



Supplemental Material to:

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**Evolutionary history of mitogen-activated protein kinase
(MAPK) genes in Lotus, Medicago, and Phaseolus**

Plant Signaling & Behavior 2013; 8(11)

<http://dx.doi.org/10.4161/psb.27189>

www.landesbioscience.com/journals/psb/article/27189

Figure S1. Maximum Parsimony analysis of MAPKs from four legume and three non-legume species.

The values above the branches represent bootstrap support of 2000 replicates. The members with phosphorylation motif TEY form clade A, B and C and those with TDY motif form clade D. Some members with the TEY motif and potentially legume specific TQY (denoted by *) and one of the gene members of soybean with TVY (denoted by **) motif are nested in clade E and B, respectively. The MAPK gene models were accepted for phylogenetic analysis using protein sequences of Serine/Threonine kinase subfamily having conserved Aspartate and Lysine residues in their catalytic domain with (D[L/I/V]K) motif and TXY phosphorylation motif in their activation loop.

Figure S2. Phylogram showing the Maximum Likelihood analysis of MAPKs of four legumes (Soybean, *Lotus*, *Medicago* and *Phaseolus*) and their orthologs in three non-legumes (*Arabidopsis*, rice and poplar).

The values above the branches are bootstrap support of 100 replicates. The JTT+G+I evolutionary model was employed to perform Maximum Likelihood analysis. The members with phosphorylation motif TEY are nested in clade A, B and C, those with the TDY motif in clade D, and members with legume specific TQY (denoted by *) and one of the gene members of soybean with TVY (denoted by **) motif in clade E and B, respectively. The MAPK gene models were accepted for phylogenetic analysis using protein sequences of Serine/Threonine kinase subfamily having conserved Aspartate and Lysine residues in their catalytic domain with (D[L/I/V]K) motif and TXY phosphorylation motif in their activation loop.

Figure S3. Gene structure showing intron/exon distribution in MAPK gene members.

The gene structure for all the MAPK gene members used in the analyses were constructed to visualize the evolutionary trend in the homologs of MAPKs in legume and non-legume species.

Figure S4. Maximum Parsimony analysis of MAPKKs from four legume and three non-legume species.

