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MAP kinases in plant signal transduction

C. Jonak, W. Ligterink and H. Hirt*

Institute of Microbiology and Genetics, Vienna Biocenter, Dr. Bohrgasse 9, A-1030 Vienna (Austria),
Fax + 43 1 4277 9546, e-mail: hehi@gem.univie.ac.at

Abstract. Mitogen-activated protein kinase (MAPK) pathways are modules involved in the transduction of extracellular signals to intracellular targets in all eukaryotes. Distinct MAPK pathways are regulated by different extracellular stimuli and are implicated in a wide variety of biological processes. In plants there is evidence for MAPKs playing a role in the signaling of abiotic stresses, pathogens and plant hormones. The large number and divergence of plant MAPKs indicates that this ancient mechanism of bioinformatics is extensively used in plants and may provide a new molecular handle on old questions.

Key words. Signal transduction; MAP kinase; protein kinase; phosphorylation.

MAPKs modules form basic units of eukaryotic signal transduction

Mitogen-activated protein kinases (MAPKs) are encoded by a large family of serine/threonine protein kinases that are found in all eukaryotes. Activation of MAPKs is brought about by upstream MAPK kinases (denoted as MAPKKs) through phosphorylation of the conserved threonine and tyrosine residues that are located close to kinase domain VIII in all MAPKs [1] (fig. 1). A given dual-specificity MAPKK can only activate a specific MAPK and cannot functionally substitute other MAPKKs. MAPKKs are themselves activated by phosphorylation through upstream kinases that belong to the class of MAPKK kinases (MAPKKKs), raf and mos proteins [1]. A specific set of three functionally interlinked protein kinases (MAPKKK-MAPKK-MAPK) forms the basic module of a MAPK pathway (fig. 1). MAPK pathways may integrate a variety of upstream signals through interaction with other kinases or G proteins, such as ras or heterotrimeric complexes. The latter factors often function as coupling agents between a plasma membrane-located receptor protein that senses an extracellular stimulus and a MAPK module.

At the downstream end of the module, activation of the cytoplasmic MAPK module often induces the translocation of the MAPK into the nucleus, where the kinase activates certain sets of genes through phosphorylation of specific transcription factors (fig. 1). In other cases, a given MAPK may translocate to other sites in the cytoplasm to phosphorylate specific enzymes (protein kinases, phosphatases, lipases etc.) or cytoskeletal components. By tight regulation of MAPK localization and through expression of certain signaling components and substrates in particular cells, tissues or organs, particular MAPK pathways can mediate signaling of a multitude of extracellular stimuli and bring about a large variety of specific responses.

Different MAPK pathways exist for different signals

MAPK pathways are best understood in yeast, and this organism can be viewed as a model to understand the role of MAPK cascades in more complex multicellular systems. Of the six MAPK genes that are present in the yeast genome, functions for five MAPKs have been identified [2]. During mating, the FUS3 and KSS1 MAPKs are activated by exposure of yeast to pheromone, and mutants of this pathway are sterile. Some of the components of the pheromone pathway, but not the FUS3 kinase, are involved in pseudohyphal growth control in response to nitrogen starvation in diploid cells and invasive growth in haploid cells. Whereas the MPK1 MAPK is required for adaptation

* Corresponding author.

to a hypoosmolar environment, the HOG1 MAPK is activated under hyperosmolar conditions and results in increased biosynthesis of glycerol, which acts as an osmotic stabilizer. The SMK1 MAPK is involved in the control of spore formation. From studies in yeast it is known that different MAPKs cannot substitute for each other and that specific interactions with other proteins are brought about by scaffold proteins that serve as interaction platforms.

In mammals, MAPKs or ERKs (for extracellular signal-regulated kinases) were originally identified as transducers of mitogens. Later, MAPKs were also shown to be involved in signaling hormones, neurotransmitters and signals for differentiation [1]. Recently, two new groups of protein kinases have been added to the family of mammalian MAPKs [1]. The stress-activated protein kinases (SAPKs) or Jun kinases (JNKs) were identified by their ability to specifically phosphorylate the transcription factor c-jun mediating transcription of specific genes following exposure to ultraviolet radiation, proinflammatory cytokines and environmental stress. The second family, the p38 kinases, are activated in response to endotoxin from Gram-negative bacteria, interleukin-1 and hyperosmolar stress. A p38 kinase is also induced by heat shock activating yet another protein kinase that phosphorylates small heat shock proteins. The JNK and p38 kinases not only share functional similarities in terms of stress signaling but can also complement HOG1-deficient mutants of yeast.

