

## **Signaling specificity and complexity of MAPK cascades in plant innate immunity**

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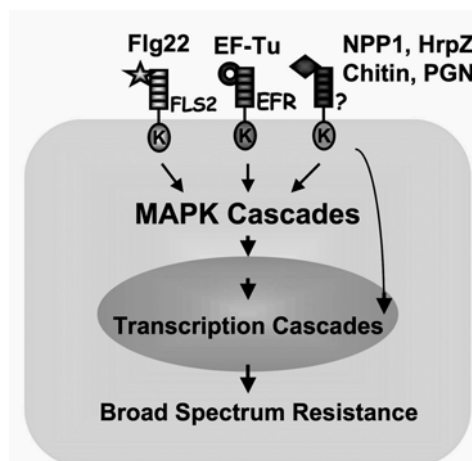
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The desire to understand and control plant diseases has been a major drive to study host-microbe interactions. In addition to the molecular basis of diseases and disease resistance, another fundamental question is how plants manage to stay healthy and survive constant exposure to a wide range of microbes. The innate immune response is the first line of defense, and is likely critical for the survival and fitness of plants in the ubiquitous presence of microbes. Research in the past decade has revealed remarkable convergent evolution in the recognition of pathogen- or microbe-associated molecular patterns (PAMPs or MAMPs) and the activation of innate immune responses in plants, insects and mammals (Nürnberger et al., 2004; Ausubel, 2005; Zipfel and Felix, 2005; Akira et al., 2006; Ferrandon et al., 2007). We are interested in dissecting the plant signal transduction networks that are responding to or manipulated by microbial signals and effectors.

For a robust innate immune system, both signaling specificity and complexity are required. Currently, the sequenced plant model *Arabidopsis thaliana* provides the most advanced and sophisticated molecular, cellular, genetic, genomic, proteomic, and bioinformatic tools for studying signaling mechanisms and transcription controls in plant innate immunity. The primary cell assays using protoplasts freshly isolated from leaves support high-throughput and versatile analyses of diverse MAMP and microbial effector signal responses (Asai et al., 2002; He et al., 2006a, 2006b; Yoo et al., 2007). The virus-induced gene silencing (VIGS) method (Burch-Smith et al., 2006), inducible RNAi capability, saturating insertional mutant resources (Sessions et al., 2002; Alonso et al., 2003), and whole-genome microarray databases

(Navarro et al., 2004; Zipfel et al., 2004; 2006; He et al., 2006a) have facilitated the use of an integrated approach that is especially powerful to overcome lethality and functional redundancy problems that hindered the advance of research in regulatory networks.

Using the *Arabidopsis* fresh leaf cell system based on the induction of early defense gene transcription by the conserved 22 amino acid peptide of bacterial flagellin (flg22) (Felix et al., 1999), we have identified the evolutionarily conserved MAP kinase (MAPK) cascades and transcription factors (Asai et al., 2002) that function downstream of the flg22 receptor FLS2, a leucine-rich-repeat (LRR) receptor kinase (Gomez-Gomez and Boller, 2000). The pattern recognition receptors (PRRs) in plant innate immunity share similarity with the mammalian toll-like receptors (TLRs) and downstream IRAK (IL-1 Receptor-Associated Kinase), which also activate multiple MAPK cascades besides the NF- $\kappa$ B pathway (Shiu and Bleecker, 2003; Zipfel and Felix, 2005; Akira et al., 2006). Significantly, activation of the plant MAPK cascade confers resistance to a broad spectrum of bacterial and fungal pathogens (Asai et al., 2002; Yamamizo et al., 2006; Brader et al., 2007), suggesting that signaling events initiated by diverse microbes converge into conserved MAPK cascades (Fig. 1). An integrated functional genomic approach has been taken to elucidate the complex and intertwined MAPK cascades that mediate MAMP, stress and hormonal signaling, and modulate plant innate immune responses (Kovtun et al., 2000; Tena et al., 2000; Asai et al., 2002; Zhang and Klessig, 2001; Soyano et al., 2003; Droillard et al., 2004; Nakagami et al., 2005; Pedley and Martin, 2005; Doczi et al., 2007; Seo et al., 2007; Takahashi et al., 2007).