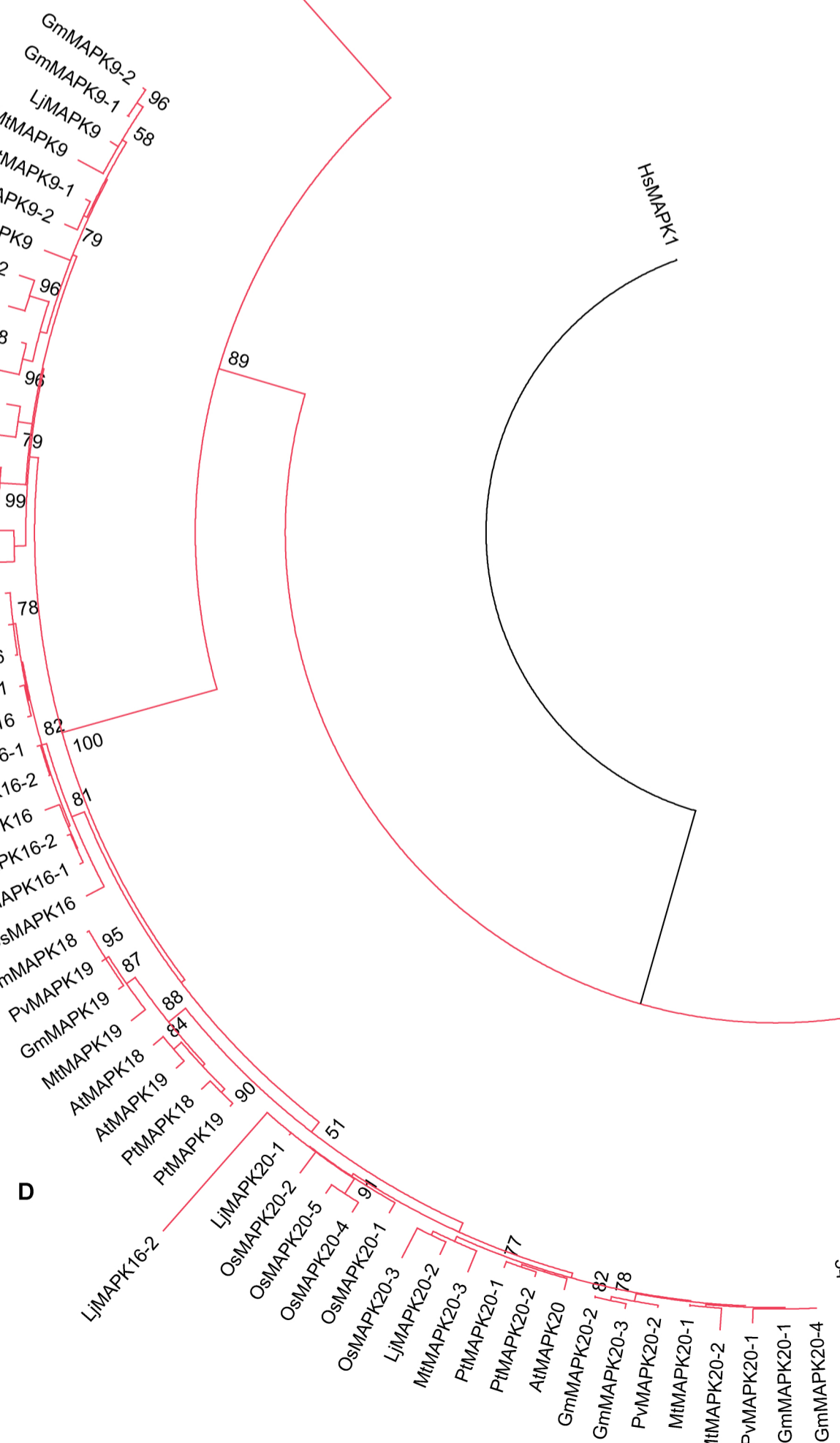
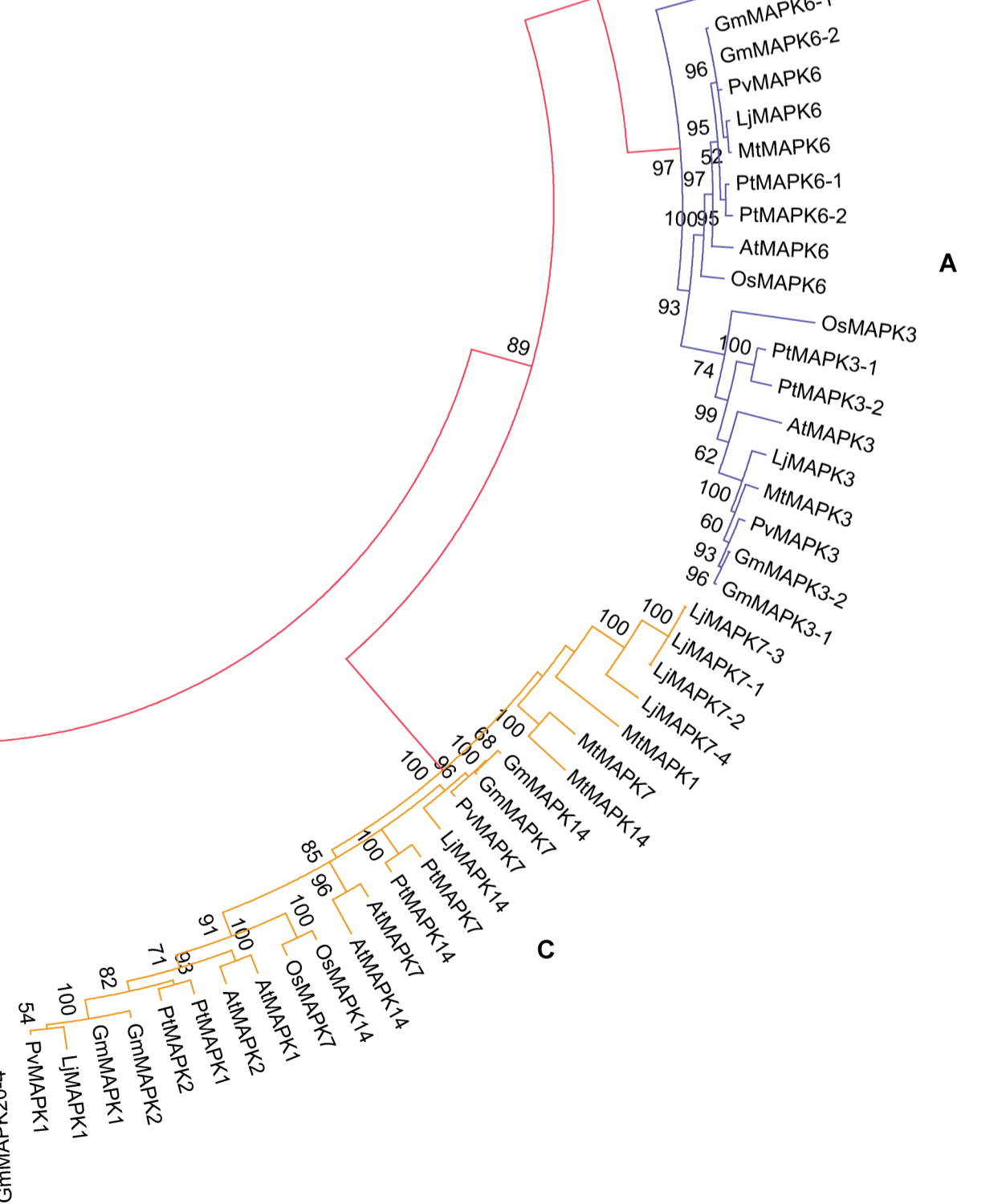
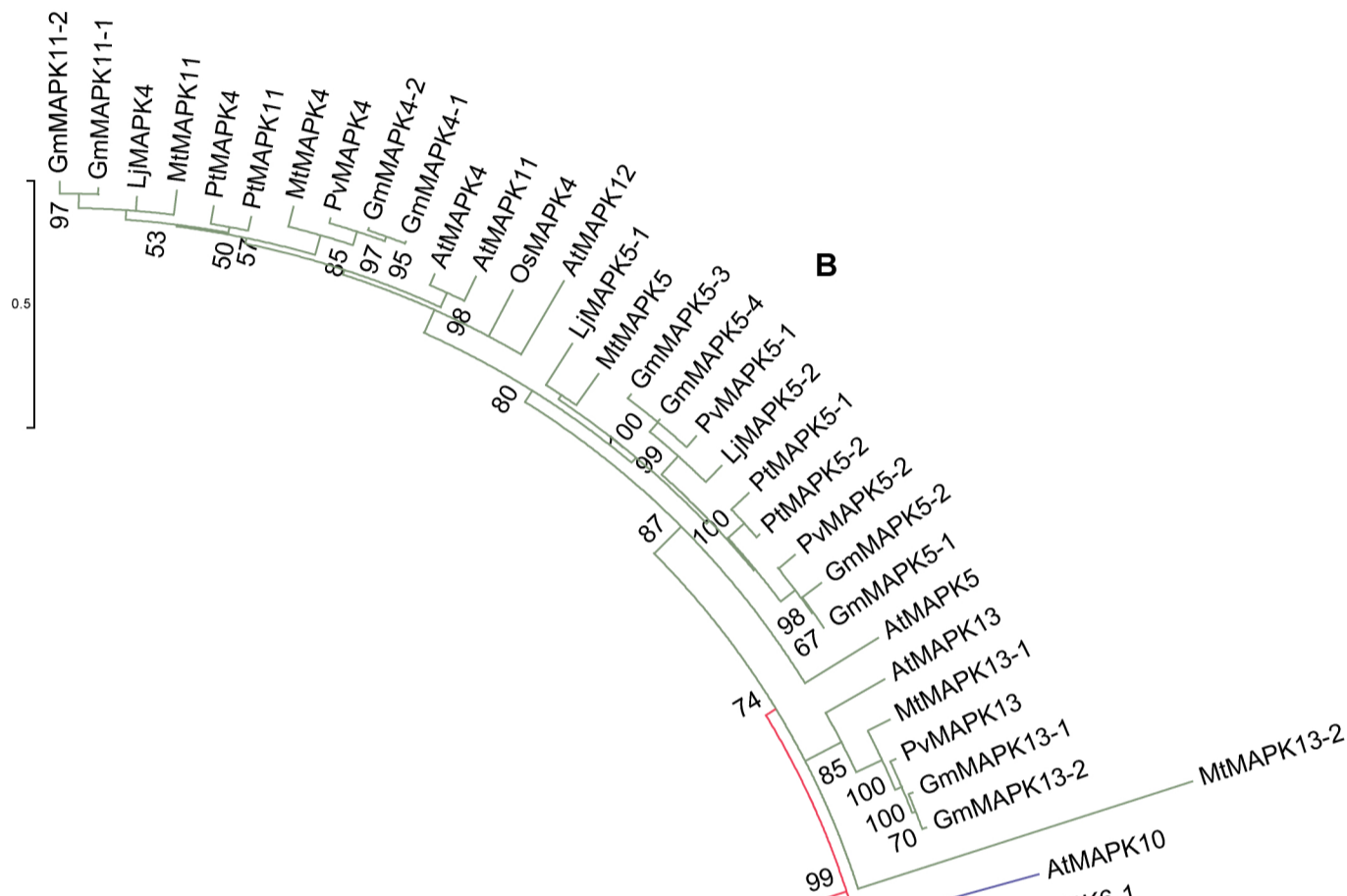
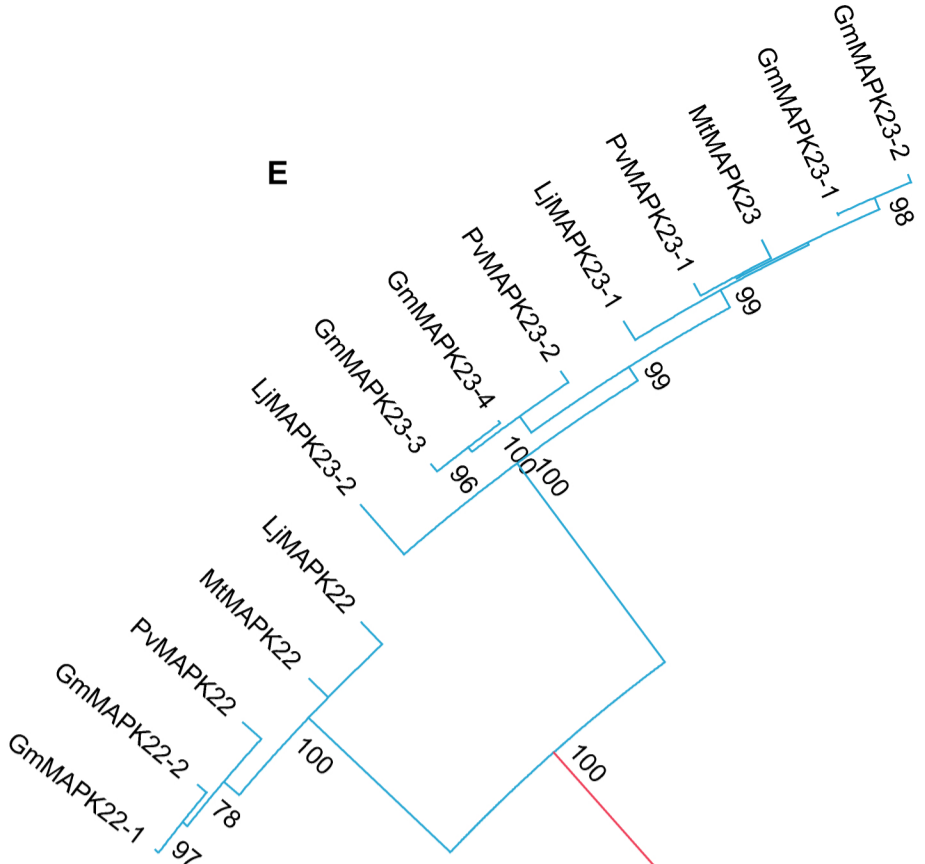
The values above the branches represent bootstrap support of 2000 replicates. The MAPKK gene models were accepted for phylogenetic analysis using dual-specificity protein kinases having conserved Aspartate and Lysine residues in their catalytic domain with (D[L/I/V]K) motif and S-X5-T phosphorylation motif along their activation loop.

