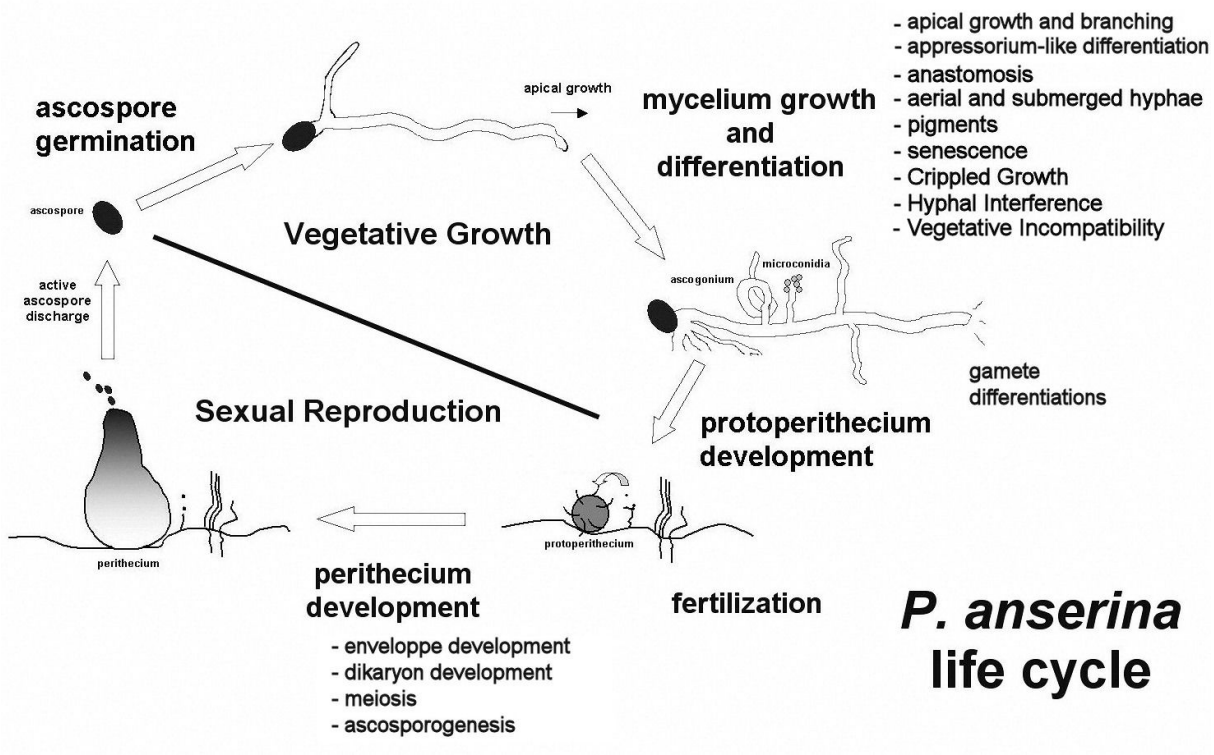

Figure S1 Major developmental steps of the *P. anserina* life cycle and features that have been assessed in the MAPK mutants.