**Fig. 1.** MAMP signaling converges on MAPK cascades.

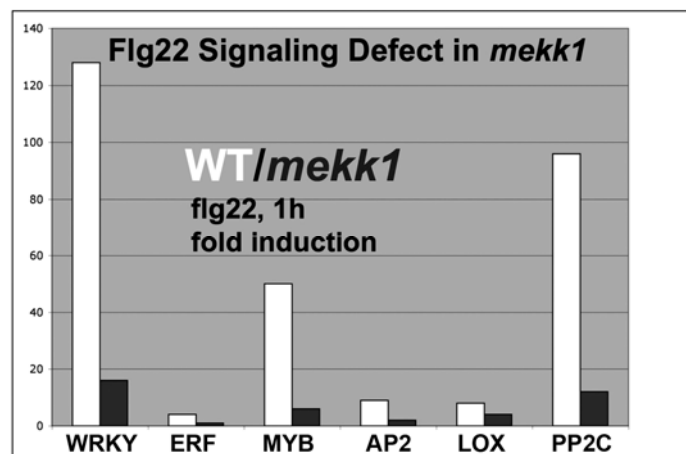
MAPK cascades are pivotal and evolutionarily conserved regulatory modules controlling diverse signal transduction pathways in eukaryotic cells from yeast to human and plants. Plant genomes encode the largest number of putative MAPK cascade genes (e.g., *Arabidopsis*: more than 100; yeast: 14; human: 34) in all sequenced eukaryotes (Ichimura et al., 2002; Hamel et al., 2006). Very limited information on MAPK cascade functionality and regulatory mechanisms has surfaced from classical genetic screens in plants. Extensive phenotype-based genetic screens in the past two decades have yielded three mutants (*ctr1*: constitutive ethylene triple response, *edr1*: enhanced disease resistance, and *yda*: YODA for defects in asymmetric cell division in zygotes and epidermis) with defects in putative MAPKKK (MKKK/MTK) genes in *Arabidopsis* (Kieber et al., 1993; Frye et al., 2001; Lukowitz et al., 2004). Their downstream signaling components and regulatory mechanisms remain to be fully elucidated in *Arabidopsis* and other plants (Wang et al., 2007). Biochemical, molecular, reverse genetic and transgenic studies have indicated that plant MAPK cascades are important for controlling broad and essential plant processes, including cytokinesis, cell death, various developmental programs, and hormone, stress and innate immune signaling in diverse plant species, including *Arabidopsis*, tobacco, tomato, alfalfa, rice, maize and poplar (Cardinale et al., 2000; Kovtun et al., 2000; Petersen et al., 2000; Tena et al., 2000; Lee et al., 2001; Zhang and Klessig, 2001; Asai et al., 2002; Fellbrich et al., 2002; Ichimura et al., 2002; Jin et al., 2002; Soyano et al., 2003; Wan et al., 2004; Nakagami et al., 2005; Pedley and Martin, 2005; Brodersen et al., 2006; Hamel et al., 2006; Ichimura et al., 2006; Meszaros et al., 2006; Nakagami et al., 2006; Doczi et al., 2007; Miao et al., 2007; Seo et al., 2007; Su et al., 2007; Suarez-Rodriguez et al., 2007; Takahashi et al., 2007; Wang et al., 2007; Zhang et al., 2007). However, how MAPKs/MPKs and their immediate upstream regulators, MAPKKs/MKKs and MAPKKKs/MTKs, are integrated into the plant signaling networks connecting upstream signals and downstream transcription factors and target genes remains a major challenge in plant biology.