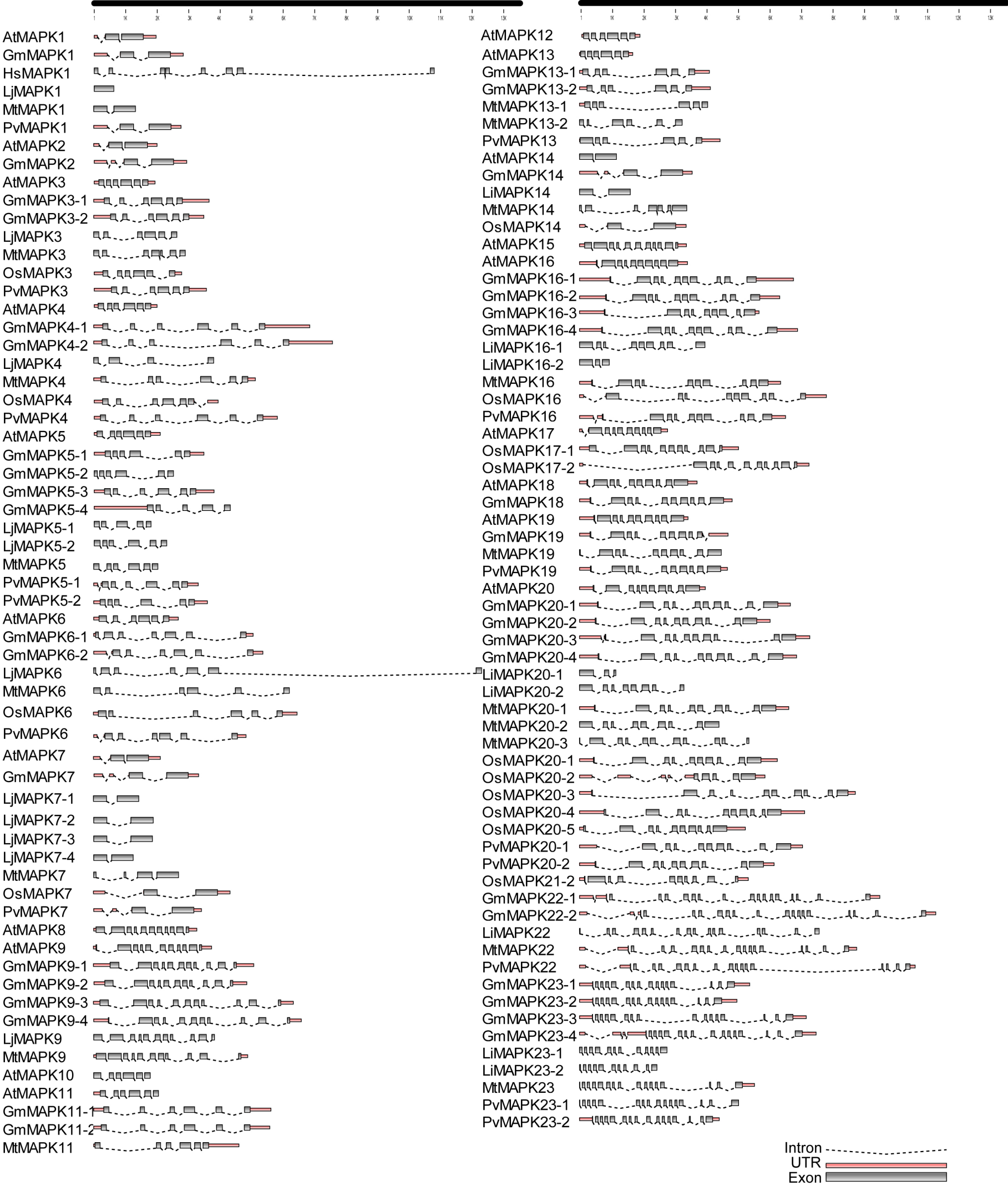
Figure S5. Maximum Likelihood analysis of GmMAPKKs and their orthologs in *Arabidopsis*, poplar and rice.

