

SUPPLEMENTAL DATA

TABLE S1. Gene-specific oligonucleotides used for the generation of DIG-dUTP-labeled hybridisation probes.

primer	sequence	gene	protein
AnesmAF	5'-CCACAGACTTGAGCGTAG-3'	<i>esmA</i>	chitinsynthase
AnesmAR	5'-CCGTATCCATCCCAGAG-3'		
AnchiAF	5'-CCTGTATGACCTTGACCC-3'	<i>chiA</i>	chitinase A
AnchiAR	5'-GTTGAGGTTTCCGTGTGG-3'		
AnchsAF	5'-GTAAGACCACCATCAGCG-3'	<i>chsA</i>	chitin synthase A
AnchsAR	5'-CGATACCTTCTCAGCCC-3'		
AnGs1F	5'-ATCATCCCTCTCCTGTCC-3'	<i>agsB</i>	α -1,3-glucan synthase B
AnGs1R	5'-GCTTGCCGTACTTCTTCG-3'		
Anb1,3GsF	5'-GGACGAACGGTACAACAG-3'	<i>fksA</i>	β -1,3-glucan synthase A
Anb1,3GsR	5'-GCAGTGGGAATGTCAACG-3'		
orlmAnF	5'-ATCCACAACAGCATCACCC-3'	<i>rlmA</i>	ortholog of the MADS-box
orlmAnR	5'-ACAGCCTTATTTCCCGCAG-3'		transcription factor RlmA of <i>A. niger</i>

Figure S1

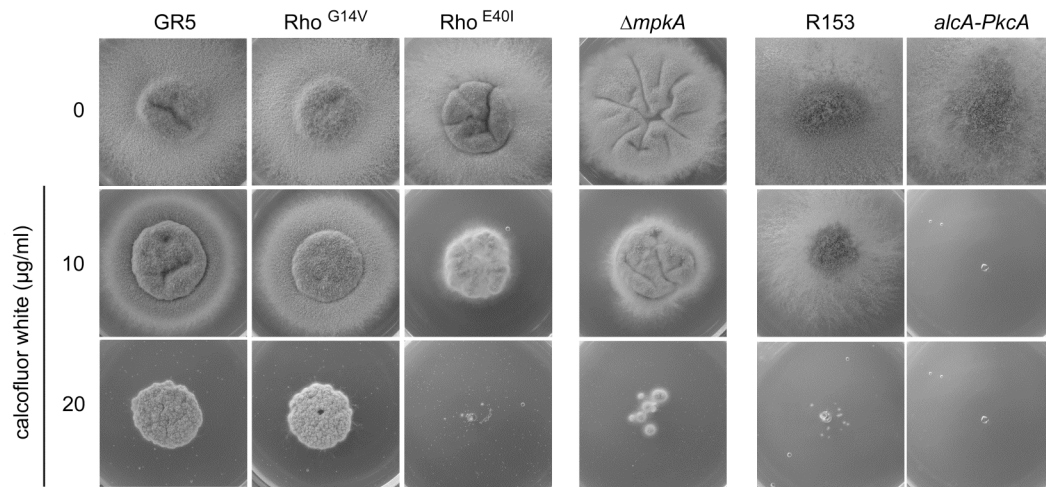


Fig. S1: Effect of CFW on *A. nidulans* strains defective in the activation of the components of the CWI pathway, (*rhoA*^{G14R}, *rhoA*^{E40I}, $\Delta mpkA$ and *alcA-PkcA*) in comparison to the respective recipient strains (GR5 for the *rhoA*^{G14R}, *rhoA*^{E40I}, $\Delta mpkA$ mutant strains, R153 for the conditional mutant *alcA-PkcA*). 2×10^3 conidia were point inoculated on appropriately supplemented agar plates containing 0-20 $\mu\text{g/ml}$ CFW. Plates were incubated at 37°C for 48 h (RhoA and MpkA mutant strains) or for 72 h (PkcA mutant strain), respectively.