A key question in MAPK cascade signaling has been how similar or even identical components control different biological responses. In yeast and mammals, specificity for distinct signaling pathways with shared components is determined by scaffolding proteins and specific MAPKs. In plants, more complex roles of MAPKs have been observed. For instance, diverse stresses, microbial elicitors, plant hormones and developmental programs activate the same conserved MPK3, MPK4 and MPK6 in *Arabidopsis* and their orthologs in other plant species (Tena et al., 2000; Zhang and Klessig, 2001; Ichimura

et al., 2002; Nakagami et al., 2005; Wang et al., 2007). It is unclear whether and how signaling specificity is regulated by MAPK cascades in the genetic framework of the plant signal transduction network. There are also many unresolved puzzles and confusing information regarding the precise physiological roles of MPKs, MKKs and MTKs in stress, defense, and hormonal signaling.

Although many mutant lines for the MAPK cascade genes are now available from the large collection of Arabidopsis genome resources, most single mutants exhibit no overt phenotypes, suggesting functional redundancy. In contrary, some single mutants (e.g., *mpk4* & *mekk1*) display severe and long-term growth defects not associated with their dynamic and often transient functions in signal transduction, likely due to multiple functional roles in unrelated pathways (Petersen et al., 2000; Brodersen et al., 2006; Ichimura et al., 2006; Meszaros et al., 2006; Nakagami et al., 2006; Su et al., 2007; Suarez-Rodriguez et al., 2007). In yeast, extensive studies have placed a single MAPKKK (Ste11) in three unrelated signal transduction pathways. In Arabidopsis, MEKK1 is involved in distinct functions, including positive roles in flg22 signaling (Figs. 2, 3). Some double or triple Arabidopsis mutants are embryonic or seedling lethal or show constitutive stress stimulation, thus impeding thorough functional characterization (Wang et al., 2007).

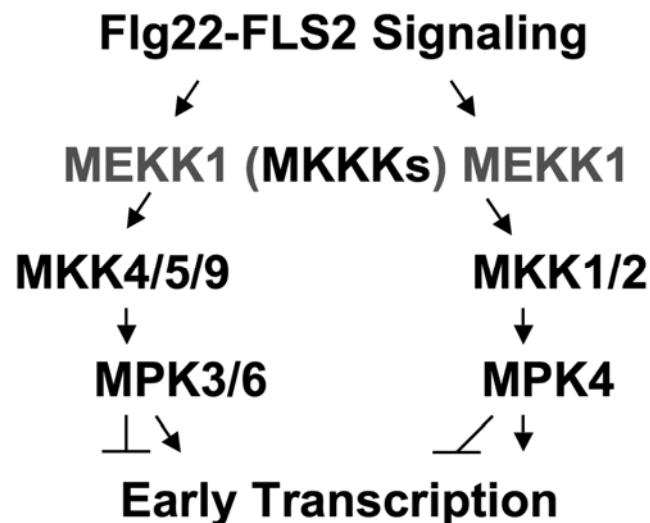
To circumvent the limitations in classical and reserve genetic analyses, and to examine dynamic and complex actions in MAPK cascade signaling, we have developed MAPK cascade genomic resources and performed extensive cell-autonomous and systematic screens for *Arabidopsis* MAPKs acting



**Fig. 2.** The *mekk1* mutant showed defect in flg22 inducible transcription.

downstream of MKKs and MTKs and various signals that activate putative endogenous MPKs in the cell-based transient expression systems. Arabidopsis mesophyll protoplasts have been used to show the conservation of MAPK cascade signaling and similar physiological responses in isolated cells and in intact plants. In the functional genomic analysis of Arabidopsis MAPK cascade signaling, 20 MPK, 10 MKK and 68 putative MTK genes have been analyzed in mesophyll protoplasts to establish a blueprint for potential MAPK cascade functions and connections. The information will serve as the foundation to launch new genome-wide studies linking dynamic and overlapping signal transduction pathways.