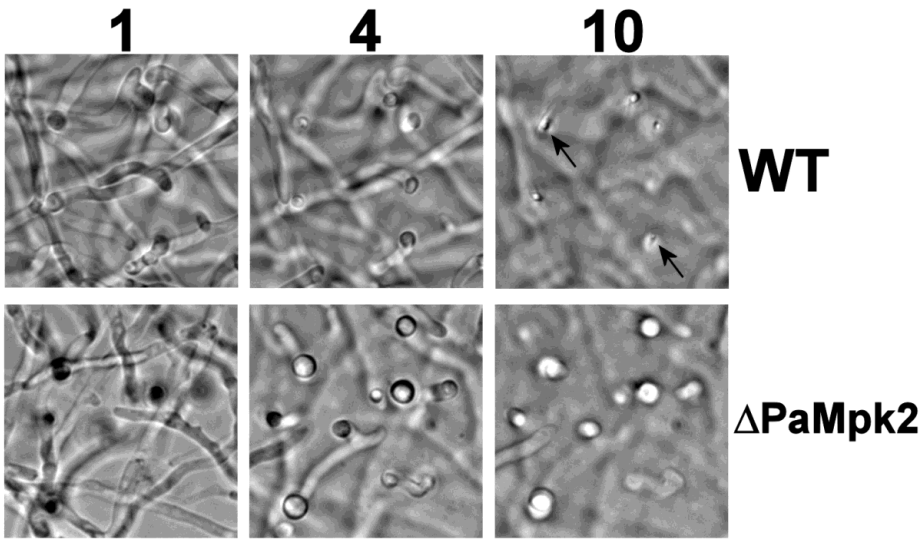

Figure S2 Lack of appressorium-like structures in the *ΔPaMpk2* mutants. Mycelia of the indicated strains were grown on cellophane overlaying M2 medium. Mycelium was observed after two days of growth at different focal plans. **1**, hyphae running parallel to the cellophane are observed. **4**, corresponds to a plan 4 mm below that of **1**; hyphae had reoriented in both wild type and the *ΔPaMpk2* mutants and made bulging contacts with cellophane. **10**, i.e., 10 μm below plan 1, needle like hyphae (arrows) have breached cellophane in wild type but not in mutants.

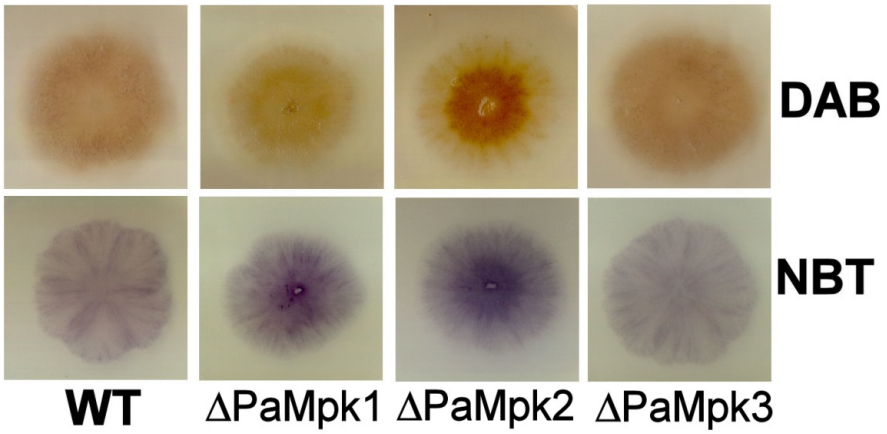

Figure S3 DAB and NBT staining. Assays were performed on three-day old mycelia.

