



HAL
open science

CMLS control host-plant interactions

Christian Mazars, Benoît Ranty, Didier Aldon, Ralf Oelmüller, Jean-Philippe Galaud, Axel Mithöfer

► **To cite this version:**

Christian Mazars, Benoît Ranty, Didier Aldon, Ralf Oelmüller, Jean-Philippe Galaud, et al.. CMLS control host-plant interactions. *Endocytobiosis and Cell Research*, 2016, 27 (1), pp.1-13. hal-03763120

HAL Id: hal-03763120

<https://ut3-toulouseinp.hal.science/hal-03763120v1>

Submitted on 29 Aug 2022

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution 4.0 International License



CMLs control host-plant interactions

Christian Mazars^{1,2*}, Benoit Ranty^{1,2}, Didier Aldon^{1,2}, Ralf Oelmüller³, Jean-Philippe Gaulaud^{1,2} and Axel Mithöfer⁴

¹Université de Toulouse, UPS, UMR 5546, Laboratoire de Recherche en Sciences Végétales, BP 42617, F-31326, Castanet-Tolosan, France; ²CNRS, UMR 5546, BP 42617, F-31326, Castanet-Tolosan, France; ³Friedrich-Schiller-Universität Jena, Institut für Allgemeine Botanik und Pflanzenphysiologie, Dornburgerstr. 159, D-07743 Jena, Germany; ⁴Max-Planck-Institute for Chemical Ecology, Bioorganic Chemistry Department, Hans-Knöll-Straße 8, D-07745 Jena, Germany; *correspondence to: mazars@lrsv.ups-tlse.fr

In the perspective of world-wide climate changes that likely will affect the outcomes of plant interactions with pathogens, pests and symbionts, the contribution of a certain class of plant-specific calcium sensors, referred to as calmodulin-like proteins (CMLs), will play an important role in the future. Functional, genetic and biotechnological approaches in the model plant *Arabidopsis* and, subsequently as a proof of concept, in agricultural important plant species will be necessary to elucidate their function. Several members of CMLs may act as key players in both abiotic and biotic responses including plant/microbe and plant/herbivory interactions. Interestingly, using gain and loss of function strategies, these interactions can be investigated and manipulated by modulating CMLs expression in model plants. It will be important to understand how CMLs translate intracellular calcium signatures generated by various stimuli from pathogenic and beneficial microbes or pests into appropriate biological responses through the regulation of downstream components. The comprehensive analyses of CMLs will first improve our knowledge on Ca²⁺/CML-mediated signaling pathways but will also facilitate their application in future plant-breeding programs in order to develop highly productive crops.

Journal of Endocytobiosis and Cell Research (2016) 27(1):13-19

Category: Mini Review

Keywords: CMLs, calcium

Accepted: 10 November 2015

Introduction

Global climate changes (increased temperatures, CO₂ and O₃ levels, drought or raining periods, *etc.*) will have strong influences on plant performance and plant-host interactions, and will generate novel communities composed of new combinations of species (Lurgi *et al.* 2012). Elevated CO₂ levels

in the atmosphere result in higher CO₂ fixation rates, biomass production and carbohydrate metabolism in plants, and consequently will impact the microbial communities associated with them. The altered plant metabolism influence the biotic interactions, local species distribution (Buckley 2013, HilleRisLambers *et al.* 2013), the structure and function of roots, the diversity of rhizobial communities (Rajkumar *et al.* 2013), and the defense capacity of the plants against biotic and abiotic threats. Various models predict that long-term changes of the biological interactions of plants with their environment as well as their microbial communities have both positive and negative impacts on ecological systems, forestry and agricultural productivity. For example, a recent meta-analysis of more than 400 studies has shown that an increase in microbial and plant biomass in response to global changes should result in an alleviation of drought stress responses in both kingdoms (Kivlin *et al.* 2013). Thus, global climate warming will have unpredictable long-term consequences for crop yields, and the available studies predict predominantly negative effects what will have impacts on food productivity and security in the future. The global food production should increase by 50% to satisfy the world's population demand by 2050, but this goal appears to be difficult to achieve. Indeed, a recent review points out that crop losses due to fungal or oomycete infections, abiotic stress, injuries to plants and other pests will increase in the future due to the expected climate changes (Fisher *et al.* 2012). Selection of suitable crop plants for enhanced yield production and better tolerance to multi-stress conditions is a major task for a future world with an altered global climate (Rajkumar *et al.* 2013). These few findings highlight the importance of plant-host interactions in the context of food production and stability of ecosystems in a changing climate environment, and force scientists to (1) understand the climate change-induced effects on biological interactions at the scientific level (Weckwerth 2011), (2) decipher and enhance resistance mechanisms against pests and pathogens to preserve crop yields, and (3) to develop strategies which stabilize or even promote beneficial symbiotic plant-microbe interaction in a future world with a global climate change.

The calcium (Ca²⁺) toolkit include CMLs

Among the multiple possibilities to thwart the climate change-induced effects on biological interactions and plant cell signaling, investigations of the plant «calcium (Ca²⁺) toolkit» are highly promising because they take part in one of the earliest events occurring in response to environmental changes, namely the free cellular Ca²⁺ increases.

