

Identifying essential components of a plant LINC complex

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Introduction

The higher plant NE contains a functional Linker of Nucleoskeleton and Cytoskeleton (LINC) complex based on conserved Sad1-Unc84 (SUN) domain proteins and plant specific Klarsicht/Anc1/Syne homology (KASH) domain proteins (Figure 1). Recent evidence suggests the presence of a plant lamina underneath the inner membrane and various coiled-coil proteins have been hypothesised to be associated with it including Crowded Nuclei (CRWN), Nuclear Envelope Associated Protein (NEAP) protein families as well as the CRWN binding protein KAKU4. In this study, we explore the presence of proteins of these nuclear envelope (NE) proteins from the most ancestral plant species to advanced angiosperms.

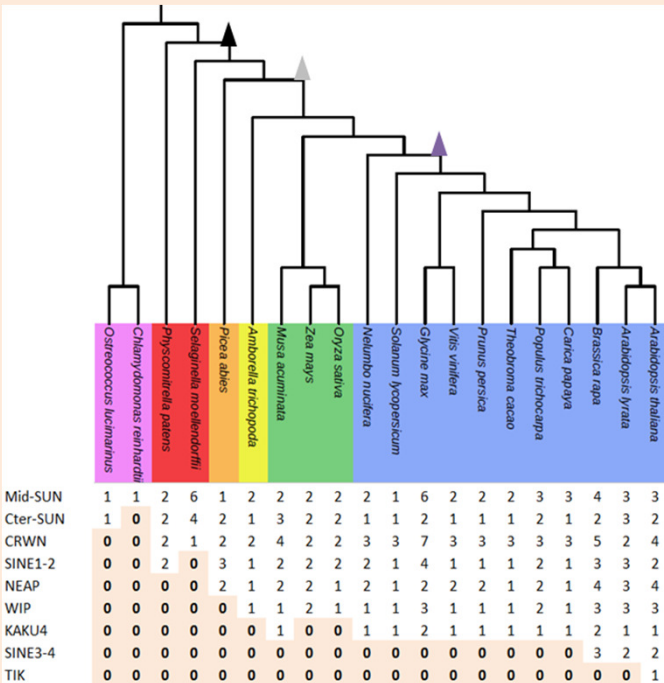


Figure 2: Distribution of components of plant nuclear envelope in the plant kingdom.

Selected plant lineages used in this study from left to right: Unicells Algae (pink), Moss and Club Moss (red), Gymnosperm (orange), Basal Angiosperms (yellow), Monocots (green) and Eudicots (blue). zeta epsilon and gamma WGDs are indicated as arrow heads respectively in black, grey and purple.

Distribution of the 9 protein families (rows) in the 20 species (columns). Absence (0) of a given protein is highlighted in light orange.

Methods were as described in Poulet et al., 2017.

Phylogenetic reconstruction

Selected sequences were first aligned with MUSCLE, a multiple sequence alignment tool (Edgar, 2004), using default parameters. The alignment was then refined using Gblocks (Talavera and Castresana, 2007). Fast-Tree was then applied with default parameters, for the construction of the phylogenetic tree (Price et al., 2010). Fast-Tree infers approximately-maximum-likelihood phylogenetic trees from alignments. Finally, phylogenetic trees were drawn using the Interactive Tree Of Life ITOL (Letunic and Bork, 2011).

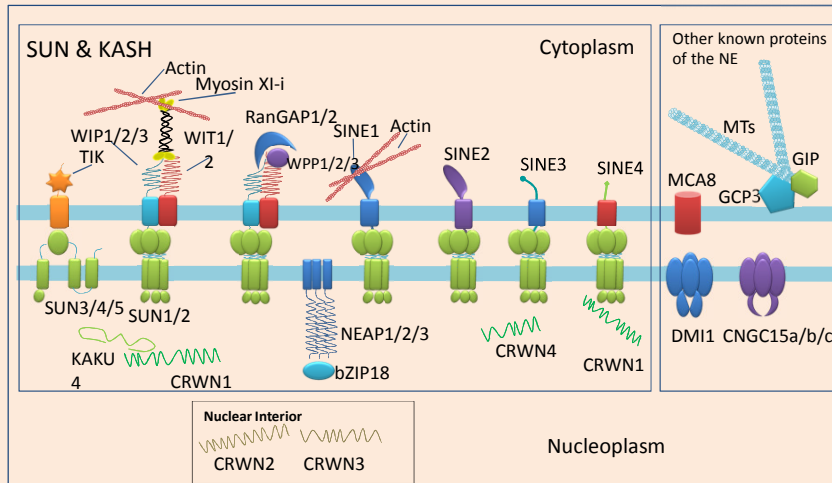


Figure 1 Plant nuclear envelope-associated proteins. SUN1/2 are shown as trimers. TIK is shown to interact with SUN3/4/5, but also interacts with SUN1/2. WIP/WIT/RanGAP and WIP/WIT/Myosin XI-i complexes are drawn separately, but all proteins might reside in a single complex. KAKU4-CRWN1, CRWN1-SUN1/2, and NEAP1/2/3-bZIP18 interactions are indicated.

Results and Discussion

Mid-SUN proteins were present in all species studied while classical C-ter SUN domain proteins were absent in Chlamydomonas, but appear throughout the multicellular plant species studied, suggesting a key role for mid-SUN proteins. Evolution of KASH domain proteins has resulted in increasing complexity, with some appearing in all species considered, while other KASH proteins are progressively gained during evolution. Failure to identify CRWN homologs in unicellular organisms included in the study and their presence in higher plants leads us to speculate that convergent evolution may have occurred in the formation of the lamina with each kingdom having new proteins such as the Lamin B receptor (LBR) and Lamin-Emerin-Man1 (LEM) domain proteins (animals) or NEAPs and KAKU4 (plants). Our data support a model in which increasing complexity at the nuclear envelope occurred through the plant lineage and suggest a key role for mid-SUN proteins as an earliest and essential component of the nuclear envelope.

References

- Edgar, R.C. (2004). MUSCLE: a multiple sequence alignment method with reduced time and space complexity. *BMC Bioinformatics* 5, 113
- Letunic, I., and Bork, P. (2011). Interactive Tree Of Life v2: online annotation and display of phylogenetic trees made easy. *Nucleic Acids Res.* 39, W475–W478.
- Poulet, A., Probst, A.V., Graumann, K. Tatout, C. and Evans, D.E. (2017) Exploring the evolution of the proteins of the plant nuclear envelope. *Nucleus*. 8: 46–59.
- Talavera, G., and Castresana, J. (2007). Improvement of phylogenies after removing divergent and ambiguously aligned blocks from protein sequence alignments. *Syst. Biol.* 56, 564–577.

ACKNOWLEDGEMENTS

The work was supported by the CNRS, INSERM, Clermont Auvergne and Oxford Brookes Universities.



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