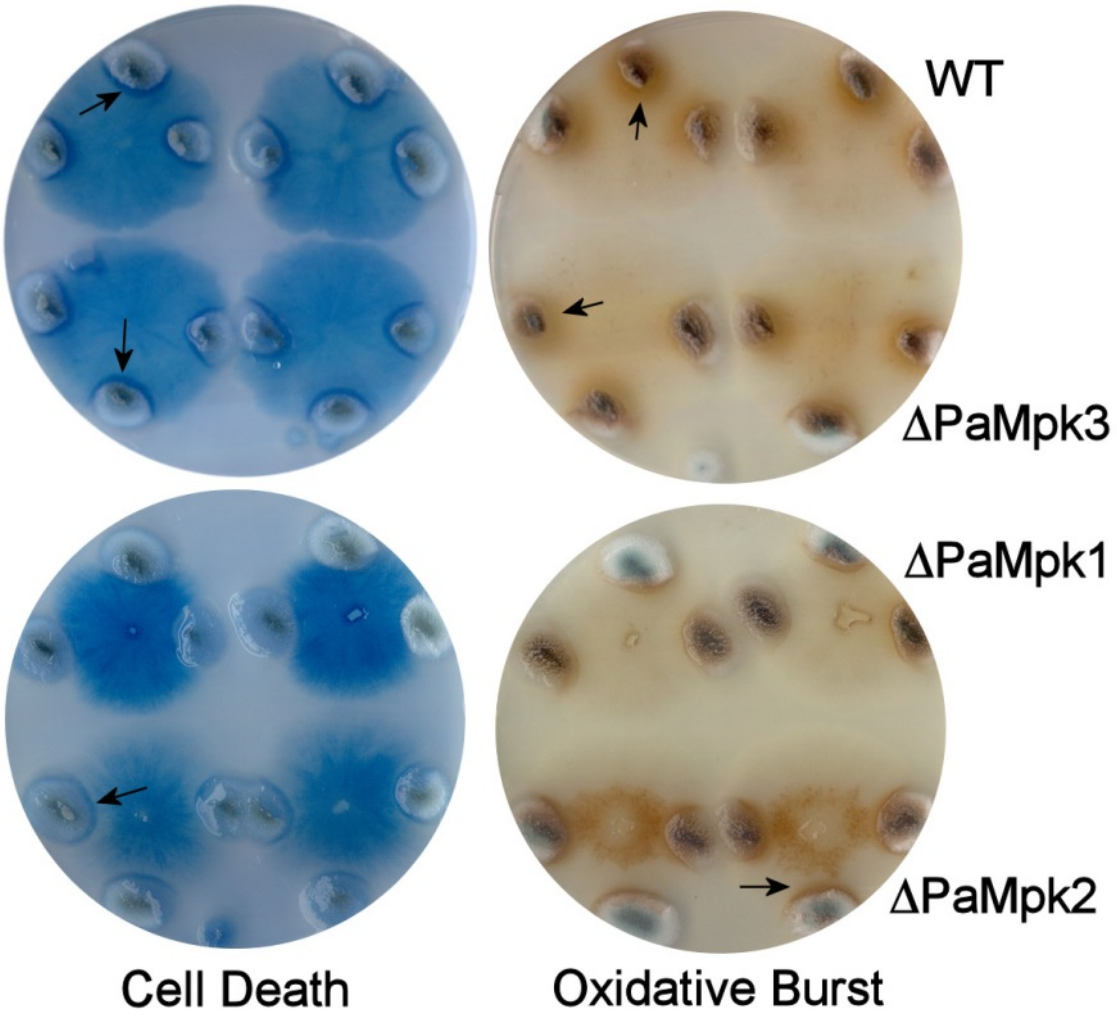

Figure S4 Hyphal Interference in the MAPK mutants. Cell death and Oxidative Burst were revealed by the accumulation of Trypan blue and precipitation of DAB, respectively, at the confrontation between *P. anserina* and *Penicillium chrysogenum*. For each strain, two mycelia of *P. anserina* were inoculated with three neighbouring *P. chrysogenum* thalli. After three days of growth, the mycelia of the two competing species confront for at least 24 hours at which time the assays begin. Dead cells and oxidative burst (arrows) are clearly visible on wild type (WT) and the *ΔPaMpk3* mutants, while they are reduced on the *ΔPaMpk2* mutants and completely abolished in the *ΔPaMpk1* ones. Note that as previously described (4) the *ΔPaMpk1* mutant exhibit numerous dead cell all over the thallus.