The MAPK functional genomic project combines global gene expression profiling and bioinformatics tools to dissect distinct and overlapping MAPK cascades with gain-of-function and loss-of-function mutant analyses in response to stress, elicitor, and hormonal signals. The experiments have first been carried out in the well-established mesophyll protoplasts, which respond to the majority of physiological signals and regulate transcription, and are suitable for studying quantitative and global processes and for genetic and molecular manipulations. The aims are to integrate broad resources and information on stress, defense, and plant hormone signaling and gene regulation to facilitate comprehensive and molecular understanding of the signaling specificity and complexity of the evolutionarily conserved MAPK



**Fig. 3.** Multiple MAPK cascades are activated by flg22 through the FLS2 receptor.

cascade signaling networks in *Arabidopsis*. Recent discoveries have unraveled multiple functions of MAPK signaling cascade components in innate immunity (Fig. 3) and antagonistic MAPK cascades in the ethylene signal transduction pathway. The most unexpected findings include the complexity of the MTKs functionality in distinct or shared signaling regulation without or with PK activities, and the specificity and redundancy of MKKs and MPKs in different signaling pathways (Fig. 3). The unique advantage of the integrated approach is to carry out whole plant studies guided by hypotheses established using simplified cell and molecular assays scaled up to the whole genome level. The integrated analyses establish new paradigms for linking intertwined MAPK cascades in controlling quantitative and dynamic responses and signaling specificity and complexity.

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### Literature cited

- Akira, S., Uematsu, S., and Takeuchi, O. 2006. Pathogen recognition and innate immunity. *Cell* 124:783-801.
- Alonso, J.M., Stepanova, A.N., Leisse, T.J., Kim, C.J., Chen, H., Shinn, P., Stevenson, D.K., Zimmerman, J., Barajas, P., Cheuk, R., Gadrinab, C., Heller, C., Jeske, A., Koesema, E., Meyers, C.C., Parker, H., Prednis, L., Ansari, Y., Choy, N., Deen, H., Geralt, M., Hazari, N., Hom, E., Karnes, M., Mulholland, C., Ndbuku, R., Schmidt, I., Guzman, P., Aguilar-Henonin, L., Schmid, M., Weigel, D., Carter, D.E., Marchand, T., Risseuw, E., Brogden, D., Zeko, A., Crosby, W.L., Berry, C.C., and Ecker, J.R. 2003. Genome-wide insertional mutagenesis of *Arabidopsis thaliana*. *Science* 301:653-657.
- Asai, T., Tena, G., Plonikova, J., Willmann, M., Chiu, W.L., Gomez-Gomez, L., Boller, T., Ausubel, F.M., and Sheen, J. 2002. MAP kinase signaling cascade in *Arabidopsis* innate immunity. *Nature* 415:977-983.
- Ausubel, F.M. 2005. Are innate immune signaling pathways in plants and animals conserved? *Nat Immunol.* 6:973-979.
- Brader, G., Djamei, A., Teige, M., Palva, E.T., and Hirt, H. 2007. The MAP kinase kinase MKK2 affects disease resistance in *Arabidopsis*. *MPMI* 20:589-596.
- Brodersen, P., Petersen, M., Nielsen, H.B., Zhu, S., Newman, M.A., Shokat, K.M., Rietz, S., Parker, J., and Mundy, J. 2006. *Arabidopsis* MAP kinase 4 regulates salicylic acid- and jasmonic acid/ethylene-dependent responses via EDS1 and PAD4. *Plant J.* 47:5332-546.
- Burch-Smith, T.M., Schiff, M., Liu, Y., and Dinesh-Kumar, S.P. 2006. Efficient virus induced gene silencing in *Arabidopsis thaliana*. *Plant Physiol.* 142:21-27.

