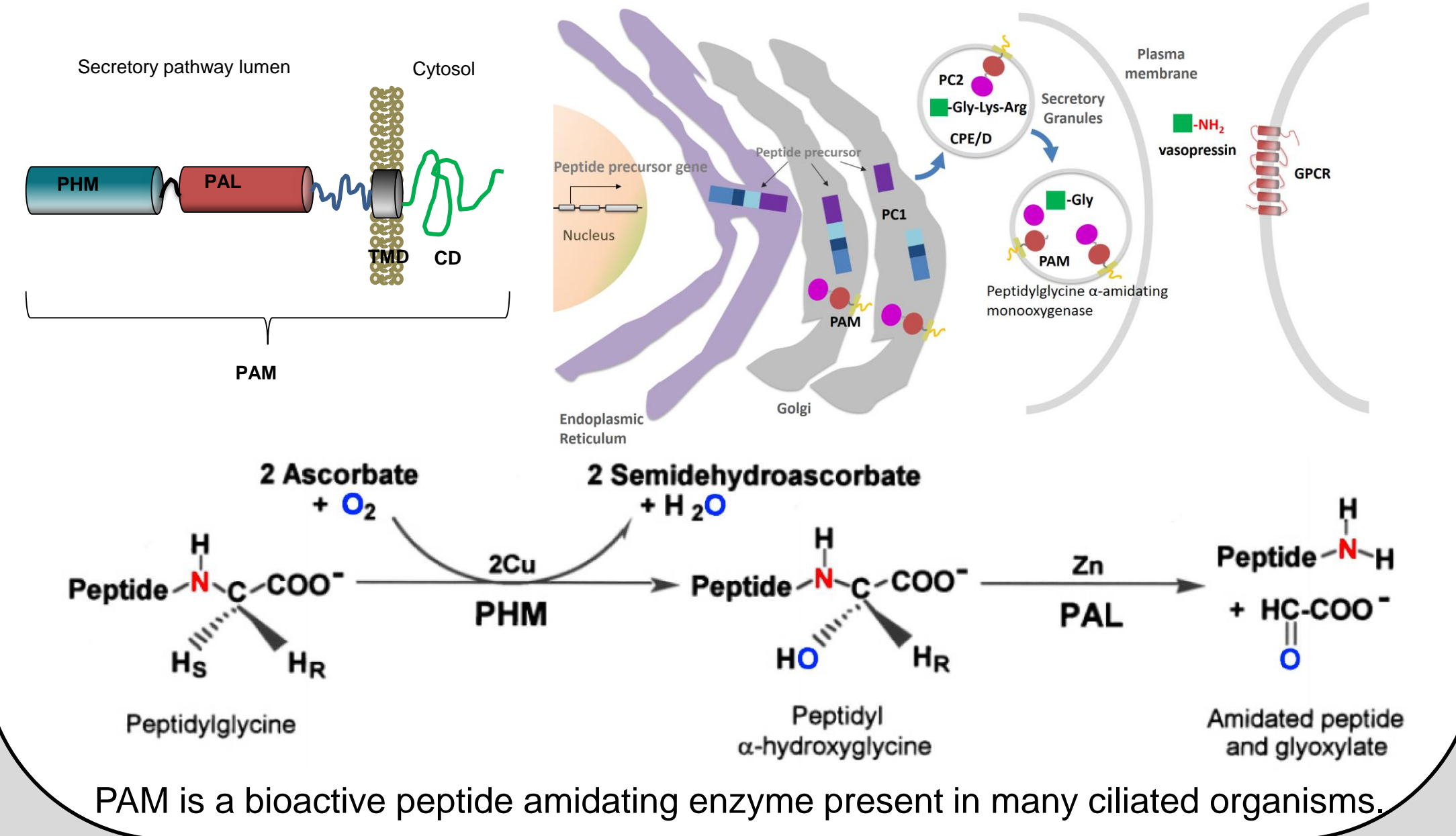


A Bioactive Peptide Amidating Enzyme is Specifically Released in Ciliary Ectosomes during Mating in *Chlamydomonas*