These Ca²⁺ increases initiate appropriate signaling pathways leading to specific adaptive responses according the nature and strength of the stimulus or environmental modification perceived by the plant (Hetherington and Brownlee

2004; Hong-Bo et al. 2008; Lecourieux et al. 2006; Poovaiah and Reddy 1993; Rudd and Franklin-Tong 1999; Sanders et al. 2002; Sarwat et al. 2013). In the pathogenic interaction between *Arabidopsis thaliana* and *Pseudomonas syringae*, such Ca²⁺ signatures are different according to whether a plant has been challenged by a virulent or avirulent strain (Grant et al. 2000). These differences in the Ca²⁺ signatures contribute to initiate a complex signaling network leading to either susceptibility (virulent strain) or defense (avirulent strain). Indeed, if these Ca²⁺ signatures have been proven by their spatio-temporal properties to encode a first layer of specificity (McAinsh and Pittman 2009; Ng and McAinsh 2003), they need to be decoded by downstream Ca²⁺-binding proteins possessing the canonical Ca²⁺-binding motif called EF-hand (Nakayama and Kretsinger 1994). The *Arabidopsis* genome encodes at least 250 EF-hand proteins, which is a much higher number than in mammals, arguing for higher importance of these class of proteins in plants (Day et al. 2002). These Ca²⁺ sensors can be classified into 4 major groups, the calcineurin-B like, the calcium-dependent kinases, the calmodulin group and a closely related group, the calmodulin-like protein (CML) family, which is specific to plants and encompasses the highest number of members (50 in *Arabidopsis*; Figure 1). The sequence similarity of these Ca²⁺ sensors with the evolutionary conserved AtCaM2 (CALMODULIN2) varies from 16% to 74.5% (McCormack et al. 2005) and the CML family members contain between one to six EF-hands. Data currently available for CML members from different plant species indicate that they participate in development but also in stress responses (cf. references in Table 1). For instance, CML9 acts as a negative regulator of drought stress showing that *cml9* mutants are able to withstand water deficit and achieve their full life cycle (Magnan et al. 2008). On the contrary, the rice CML OsMSR2 behaves as a positive regulator of drought and salinity responses (Xu et al. 2011). Overexpression of this CML in *Arabidopsis* results in enhanced tolerance to drought and salinity and increased sensitivity to exogenous ABA (Xu et al. 2011).

It has also been reported that others CMLs are responsive to different abiotic stress such as the gene CML8 which is induced by salicylic acid and NaCl (Park et al. 2010), CML24 whose transcripts level increase in response to high or low temperature (Delk et al. 2005) and CML18 reported to be involved in salt stress signaling through its interaction with the Na⁺/H⁺ antiporter AtNHX1 (Yamaguchi et al. 2005).

CMLs also contribute to several aspects of plant development including CML42 involved in trichome branching (Dobney et al. 2009), CML25 and -7 in root hair elongation (Lin et al. 2011; Won et al. 2009), CML39 in early seedling establishment (Bender et al. 2013), CML23 and -24 in flowering (Tsai et al. 2007), CML24 in regulating pollen tube growth by modulating the actin cytoskeleton and controlling the cytosolic Ca²⁺ concentration (Yang et al. 2014) and CML20 regulating the cortical microtubule organization (Azimzadeh et al. 2008).

In addition, several CMLs have also been reported to participate in plant-host interactions. CML9, a stress-responsive member of the CML family in *Arabidopsis* previously shown to be a negative regulator of plant tolerance to drought (Magnan et al. 2008) is also a positive regulator of salicylic acid-dependent defense responses against the bacterial pathogen *P. syringae* pv tomato (Pst) DC 3000 (Leba et al. 2012). The fact that CML9 is involved both in abiotic and biotic stresses indicate that CMLs can be at the crossroads of development, biotic and abiotic stress signaling pathways

(Leba et al. 2012). Thus, as like CML9, CML24 which was already described to be involved in abiotic stress responses (Delk et al. 2005) takes also part in innate immune responses including the hypersensitive response (Ma et al. 2008), and both CML9 and CML24 play a role in the beneficial interaction of the endophyte *Piriformospora indica* with *Arabidopsis* roots in addition to two others CMLs, CML37 and CML43 (Bender and Snedden 2013; Yang et al. 2014). Expression of three of the above cited CML genes (CML42, CML37 and CML9), is also induced by a recently identified MAMP exuded by *Mortierella hyalina* which triggers cytoplasmic Ca²⁺ elevation (R. Oelmüller, unpublished data).

Interestingly, if we consider a third type of interaction concerning pests, *Arabidopsis* defense mechanisms in response to attacks by herbivorous insects involve also Ca²⁺ signaling. During these interactions, at least eight CML genes appeared to be up-regulated by treatments with oral secretions (OS) from the herbivorous lepidopteran insect *Spodoptera littoralis* (Scholz et al. 2014; Vadassery et al. 2012b). Among them, again CML42 and CML37 are involved but act antagonistically: while CML42 acts as a negative regulator of plant defense against herbivorous insects, CML37 acts as a positive regulator (Scholz et al. 2014; Vadassery et al. 2012a; Vadassery et al. 2012b). The latter CML is connecting Ca²⁺ and jasmonate (JA) signaling by affecting the synthesis of jasmonic acid-isoleucine (Scholz et al. 2014). Interestingly CML37 and CML42 also act as positive and negative regulators on drought stress (Vadassery et al. 2012a; Scholz et al. 2015).

It is clear from literature that most of the above mentioned CMLs participate in growth regulation and developmental processes, in addition to their involvement in defense/adaptation processes. This pleiotropic role makes them interesting candidates to understand how calcium can control the final adaptive responses. As shown above, some of them are simultaneously involved in defense against pathogens and in beneficial interactions such as the symbiotic interaction which reinforces the plant defenses against environmental stress. Indeed, we and others have shown that symbiosis strongly promotes plant performance (which results in higher biomass and seed production), resistance to biotic and abiotic stress (Sun et al. 2014), and nutrient uptake, in particular phosphate, nitrate and sulfate (Camehl et al. 2011; Vadassery et al. 2009) by the host. The beneficial effects of *P. indica* in *Arabidopsis*, rice, barley and Chinese cabbage are strongly correlated with the promotion of lateral root development, which – in turn – increases with decrease in the phosphate concentration in the medium (Dong et al. 2013; Lee et al. 2011). In the inoculated plants, the root phenotype becomes bushier in the presence of the fungus. Genetic analyses demonstrated that benefits for the plants during the symbiosis with *P. indica* are associated with cytoplasmic Ca²⁺ elevation which stimulates the expression of a specific subset of CML genes in *Arabidopsis* root (Vadassery and Oelmüller 2009, and unpublished data). Interestingly, in another beneficial symbiotic interaction system, the basidiomycete *M. hyalina* also promotes *Arabidopsis* performance when the fungus colonizes the roots. Strong up-regulation of the CML42, CML37 and CML9 mRNA levels both in roots and shoots suggests an important role of these proteins in the symbiotic interaction. Overall, scientific evidence makes CMLs interesting targets to develop new concepts to improve plant defense and performance which are worth to be investigated.