- Cardinale, F., Jonak, C., Ligterink, W., Niehaus, K., Boller, T., and Hirt, H. 2000. Differential activation of four specific MAPK pathways by distinct elicitors. *J. Biol. Chem.* 275:36734-36740.
- Doczi, B., Brader, G., Pettko-Szandtner, A., Rajh, I., Djamei, D., Pitzschke, A., Teige, M., and Hirt, H. 2007. The Arabidopsis Mitogen-Activated Protein Kinase Kinase MKK3 Is Upstream of Group C Mitogen-Activated Protein Kinases and Participates in Pathogen Signaling. *Plant Cell* 19:3266 – 3279.
- Droillard, M.J., Boudsocq, M., Barbier-Brygoo, H., and Lauriere, C. 2004. Involvement of MPK4 in osmotic stress response pathways in cell suspensions and plantlets of *Arabidopsis thaliana*: activation by hypoosmolarity and negative role in hyperosmolarity tolerance. *FEBS Lett.* 574:42-48.
- Felix, G., Duran, J.D., Volko, S., and Boller, T. 1999. Plants have a sensitive perception system for the most conserved domain of bacterial flagellin. *Plant J.* 18:265-276.
- Fellbrich, G., Romanski, A., Varet, A., Blume, B., Brunner, F., Engelhardt, S., Felix, G., Kemmerling, B., Krzymowska, M., and Nürnberger, T. 2002. NPP1, a *Phytophthora*-associated trigger of plant defense in parsley and *Arabidopsis*. *Plant J.* 32:375-390.
- Ferrandon, D., Imler, J.L., Hetru, C., and Hoffmann, J.A. 2007. The *Drosophila* systemic immune response: sensing and signaling during bacterial and fungal infections. *Nat. Rev. Immunol.* 7:862-874.
- Frye, C.A., Tang, D., and Innes, R.W. 2001. Negative regulation of defense responses in plants by a conserved MAPKK kinase. *Proc. Natl. Acad. Sci. U.S.A.* 98:373-378.
- Gomez-Gomez, L., and Boller, T. 2000. FLS2: an LRR receptor-like kinase involved in the perception of the bacterial elicitor flagellin in *Arabidopsis*. *Mol. Cell* 5:1003-1011.
- He, P., Shan, L., and Sheen, J. 2006. The use of protoplasts to study innate immune responses. *Methods Mol. Biol.* 354:1-10.
- Hamel, L.P., Nicole, M.C., Sritubtim, S., Morency, M.J., Ellis, M., Ehlting, J., Beaudoin, N., Barbazuk, B., Klessig, D., Lee, J., Martin, G., Mundy, J., Ohashi, Y., Scheel, D., Sheen, J., Xing, T., Zhang, S., Seguin, A., and Ellis, B.E. 2006. Ancient signals: Comparative genomics of plant MAPK and MAPKK gene families. *Trends Plant Sci.* 11:192-198.
- He, P., Shan, L., Lin, N.C., Martin, G.B., Kemmerling, B., Neurberger, T., and Sheen, J. 2006. Specific bacterial suppressors of MAMP signaling upstream of MAPKKK in *Arabidopsis*. *Cell* 125:563-575.
- He, P., Shan, L., and Sheen, J. 2007. Elicitation and suppression of microbe-associated molecular pattern-triggered immunity in plant-microbe interactions. *Cell. Microbiol.* 9:1385-1396.
- Ichimura, K., Shinozaki, K., Tena, G., Sheen, J., Henry, Y., Champion, A., Kreis, M., Zhang, S., Hirt, H., Wilson, C., Heberle-Bors, E., Ellis, B.E., Morris, P.C., Innes, R.W., Ecker, J.R., Scheel, D., Klessig, D.F., Machida, Y., Mundy, J., Ohashi, Y., and Walker, J.C. 2002. Mitogen-activated protein kinase cascades in plants: a new nomenclature. *Trends in Plant Sci.* 7:301-308.