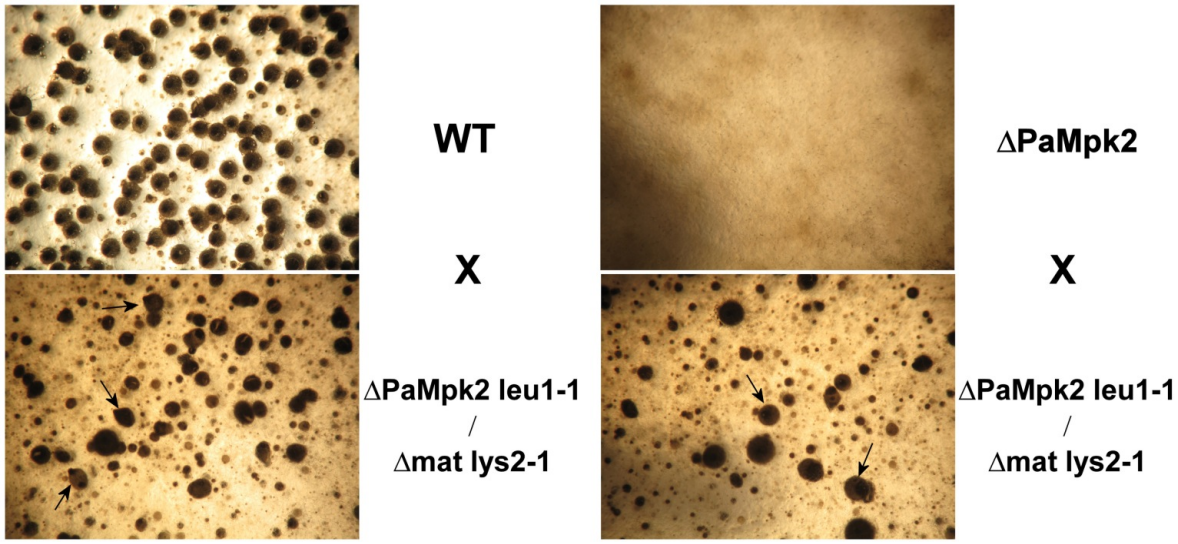

Figure S5 Fertility rescue in heterokaryons. *ΔPaMpk2 leu1-1/Δmat lys2-1* heterokaryons were crossed with wild type (WT) or *ΔPaMpk2* mutants. When wild type was the female parent (top left), numerous normal perithecia matured. When *ΔPaMpk2* mutants were the female parent, no perithecium was differentiated (Top right). When the heterokaryons were the female parent (bottom), a few normal-looking ascospore-producing perithecia (arrows) were obtained among numerous abnormal looking ones in both types of cross.