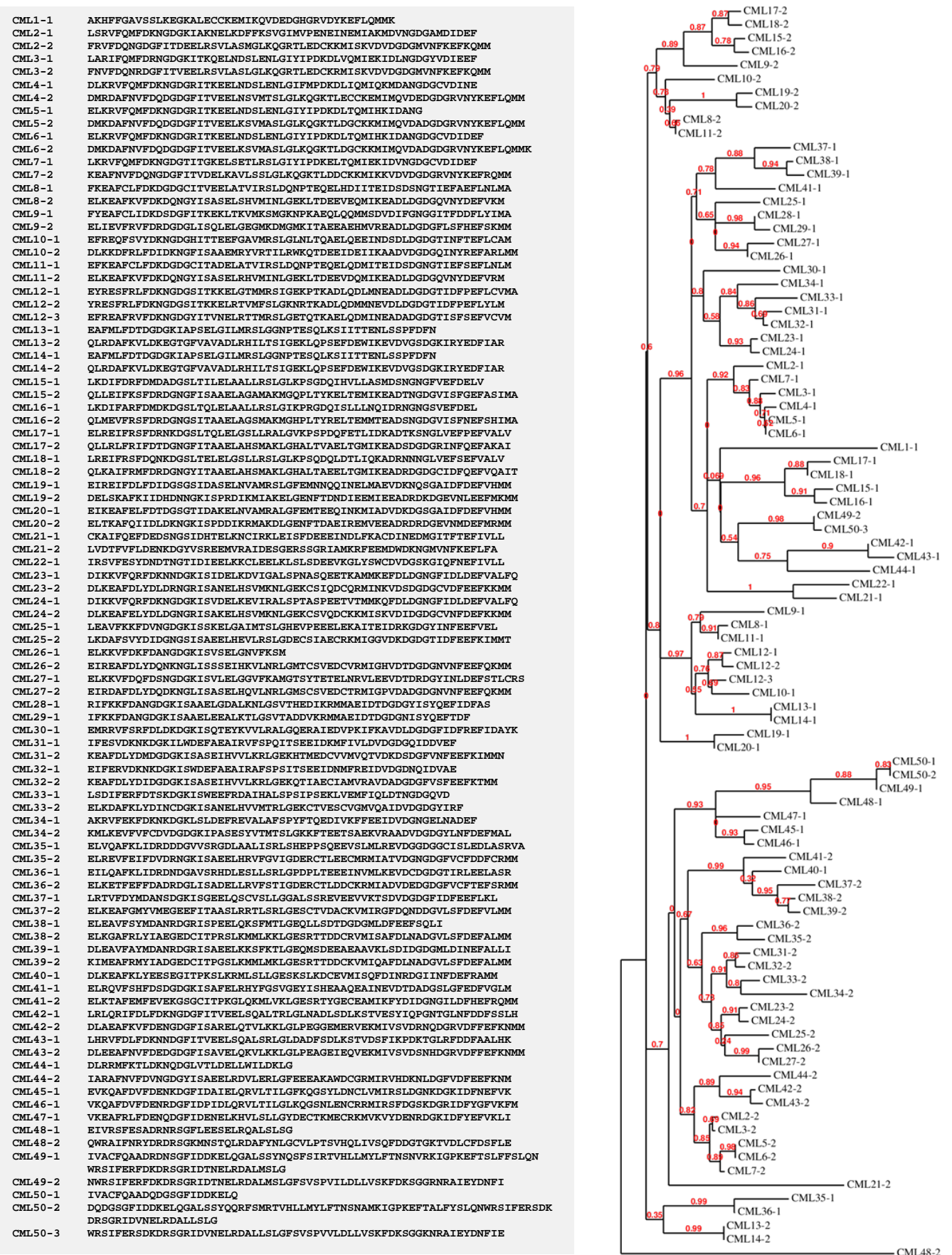


Figure 1: Phylogenetic tree built with the EF-hand motifs found in CML from Arabidopsis. (left) amino acid sequence covering EF hand pairs are shown (prediction according to PROTEIN BLAST). The first number after CML indicates the CML corresponding protein (from 1 to 50 in Arabidopsis) and the second number (1, 2 or 3) the EF-hand pair. As reported by Zhu et al. (2015), CML1 is certainly a pseudogene and cannot be considered as a CML but was conserved for the phylogenetic analysis. (right) Phylogenetic tree of the sequences.

Ideas for future strategies

The role of CMLs in deleterious and beneficial interactions of Arabidopsis with plant pathogens, beneficial microbes and herbivores needs to be investigated in greater details. Indeed, CMLs take part in multiple interactions between Arabidopsis and bacterial [*P. syringae* (Grant et al. 2000)] and fungal pathogens such as *Alternaria brassicae* (Michal Johnson et al. 2014) and *Verticillium dahliae* (Sun et al. 2014), the herbivorous insect *S. littoralis* (Vadassery et al. 2012b) and the root-colonizing symbiotic fungi *P. indica* (Vadassery et al. 2009) and *M. hyalina* (Ludwig and Oelmüller, unpublished). The aims should be to identify (a) the CMLs involved in the decoding of the Ca²⁺ signatures induced in the above mentioned model systems, and (b) the downstream signaling components (CMLs targets and their downstream components and effectors) required for the physiological stress adaptive responses leading to resistance (bacteria, insects) or improved symbiosis (*P. indica* and *M. hyalina*).

In each of the interactions, the participation of Ca²⁺ variations and CMLs has already been shown. In some cases, CMLs were identified as downstream components of Ca²⁺ variations and upstream of regulatory hormonal pathways [SA pathway in the case of *P. syringae* (Leba et al. 2012), JA pathway in the case of *S. littoralis* (Scholz et al. 2014), and putatively auxin in the case of *P. indica* (Lee et al. 2011)]. As outlined above, CMLs appear as versatile tools to integrate at the cellular levels diverse inputs (i.e. stress, pathogen attacks, growth and developmental programs). This raises the

question of their contribution in a single or several signaling pathways. Since they do not have by themselves intrinsic activities they have to play a role by modulating the activity of their downstream targets.