- Ichimura, K., Casais, C., Peck, S.C., Shinozaki, K., and Shirasu, K. 2006. MEKK1 is required for MPK4 activation and regulates tissue-specific and temperature-dependent cell death in *Arabidopsis*. *J. Biol. Chem.* 281:36969-36976.
- Jin, H., Axtell, M.J., Dahlbeck, D., Ekwenna, O., Zhang, S., Staskawicz, B., and Baker, B. 2002. NPK1, an MEKK1-like mitogen-activated protein kinase kinase, regulates innate immunity and development in plants. *Dev. Cell* 3:291-297.
- Kieber, J.J., Rthenberg, M., Roman, G., Feldmann, K.A., and Ecker, J.R. 1993. CTR1, a negative regulator of the ethylene response pathway in *Arabidopsis*, encodes a member of the raf family of protein kinases. *Cell* 72:427-441.
- Kovtun, Y., Chiu, W.L. Tena, G., and Sheen, J. 2000. Functional analysis of oxidative stress-activated MAPK cascade in plants. *Proc. Natl. Acad. Sci. U.S.A.* 97:2940-2945.
- Lee, J., Klessig, D.F. and Nurnberger, T. 2001. A harpin binding site in tobacco plasma membranes mediates activation of the pathogenesis-related gene HIN1 independent of extracellular calcium but dependent on mitogen-activated protein kinase activity. *Plant Cell.* 13:1079-1093.
- Lukowitz, W., Roeder, A., Parmenter, D., and Somerville, C. 2004. A MAPKK kinase gene regulates extra-embryonic cell fate in *Arabidopsis*. *Cell* 116:109-119.
- Mészáros, T., Helfer, A., Hatzimasoura, E., Magyar, Z., Serazetdinova, L., Rios, G., Bardóczy, V., Teige, M., Koncz, C., Peck, S., and Bögre, L. 2006. The *Arabidopsis* MAP kinase kinase MKK1 participates in defence responses to the bacterial elicitor flagellin. *Plant J.* 48:485-498.
- Miao, Y., Laun, T.M., Smykowski, A., and Zentgraf, U. 2007. *Arabidopsis* MEKK1 can take a short cut: it can directly interact with senescence-related WRKY53 transcription factor on the protein level and can bind to its promoter. *Plant Mol Biol.* DOI 10.1007/s11103-007-9198-z
- Nakagami, H., Pitzschke, A., and Hirt, H. 2005. Emerging MAP kinase pathways in plant stress signaling. *Trends Plant Sci.* 10:339-346.
- Nakagami, H., Soukupova, H., Schikora, A., Zarsky, V., and Hirt, H. 2006. A mitogen-activated protein kinase kinase kinase mediates reactive oxygen species homeostasis in *Arabidopsis*. *J. Biol. Chem.* 281:38697-38704.
- Navarro, L., Zipfel, C., Rowland, O., Keller, I., Robatzek, S., Boller, T., and Jones, J.D. 2004. The transcriptional innate immune response to flg22. Interplay and overlap with Avr gene-dependent defense responses and bacterial pathogenesis. *Plant Physiol.* 135:1113-1128.
- Nürnbergger, T., Brunner, F., Kemmerling, B., and Piater, L. 2004. Innate immunity in plants and animals: striking similarities and obvious differences. *Immunol. Rev.* 198:249-266.
- Petersen, M., Brodersen, P., Naested, H., Andreasson, E., Lindhart, U., Johansen, B., Nielsen, H.B., Lacy, M., Austin, M.J., Parker, J.E., Sharma, S.B., Klessig, D.F., Martienssen, R., Mattsson, O., Jensen, A.B., and Mundy, J. 2000. *Arabidopsis* map kinase 4 negatively regulates systemic acquired resistance. *Cell* 103:1111-1120.
- Pedley, K.F., and Martin, G.B. 2005. Role of mitogen-activated protein kinases in plant immunity. *Curr. Opin. Plant Biol.* 8:541-547.