Dhivya Kumar¹, Raj Luxmi², Myah Bartolotta³, Richard E. Mains², Stephen M. King¹ and Betty A. Eipper^{1,2}

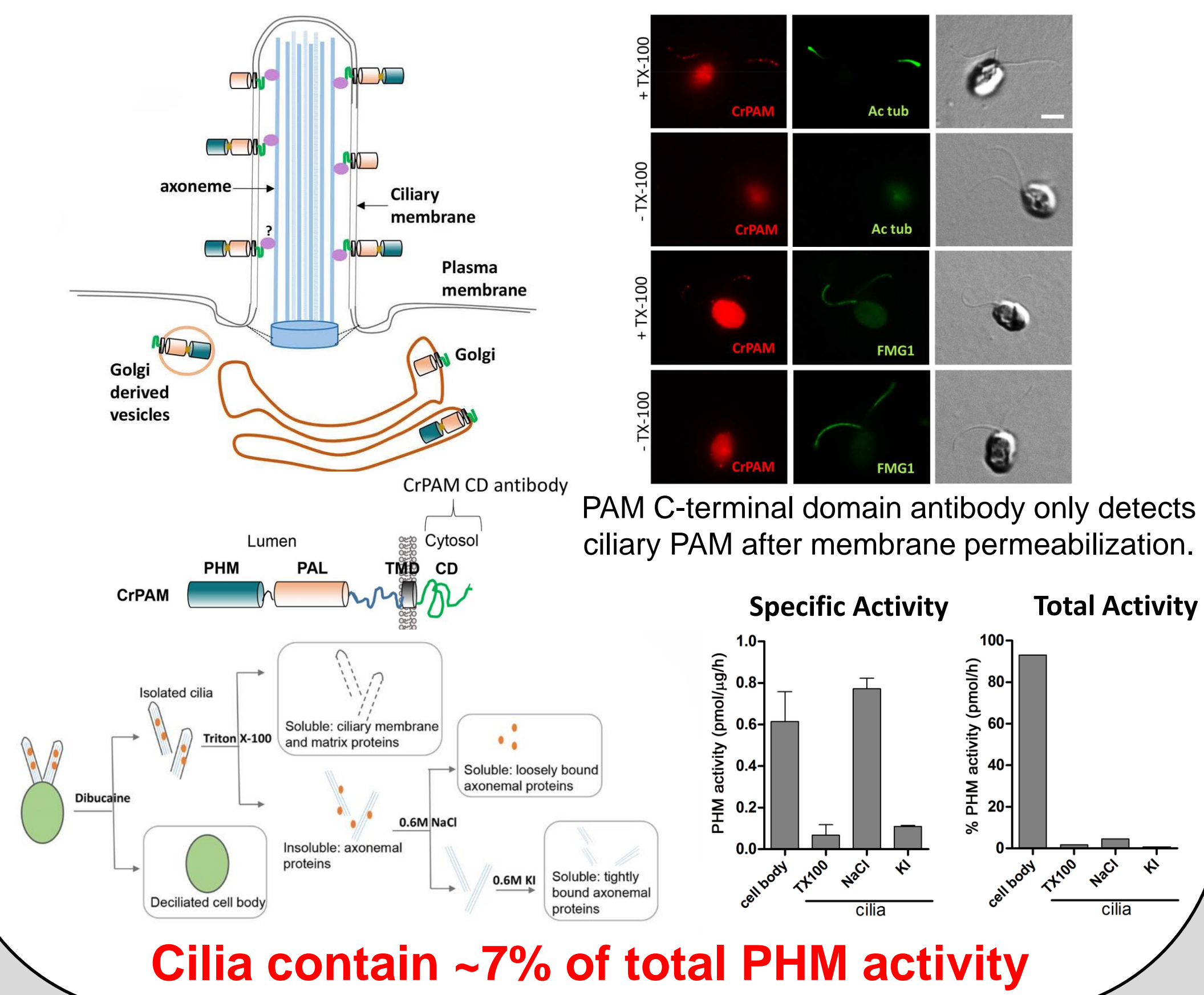
Departments of ¹Molecular Biology and Biophysics and ²Neuroscience, University of Connecticut Health Center, Farmington, CT, USA and ³Wheaton College, Norwood, MA, USA