This finding demonstrates that CMLs play an essential and so far little recognized role in various biotic interactions of plants. Their involvement in both pathogenic and beneficial interactions may allow them to balance plant defense responses and beneficial developmental strategies. The trade-off between defense and growth is probably central and often irreversible for the plant and will have a strong influence on its performance and therefore also for agricultural yields. Investment in beneficial symbiotic interactions will stimulate growth and development, which is consistent with the observation that CMLs also participate in these regulatory processes. Therefore, the future goal should be to improve our knowledge about the role of CMLs as Ca²⁺ sensors, considering that pathogenic and beneficial plant interactions that form our ecosystems are important determinants for the agricultural productivity and breeding strategies. The central role of CMLs for both defense and developmental strategies makes them interesting candidates in plant breeding programs. The ultimate goal is to increase resistance to pests and pathogens by down-regulating interactions with these deleterious hosts and/or by increasing plant-endosymbiotic interactions. Both reflect processes in which CMLs are involved at central signaling positions and known to be linked to each other (Sun et al. 2014).

Table 1: Arabidopsis CML proteins and related publications for their gene expression or/and functions.

Gene	Name	Publications
At3g59450	CML1	-
At4g12860	CML2	Wang et al. 2008; Pagnussat et al. 2005
At3g07490	CML3	Dolze et al. 2013; Chigri et al. 2012
At3g59440	CML4	Che et al. 2006; Lee et al. 2005a
At2g43290	CML5	Lee et al. 2005a; Lee et al. 2005b; Goda et al. 2004; Gepstein et al. 2003
At4g03290	CML6	Lee et al. 2005a; Bae et al. 2003; Wang et al. 2008; Becker et al. 2003; Armengaud et al. 2004
At1g05990	CML7	Lee et al. 2005a; Lin et al. 2011; Won et al. 2009
At4g14640	CML8	Hartmann et al. 2014; Park et al. 2010
At3g51920	CML9	Hartmann et al. 2014; Fischer et al. 2013; Leba et al. 2012a; Leba et al. 2012b; Vadassery et al. 2012; Magnan et al. 2008
At2g41090	CML10	Cho et al. 2015; Franco-Zorilla et al. 2005
At3g22930	CML11	Reddy et al. 2004; Oh et al. 2012
At2g41100	CML12	Antosiewicz et al. 1995; Ito et al. 1995; Rairdan et al. 2001; Wright et al. 2002; Benjamins et al. 2003; Gleeson et al. 2012; Cazzonelli et al. 2014
At1g12310	CML13	-
At1g62820	CML14	-
At1g18530	CML15	-
At3g25600	CML16	-
At1g32250	CML17	-
At3g03000	CML18	Yamaguchi et al. 2005
At4g37010	CML19	Liang et al. 2006; Azimzadeh et al. 2008
At3g50360	CML20	Liang et al. 2006; Azimzadeh et al. 2008
At4g26470	CML21	-
At3g24110	CML22	-
At1g66400	CML23	Tsai et al. 2007
At5g37770	CML24	Tsai et al. 2007; Yang et al. 2014; Ma et al., 2014; Abu-Abied et al. 2006; Delk et al. 2005
At1g24620	CML25	Wang et al. 2015; Lin et al. 2011; Won et al. 2009; Bae et al. 2003; Lee et al. 2005
At1g73630	CML26	Lee et al. 2005; Wang et al. 2008; Zhou et al. 2009
At1g18210	CML27	Lee et al. 2005; Hampton et al. 2004; Armengaud et al. 2004; Carter et al. 2004; Gepstein et al. 2003
At3g03430	CML28	Zhou et al. 2009; Wang et al. 2008; Lee et al. 2005
At5g17480	CML29	Rozwadowski et al. 1999; Zhou et al. 2009; Wang et al. 2008; Lee et al. 2005

At2g15680	CML30	Chigri et al. 2012; Lee et al. 2005
At2g36180	CML31	Lee et al. 2005
At5g17470	CML32	Lee et al. 2005
At3g03400	CML33	Wuest et al. 2010; Lee et al. 2005
At3g03410	CML34	Lee et al. 2005
At2g41410	CML35	Ascencio-Ibáñez et al. 2008; Ma et al. 2007; Wang et al. 2008; Peng et al. 2007; Lee et al. 2005
At3g10190	CML36	Le et al. 2014; Lee et al. 2005; Keates et al. 2003
At5g42380	CML37	Scholz et al. 2014; Scholz et al. 2015
At1g76650	CML38	Lee et al. 2005; Kim et al. 2008; Abercrombie et al. 2008; Marri et al. 2005
At1g76640	CML39	Bender et al. 2013; Vanderbeld et al. 2007
At3g01830	CML40	Ascencio-Ibáñez et al. 2008
At3g50770	CML41	Naydenov et al. 2015
At4g20780	CML42	Vadassery et al. 2012a; Dobney et al. 2009
At5g44460	CML43	Bender et al. 2014; Chiasson et al. 2005
At1g21550	CML44	Lee et al. 2005
At3g29000	CML45	Cho et al. 2015
At5g39670	CML46	Magnan et al. 2008
At3g47480	CML47	-
At2g27480	CML48	Abu-Abied et al. 2006
At3g10300	CML49	-
At5g04170	CML50	-