Figure S2

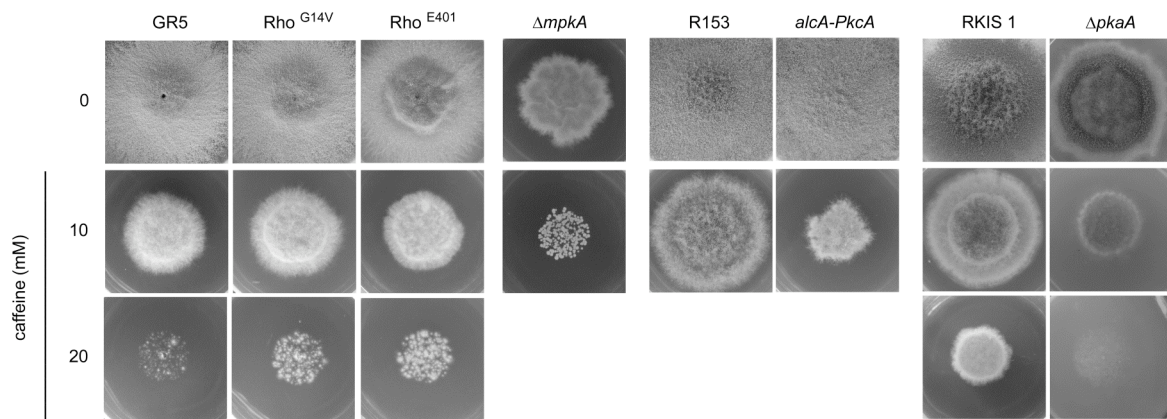


Fig. S2: Effect of caffeine on the growth of RhoA mutants, $\Delta mpkA$, *alcA-PkcA* and $\Delta pkaA$ compared to the respective recipient strains (GR5 for both RhoA mutants and $\Delta mpkA$, R153 for *alcA-PkcA* and RKIS1 for $\Delta pkaA$) on solid medium. 2×10^3 conidia were point inoculated on CM containing the appropriate supplements, except for the *alcA-PkcA* mutant and its respective control strain which were grown on repressive minimal medium (MM) containing 1% glucose (Ronen *et al.*, 2007). Caffeine concentrations of 0-20 mM were tested. The *alcA-PkcA* mutant and R153 were incubated for 72 h, all other strains for 48 h.

Figure S3

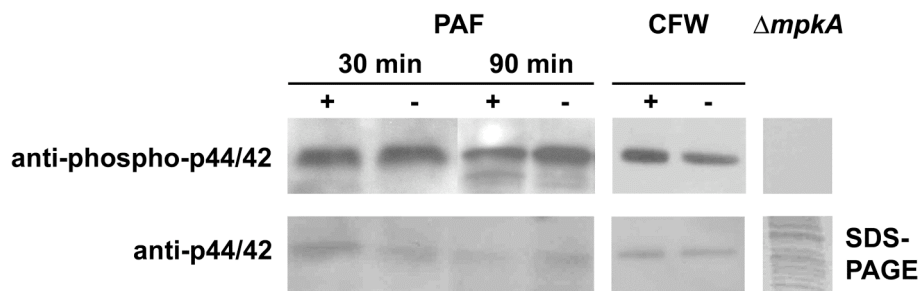


Fig. S3: Immunoblotting of *A. nidulans* cellular extracts with polyclonal anti-phospho p44/42 Mpk antibodies (anti-phospho-p44/42, upper panels). The lower panels show the results of immunoblotting of the PAF and CFW-treated samples with polyclonal anti-Mpk antibodies (anti-p44/42) as a loading control. Total proteins were extracted from *A. nidulans* without treatment (-), or after exposure to 50 $\mu\text{g/ml}$ PAF (+) for 30 min and 90 min, respectively, or to 20 $\mu\text{g/ml}$ CFW (+) for 90 min. Cellular extract from the $\Delta mpkA$ mutant strain was used to prove anti-phospho-p42/44 antibody specificity (upper panel). Coomassie-stained cellular extract of the $\Delta mpkA$ mutant after SDS-PAGE is shown as loading control (lower panel). 10-20 μg protein extract was loaded per lane on a 10% SDS-polyacrylamide gel.

Figure S4

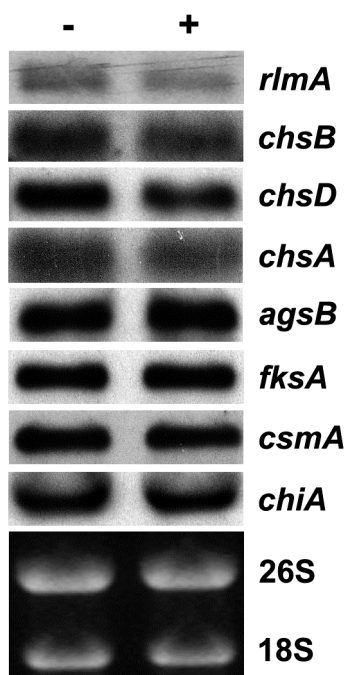


Fig. S4: Northern blot analysis of genes involved in cell wall biogenesis and remodeling. Sixteen hours old *A. nidulans* cultures were exposed to 0 (-) and 50 µg/ml PAF (+) for 3 h. Ten µg total RNA was loaded per lane and gene expression was detected by hybridizing the blotted samples with the respective DIG-dUTP-labeled probe. The lowest panel shows the 26S and 18S rRNA stained with ethidium bromide as loading control.

