (2016). Y-box protein 1 is required to sort microRNAs into exosomes in cells and in a cell-free reaction. Elife. Aug 25;5. pii: e19276. doi: 10.7554/eLife.19276. PMID: 27559612

Shurtleff, M., Yao, J., Qin, Y, Nottingham, R., Temoche-Diaz, M., Schekman, R and Lambowitz, A. (2017) A broad role for YBX1 in defining the small non-coding RNA composition of exosomes PNAS 2017 October, 114 (43) E8987-E8995. https://doi.org/10.1073/pnas.1712108114

Shurtleff, M., Temoche-Diaz, M. and Schekman, R. (2018) Extracellular Vesicles and Cancer: Caveat Lector Ann. Rev. Cancer Biology https://doi.org/10.1146/annurev-cancerbio-030617-050519

Temoche-Diaz, M., Shurtleff, M.J., Nottingham, R., Yao, J., Fadadu, R. P., Lambowitz, A.and Schekman, R. 2019) Distinct mechanisms of microRNA sorting into cancer-derived extracellular vesicle subtypes. (preprint posted on bioRxiv)