In plants, a variety of genes encoding MAPKs have been identified from alfalfa [3–6], *Arabidopsis* [7, 8], *Avena* [9], parsley [10], pea [11], petunia [12] and tobacco [13–16] (fig. 2). The predicted amino acid sequences show high conservation over the entire lengths with highest similarity in the 11 domains that are necessary for the catalytic function of serine/threonine protein kinases. Whereby the sequences outside the 11 subdomains may show little homology within different MAPKs of a given species, these regions are often found to be highly conserved in a specific MAPK of another species, indicating that these sequences have important biological functions possibly with respect to substrate specificity or interaction with other proteins. The threonine and tyrosine residues whose phosphorylation is necessary for activation of MAPKs are found in all plant MAPKs between subdomains VII and VIII of the catalytic core (stars in fig. 2).

From an analysis of sequence homology of the predicted amino acid sequences, plant MAPKs can be grouped into at least four distinct families (fig. 3). The significance of the branching into different families is not yet fully understood, but so far suggests that MAPKs within one branch serve similar functions in different species. According to the available information, MAPKs of families I and II are mostly involved in signaling pathogens and abiotic stress, whereas at least some of the MAPKs of family III are involved in cell cycle regulation. Therefore, the sequence divergence most likely reflects different substrate specificities and functions. This idea is supported by a recent analysis of four alfalfa MAPKs showing that the bacterially expressed kinases have different substrate specificities in vitro and only one of the kinases was able to substitute for a defective MAPK in yeast [5].

Involvement of MAPKs in intracellular transmission of diverse stresses in plants: MAPKs as mediators of mechanical stress

Due to their sessile habit, plants are exposed to a variety of environmental stresses, including changes in temperature, water conditions, radiation and wind.

Wind is a mechanical stress and can lead to major changes in the growth pattern of plants, diverting energy into strengthening the plants stature, which is nicely seen on the short stature of wind-exposed trees in the mountains or in coastal areas. Experiments, showing that mechanical manipulation of *Arabidopsis* leaves induces transcription of particular MAPK and MAPKK genes [17], suggested that a MAPK pathway might signal mechanical stimuli. Direct evidence for

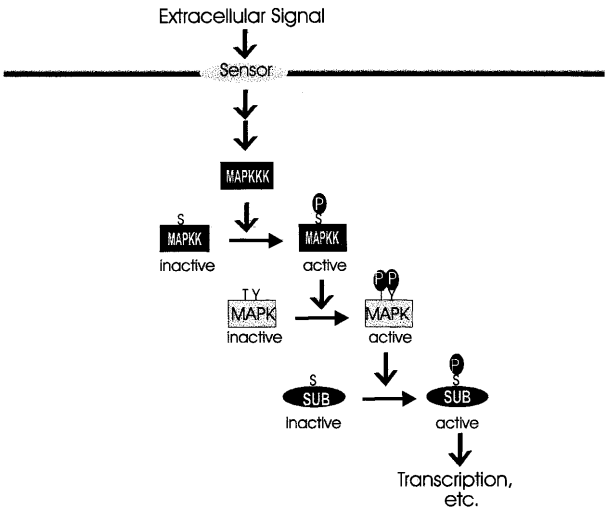

Figure 1. Simplified scheme for MAP kinase signal transduction cascades. An extracellular signal is received by a membrane-located receptor. The activation of the MAP kinase module (MAPKKK activates MAPKK, activates MAPK) by the receptor may occur via several intermediate steps and by different routes. The active MAP kinase may activate other protein kinases, phosphorylate cytoskeletal components or translocate to the nucleus and activate transcription factors giving rise to expression of specific genes. Arrows indicate activation.

such a role consisted in showing that touching alfalfa leaves for 2 s is sufficient to induce a transient activation of a MAPK [18]. Whereas constantly shaken suspension-cultured alfalfa cells were found to have constitutive levels of activated MAPK, this kinase activity vanished when cells were allowed to rest for 1 h, but shaking for a few seconds restored activity.

MAPK plays a role in abiotic stress signaling

Water availability and extreme temperatures are limiting factors for the development and growth of all plants. Plants in different climatic zones have developed specific mechanisms to withstand these stresses. The adaptive strategies mainly depend on the expression of specific sets of genes that result in changes in the composition of the major cell components. A MAPK has recently been shown to be involved in the transmission of drought and cold signals [6]. Using specific antibodies that differentiate between different members of the alfalfa MAPK family, it was shown that only one specific MAPK is activated by cold and drought stress in alfalfa plants. Moreover, the only gene to be transcriptionally induced is the gene encoding the MAPK that is activated. Despite these changes in MAPK activity and transcript levels, no changes of MAPK protein amounts were detected by Western blotting. The activation of the MAPK is not a general stress response, because heat or hypo- and hyperosmolar stress were unable to induce the kinase.