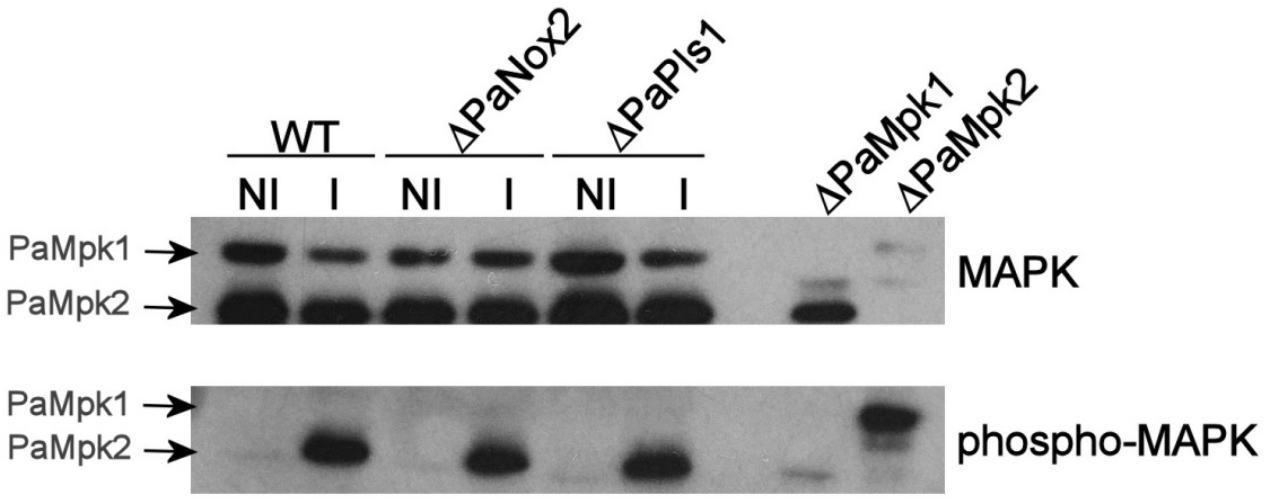

Figure S6 Phosphorylation of PaMpk1 and PaMpk2 in ascospores. Ascospores non-induced (NI) and induced (I) for germination were assessed for the phosphorylation of PaMpk1 and PaMpk2. Ascospores were obtained from homozygous wild-type, *ΔPaNox2* and *ΔPaPls1* crosses. Protein extract from *ΔPaMpk1* and *ΔPaMpk2* mycelia were loaded as control.

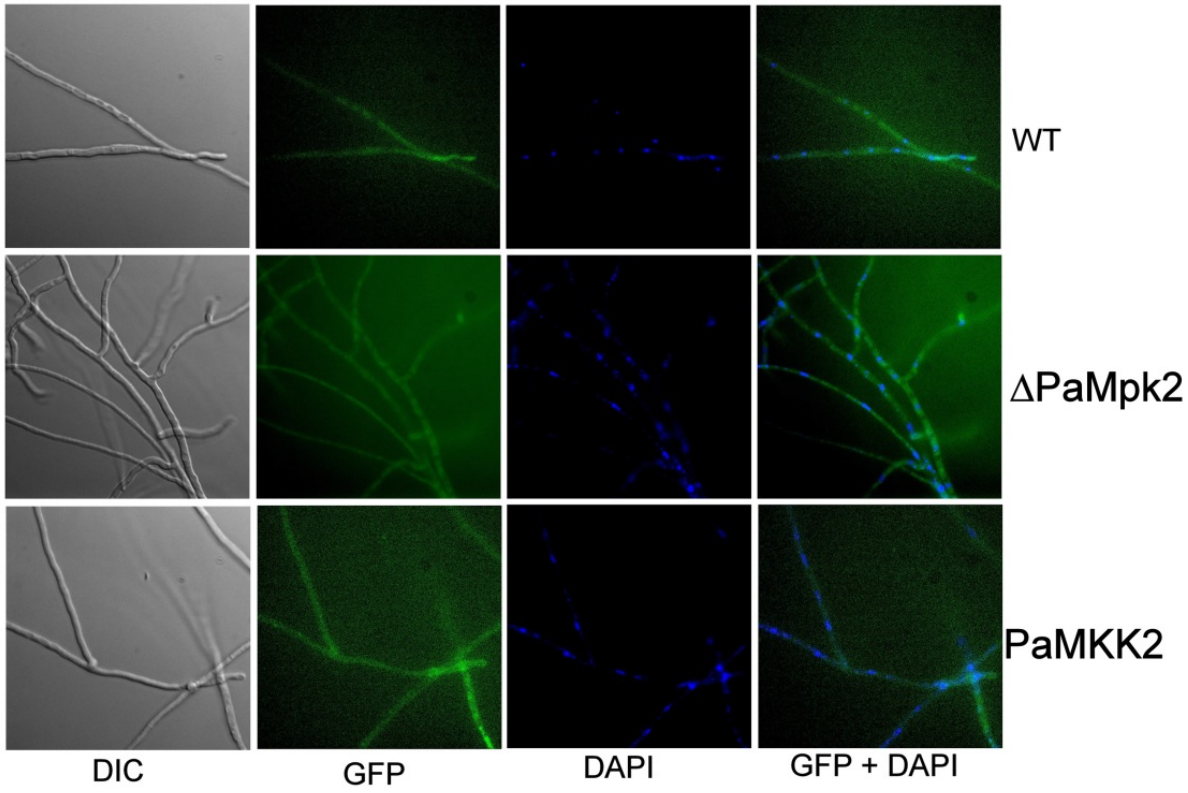

Figure S7 Localization of PaMpk1 in apical hyphae. Hyphae from the growing edge of cultures of the indicated strains carrying the PaMpk1-GFP transgene presents only diffuse fluorescence.

