

## Aberystwyth University

### *The Yin-Yang of the Green Fluorescent Protein*

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*Published in:*

Journal of Photochemistry and Photobiology B: Biology

*DOI:*

[10.1016/j.jphotobiol.2022.112603](https://doi.org/10.1016/j.jphotobiol.2022.112603)

*Publication date:*

2023

*Citation for published version (APA):*

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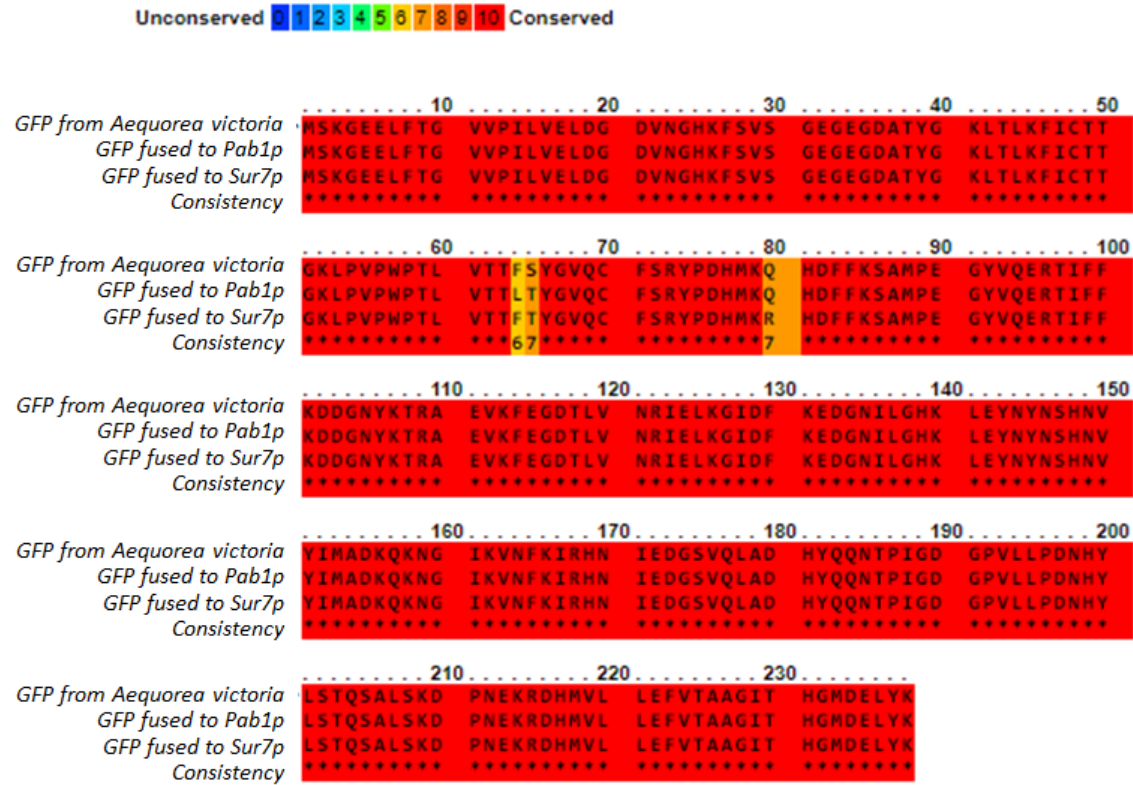
## Supplementary data

**Table S1.** Yeast strains used in this study.

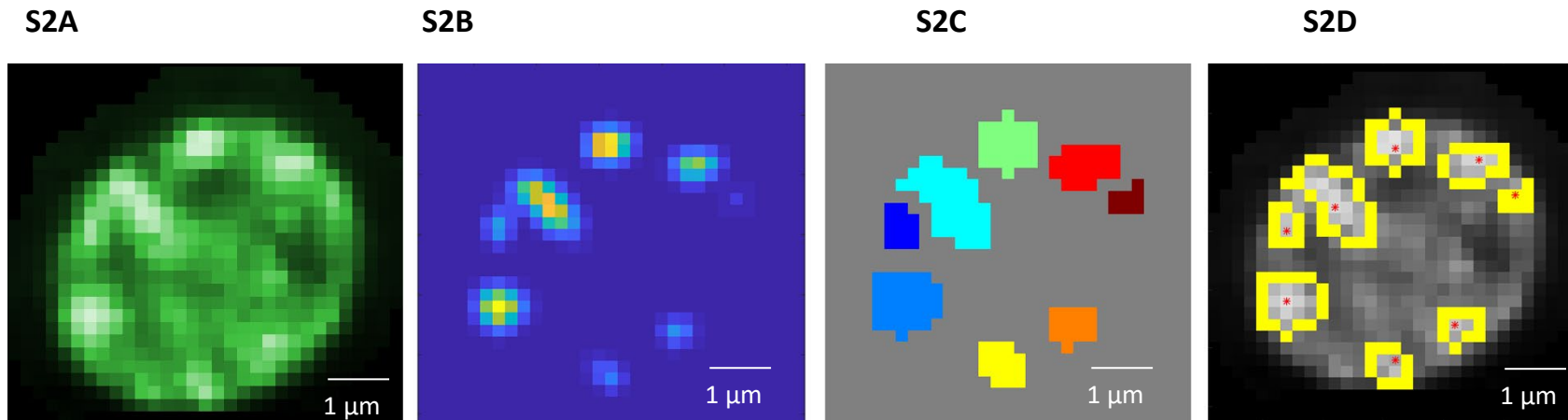
Wild-type strain				
	BY4742		MAT $\alpha$ ; his3 $\Delta$ 1; leu2 $\Delta$ 0; lys2 $\Delta$ 0; ura3 $\Delta$ 0	EUROSCARF
Knockout mutants				
	<i>erg6</i> $\Delta$	YML008C	BY4742; MAT $\alpha$ ; his3 $\Delta$ 1; leu2 $\Delta$ 0; met15 $\Delta$ 0; ura3 $\Delta$ 0; YML008C::KanMX4	converts zymosterol to fecosterol in the ergosterol biosynthetic pathway
Insertion mutants				
	<i>SUR7-GFP</i>	YML052W	BY4742; MAT $\alpha$ ; his3 $\Delta$ 1; leu2 $\Delta$ 0; met15 $\Delta$ 0; ura3 $\Delta$ 0; YML052W::GFP	plasma membrane protein in microdomains Dupont <i>et al.</i> (2010) <sup>61</sup>
	<i>erg6</i> $\Delta$ / <i>SUR7-GFP</i>		BY4742; MAT $\alpha$ ; his3 $\Delta$ 1; leu2 $\Delta$ 0; met15 $\Delta$ 0; ura3 $\Delta$ 0; YML008C::KanMX4; YML052W::GFP	see above
	<i>PAB1-GFP</i>	YER165W	BY4742; MAT $\alpha$ ; his3 $\Delta$ 1; leu2 $\Delta$ 0; met15 $\Delta$ 0; ura3 $\Delta$ 0; YER165W::GFP	stress granules and P-bodies This study
	<i>PBP1</i> (+)	YGR178C	BY4742; MAT $\alpha$ ; his3 $\Delta$ 1; leu2 $\Delta$ 0; lys2 $\Delta$ 0; ura3 $\Delta$ 0 / PBP1(+)	poly(A)-binding protein (Pab1)-binding protein and a component of stress granules This study, plasmid donated by Pr Tatsuya Maeda [Takahara and Maeda, 2012] <sup>31</sup>
	<i>PBP1</i> (+)/ <i>PAB1-GFP</i>	YGR178C/YER165W	BY4742; MAT $\alpha$ ; his3 $\Delta$ 1; leu2 $\Delta$ 0; met15 $\Delta$ 0; ura3 $\Delta$ 0; YER165W::GFP/PBP1(+)	see above