Ntf4, a tobacco MAPK, was recently shown to be expressed and activated in pollen [19]. Although both the expression and the activity of Ntf4 are developmentally controlled during pollen maturation, hydration of the mature dry pollen can stimulate the activity of Ntf4 much further.

The idea that a MAPK is involved in osmotic stress adaptation might find support in the report where the pea PsD5 MAPK was shown to complement HOG1-deficient yeast for their ability to grow on a hyperosmolar medium [20]. However, so far it is unclear whether hyperosmolar stress can also activate this or any MAPK in plants. Moreover, care must be taken in extending functional complementation data from yeast to other organsims. A good example is the ability of alfalfa MMK2 to complement the MPK1 pathway that is necessary for hypoosmolar signaling in yeast. Although these data could be taken as evidence that MMK2 should be involved in hypoosmolar stress signaling in plants, no evidence for such a role was found [5].

Assuming induction of MAPK gene expression by a particular stimulus as evidence for a role in signal transduction, it is likely that MAPK pathways also play a role in response to other physical stresses, because transcript amounts of specific *Arabidopsis* MAPK and MAPKK genes increased upon exposure to water stress, cold, touch and high salt [17].

MAPKs as mediators of wound signaling

In addition to these abiotic challenges, plants are also in constant contact with a variety of potential pathogens. Survival of plants in this environment was only possible by the development of sophisticated defense and adaptation strategies, and it comes as no surprise to find that MAPKs are involved in signal transduction of pathogen attack.

One of the most severe environmental stresses to which plants can be subjected is wounding, which may be caused by mechanical injury, pathogen or herbivore attack. To counter this challenge, plants have developed defense systems that are mostly based on activation of particular sets of genes encoding a variety of enzymes, pathogen response (PR) proteins or proteinase inhibitors (PIs). Whereas some of these genes are only induced locally at the site of attack, others are expressed systemically throughout the plant and protect the plant from attack at distant sites. Several of the genes involved in defense response have been identified and studied, but relatively little is known about how a plant senses wounding and transmits the signal to the nucleus before induction of the respective defense response genes. Several reports have implicated a MAPK in this process. A protein kinase with all the properties of a MAPK is induced by wounding of leaves from a variety of species, including both monocots and dicots [21]. Activation of this MAPK occurs within less than 1 min, placing this process in the very first line of responses to a wound signal. In a separate study, it was shown that wounding tobacco leaves also leads to the rapid accumulation of transcripts of a particular MAPK gene, termed WIPK for wound-induced protein kinase [14]. Overexpression of the *WIPK* gene in transgenic tobacco led to inactivation of the endogenous copies and as a consequence to suppression of the wound response. Wounding of leaves of the transgenic lines did not lead to increased levels of jasmonic acid and its methyl ester, substances that are normally induced by wounding and that are involved in mediating the systemic wound response. These results indicate that the MAPK pathway is upstream of jasmonic acid and by analogy to prostaglandin synthesis in mammalian cells, suggesting further that one of the targets of the MAPK pathway may be a phospholipase that is involved in jasmonic acid synthesis.

Further insight into the role of MAPKs in wound signaling was provided by the finding that systemin, an

18-amino acid peptide that confers a systemic wound response to tomato, activates a 48-kDa MBP kinase that is also tyrosine-phosphorylated upon wounding [22]. Two other substances that are released during wounding by pathogens, chitosan and polygalacturonic acid, were able to induce a similar kinase activation pattern. As shown by the *def1* mutant that is defective in the jasmonic acid pathway, wound defense gene signaling in tomato is mediated by jasmonic acid. The jasmonic acid pathway is not necessary for wound-induced kinase activation because *def1* mutant tomato plants still showed MBP kinase activation in response to systemin, chitosan and polygalactuonic acid, but not to jasmonic acid [22]. Taken together, these results are consistent with the model that WIPK in tobacco and its homolog in tomato are involved in translating the wound signal into the synthesis of jasmonic acid [14, 22].