Peptidylglycine α -Amidating Monooxygenase (PAM) Organization and Chemistry of the Amidation Reaction

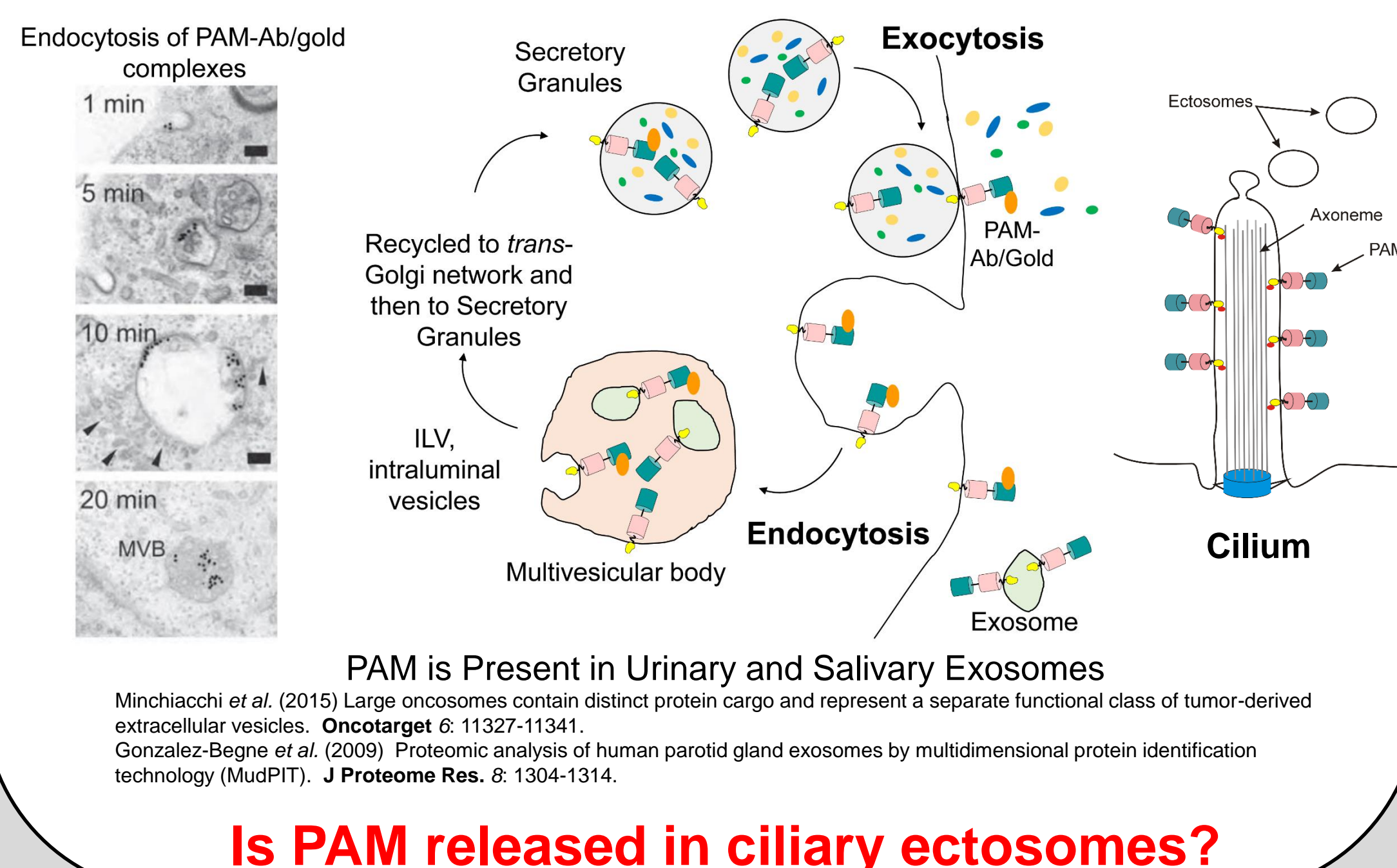