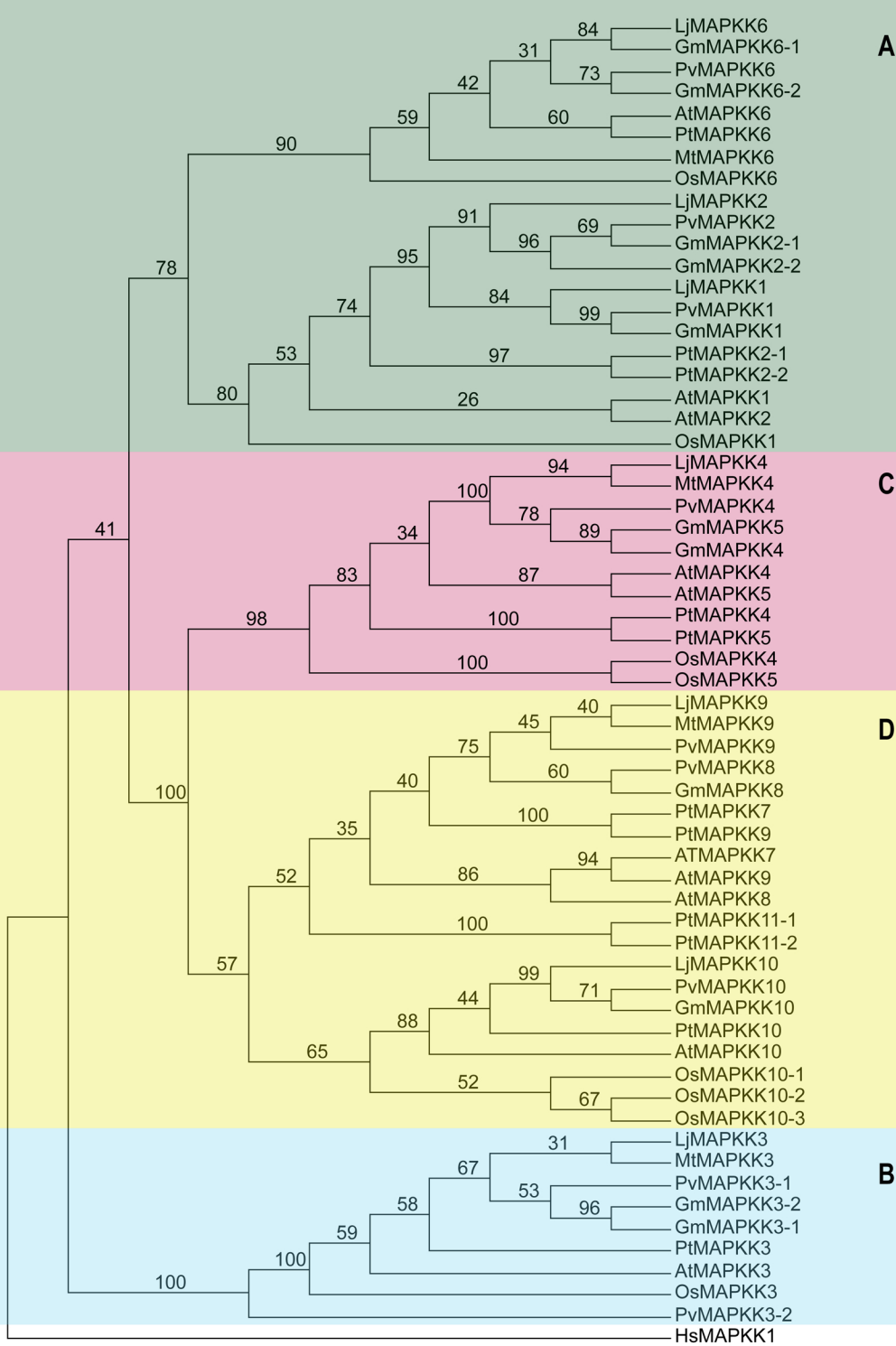
In the ML phylogram, the values above the branches are bootstrap support of 100 replicates. The JTT+G+I evolutionary model was employed to perform Maximum Likelihood analysis. The MAPKK gene models were accepted for phylogenetic analysis using dual-specificity protein