Further evidence for the role of WIPK in wound signaling comes from studies in alfalfa, where a specific 46-kDa protein kinase was shown to be activated in leaves by wounding [23]. Using specific antisera, the authors could show that the protein kinase is MMK4, a specific alfalfa MAPK that is most closely related to tobacco WIPK.

In all plant species investigated, wounding induces a transient activation of the respective MAPKs [14, 21–23]. Whereas the activation of wound-induced MAPKs is a posttranslational process, inactivation is dependent on de novo transcription and protein synthesis [21, 23]. One of these newly synthesized proteins was recently identified to be MP2C, a protein phosphatase of type 2C [24]. MP2C is part of a negative feedback loop, whereby MP2C expression is tightly regulated through the activity of the wound-activated MAPK. By inactivating the MAPK pathway, the phosphatase shuts down its own synthesis and thereby helps to reset the pathway. This feedback loop provides the cell with a mechanism for using the components of a signaling pathway repeatedly for monitoring whether more pathogens line up in front of the city walls [24].

Different MAPKs are activated by microbial elicitors

While plants may sense the presence of a pathogen through wounding, pathogens are also recognized by other means involving cell wall components and microbial elicitors. Previous studies indicated that plant cells respond to elicitors by rapid changes in the phosphorylation status of proteins [25, 26], and it was demonstrated that treating tobacco cells with an elicitor, derived from cell walls of the fungus *Phytophthora infestans*, activates a protein kinase which has properties of a MAPK [27]. Stauroporine, a protein kinase inhibitor, and $Gd3^+$, a calcium ion channel blocker, were found to block activation of the kinase, suggesting that upstream kinases and calcium might be involved in activating this MAPK. Inactivation of the kinase was found to be blocked by calyculin A, a potent inhibitor of phosphatases 1 and 2A, and cycloheximide, indicating the involvement of phosphatases and protein synthesized de novo in the resetting of the pathway. Recently, three elicitors from *Phythophthora parasitica* were shown to be able to differentially activate three kinases [28]. All three kinases have similarities with MAPKs, but only one of them was unequivocally identified as SIPK, a MAPK that was previously identified to be activated by salicylic acid [16].

A peptide elicitor derived from a secreted glycoprotein of *P. sojae* activates ERMK (elicitor-regulated MAP kinase) in parsley cells [10]. In this plant pathogen model system, extensive studies have charted a presumptive signal transduction pathway, whereby peptide elicitor binding to a plasma membrane-located receptor sequentially activates various ion channels, an nicotinamide adenine dinucleotide phosphate (NADPH) oxidase (resulting in an oxidative burst), and induction of PR (pathogen related) genes and phytoalexin synthesis [29]. The studies by Ligterink et al. [10] showed that within this pathway, ERMK is downstream of the receptor and the elicitor-activated ion channels but acts upstream or independently of the NADPH oxidase. Interestingly, after treating parsley cells with elicitor, ERMK becomes rapidly translocated from the cytoplasm into the nucleus, suggesting a direct role of ERMK in the regulation of elicitor-induced gene transcription [10].

When tobacco leaves are treated with harpin, protein elicitors obtained from the bacterial pathogen *Erwinia amylovora*, necrosis occurs in the infiltrated regions within 24 h. A 49-kDa MBP kinase that also becomes tyrosine-phosphorylated is activated under these conditions within 15 min [30]. These results indicate that not only fungal but also bacterial elcitors are able to activate MAPK pathways in plants.

Evidence for a role of MAPKs in intracellular signaling of plant hormones

Ever since the discovery of plant hormones, their roles in development and physiological responses have fascinated and puzzled plant biologists. Despite intense efforts to understand how plant hormone signals are perceived and transmitted, many of the molecular mechanisms involved have still remained unclear. Increasing evidence now suggests that MAPK pathways are involved in mediating abscisic acid, auxin and ethylene responses.