References

- Abercrombie JM, Halfhill MD, Ranjan P, Rao MR, Saxton AM, Yuan JS, Stewart CN Jr. (2008) Transcriptional responses of *Arabidopsis thaliana* plants to As (V) stress. *BMC Plant Biol.* 8:87.
- Abu-Abied M, Golomb L, Belausov E, Huang S, Geiger B, Kam Z, Staiger CJ, Sadot E. (2006) Identification of plant cytoskeleton-interacting proteins by screening for actin stress fiber association in mammalian fibroblasts. *Plant J.* 48:367-79.
- Antosiewicz DM, Polisensky DH, Braam J. (1995) Cellular localization of the Ca²⁺ binding TCH3 protein of *Arabidopsis*. *Plant J.* 8:623-36.
- Armengaud P1, Breitling R, Amtmann A. (2004) The potassium-dependent transcriptome of *Arabidopsis* reveals a prominent role of jasmonic acid in nutrient signaling. *Plant Physiol.* 136:2556-76.
- Ascencio-Ibáñez JT, Sozzani R, Lee TJ, Chu TM, Wolfinger RD, Cella R, Hanley-Bowdoin L. (2008) Global analysis of *Arabidopsis* gene expression uncovers a complex array of changes impacting pathogen response and cell cycle during geminivirus infection. *Plant Physiol.* 148:436-54.
- Azimzadeh J, Nacry P, Christodoulidou A, Drevensek S, Camilleri C, Amieur N, Parcy F, Pastuglia M, Bouchez D. (2008) *Arabidopsis* TONNEAU1 proteins are essential for preprophase band formation and interact with centrin. *Plant Cell.* 20:2146-2159.
- Bae MS, Cho EJ, Choi EY, Park OK. (2003) Analysis of the *Arabidopsis* nuclear proteome and its response to cold stress. *Plant J.* 36:652-63.
- Becker JD, Boavida LC, Carneiro J, Haury M, Feijó JA. (2003) Transcriptional profiling of *Arabidopsis* tissues reveals the unique characteristics of the pollen transcriptome. *Plant Physiol.* 133:713-25.
- Bender KW, Dobney S, Ogunrinde A, Chiasson D, Mullen RT, Teresinski HJ, Singh P, Munro K, Smith SP, Snedden WA. (2014) The calmodulin-like protein CML43 functions as a salicylic-acid-inducible root-specific Ca²⁺ sensor in *Arabidopsis*. *Biochem J.* 457:127-36.
- Bender KW, Rosenbaum DM, Vanderbeld B, Ubaid M, Snedden WA. (2013) The *Arabidopsis* calmodulin-like protein, CML39, functions during early seedling establishment. *Plant J.* 76:634-647.
- Benjamins R, Ampudia CS, Hooykaas PJ, Offringa R. (2003) PINOID-mediated signaling involves calcium-binding proteins. *Plant Physiol.* 132:1623-30.
- Buckley LB. (2013) Get real: putting models of climate change and species interactions in practice. *Ann NY Acad Sci.* 1297:126-138.
- Camehl I, Drzewiecki C, Vadassery J, Shahollari B, Sherameti I, Forzani C, Munnik T, Hirt H, Oelmüller R. (2011) The OX11 kinase pathway mediates *Piriformospora indica*-induced growth promotion in *Arabidopsis*. *PLoS Pathog.* 7:e1002051.
- Carter C, Pan S, Zouhar J, Avila EL, Girke T, Raikhel NV. (2004) The vegetative vacuole proteome of *Arabidopsis thaliana* reveals predicted and unexpected proteins. *Plant Cell.* 16:3285-303.
- Cazzonelli CI, Nisar N, Roberts AC, Murray KD, Borevitz JO, Pogson BJ. (2014) A chromatin modifying enzyme, SDG8, is involved in morphological, gene expression, and epigenetic responses to mechanical stimulation. *Front Plant Sci.* 5:533.
- Che P, Lall S, Nettleton D, Howell SH. (2006) Gene expression programs during shoot, root, and callus development in *Arabidopsis* tissue culture. *Plant Physiol.* 141:620-37.
- Chiasson D, Ekengren SK, Martin GB, Dobney SL, Snedden WA. (2005) Calmodulin-like proteins from *Arabidopsis* and tomato are involved in host defense against *Pseudomonas syringae* pv. tomato. *Plant Mol Biol.* 58:887-97.
- Chigri F, Flsdorff S, Pilz S, Kölle E, Dolze E, Gietl C, Vothknecht UC. (2012) The *Arabidopsis* calmodulin-like proteins AtCML30 and AtCML3 are targeted to mitochondria and peroxisomes, respectively. *Plant Mol Biol.* 78:211-22.
- Cho KM, Nguyen HT, Kim SY, Shin JS, Cho DH, Hong SB, Shin JS, Ok SH. (2015) CML10, a variant of calmodulin, modulates ascorbic acid synthesis. *New Phytol.* doi:10.1111/nph.13612.
- Day IS, Reddy VS, Shad Ali G, Reddy AS. (2002) Analysis of EF-hand-containing proteins in *Arabidopsis*. *Genome Biol.* 3:RESEARCH0056.
- Delk NA, Johnson KA, Chowdhury NI, Braam J. (2005) CML24, regulated in expression by diverse stimuli, encodes a potential Ca²⁺ sensor that functions in responses to abscisic acid, daylength, and ion stress. *Plant Physiol.* 139:240-253.
- Dobney S, Chiasson D, Lam P, Smith SP, Snedden WA. (2009) The calmodulin-related calcium sensor CML42 plays a role in trichome branching. *J Biol Chem.* 284:31647-31657.
- Dolze E, Chigri F, Höwing T, Hierl G, Isono E, Vothknecht UC, Gietl C. (2013) Calmodulin-like protein AtCML3 mediates dimerization of peroxisomal processing protease AtDEG15 and contributes to normal peroxisome metabolism. *Plant Mol Biol.* 83:607-24.
- Dong S, Tian Z, Chen PJ, Senthil Kumar R, Shen CH, Cai D, Oelmüller R, Yeh KW. (2013) The maturation zone is an important target of *Piriformospora indica* in Chinese cabbage roots. *J Exp Bot.* 64:4529-4540.
- Fischer C, Kugler A, Hoth S, Dietrich P. (2013) An IQ domain mediates the interaction with calmodulin in a plant cyclic nucleotide-gated channel. *Plant Cell Physiol.* 54:573-84.
- Fisher MC, Henk DA, Briggs CJ, Brownstein JS, Madoff LC, McCraw SL, Gurr SJ. (2012) Emerging fungal threats to animal, plant and

