CORTICAL ER – PM CONTACT SITES IN PLANTS

Where Cell Biology meets abiotic stress tolerance

EUROPEAN NETWORK FOR PLANT ENDOMEMBRANE RESEARCH

Jessica Pérez-Sancho
August 27-30, 2013
Ghent, Belgium.
The ER requires physical interactions with different organelles to be fully functional.

- cER interacts with PM through membrane contact sites: ER-PMCS.
- The establishment and the functions of this ER-PMCS are unknown in plants.
cER – PM Contact Sites (ER-PMCS)

- Close apposition between cER and PM
- 10-30 nm distance between membranes
- Ribosomal exclusion

Pérez-Sancho et al., Unpublished
ER-PMCS establishment relies on Ca\(^{2+}\)-dependent phospholipid binding proteins

**Yeast : Tricalbins**

**Mammals : Extended Synaptotagmins**

**Plants : Synaptotagmins**
ER-PMCS establishment mechanism in yeast and mammals
ER-PMCS establishment mechanism in yeast and mammals
Are Arabidopsis SYTs localized in ER-PMCS?

Pérez-Sancho et al., Unpublished
SYT1 is closely associated to the cortical ER

Pérez-Sancho et al., Unpublished
SYT1 partially colocalizes with PM markers

Pérez-Sancho et al., Unpublished
SYT1 and cortical microtubules signals are mutually exclusive

Pérez-Sancho et al., Unpublished
Different SYTs are located in plasmodesmata

SYTs localization in plants is compatible with function in ER-PMCS

Pérez-Sancho et al., Unpublished
Known functions of ER-PMCS in yeast and mammals

1. Ca\(^{2+}\) homeostasis
2. Lipid homeostasis
3. Stress response
Known functions of ER-PMCS in yeast and mammals

1. Ca$^{2+}$ homeostasis
2. Lipid homeostasis
3. Stress response
syt1 has altered Ca\(^{+2}\) sensitivity under stress conditions

<table>
<thead>
<tr>
<th>Condition</th>
<th>Plant 1</th>
<th>Plant 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>MS (1/10) + NaCl 100mM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca(^{2+}) 3mM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Col</td>
<td></td>
<td></td>
</tr>
<tr>
<td>syt1</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Schapire et al., 2008 Plant Cell
**sy**t1 has altered Ca$^{2+}$ sensitivity under stress conditions

SYT1 might be involved Ca$^{2+}$ homeostasis during stress episodes.

Schapire et al., 2008 Plant Cell
Known functions of ER-PMCS in yeast and mammals

1. Ca$^{2+}$ homeostasis
2. Lipid homeostasis
3. Stress response
SYT1 binds PtdIns(P)s and PGs
Lipid profile is altered in *syt1*

<table>
<thead>
<tr>
<th>LIPIDS</th>
<th>Col</th>
<th>syt1</th>
</tr>
</thead>
<tbody>
<tr>
<td>DGDG</td>
<td>1495.1 ± 67.1</td>
<td>1567.6 ± 104.3</td>
</tr>
<tr>
<td>MGDG</td>
<td>3206.4 ± 226.5</td>
<td>3218.0 ± 209.8</td>
</tr>
<tr>
<td>PC</td>
<td>3726.3 ± 264.6</td>
<td>3687.0 ± 243.7</td>
</tr>
<tr>
<td>PE</td>
<td>337.7 ± 19.2</td>
<td>346.5 ± 19.3</td>
</tr>
<tr>
<td>TAG</td>
<td>326.4 ± 36</td>
<td>326.5 ± 17.4</td>
</tr>
<tr>
<td>DAG</td>
<td>19.6 ± 1.4</td>
<td>21.1 ± 1.2</td>
</tr>
<tr>
<td>SQDG</td>
<td>194.2 ± 9.8</td>
<td>171.5 ± 13.0</td>
</tr>
<tr>
<td>PG</td>
<td>79.5 ± 18.4</td>
<td>132.7 ± 22.9</td>
</tr>
<tr>
<td>PS</td>
<td>474 ± 41.3</td>
<td>459.7 ± 30.0</td>
</tr>
<tr>
<td>PI</td>
<td>1 ± 0.2</td>
<td>2.2 ± 0.4</td>
</tr>
</tbody>
</table>

*SYT1 might be involved in lipid homeostasis maintenance*

Rosado et al., Unpublished
Known functions of ER-PMCS in yeast and mammals

1. Ca\(^{2+}\) homeostasis
2. Lipid homeostasis
3. Stress response
The *syt1* lipid changes are associated with cold stress sensitivity

<table>
<thead>
<tr>
<th></th>
<th>Col</th>
<th>syt1</th>
</tr>
</thead>
<tbody>
<tr>
<td>DGDG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MGDG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PE</td>
<td></td>
<td></td>
</tr>
<tr>
<td>TAG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>DAG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MAG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SQDG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PG</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PI</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Fold Reduction</th>
<th>Fold accumulation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cold Stressed</td>
<td>&lt;12 8 4 3 2 1.5 1 1.5 2 2.5 3 3.5 5 10 &gt;15</td>
<td></td>
</tr>
</tbody>
</table>

**Cold Treatment**

**Survival (%)**

- WT
- syt1
- Compl 5/1
- Compl 9/2

**SYT1 is involved in cold tolerance**

Rosado et al., Unpublished
Are Arabidopsis SYTs the orthologs of tricalbins and extended synaptotagmins?

- SYT1 has the proper subcellular localization.
- SYT1 has equivalent functions.

It is required for Ca$^{2+}$ and lipid homeostasis.
- It binds phospholipids.
- It has roles in stress tolerance.
Acknowledgements

Universidad de Málaga
Abel Rosado
Arnaldo Schapire
Miguel Ángel Botella

CIB-CSIC Madrid
Carlos Perea
Julio Salinas

Gent University
Steffen Vanneste
Jiří Friml

MPI (Potsdam-Golm)
Sonia Osorio
Lothar Willmitzer

UPSC Umea
Stephanie Robert
Thank you!
The *syf1* lipid changes are associated with cold stress sensitivity

Rosado et al., Unpublished
SYT1 BINDING TO PS: Ca^{+2} DEPENDENCY

Biochemical Properties of the Arabidopsis Synaptotagmin1 (SYT1)

Schapire et al., 2008. Plant Cell
Biochemical Properties of the Arabidopsis Synaptotagmin1 (SYT1)

SYT1 Binding to PS: Ionic Competition

10µM Ca^{2+}

Rosado et al., Unpublished
Plant synaptotagmin families
SYT1 binds PtdIns(P)s and PGs in a Ca^{+2} independent manner

Rosado et al., Unpublished