Figure S5

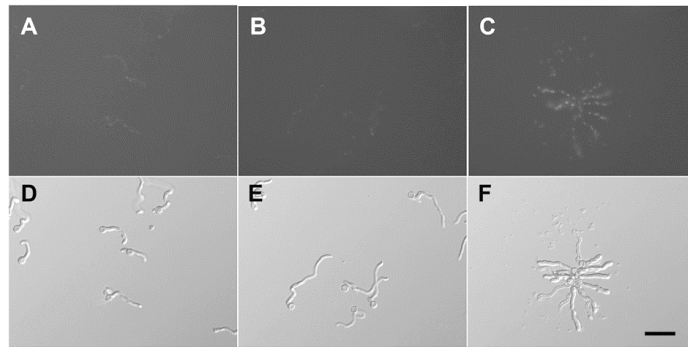


Fig. S5: Microscopic analysis of *A. niger* strain RD6.47 expressing a nucleus-targeted GFP under the control of the *A. niger agsA* promoter. Five hours old germlings were treated with 100 µg/ml PAF (**B**, **E**) or 10 µg/ml caspofungin (**C**, **F**) as a positive control for additional 2 h. Untreated germlings served as negative control (**A**, **D**). Nuclear staining was monitored by fluorescence microscopy (**A-C**) as described in Materials and Methods. (**D-F**) represent differential interference contrast images of the respective germlings. Scale bar, 20 µm.

Figure S6

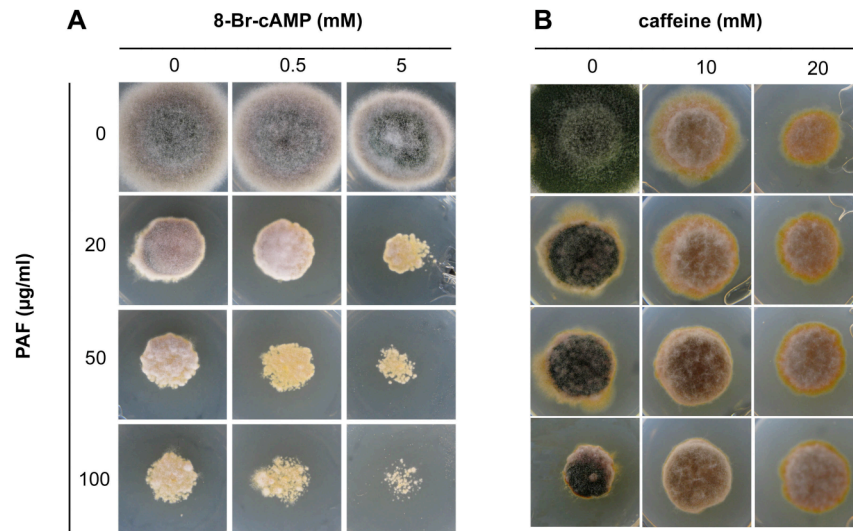


Fig. S6: Effect of the cAMP/PkaA signaling pathway modulating substances 8-Br-cAMP **(A)** and caffeine **(B)** in combination with PAF on the growth of *A. nidulans* FGSC4A. 2×10^3 conidia were point inoculated on CM agar plates containing 0-100 µg/ml PAF combined with 0-5 mM 8-Br-cAMP or 0-20 mM caffeine. Plates were incubated at 37°C for 48 h in **(A)** and for 72 h in **(B)**.

Figure S7

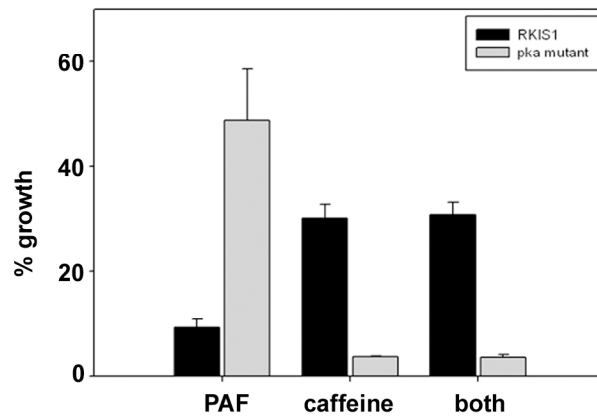


Fig. S7: Liquid growth inhibition assay of *A. nidulans* $\Delta pkaA$ and *RKIS1* exposed to PAF (5 $\mu\text{g/ml}$), caffeine (10 mM) or to a combination of both substances. The % growth was calculated from % changes in OD_{620} of compound-treated conidia compared to the untreated control (=100%). Note that for growth assays in liquid culture (96-well plates) less concentration of growth inhibitory compounds is needed compared to agar plate assays.