Table S1 The full set of MAP kinases in *P. anserina*

	Mpk1-like	Fus3/Kss1-like	Hog1-like
MAPKKK	**PaASK1**	**PaTLK2**	**PaHOK3**
	(Pa_5_9370)	(Pa_7_8030)	(Pa_1_12080)
	1832 aa	926 aa	1360 aa
	0 EST	3 EST	6 EST
MAPKK	**PaMKK1**	**PaMKK2**	**PaMKK3**
	(Pa_7_10270)	(Pa_2_820)	(Pa_1_15320)
	527 aa	416 aa	681 aa
	1 EST	1 EST	10 EST
MAPK	**PaMpk1**	**PaMpk2**	**PaMpk3**
	(Pa_2_13340)	(Pa_5_5680)	(Pa_1_23930)
	413 aa	353 aa	357 aa
	17 EST	9 EST	9 EST

The table gives the coding sequences (CDS) identifier as defined by the *P. anserina* genome project (http://podospora.igmors.u-psud.fr), the size of the proteins and the number of Expressed Sequence Tag (EST) found in the collection.

Table S2 Primers

fus3gauche	TTATTTTGGCGCTCGTTG
fus3G-sphI	ATGCATGCGTTGGATTAGGGAGAGTGGATT
fus3D-notI	ATGCGGCCGCCCGTTTGTTGGAAGTGGCTTG
fus3droit	GCTGTCGAGGCTTTCACTAA
MKK2A	GGCCCGCAAGGTTAGTCT
MKK2B	CGGTGCATGATGTGGTGT
TLK2-1F	AACGGTGACACAGCATCGTTAGTAGAGAGG
Mk_TLK2-2R	ctatttaacgaccctgccctgaaccgGGCGGAGCCTTGGTTTTCTCTTTG
TLK2_Mk-2F	CAAAGAGAAAACCAAGGCTCCGCCcggttcagggcagggtcgttaaatag
TLK2_Mk-3R	CAAGCACGGCTCTCCCAAGAATCAATTATcatcgaactggatctcaacagcggtaag
Mk_TLK2-3F	cttaccgctgttgagatccagttcgatgATAATTGATTCTTGGGAGAGCCGTGCTTG
TLK2-4R	GTTCTAGCGCAAACGCAAGTCAATTTATCC
MKK2_GF_CH1_for	GTGAGCTCATCAACGACATCGCTGACGACTTTGTCGGTACGTC
MKK2_GF_CH1_rev	GACGTACCGACAAAGTCGTCAGCGATGTCGTTGATGAGCTCAC
Mpk2_for	CGGCGACGTAACTCGACCT
Mpk2_rev	GCAGGCGAGCATGGTTAT
HOG1AML	GTCGACATGTGTGGTTCGGGCAGT
HOG1AMR	GGGCCCTGTGGCGGGGAAGATAGA
HOG1VML	GCGGCCGCCCGATATGGGTGGTCCTG
HOG1VMR	GTCGACAAAGCTGTTGACGCTTTGC
M3K2GF	GATATCTCCATGGTTTTTGGAGCTG
M3K2GR	TCTAGATGTGTGGTTGAGGTTGCAC
M3K2DF	CTCGAGGTGCGAGCAACAAACAGC
M3K2DR	GATATCTCGTGTGCAATCATCCTATCA
HOK3GF	GATATCTCATTCCTTTCCTGAAACAGC
HOK3GR	TCTAGATCGATCGTATGCCCACTG
HOK3DF	CTCGAGCGAAATGGATACCCCTTGG
HOK3DR	GATATCTCTCATTCACCACCAACAACAA

Table S3 Stress resistance phenotypes associated with the PaMpk1, PaMpk2 and PaMpk3 MAPK modules inactivation

