

<i>A. nidulans</i> AtfA 485 aa	1	-----MSAAVASAVSTTLPSNP-TAHSSPMDAKKNSVKMDN-EASSETKE	43
<i>A. oryzae</i> BAD93190 366 aa	1	-----	1
<i>C. purpurea</i> CAD21519 550 aa	1	-----MGTSPTNESKSPDLSNKTRQDPPTGKVDVVKPN-----	36
<i>S. pombe</i> BAA12194 566 aa	1	MSPSPVNTSTEPASVAASVNGNATASSTQVPENNQSDSFAPPSNNSQQNQ	50
<i>A. nidulans</i> AtfA 485 aa	44	QKTDGEPQTSLAPPSPRNPSAATDTP-DYFN-SVHNPFALEPNPFEQSFG	91
<i>A. oryzae</i> BAD93190 366 aa	1	-----	1
<i>C. purpurea</i> CAD21519 550 aa	37	GASNAEPTKPLAPPSPRPHQQPQGNNTTSDFAGQVAGSLSLEPNPFEQSFG	86
<i>S. pombe</i> BAA12194 566 aa	51	QSSTIAPNGGAGSVANANPADQSDGVTPSFVGSLK--LDYEPNPFEHSFG	98
<i>A. nidulans</i> AtfA 485 aa	91	-----GGS-GETPGKSILPPVASITSPALPG---	116
<i>A. oryzae</i> BAD93190 366 aa	1	-----	1
<i>C. purpurea</i> CAD21519 550 aa	86	-----GGGGPTEPGGKTLPSVASLTSPSSL---	112
<i>S. pombe</i> BAA12194 566 aa	99	STASVGQGNPSLNRNPSLSNIPSGVPPAFARTLLPPVSSIASPDLISGAP	148
<i>A. nidulans</i> AtfA 485 aa	116	-----TSSAGGAYNWSN-SLRSGPLSPAM <ins>L</ins> AGPAGGS D YFDS-	152
<i>A. oryzae</i> BAD93190 366 aa	1	-----MLAGPTGSS D YFDS-	14
<i>C. purpurea</i> CAD21519 550 aa	112	-----PGGNSTPFNWGGGSLRTGPLSPAM <ins>L</ins> SGPAN--DYFSDS	148
<i>S. pombe</i> BAA12194 566 aa	149	GIASPLGYPAWSAFTRGTMHNPLSPA <ins>I</ins> YDATLRPDYLNNPSDASAARFS	198
<i>A. nidulans</i> AtfA 485 aa	152	--IGRG <ins>F</ins> PTPNESSL <ins>R</ins> TGLTPGGGSMFPAP <ins>S</ins> PNSQ A LLNQLQNGGATE <ins>S</ins>	200
<i>A. oryzae</i> BAD93190 366 aa	14	--IGRG <ins>F</ins> PTPNESSL <ins>R</ins> TGLTPGGGSMFPAP <ins>S</ins> PNSQ A LLQQLQSGGATE <ins>S</ins>	62
<i>C. purpurea</i> CAD21519 550 aa	149	HHLRG <ins>G</ins> PTPNESSL <ins>R</ins> SGLTPGGGSMFPAP <ins>T</ins> PTSQ A LFSQLASGGATE <ins>S</ins>	198
<i>S. pombe</i> BAA12194 566 aa	198	--SGTG <ins>F</ins> PTGVNEPFR <ins>S</ins> LLPTGAG--FPAP <ins>S</ins> PGTAN <ins>L</ins> GFHTFD S QFPD	244
<i>A. nidulans</i> AtfA 485 aa	201	TIEFHRTALN-----VKKNGIA-PTSNP-----T	223
<i>A. oryzae</i> BAD93190 366 aa	63	TIEFHRTALNA-----AKKNALNGPTSNP-----T	87
<i>C. purpurea</i> CAD21519 550 aa	199	AIDFHRTAINAAA K REQMPPRQS Q HQPTQS Q QLQHSQQQSHQQHQQH	248
<i>S. pombe</i> BAA12194 566 aa	245	QYRF <ins>T</ins> PRDGKP-----PVVNGDQSDYFGANA A VHG L CLLSQV	284
<i>A. nidulans</i> AtfA 485 aa	224	GEGD <ins>Q</ins> V <ins>Q</ins> ONITTT-----MDIKPAQP-ATVDFGPHAA A ANGLFML	264
<i>A. oryzae</i> BAD93190 366 aa	88	SDPE <ins>Q</ins> ASQNTN-----MDMKPNQP-----DFPGHHDAAD A ANGLFML	124
<i>C. purpurea</i> CAD21519 550 aa	249	QHQ Q GAQSATSAPPDITNGVPAVKLEQKQPSGPFD H DN-DA A NGLFML	297