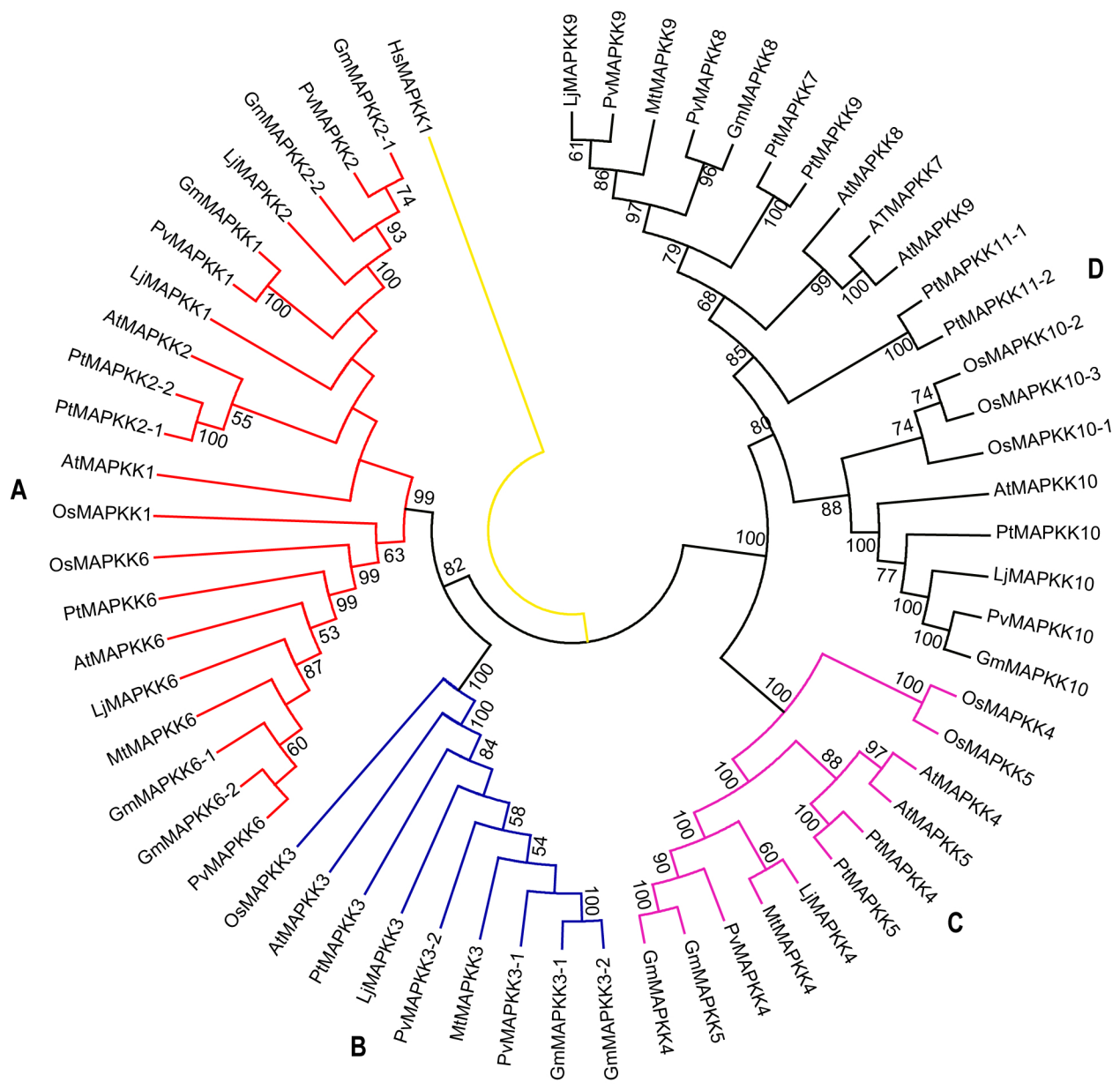
kinases having conserved Aspartate and Lysine residues in their catalytic domain with (D[L/I/V]K) motif and S-X5-T phosphorylation motif along their activation loop.