Topology and Axonemal Association of Ciliary PAM

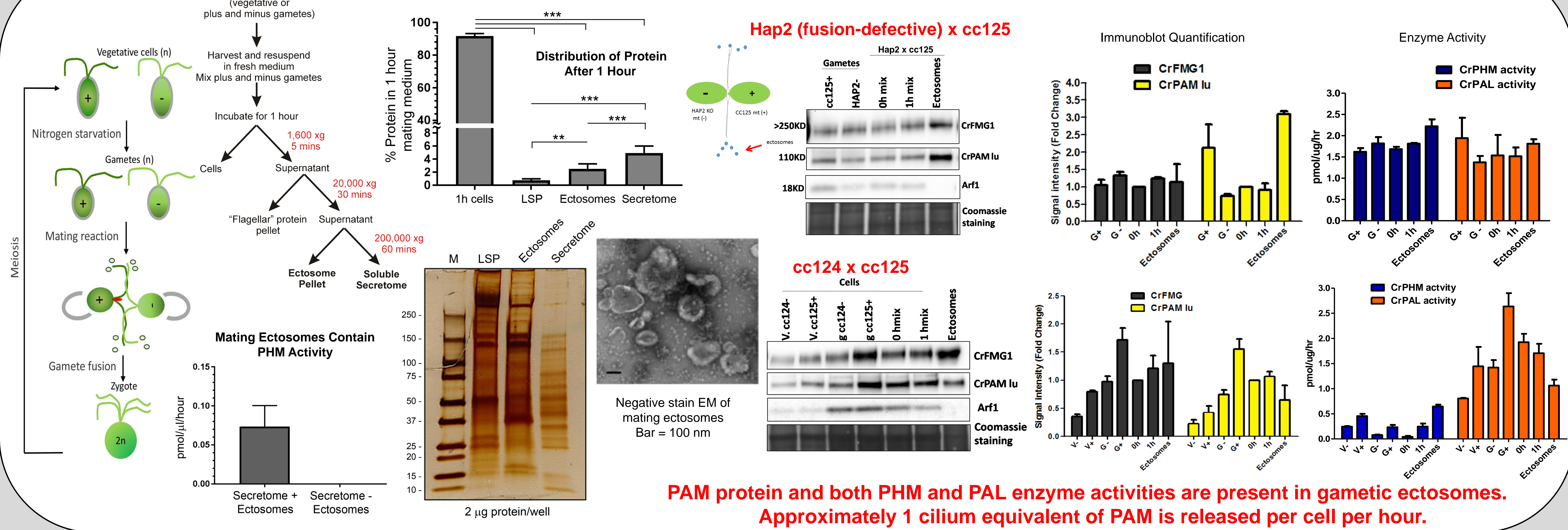
Data from Kumar et al. (2016) J Cell Sci 129: 943-956 and Kumar et al. (2017) eLife 6:e25728.