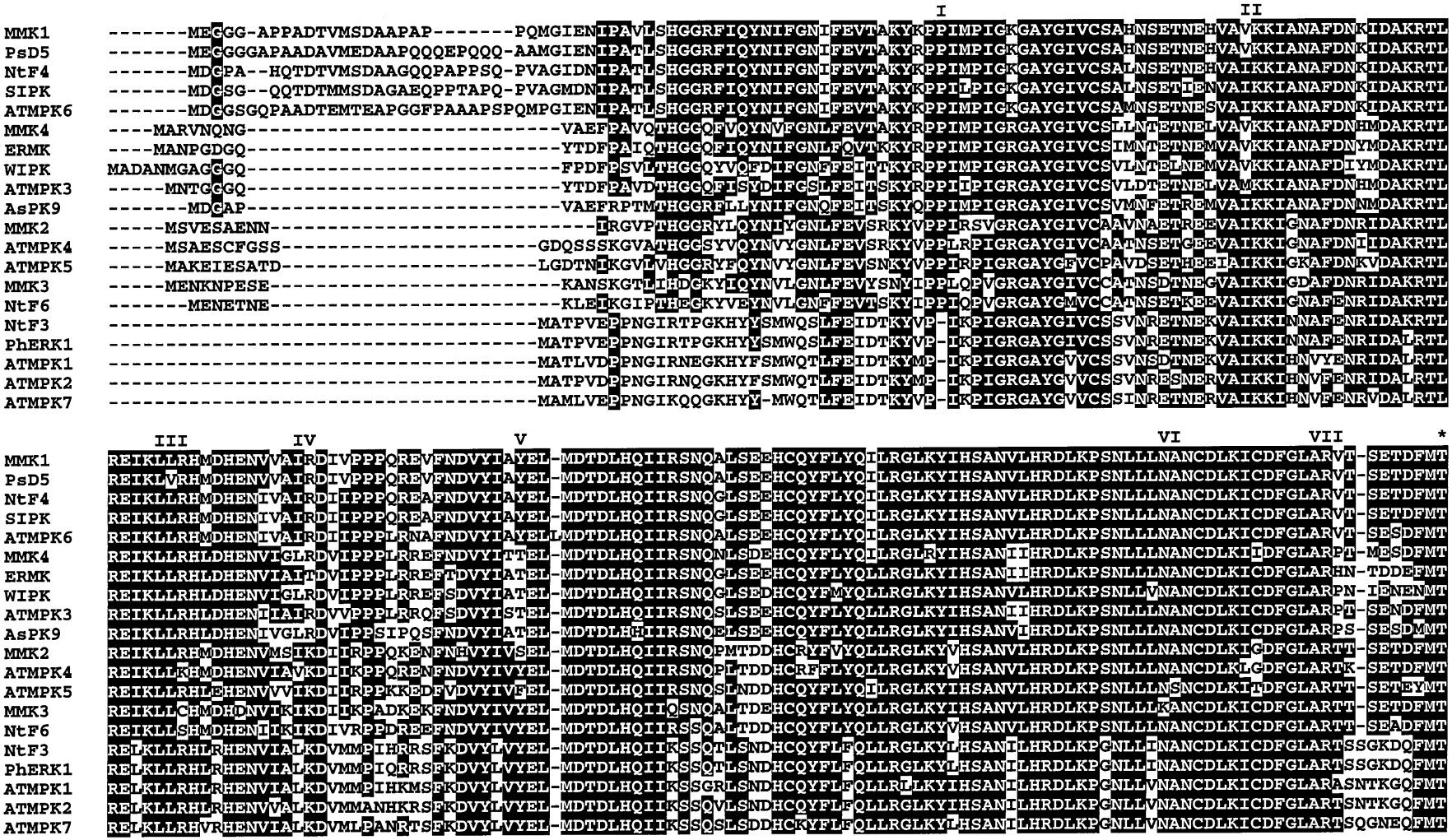

Fig. 2 Alignment of predicted amino acid sequences of plant MAPKs from alfalfa (MMK1 [3, 4], MMK2 [5], MMK3, MMK4 [6]), *Arabidopsis* (ATMPK1-7 [7, 8]), *Avena* (AsPK9 [9]), parsley (ERMK [10]), pea (PsD5 [11]), petunia (PhERK1 [12]) and tobacco (Ntf3 [13], Ntf4, Ntf6 [15], WIPK [14] and SIPK [16]). Amino acids are shown in the single letter code. Identical amino acids are shown in black. For optimal alignment, gaps were introduced and are shown by dashes.