<i>S. pombe</i> BAA12194 566 aa	285	PDQQ <ins>Q</ins> KLQ <ins>Q</ins> PISSEND----QA A STANNLLKQTQQQT F PD <ins>S</ins> IRPSFTQ	329
<i>A. nidulans</i> AtfA 485 aa	265	AKGG <ins>Q</ins> STANQFAAVSNQ <ins>T</ins> AI <ins>P</ins> QTLQTSEILQDQNAARRQSVNVNG-VAN	313
<i>A. oryzae</i> BAD93190 366 aa	125	AKGG <ins>Q</ins> ANPNQFA-VSNQ <ins>S</ins> IPPQN I QNND--QARDSDRTS---NG---	164
<i>C. purpurea</i> CAD21519 550 aa	298	AQGA <ins>Q</ins> NRNGAQAPSHYN T SAP S HAHPAPISSSQNPSPQMSGKAVSR	347
<i>S. pombe</i> BAA12194 566 aa	330	NTNP <ins>Q</ins> AVTGTMN P QASR <ins>T</ins> QQQ <ins>P</ins> MYFMGSQQFNGMPSVYGD T VNPADPLT	379
<i>A. nidulans</i> AtfA 485 aa	314	TREPSGDGSE-QSEQ A KP-ARGRGKRNT T KASSTGN-RRKT T DDS-TQGS	359
<i>A. oryzae</i> BAD93190 366 aa	165	GRETSGDVSDVQGEQ A KPAT K G-KKNTAT K TSGAANNRRKA D APVKGS	213
<i>C. purpurea</i> CAD21519 550 aa	348	GVSEATNGSG-DSEQ A KPLPKGKGKTAAS---ATNGRRKAEDMPTKAP	392
<i>S. pombe</i> BAA12194 566 aa	380	LRQT T DFSGQNAENG S TNL P OKTSNSDMPTANSMPVKLENGT D YSTS Q E P	429
<i>A. nidulans</i> AtfA 485 aa	360	NKRTKLNNGA A STESPSE-GESEEEEQQPAQKKAGDTKKM <ins>T</ins> DEEKRKNF	408
<i>A. oryzae</i> BAD93190 366 aa	214	NKKAK--LSSG S TEPPSDAGDSEEEEQ--KKKSQSDSKKM <ins>T</ins> DEEKRKNF	259
<i>C. purpurea</i> CAD21519 550 aa	393	PTKKAKGMPETMNGSVNEEDSDDDDDDM--MGEDGNPKVKMTDEEKRKNF	440
<i>S. pombe</i> BAA12194 566 aa	430	SSNANNQSSPTSSINGKASSESANGT S YS-KGSSRRNSKNET <ins>T</ins> DEEKRKNF	478
<i>A. nidulans</i> AtfA 485 aa	409	bZIP LERNRVAALKCRQRKKQWL N QAKVELFTSENDAL T TTVTQLREE I VNL	458
<i>A. oryzae</i> BAD93190 366 aa	260	LERNRVAALKCRQRKKQWL N QAKVELFTSENDAL T ATATV T QLREE I VNL	309
<i>C. purpurea</i> CAD21519 550 aa	441	LERNRVAALKCRQRKKQWL N QAKVELFTSENDALTAQ I QLREE V VNL	490
<i>S. pombe</i> BAA12194 566 aa	479	LERNRQAAALKCRQRKKQWL N QAKVELFYGNENE I LSAQVSALREE I VSL	528
<i>A. nidulans</i> AtfA 485 aa	459	KTLL I LAHKDCPVSQAQGL-----IWN-----PNPP	483
<i>A. oryzae</i> BAD93190 366 aa	310	KTLL I LAHKDCPVSQAQGL G PLMMNGMSAGF--DPHPYNI P NNMGMQPGAP	357
<i>C. purpurea</i> CAD21519 550 aa	491	KTLL I LAHKDCPVTQQQGL H GPYMAQV V PYNAQMNPYGMAGPIP-NQQQV	539
<i>S. pombe</i> BAA12194 566 aa	529	KTLL I LAHKDCPVAKS N XAAVATS-----VIGSGDLAQRIX	563
<i>A. nidulans</i> AtfA 485 aa	484	YT-----485	
<i>A. oryzae</i> BAD93190 366 aa	358	IPTQGLRRQ--366	
<i>C. purpurea</i> CAD21519 550 aa	540	MAGQGVQRRFS 550	
<i>S. pombe</i> BAA12194 566 aa	564	LGY-----566	

Fig. 1S. The *atfA* gene encodes a putative transcription factor of ATF/CREB family.