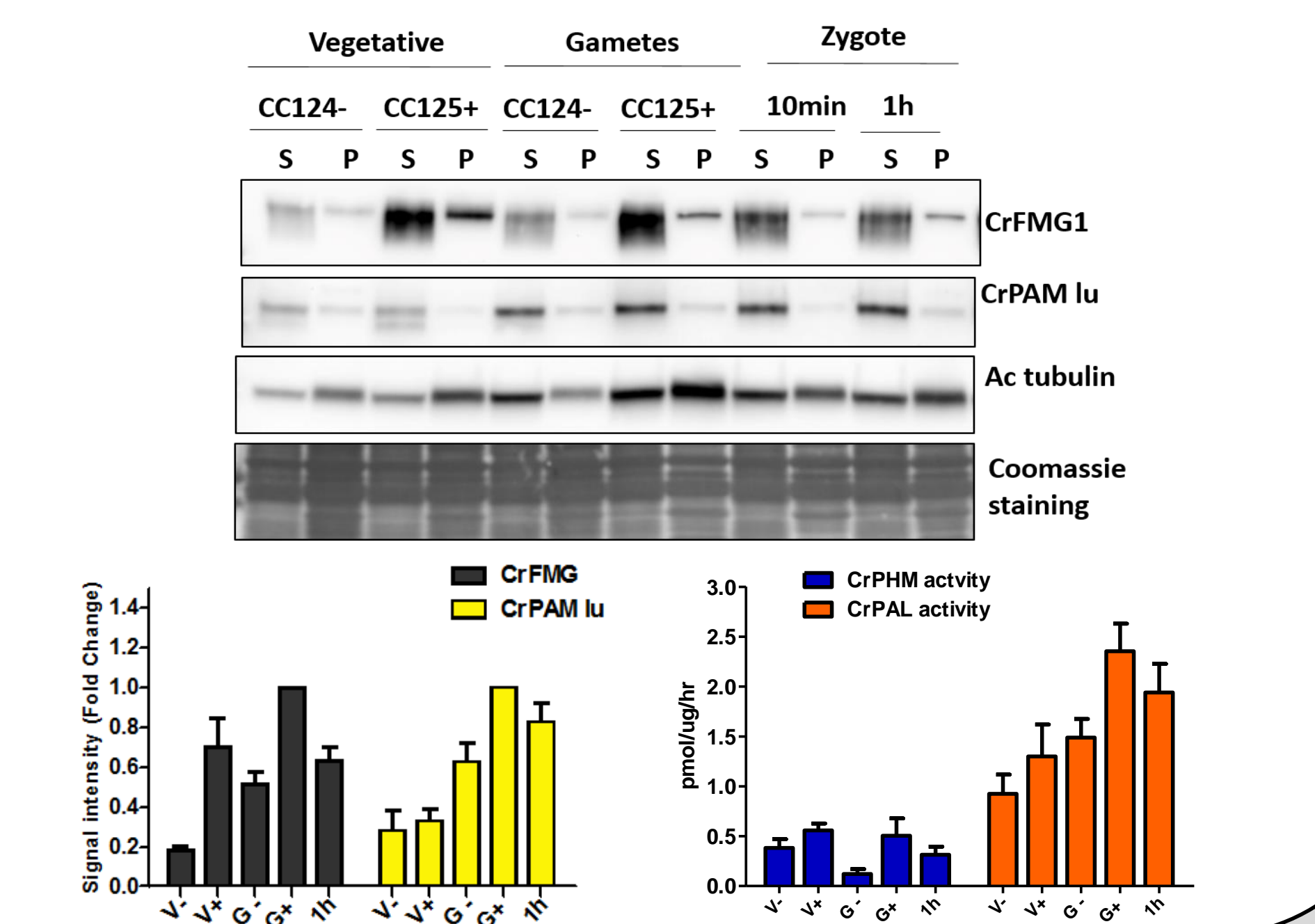
PAM Traverses the Biosynthetic and Endocytic Pathways in Rodent Neuroendocrine Cells