**Table S2.** Primers used in this study.

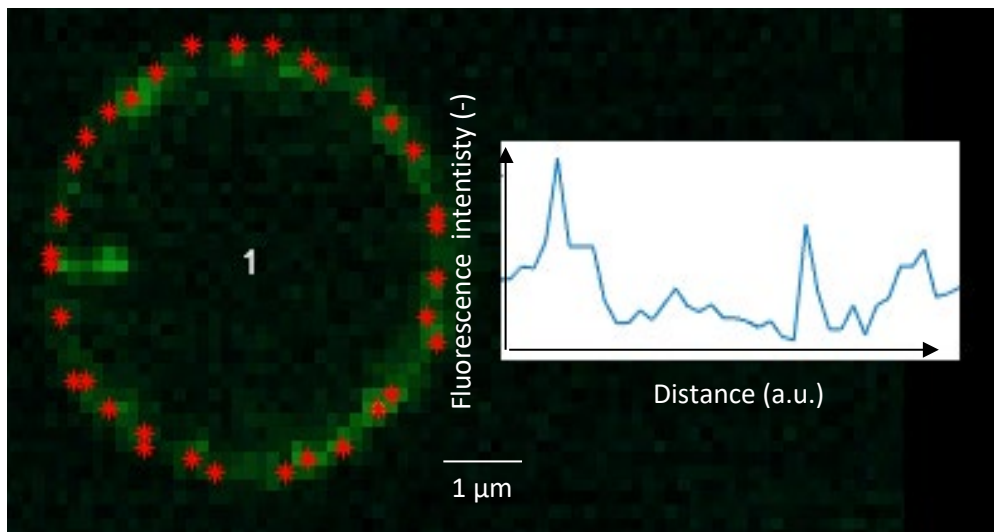
<i>Primers</i>	<i>Description</i>
RH-Pab1-GFP-F	GTCTTTCAAAAAGGAGCAAGAACAACAAACTGAGCAAGCTGGTGACGGTGCTGGTTTA
RH-Pab1-GFP-R	TAAGTTTGTTGAGTAGGGAAGTAGGTGATTACATAGAGCATCGATGAATTCGAGCTCG



**Figure S1.** Multiple sequence alignment of the GFP used in this study and the GFP from the jellyfish *A. victoria*. Alignment was performed using PRALINE, a multiple sequence alignment toolkit <sup>69</sup>.



**Figure S2.** Image processing steps to isolate and characterize RNP granules at the level of the single cell. **(A)** Cell isolated and denoised, **(B)** Region of interest with RNP granules **(C)** RNP granules segmented, **(D)** Granules centroids (red stars) and perimeter (yellow lines) calculated and overlay on cell image.



**Figure S3.** Image processing steps to isolate and characterize Sur7p-GFP at the level of the single cell. Each red star is a retained profile point. Yeast fluorescence profile is showed on the right insert.

## References

[31] T. Takahara, T. Maeda, Transient sequestration of TORC1 into stress granules during heat stress, *Mol. Cell* 47 (2012) 242–252.

[61] S. Dupont, L. Beney, J.F. Ritt, J. Lherminier, P. Gervais, Lateral reorganization of plasma membrane is involved in the yeast resistance to severe dehydration, *Biochim. Biophys. Acta* 1798 (2010) 975–985.

[69] V. A. Simossis, J. Heringa, PRALINE: a multiple sequence alignment toolbox that integrates homology-extended and secondary structure information, *Nucleic Acids Res.* 33 (2005) W289–W294.