- ecosystem health. *Nature*. 484:186-194.
- Franco-Zorrilla JM, Martín AC, Leyva A, Paz-Ares J. (2005) Interaction between phosphate-starvation, sugar, and cytokinin signaling in Arabidopsis and the roles of cytokinin receptors CRE1/AHK4 and AHK3. *Plant Physiol*. 138:847-57.
- Gepstein S, Sabehi G, Carp MJ, Hajouj T, Neshar MF, Yariv I, Dor C, Bassani M. (2003) Large-scale identification of leaf senescence-associated genes. *Plant J*. 36:629-42.
- Gleeson L, Squires S, Bisgrove SR. (2012) The microtubule associated protein END BINDING 1 represses root responses to mechanical cues. *Plant Sci*. 187:1-9.
- Goda H, Sawa S, Asami T, Fujioka S, Shimada Y, Yoshida S. (2004) Comprehensive comparison of auxin-regulated and brassinosteroid-regulated genes in Arabidopsis. *Plant Physiol*. 134:1555-73.
- Grant M, Brown I, Adams S, Knight M, Ainslie A, Mansfield J. (2000) The *RPM1* plant disease resistance gene facilitates a rapid and sustained increase in cytosolic calcium that is necessary for the oxidative burst and hypersensitive cell death. *Plant J*. 23:441-450.
- Hampton CR, Bowen HC, Broadley MR, Hammond JP, Mead A, Payne KA, Pritchard J, White PJ. (2004) Cesium toxicity in Arabidopsis. *Plant Physiol*. 136:3824-37.
- Hartmann J, Fischer C, Dietrich P, Sauter M. (2014) Kinase activity and calmodulin binding are essential for growth signaling by the phytosulfokine receptor PSKR1. *Plant J*. 78:192-202.
- Hetherington AM, Brownlee C. (2004) The generation of Ca²⁺ signals in plants. *Annu Rev Plant Biol*. 55:401-427.
- HilleRisLambers J, Harsch MA, Ettinger AK, Ford KR, Theobald EJ. (2013) How will biotic interactions influence climate change-induced range shifts? *Ann NY Acad Sci*, 1297:112-125.
- Hong-Bo S, Li-Ye C, Ming-An S. (2008) Calcium as a versatile plant signal transducer under soil water stress. *BioEssays*. 30:634-641.
- Hruz T, Laule O, Szabo G, Wessendorp F, Bleuler S, Oertle L, Widmayer P, Gruissem W, Zimmermann P. (2008) Genevestigator v3: a reference expression database for the meta-analysis of transcriptomes. *Adv Bioinformatics*. 420747. doi:10.1155/2008/420747
- Ito T, Hirano M, Akama K, Shimura Y, Okada K. (1995) Touch-inducible genes for calmodulin and a calmodulin-related protein are located in tandem on a chromosome of *Arabidopsis thaliana*. *Plant Cell Physiol*. 36(7):1369-73.
- Johnson MJ, Reichelt M, Vadassery J, Gershenzon J, Oelmüller R. (2014) An Arabidopsis mutant impaired in intracellular calcium elevation is sensitive to biotic and abiotic stress. *BMC Plant Biol*, 14:162.
- Keates SE, Kostman TA, Anderson JD, Bailey BA. (2003) Altered gene expression in three plant species in response to treatment with Nep1, a fungal protein that causes necrosis. *Plant Physiol*. 132(3):1610-22.
- Kim CY, Bove J, Assmann SM. (2008) Overexpression of wound-responsive RNA-binding proteins induces leaf senescence and hypersensitive-like cell death. *New Phytol*. 180(1):57-70.
- Kivlin SN, Emery SM, Rudgers JA. (2013) Fungal symbionts alter plant responses to global change. *Am J Bot*. 100:1445-1457.
- Le MH, Cao Y, Zhang XC, Stacey G. (2014) LIK1, a CERK1-interacting kinase, regulates plant immune responses in Arabidopsis. *PLoS One*. 9(7):e102245.
- Leba LJ, Cheval C, Ortiz-Martin I, Ranty B, Beuzon CR, Galaud JP, Aldon D. (2012a) CML9, an Arabidopsis calmodulin-like protein, contributes to plant innate immunity through a flagellin-dependent signalling pathway. *Plant J*. 71:76-989.
- Leba LJ, Perochon A, Cheval C, Ranty B, Galaud JP, Aldon D. (2012b) CML9, a multifunctional *Arabidopsis thaliana* calmodulin-like protein involved in stress responses and plant growth? *Plant Signal Behav*. 7(9):1121-4.
- Lecourieux D, Ranjeva R, Pugin A. (2006) Calcium in plant defence-signalling pathways. *New Phytol*. 171:249-269.
- Lee D, Polisensky DH, Braam J. (2005a) Genome-wide identification of touch- and darkness-regulated Arabidopsis genes: a focus on calmodulin-like and XTH genes. *New Phytol*. 165(2):429-44.
- Lee BH, Henderson DA, Zhu JK. (2005b) The Arabidopsis cold-responsive transcriptome and its regulation by ICE1. *Plant Cell*. 17(11):3155-75.