PAM is Released in Ciliary Ectosomes from Mating Gametes

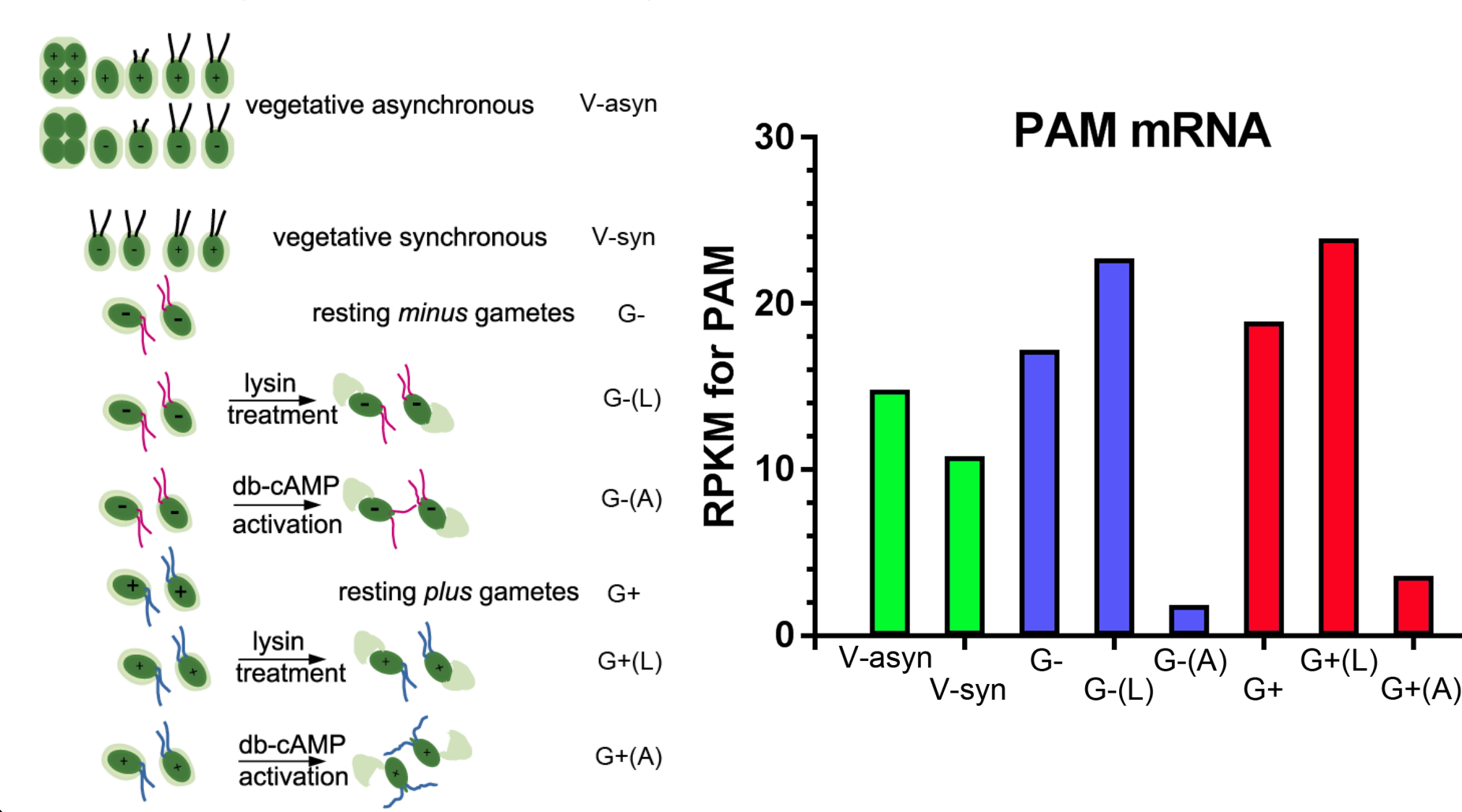


PAM Protein is Upregulated During the Sexual Cycle of *Chlamydomonas*