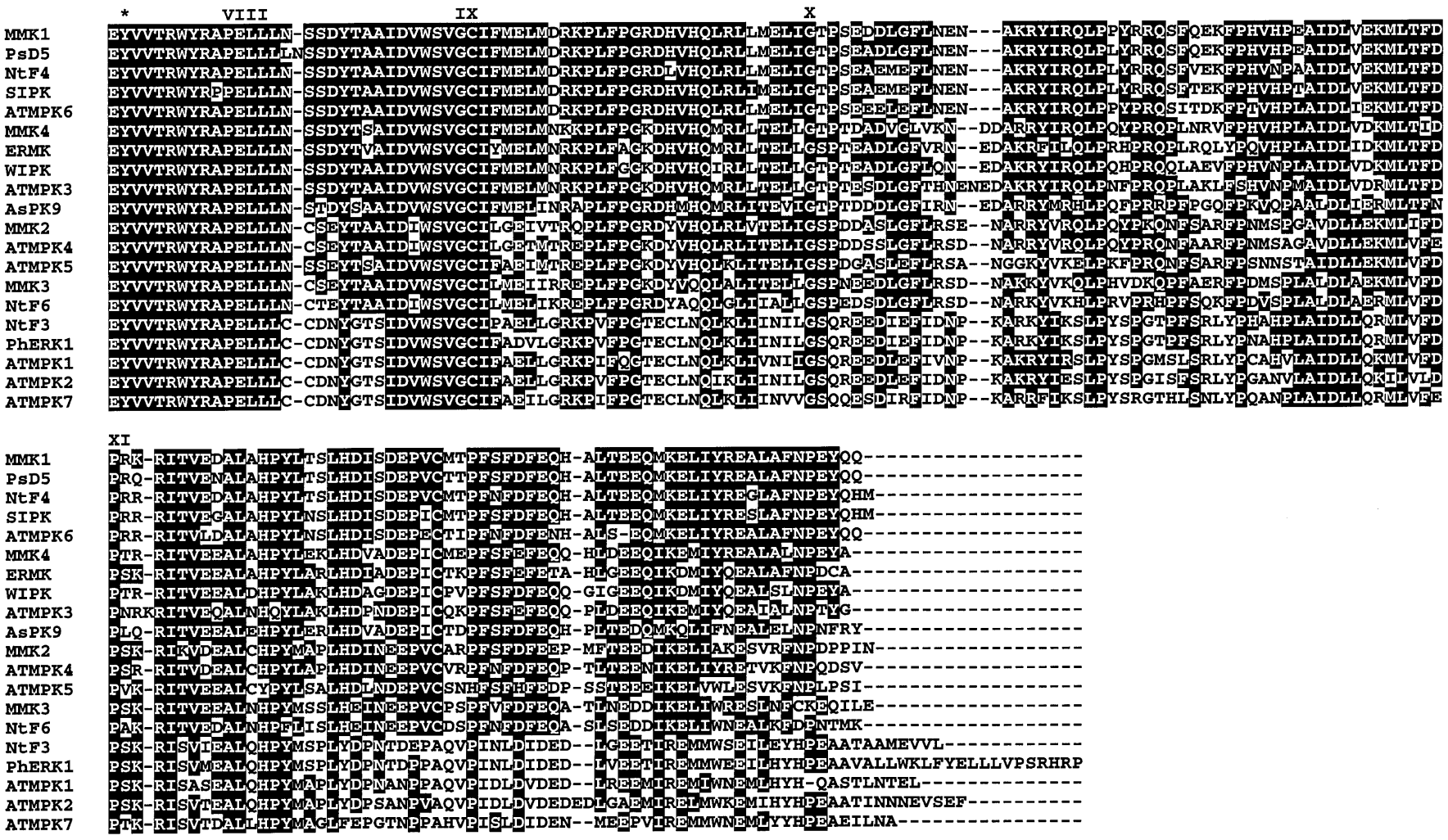

Figure 2. (Continued).

ABA influences many processes in plant physiology, such as embryo development, seed germination and abiotic stress responses, including adaptation to drought and salt stress. ABA is able to induce a MAPK-like activity in barley aleurone protoplasts [31]. Although the MAPK in barley aleurone cells has yet to be isolated and characterized, these results suggest that MAPK pathways may mediate hormone signaling in a tissue-specific way. Besides inducing specific genes, ABA is known to inhibit giberellic acid (GA)-induced effects in aleurone cells, such as the expression of hydrolytic enzymes, stimulation of protein synthesis and breakdown of storage reserves. Whereas ABA stimulates MAPK activation in aleurone cells, GA may do the reverse, as indicated by the negative effect of GA on transcript accumulation of a MAPK gene in oat aleurone cells [9].

Most plant cell cultures require auxins for proliferation, which suggests that auxin may act as a mitogen under certain conditions. Whereas auxin starvation arrests cell division in a tobacco cell suspension culture, readdition starts the cell cycle. During this process, a MAPKK and a protein kinase that has the properties of a MAPK are activated, suggesting that a MAPK pathway is involved in signal transduction of auxin [8].