- Lee YC, Johnson JM, Chien CT, Sun C, Cai D, Lou B, Oelmüller R, Yeh KW. (2011) Growth promotion of Chinese cabbage and Arabidopsis by *Piriformospora indica* is not stimulated by mycelium-synthesized auxin. *Mol Plant-Microbe Interact*. 24:421-431.
- Liang L, Flury S, Kalck V, Hohn B, Molinier J. (2006) CENTRIN2 interacts with the Arabidopsis homolog of the human XPC protein (AtRAD4) and contributes to efficient synthesis-dependent repair of bulky DNA lesions. *Plant Mol Biol*. 61(1-2):345-56.
- Lin WD, Liao YY, Yang TJ, Pan CY, Buckhout TJ, Schmidt W. (2011) Coexpression-based clustering of Arabidopsis root genes predicts functional modules in early phosphate deficiency signaling. *Plant Physiol*. 155:1383-1402.
- Lurgi M, Lopez BC, Montoya JM. (2012) Novel communities from climate change. *Philos Trans R Soc Lond B Biol Sci*. 367:2913-2922.
- Ma W, Smigel A, Tsai YC, Braam J, Berkowitz GA. (2008) Innate immunity signaling: cytosolic Ca²⁺ elevation is linked to downstream nitric oxide generation through the action of calmodulin or a calmodulin-like protein. *Plant Physiol*. 148(2):818-28.
- Magnan F, Ranty B, Charpenteau M, Sotta B, Galaud JP, Aldon D. (2008) Mutations in AtCML9, a calmodulin-like protein from *Arabidopsis thaliana*, alter plant responses to abiotic stress and abscisic acid. *Plant J*. 56:575-589.
- Marri L, Trost P, Pupillo P, Sparla F. (2005) Reconstitution and properties of the recombinant glyceraldehyde-3-phosphate dehydrogenase/CP12/phosphoribulokinase supramolecular complex of Arabidopsis. *Plant Physiol*. 139(3):1433-43.
- McAinsh MR, Pittman JK. (2009) Shaping the calcium signature. *New Phytol*. 181:275-294.
- McCormack E, Tsai YC, Braam J. (2005) Handling calcium signaling: Arabidopsis CaMs and CMLs. *Trends Plant Sci*, 10:383-389.
- Mithöfer A, Mazars C, Maffei ME. (2009) Probing spatio-temporal intracellular calcium variations in plants. *Methods Mol Biol*. 479:79-92.
- Nakayama S, Kretsinger RH. (1994) Evolution of the EF-hand family of proteins. *Annu Rev Biophys Biomol Struct*. 23:473-507.
- Naydenov M, Baev V, Apostolova E, Gospodinova N, Sablok G, Gozmanova M, Yahubyan G. (2015) High-temperature effect on genes engaged in DNA methylation and affected by DNA methylation in Arabidopsis. *Plant Physiol Biochem*. 87:102-8.
- Ng CK, McAinsh MR. (2003) Encoding specificity in plant calcium signalling: hot-spotting the ups and downs and waves. *Ann Bot*. 92:477-485.
- Oh MH, Kim HS, Wu X, Clouse SD, Zielinski RE, Huber SC. (2012) Calcium/calmodulin inhibition of the Arabidopsis BRASSINOSTEROID-INSENSITIVE 1 receptor kinase provides a possible link between calcium and brassinosteroid signalling. *Biochem J*. 443(2):515-23.
- Pagnussat GC, Yu HJ, Ngo QA, Rajani S, Mayalagu S, Johnson CS, Capron A, Xie LF, Ye D, Sundaresan V. (2005) Genetic and molecular identification of genes required for female gametophyte development and function in Arabidopsis. *Development*. 132(3):603-14.
- Park HC, Park CY, Koo SC, Cheong MS, Kim KE, Kim MC, Lim CO, Lee SY, Yun DJ, Chung WS. (2010) AtCML8, a calmodulin-like protein, differentially activating CaM-dependent enzymes in *Arabidopsis thaliana*. *Plant Cell Rep*. 29:1297-1304.
- Peng M, Bi YM, Zhu T, Rothstein SJ. (2007) Genome-wide analysis of Arabidopsis responsive transcriptome to nitrogen limitation and its regulation by the ubiquitin ligase gene *NLA*. *Plant Mol Biol*. 65(6):775-97.
- Perochon A, Aldon D, Galaud JP, Ranty B. (2011) Calmodulin and calmodulin-like proteins in plant calcium signaling. *Biochimie*. 93:2048-2053.
- Perruc E, Charpenteau M, Ramirez BC, Jauneau A, Galaud JP, Ranjeva R, Ranty B. (2004) A novel calmodulin-binding protein functions as a negative regulator of osmotic stress tolerance in *Arabidopsis thaliana* seedlings. *Plant J*. 38:410-420.
- Poovaiah BW, Reddy AS. (1993) Calcium and signal transduction in plants. *CRC Crit Rev Plant Sci*. 12:185-211.
- Rairdan GJ, Donofrio NM, Delaney TP. (2001) Salicylic acid and *NIM1/NPR1*-independent gene induction by incompatible *Peronospora parasitica* in Arabidopsis. *Mol Plant-Microbe Interact*. 14(10):1235-46.