Table S1. MAPK gene nomenclature and homology assessment in Glycine, Lotus, Medicago and Phaseolus based on their sequence identity, genetic distance and phylogenetic placement.









Additional file 3 – MAPK gene nomenclature and homology assessment in *Glycine* , *Lotus* , *Medicago* and *Phaseolus* based on their sequence identity, genetic distance and phylogenetic placement

MAPK	MAPK new name	Orthologs	% Identity
<i>Glycine max</i>			
Glyma04g03210	GmMAPK1	AtMAPK1	85
Glyma06g03270	GmMAPK2	AtMAPK2	84
Glyma11g15700	GmMAPK3-1	AtMAPK3	81.9
Glyma12g07770	GmMAPK3-2	AtMAPK3	81.4
Glyma07g07270	GmMAPK4-1	AtMAPK4	86
Glyma16g03670	GmMAPK4-2	AtMAPK4	85
Glyma01g43100	GmMAPK5-1	AtMAPK5	76.2
Glyma11g02420	GmMAPK5-2	AtMAPK5	75
Glyma08g02060	GmMAPK5-3	AtMAPK5	75
Glyma05g37480	GmMAPK5-4	AtMAPK5	74
Glyma07g32750	GmMAPK6-1	AtMAPK6	87.6
Glyma02g15690	GmMAPK6-2	AtMAPK6	87.6
Glyma05g28980	GmMAPK7	AtMAPK7	85
Glyma05g33980	GmMAPK9-1	AtMAPK9	82.7
Glyma08g05700	GmMAPK9-2	AtMAPK9	81.9
Glyma07g11470	GmMAPK9-3	AtMAPK9	79
Glyma09g30790	GmMAPK9-4	AtMAPK9	79
Glyma18g47140	GmMAPK11-1	AtMAPK11	92
Glyma09g39190	GmMAPK11-2	AtMAPK11	92
Glyma12g07850	GmMAPK13-1	AtMAPK13	78
Glyma11g15590	GmMAPK13-2	AtMAPK13	78
Glyma08g12150	GmMAPK14	AtMAPK14	80
Glyma13g28120	GmMAPK16-1	AtMAPK16	80
Glyma15g10940	GmMAPK16-2	AtMAPK16	80
Glyma17g02220	GmMAPK16-3	AtMAPK16	76
Glyma07g38510	GmMAPK16-4	AtMAPK16	76
Glyma15g38490	GmMAPK18	AtMAPK18	70
Glyma13g33860	GmMAPK19	AtMAPK19	68
Glyma18g12720	GmMAPK20-1	AtMAPK20	73
Glyma14g03190	GmMAPK20-2	AtMAPK20	71
Glyma02g45630	GmMAPK20-3	AtMAPK20	71

Glyma08g42240	GmMAPK20-4	AtMAPK20	73
Glyma03g21610	GmMAPK22-1	No At ortholog	95 with GmMAPK22
Glyma16g10820	GmMAPK22-2	No At ortholog	95 with GmMAPK22
Glyma01g35190	GmMAPK23-1	No At ortholog	>80 with GmMAPK23
Glyma09g34610	GmMAPK23-2	No At ortholog	>80 with GmMAPK23
Glyma16g08080	GmMAPK23-3	No At ortholog	>80 with GmMAPK23
Glyma16g17580	GmMAPK23-4	No At ortholog	>80 with GmMAPK23

Lotus Japonicus

LjSGA_048326.1	LjMAPK1	AtMAPK1	84.4
chr3.CM0616.20.r2.d	LjMAPK3	AtMAPK3	82.4
chr1.LjT10B06.60.r2.d	LjMAPK4	AtMAPK4	89.5
LjSGA_018738.1	LjMAPK5-1	AtMAPK5	72.8
chr4.CM0004.850.r2.d	LjMAPK5-2	AtMAPK5	75.5
chr4.CM0432.3930.r2.a	LjMAPK6	AtMAPK6	78.3
chr4.CM0432.3950.r2.a	LjMAPK7-1	AtMAPK7	72.8
chr4.CM0432.3740.r2.d	LjMAPK7-2	AtMAPK7	72.6
chr4.CM0432.3910.r2.a	LjMAPK7-3	AtMAPK7	72.6
chr4.CM0432.3920.r2.a	LjMAPK7-4	AtMAPK7	70.8
chr4.CM0042.2140.r2.m	LjMAPK9	AtMAPK9	79.9
chr4.CM0046.800.r2.a	LjMAPK14	AtMAPK14	79.7
chr6.CM0139.840.r2.m	LjMAPK16-1	AtMAPK16	79.4
chr4.CM0387.480.r2.d	LjMAPK16-2	AtMAPK16	72.1
chr6.CM0836.450.r2.d	LjMAPK20-1	AtMAPK20	90.3
chr2.CM0099.20.r2.d	LjMAPK20-2	AtMAPK20	82.2
chr3.CM0423.40.r2.a	LjMAPK22	GmMAPK22	81.3
LjSGA_012212.1	LjMAPK23-1	GmMAPK23	86.7
LjSGA_018456.1	LjMAPK23-2	GmMAPK23	76.7

Medicago truncatula

Medtr3g060330.1	MtMAPK1	AtMAPK1	70.2
Medtr4g061130.1	MtMAPK3	AtMAPK3	77.3
Medtr7g038040.1	MtMAPK4	AtMAPK4	82.2
Medtr5g010030.1	MtMAPK5	AtMAPK5	73.2
Medtr4g087620.1	MtMAPK6	AtMAPK6	88.4
Medtr8g086000.1	MtMAPK7	AtMAPK7	71.4