Genetic analysis of the ethylene pathway in *Arabidopsis* indicates the possible involvement of a MAP kinase module. A number of mutants have been isolated that show a constitutive triple response (CTR) in the absence of ethylene. The *ctr1* mutant was isolated, and the affected gene cloned and analyzed [32]. The encoded CTR1 protein was found to be similar to mammalian Raf kinase, an upstream activator of MAPKKs. Upstream components in the CTR1 pathway appear to be the plasma membrane-located ethylene receptors ETR1 and ERS [33]. The *etr1* gene was isolated in a mutant screen for ethylene insensitivity and encodes a protein with homology to two-component sensor regulator proteins that are well-known signaling transducers in bacteria. Bacterial two-component systems are responsible for regulating a variety of processes, including chemotaxis, sporulation, osmolarity and nutrient availability. The sensor is usually a membrane-located kinase that autophosphorylates on a histidine residue upon receiving the extracellular signal. The regulator then becomes phosphorylated on an aspartate residue, obtaining the phosphate group from the histidine of the sensor. The regulator can either be a separate protein or part of a hybrid sensor-regulator kinase. The ETR1 protein is a hybrid sensor-regulator kinase and resembles the SLN1 protein that is responsible for signaling hyperosmotic stress via the HOG1 pathway in yeast [2]. Following stimulation, the SLN1 hybrid kinase autophosphorylates and transfers the phosphate to an aspartic acid residue of the SSK1 protein that resembles bacterial regulators. Through activation of the MAPKKKs SSK2 and SSK22, the SSK1 protein activates the HOG1 pathway, culminating in the expression of genes involved in glycerol synthesis and adaptation to hyperosmolar stress. Because two-component systems have an inbuilt short-term inactivation mechanism, these signaling components are optimally suited to respond to rapid changes of extracellular signals, and may be found in many more eukaryotic pathways than anticipated.

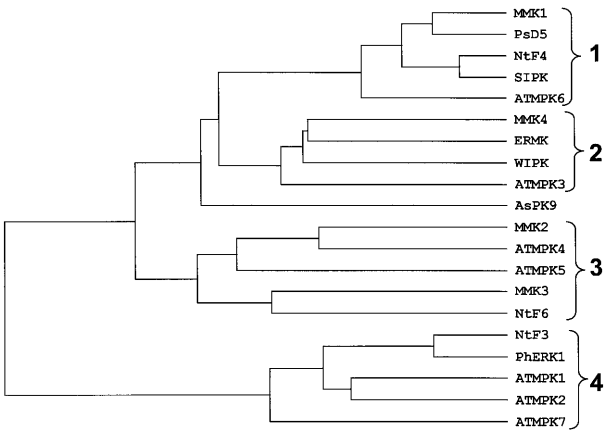

Figure 3. Phylogenetic tree constructed from plant MAPKs from alfalfa (MMK1 [3, 4], MMK2 [5], MMK3, MMK4 [6]), *Arabidopsis* (ATMPK1-7 [7, 8]), *Avena* (AsPK9 [9]), parsley (ERMK [10]), pea (PsD5 [11]), petunia (PhERK1 [12]) and tobacco (Ntf3 [13], Ntf4, Ntf6 [15], WIPK [14] and SIPK [16]).

Concluding remarks

Over the last few years we have seen a surge of results claiming an involvement of MAP kinases in a variety of signaling processes in plants (table 1). In many cases, however, only indirect proof was provided, and the responsible protein kinase/gene was not identified. Without these results, we should keep in mind that other types of protein kinases share many of the properties of MAP kinases (similar substrate specificities, similar sizes etc.). Therefore, it will be essential to obtain the proper tools to identify the specific MAP kinases and isolate the respective genes encoding the enzymes. For these purposes, biochemical and genetic approaches will be essential and should equally contribute to study the function of MAPK pathways in different processes.

Assuming that the present evidence will hold, how is it that so many signals can be transmitted by MAPK pathways? From a theoretical standpoint, there surely

Table 1. Evidence for involvement of MAP kinases in plant signal transduction.