	WT	ΔPaMpk1	PaMKK1 (IDC^{404})	PaASK1 (IDC^{118})	ΔPaMpk2	ΔPaMKK2	ΔPaTLK2	ΔPaMpk3	ΔPaMKK3	ΔPaHOK3	Δ1 Δ2	Δ1 Δ3	Δ2 Δ3	Δ1 Δ2 Δ3
thermo-sensitivity at 37°C[a]	100%	80%	80%	85%	85%	85%	80%	100%	100%	100%	100%	80%	70%	100%
cryo-sensitivity at 11°C[a]	100%	90%	90%	90%	80%	80%	80%	100%	100%	100%	80%	80%	80%	75%
neutral osmo-sensitivity (sorbitol 200g/l) [a]	100%	95%	95%	95%	95%	95%	85%	0%	0%	0%	10%	0%	0%	0%
neutral osmo-sensitivity (saccharose 200g/l) [a]	100%	95%	95%	95%	75%	75%	90%	50%	50%	50%	95%	30%	0%	0%
ionic sensitivity (KCl 0.5M) [a]	100%	80%	80%	80%	90%	90%	75%	0%	0%	0%	35%	0%	0%	0%
ionic sensitivity (NaCl 0.5M) [a]	100%	80%	80%	80%	95%	95%	75%	0%	0%	0%	70%	0%	0%	0%
ionic sensitivity (CaCl2 0.5M) [a]	100%	85%	85%	85%	30%	30%	85%	0%	0%	0%	30%	0%	0%	0%
0.5 mM EGTA [a]	100%	90%	90%	90%	80%	80%	90%	95%	95%	95%	110%	90%	75%	80%
0.04% H2O2 sensitivity [a]	100%	140%	160%	160%	180%	180%	180%	160%	160%	170%	170%	160%	175%	0%
10-4 M TBY sensitivity [a]	100%	105%	105%	100%	110%	105%	75%	100%	100%	105%	140%	95%	120%	115%
5. 10-5 M Menadione sensitivity [a]	100%	80%	85%	95%	115%	100%	95%	115%	110%	120%	125%	0%	105%	110%
50 μg/ml calcofluor sensitivity [a]	100%	90%	90%	100%	185%	175%	175%	120%	100%	120%	135%	85%	160%	140%
5mM caffeine sensitivity [a]	100%	110%	110%	115%	115%	115%	115%	100%	100%	100%	115%	115%	115%	130%

0,1 μg/ml nikkomycin sensitivity	100%	75%	75%	75%	130%	130%	115%	100%	100%	100%	130%	75%	130%	100%
5 μg/ml Iprodione [a]	100%	105%	105%	105%	180%	160%	105%	100%	100%	100%	160%	100%	170%	190%
0.01 μg/ml Fluoxonil [a]	100%	40%	70%	105%	220%	135%	50%	185%	195%	200%	165%	180%	220%	225%

[a] the table gives the percentage of growth speed as compared to wild type (100%)

Table S4 Efficiency of cellulose degradation as compared to wild type

	3d	5d	7d
ΔPaMpk1	73%	77%	93%
ΔPaMKK1	41%	69%	91%
ΔPaASK1	73%	76%	108%
ΔPaMpk2	36%	41%	52%
ΔPaMKK2	14%	39%	46%
ΔPaTLK2	42%	37%	46%
ΔPaMpk3	86%	96%	97%
ΔPaMKK3	100%	98%	102%
ΔPaHOK3	88%	99%	99%
ΔPaMpk1 ΔPaMpk2	20%	47%	70%
ΔPaMpk1 ΔPaMpk3	47%	76%	88%
ΔPaMpk2 ΔPaMpk3	7%	40%	57%
ΔPaMpk1 ΔPaMpk2 ΔPaMpk3	16%	42%	69%

The table gives the loss of dry weight of Whatman paper pieces incubated for 3, 5 and 7 days of *P. anserina* mutants as compared to wild type.