- Rajkumar M, Prasad MN, Swaminathan S, Freitas H. (2013) Climate change driven plant-metal-microbe interactions. *Environ Int.* 53:74-86.
- Ranty B, Aldon D, Galaud JP. (2006) Plant calmodulins and calmodulin-related proteins: multifaceted relays to decode calcium signals. *Plant Signal Behav.* 1:96-104.
- Reddy VS, Day IS, Thomas T, Reddy AS. (2004) KIC, a novel Ca²⁺ binding protein with one EF-hand motif, interacts with a microtubule motor protein and regulates trichome morphogenesis. *Plant Cell.* 16(1):185-200.
- Rozwadowski K, Zhao R, Jackman L, Huebert T, Burkhart WE, Hemmingsen SM, Greenwood J, Rothstein SJ. (1999) Characterization and immunolocalization of a cytosolic calcium-binding protein from *Brassica napus* and *Arabidopsis* pollen. *Plant Physiol.* 120(3):787-98.
- Rudd JJ, Franklin-Tong VE. (1999) Calcium signaling in plants. *Cell Mol Life Sci.* 55:214-232.
- Sanders D, Pelloux J, Brownlee C, Harper JF. (2002) Calcium at the crossroads of signaling. *Plant Cell.* 14 Suppl:S401-17.
- Sarwat M, Ahmad P, Nabi G, Hu X. (2013) Ca²⁺ signals: the versatile decoders of environmental cues. *Crit Rev Biotechnol.* 33:97-109.
- Scholz SS, Reichelt M, Vadassery J, Mithöfer A. (2015) Calmodulin-like protein CML37 is a positive regulator of ABA during drought stress in *Arabidopsis*. *Plant Signal Behav.* 10(6):e1011951.
- Scholz SS, Vadassery J, Heyer M, Reichelt M, Bender KW, Snedden WA, Boland W, Mithöfer A. (2014) Mutation of the *Arabidopsis* Calmodulin-Like Protein CML37 deregulates the jasmonate pathway and enhances susceptibility to herbivory. *Mol Plant.* 7:1712-1726.
- Sun C, Shao Y, Vahabi K, Lu J, Bhattacharya S, Dong S, Yeh KW, Sherameti I, Lou B, Baldwin IT, Oelmüller R. (2014) The beneficial fungus *Piriformospora indica* protects *Arabidopsis* from *Verticillium dahliae* infection by downregulation plant defense responses. *BMC Plant Biol.* 14:268.
- Tsai YC, Delk NA, Chowdhury NI, Braam J. (2007) *Arabidopsis* potential calcium sensors regulate nitric oxide levels and the transition to flowering. *Plant Signal Behav.* 2:446-454.
- Vadassery J, Oelmüller R. (2009) Calcium signaling in pathogenic and beneficial plant-microbe interactions: what can we learn from the interaction between *Piriformospora indica* and *Arabidopsis thaliana*. *Plant Signal Behav.* 4:1024-1027.
- Vadassery J, Ranf S, Drzewiecki C, Mithöfer A, Mazars C, Scheel D, Lee J, Oelmüller R. (2009) A cell wall extract from the endophytic fungus *Piriformospora indica* promotes growth of *Arabidopsis* seedlings and induces intracellular calcium elevation in roots. *Plant J.* 59:193-206.
- Vadassery J, Reichelt M, Hause B, Gershenzon J, Boland W, Mithöfer A. (2012a) CML42-mediated calcium signaling coordinates responses to *Spodoptera* herbivory and abiotic stresses in *Arabidopsis*. *Plant Physiol.* 159:1159-1175.
- Vadassery J, Scholz SS, Mithöfer A. (2012b) Multiple calmodulin-like proteins in *Arabidopsis* are induced by insect-derived (*Spodoptera littoralis*) oral secretion. *Plant Signal Behav.* 7:1277-1280.
- Vanderbeld B, Snedden WA. (2007) Developmental and stimulus-induced expression patterns of *Arabidopsis* calmodulin-like genes CML37, CML38 and CML39. *Plant Mol Biol.* 64(6):683-97.
- Walter A, Mazars C, Maitrejean M, Hopke J, Ranjeva R, Boland W, Mithöfer A. (2007) Structural requirements of jasmonates and synthetic analogues as inducers of Ca²⁺ signals in the nucleus and the cytosol of plant cells. *Angew Chem Int Ed Engl.* 46:4783-4785.
- Wang SS, Diao WZ, Yang X, Qiao Z, Wang M, Acharya BR, Zhang W. (2015) *Arabidopsis thaliana* CML25 mediates the Ca²⁺ regulation of K⁺ transmembrane trafficking during pollen germination and tube elongation. *Plant Cell Environ.* 38(11):2372-2386.
- Wang Y, Zhang WZ, Song LF, Zou JJ, Su Z, Wu WH. (2008) Transcriptome analyses show changes in gene expression to accompany pollen germination and tube growth in *Arabidopsis*. *Plant Physiol.* 148(3):1201-11.
- Weckwerth W. (2011) Green systems biology - From single genomes, proteomes and metabolomes to ecosystems research and biotechnology. *J Proteomics.* 75:284-305.
- Winter D, Vinegar B, Nahal H, Ammar R, Wilson GV, Provart NJ. (2007) An "Electronic Fluorescent Pictograph" browser for exploring and analyzing large-scale biological data sets. *PLoS One.* 2:e718.
- Won SK, Lee YJ, Lee HY, Heo YK, Cho M, Cho HT. (2009) Cis-element- and transcriptome-based screening of root hair-specific genes and their functional characterization in *Arabidopsis*. *Plant Physiol.* 150:1459-1473.
- Wright AJ, Knight H, Knight MR. (2002) Mechanically stimulated *TCH3* gene expression in *Arabidopsis* involves protein phosphorylation and EIN6 downstream of calcium. *Plant Physiol.* 128(4):1402-9.
- Wuest SE, Vijverberg K, Schmidt A, Weiss M, Gheyselinck J, Lohr M, Wellmer F, Rahnenführer J, von Mering C, Grossniklaus U. (2010) *Arabidopsis* female gametophyte gene expression map reveals similarities between plant and animal gametes. *Curr Biol.* 20(6):506-12.
- Xu GY, Rocha PS, Wang ML, Xu ML, Cui YC, Li LY, Zhu YX, Xia X. (2011) A novel rice calmodulin-like gene, *OsMSR2*, enhances drought and salt tolerance and increases ABA sensitivity in *Arabidopsis*. *Planta.* 234:47-59.
- Yamaguchi T, Aharon GS, Sottosanto JB, Blumwald E. (2005) Vacuolar Na⁺/H⁺ antiporter cation selectivity is regulated by calmodulin from within the vacuole in a Ca²⁺- and pH-dependent manner. *Proc Natl Acad Sci USA.* 102(44):16107-12.
- Yang X, Wang SS, Wang M, Qiao Z, Bao CC, Zhang W. (2014) *Arabidopsis thaliana* calmodulin-like protein CML24 regulates pollen tube growth by modulating the actin cytoskeleton and controlling the cytosolic Ca²⁺ concentration. *Plant Mol Biol.* 86:225-236.
- Zhou L, Fu Y, Yang Z. (2009) A genome-wide functional characterization of *Arabidopsis* regulatory calcium sensors in pollen tubes. *J Integr Plant Biol.* 51(8):751-61.
- Zhu X, Dunand C, Snedden W, Galaud JP. (2015) CaM and CML emergence in the green lineage. *Trends Plants Sci.* 20(8):483-89.