Medtr8g106960.1	MtMAPK9	AtMAPK9	78.2
Medtr7g078690.1	MtMAPK11	AtMAPK11	82.7
Medtr4g061320.1	MtMAPK13-1	AtMAPK13	78.5
Medtr4g061330.1	MtMAPK13-2	AtMAPK13	56.3
Medtr8g086010.1	MtMAPK14	AtMAPK14	53.7
Medtr2g021330.1	MtMAPK16	AtMAPK16	79.1
AC235677_37.1	MtMAPK19	AtMAPK19	69.5
Medtr3g089510.1	MtMAPK20-1	AtMAPK20	72.3
AC225528_53.1	MtMAPK20-2	AtMAPK20	69.2
Medtr5g091680.1	MtMAPK20-3	AtMAPK20	71.5
Medtr8g012450.1	MtMAPK22	GmMAPK22	88
Medtr5g026960.1	MtMAPK23	GmMAPK23	88

Phaseolus vulgaris

Phvul.009G061000.1	PvMAPK1	AtMAPK1	85.7
Phvul.011G071400.1	PvMAPK3	AtMAPK3	82.7
Phvul.010G102800.1	PvMAPK4	AtMAPK4	84.6
Phvul.002G292400.1	PvMAPK5-1	AtMAPK5	75.3
Phvul.002G159500.1	PvMAPK5-2	AtMAPK5	74.2
Phvul.003G059500.1	PvMAPK6	AtMAPK6	87.6
Phvul.002G237500.1	PvMAPK7	AtMAPK7	85.6
Phvul.011G070800.1	PvMAPK13	AtMAPK13	78.5
Phvul.003G095900.1	PvMAPK16	AtMAPK16	77.3
Phvul.005G069800.1	PvMAPK19	AtMAPK19	71
Phvul.006G033600.1	PvMAPK20-1	AtMAPK20	70.6
Phvul.008G191500.1	PvMAPK20-2	AtMAPK20	70.5
Phvul.010G023600.1	PvMAPK22	GmMAPK22	92.9
Phvul.002G131400.1	PvMAPK23-1	GmMAPK23	92.5
Phvul.003G279600.1	PvMAPK23-2	GmMAPK23	90

MAPKK

MAPKK new name Orthologs

%Identity

Glycine max

Glyma15g18860	AtMAPKK1	GmMAPKK1	64
Glyma13g16650	AtMAPKK2	GmMAPKK2-1	67
Glyma17g06020	AtMAPKK2	GmMAPKK2-2	67
Glyma05g08720	AtMAPKK3	GmMAPKK3-1	80

Glyma19g00220	AtMAPKK3	GmMAPKK3-2	78
Glyma07g00520	AtMAPKK4	GmMAPKK4	68
Glyma08g23900	AtMAPKK5	GmMAPKK5	72
Glyma10g15850	AtMAPKK6	GmMAPKK6-1	89
Glyma02g32980	AtMAPKK6	GmMAPKK6-2	85
Glyma09g30310	AtMAPKK8	GmMAPKK8	44
Glyma01g01980	AtMAPKK10	GmMAPKK10	54

Lotus Japonicus

chr6.CM0013.1740.r2.d	LjMAPKK1	AtMAPKK1	45.8
chr4.CM0297.320.r2.m	LjMAPKK2	AtMAPKK2	67
chr2.CM0263.270.r2.m	LjMAPKK3	AtMAPKK3	77.4
chr3.LjB21L17.150.r2.a	LjMAPKK4	AtMAPKK4	66.1
chr2.CM0065.640.r2.d	LjMAPKK6	AtMAPKK6	82.2
LjSGA_014749.1.1	LjMAPKK9	AtMAPKK9	57.4
LjSGA_023606.1	LjMAPKK10	AtMAPKK10	49.5

Medicago truncatula

Medtr6g005210.1	MtMAPKK3	AtMAPKK3	77.5
Medtr4g005830.1	MtMAPKK4	AtMAPKK4	67.2
Medtr6g090470.1	MtMAPKK6	AtMAPKK6	61.6
Medtr6g071280.1	MtMAPKK9	AtMAPKK9	56

Phaseolus vulgaris

Phvul.009G229800.1	PvMAPKK1	AtMAPKK1	61.2
Phvul.003G134100.1	PvMAPKK2	AtMAPKK2	64.8
Phvul.004G010400.1	PvMAPKK3-1	AtMAPKK3	78.6
Phvul.007G112700.1	PvMAPKK3-2	AtMAPKK3	72.7
Phvul.010G163000.1	PvMAPKK4	AtMAPKK4	66.6
Phvul.004G174500.1	PvMAPKK6	AtMAPKK6	86.8
Phvul.004G147900.1	PvMAPKK8	AtMAPKK8	41.1
Phvul.004G147800.1	PvMAPKK9	AtMAPKK9	57.3
Phvul.002G138400.1	PvMAPKK10	AtMAPKK10	49.4