Stimulus	Species	System	Protein/gene	Results	Ref.
Cold	*Arabidopsis thaliana*	in vitro-grown plants	*ATMPK3*	induction of *ATMPK3* transcripts	17
	Medicago sativa	soil-grown plants	$p44^{MMK4}$	induction of MMK4 protein kinase activity induction of *MMK4* transcripts constant MMK4 protein level	6
Drought	*Arabidopsis thaliana*	in vitro-grown plants	*ATMPK3*	induction of *ATMPK3* transcripts	19
	Medicago sativa	in vitro-grown plants	$p44^{MMK4}$	induction of MMK4 protein kinase activity induction of *MMK4* transcripts constant MMK4 protein level	6
Salt stress	*Arabidopsis thaliana*	in vitro-grown plants	*ATMPK1*	induction of *ATMPK1* transcripts	17
			ATMPK3	induction of *ATMPK3* transcripts	17
Touch	*Arabidopsis thaliana*	in vitro-grown plants	*ATMPK3*	induction of *ATMPK3* transcripts	17
	Medicago sativa	leaves suspension culture	$p44^{MMK4}$	induction of MMK4 protein kinase activity	18
Wounding	*Lycopersicon esculentum*	plants	p48	induction of MBP protein kinase activity	22
	Medicago sativa	leaves	$p44^{MMK4}$	induction of MMK4 protein kinase activity induction of *MMK4* transcripts constant MMK4 protein level	23
	Nicotiana tabacum	leaf disc	$p46^{PMSAP}$	induction of MBP protein kinase activity	21
		plants	p47	induction of MBP protein kinase activity induction of *WIPK* transcripts silencing of endogenous *WIPK* gene	14
Bacterial elicitor	*Nicotiana tabacum*	leaves	p49	induction of MBP protein kinase activity	30
Fungal elicitor	*Nicotiana tabacum*	suspension culture	$p48^{SIPK}$	induction of MBP protein kinase activity induction of SIP kinase activity constant SIPK protein level constant *SIPK* transcript level	28
	Nicotiana tabacum	suspension culture	p44	induction of MBP protein kinase activity	28
	Nicotiana tabacum	suspension culture	p40	induction of MBP protein kinase activity	28
	Nicotiana tabacum	suspension culture	p48	induction of MBP protein kinase activity	27
	Peteroselinum crispum	suspension culture	$p45^{ERMK}$	induction of MBP protein kinase activity induction of ERMK protein kinase activity induction of *ERMK* transcripts nuclear translocation	10
Chitosan	*Lycopersicon esculentum*	plants	p48	induction of MBP protein kinase activity	22
	Nicotiana tabacum	leaf disc	$p46^{PMSAPK}$	induction of MBP protein kinase activity	21
PGA	*Lycopersicon esculentum*	plants	p48	induction of MBP protein kinase activity	22
SA	*Nicotiana tabacum*	suspension culture	$p48^{SIPK}$	induction of SIP kinase activity	16
		leaf disc	$p46^{PMSAPK}$	induction of MBP protein kinase activity	21
GA	*Avena sativa*	aleurone cells	*AsPK9*	downregulation of *AsPK9* transcripts	9
ABA	*Hordeum vulgare*	aleurone protoplasts		induction of phosphptyrosine/ERK1 protein kinase	31
Ethylene	*Arabidopis thaliana*	mutant	*CTR1*	negative regulator of ethylene response	32

Table 1. (Continued).

Stimulus	Species	System	Protein/gene	Results	Ref.
Auxin	*Nicotiana tabacum*	suspension culture	p46	induction of MBP protein kinase activity	8
Pollen development	*Nicotiana tabacum*	pollen	$p45^{Ntf4}$	induction of Ntf4 protein kinase activity	19
				induction of *Ntf4* transcripts	
Pollen germination	*Nicotiana tabacum*	pollen	$p45^{Ntf4}$	increase in Ntf4 protein level	
				induction of Ntf4 protein kinase activity constant Ntf4 protein level	19

are enough sufficiently different MAPK genes in the plant genome to assign each of these genes a specific role to the presently identified pathways. However, experience from a growing number of investigations of mammalian cells tells us that things might be much more complicated. This is shown by the fact that a given extracellular signal mostly does not only activate a single but several independent pathways. A single pathway may also be activated by a number of other unrelated signals. The same signal may activate different pathways in different cells. Last but not least, the activation of a pathway is not an all-or-none process, but can be a transient or constitutive event and may differ in amplitude. Changing only one of the above parameters has been shown to dramatically affect the outcome of the cellular response. Contemplating along these lines, signal transduction is likely to be a question of pathway combinatorics, and the responses at the chromatin level may depend on which of the many protein kinase pathways are active and to what extent at a given moment.

Note added in proof. The following publications have appeared in the meantime:

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Zhang S. Q. and Klessig D. (1998) The tobacco wounding-activated mitogen-activated protein kinase is encoded by *SIPK*. Proc. Natl. Acad. Sci. USA **95:** 7225–7230

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