- Seo, S., Katou, S., Seto, H., Gomi, K., and Ohashi, Y. 2007. The mitogen-activated protein kinases WIPK and SIPK regulate the levels of jasmonic and salicylic acids in wounded tobacco plants. *Plant J.* 49:899-909.
- Sessions, A., Burke, E., Presting, G., Aux, G., McElver, J., Patton, D., Dietrich, B., Ho, P., Bacwaden, J., Ko, C., Clarke, J.D., Cotton, D., Bullis, D., Snell, J., Miguel, T., Hutchison, D., Kimmerly, B., Mitzel, T., Katagiri, F., Glazebrook, J., Law, M., and Goff, S.A. 2002. A high-throughput *Arabidopsis* reverse genetics system. *Plant Cell.* 14:2985-2994.
- Shan, L., He, P., and Sheen, J. 2007. Intercepting host MAPK signaling cascades by bacterial type III effectors. *Cell Host & Microbe* 1:167-174.
- Shiu, S.H., and Bleecker, A.B. 2003. Expansion of the receptor-like kinase/Pelle gene family and receptor-like proteins in *Arabidopsis*. *Plant Physiol.* 132:530-543.
- Soyano, T., Nishihama, R., Morikiyo, K., Ishikawa, M., and Machida, Y. 2003. NQK1/NtMEK1 is a MAPKK that acts in the NPK1 MAPKKK-mediated MAPK cascade and is required for plant cytokinesis. *Genes & Develop.* 17:1055-1067.
- Su, S.H., Suarez-Rodriguez, M.C., and Krysan, P. 2007. Genetic interaction and phenotypic analysis of the *Arabidopsis* MAP kinase pathway mutations mekk1 and mpk4 suggests signaling pathway complexity. *FEBS Letters* 581:3171-3177.
- Suarez-Rodriguez, M.C., Adams-Phillips, L., Liu, Y., Wang, H., Su, S.H., Jester, P.J., Zhang, S., Bent, A.F., and Krysan, P.J. 2007. MEKK1 is required for flg22-induced MPK4 activation in *Arabidopsis* plants. *Plant Physiol.* 143:661-669.
- Tena, G., Asai, T., Chiu, W.L., and Sheen, J. 2001. Plant mitogen-activated protein kinase signaling cascades. *Curr. Opin. Plant Biol.* 4:392-400.
- Takahashi, F., Yoshida, R., Ichimura, K., Mizoguchi, T., Seo, S., Yonezawa, M., Maruyama, K., Yamaguchi-Shinozaki, K., Shinozaki, K. 2007. The mitogen-activated protein kinase cascade MKK3-MPK6 is an important part of the jasmonate signal transduction pathway in *Arabidopsis*. *Plant Cell* 19:805-818.
- Wan, J., Zhang, S., and Stacey, G. 2004. Activation of a mitogen-activated protein kinase pathway in *Arabidopsis* by chitin. *Mol. Plant Pathol.* 5:125-135.
- Wang, H., Ngwenyama, N., Liu, Y., Walker, J.C. and Zhang, S. 2007. Stomatal development and patterning are regulated by environmentally responsive mitogen-activated protein kinases in *Arabidopsis*. *Plant Cell* 19:63-73.
- Yamamoto, C., Kuchimura, K., Kobayashi, K., Katou, S., Kawakita, K., Jones, J.D.G., Doke, N., and Yoshioka, H. 2006. Rewiring mitogen-activated protein kinase cascade by positive feedback confers potato blight resistance. *Plant Physiol.* 140:681-692.
- Yoo, S.D., Cho, Y.H., and Sheen, J. 2007. *Arabidopsis* mesophyll protoplasts: a versatile cell system for transient gene expression analysis. *Nat. Protoc.* 2:1565-1572.
- Zhang, S., and Klessig, D.F. 2001. MAPK cascades in plant defense signaling. *Trends Plant Sci.* 6:520-527.
- Zhang, X., Dai, Y., Xiong, Y., DeFraia, C., Li, J., Dong, X., and Mou, Z. 2007. Overexpression of *Arabidopsis* MAP kinase kinase 7 leads to activation of plant basal and systemic acquired resistance. *Plant J.* 52:1066-1079.
- Zipfel, C., and Felix, G. 2005. Plants and animals: a different taste for microbes? *Curr. Opin. Plant Biol.* 8:353-360.

- Zipfel, C., Kunze, G., Chinchilla, D., Caniard, A., Jones, J.D., Boller, T., and Felix, G. 2006. Perception of the bacterial PAMP EF-Tu by the receptor EFR restricts *Agrobacterium*-mediated transformation. *Cell* 125:749-760.
- Zipfel, C., Robatzek, S., Navarro, L., Oakeley, E.J., Jones, J.D., Felix, G., and Boller, T. 2004. Bacterial disease resistance in *Arabidopsis* through flagellin perception. *Nature* 428:764-767.