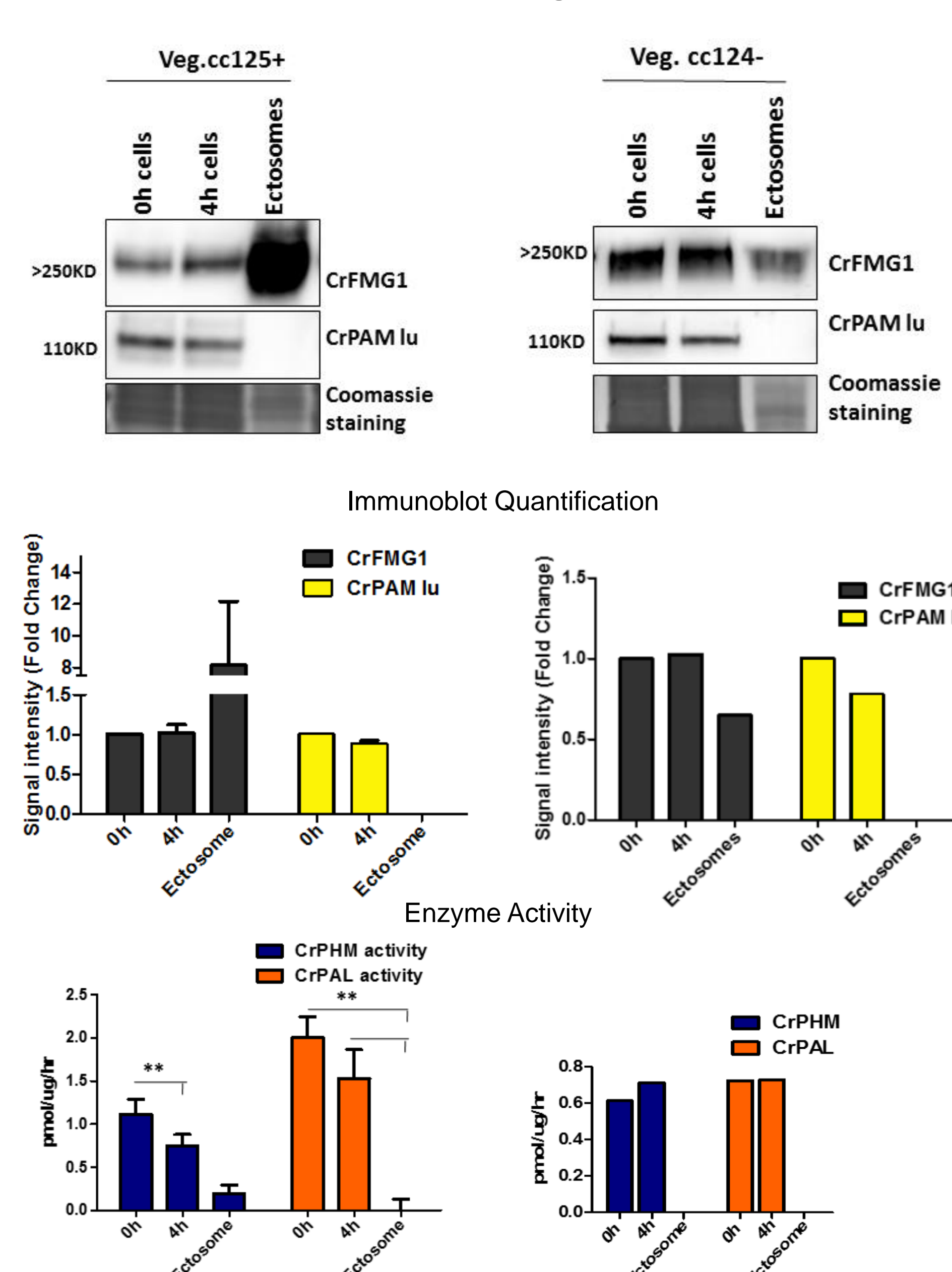


PAM Transcripts Are Dramatically Reduced upon Gamete Activation

(Diagram and data from Ning et al. (2013) *Genes Dev.* 27: 1198-1215)

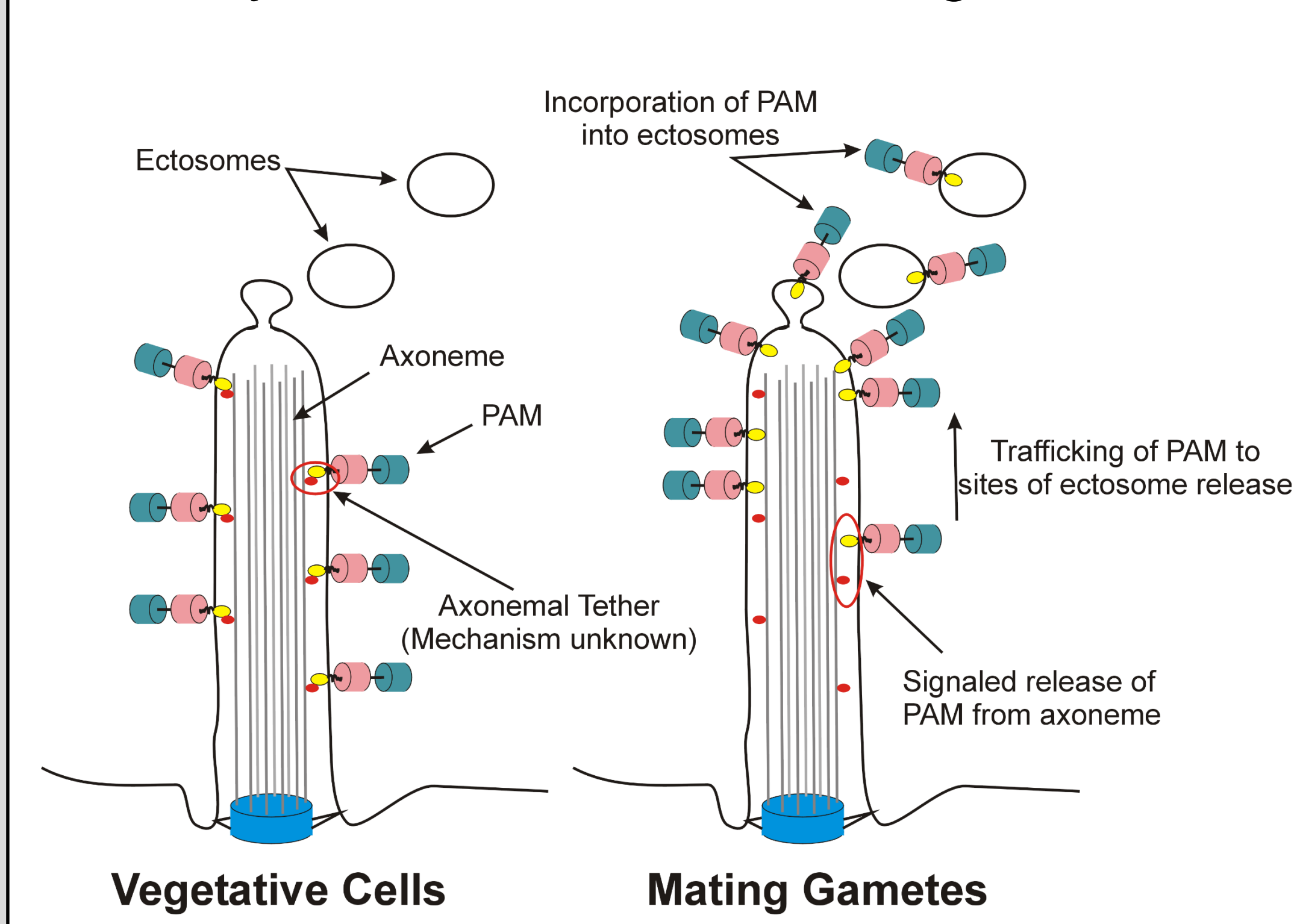


Very Little PAM is Released in Ciliary Ectosomes from Vegetative Cells



PAM protein and PHM/PAL enzyme activities were barely detectable in ectosomes from vegetative *Chlamydomonas* cells

Model for the Regulated Release of Axonemal PAM and Incorporation into Ciliary Ectosomes from Mating Gametes



Why Does *Chlamydomonas* Release PAM in Mating Ectosomes?

- Possibilities include:-
1. PAM plays a role in ectosome-based signaling or other functional processes.
 2. It provides a way to remove PAM from cilia of mating cells as it is no longer needed.
 3. Potentially ciliary localized PAM may be inhibitory to further progress in the mating reaction if not removed.
 4. Amidated proteins are present in mating ectosomes (see poster # P2047).

We are currently investigating the biochemical mechanism(s) by which PAM is released from the axoneme and its function in ectosomes.

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