The amino acid sequence of *Aspergillus nidulans* AtfA is aligned with *Schizosaccharomyces pombe* Atf1, *Claviceps purpurea* Cptf1 and *Aspergillus oryzae* AtfB. Sequences are identified by species name, GenBank accession number and protein size. Underlined sequence indicates the amino acids deleted in the Δ atfA mutant. The top red bar indicates the DNA binding domain as deduced from Expasy-Prosite database (<http://www.expasy.org/prosite/>).

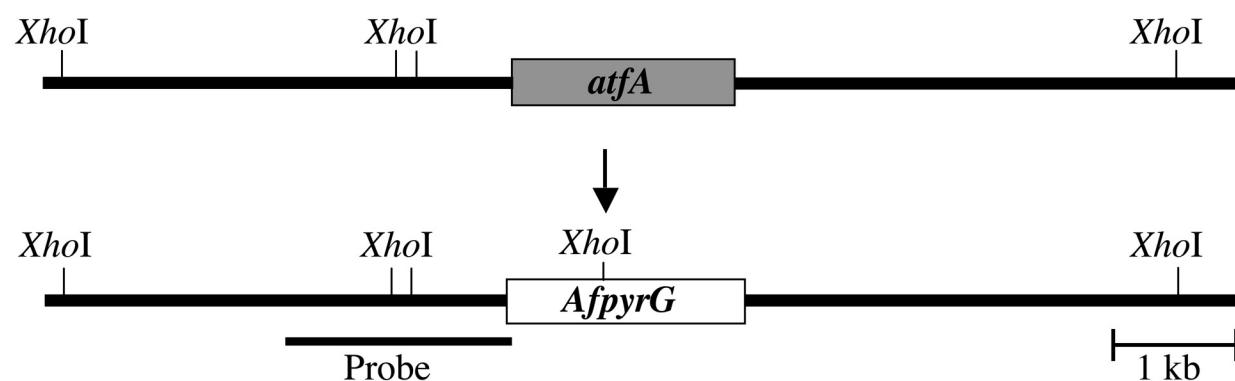
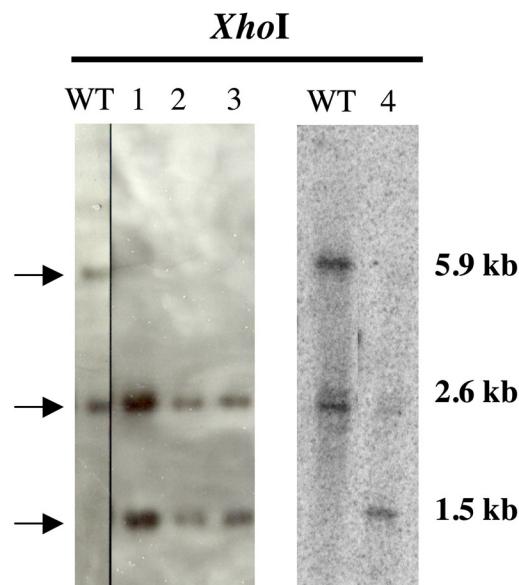
A**B**

Fig. 2S. Deletion of the *atfA* gene. (A) The *atfA* deletion construct containing the *AfpYrG* gene as selective marker was generated by double-joint PCR and used to transform strains CFL3 and 11035. Double recombination results in the replacement of wild type *atfA* by the deletion construct. (B) DNA from WT and PyrG⁺ transformants from CFL3 (1-3) and 11035 (4) strains was digested with *Xhol* and used for Soutern blot analysis with the probe indicated in (A). The wild type *Xhol* pattern correspond to bands of 5.9 and 2.6 kb while $\Delta atfA$ pattern corresponds to bands of 2.6 and 1.5 kb.

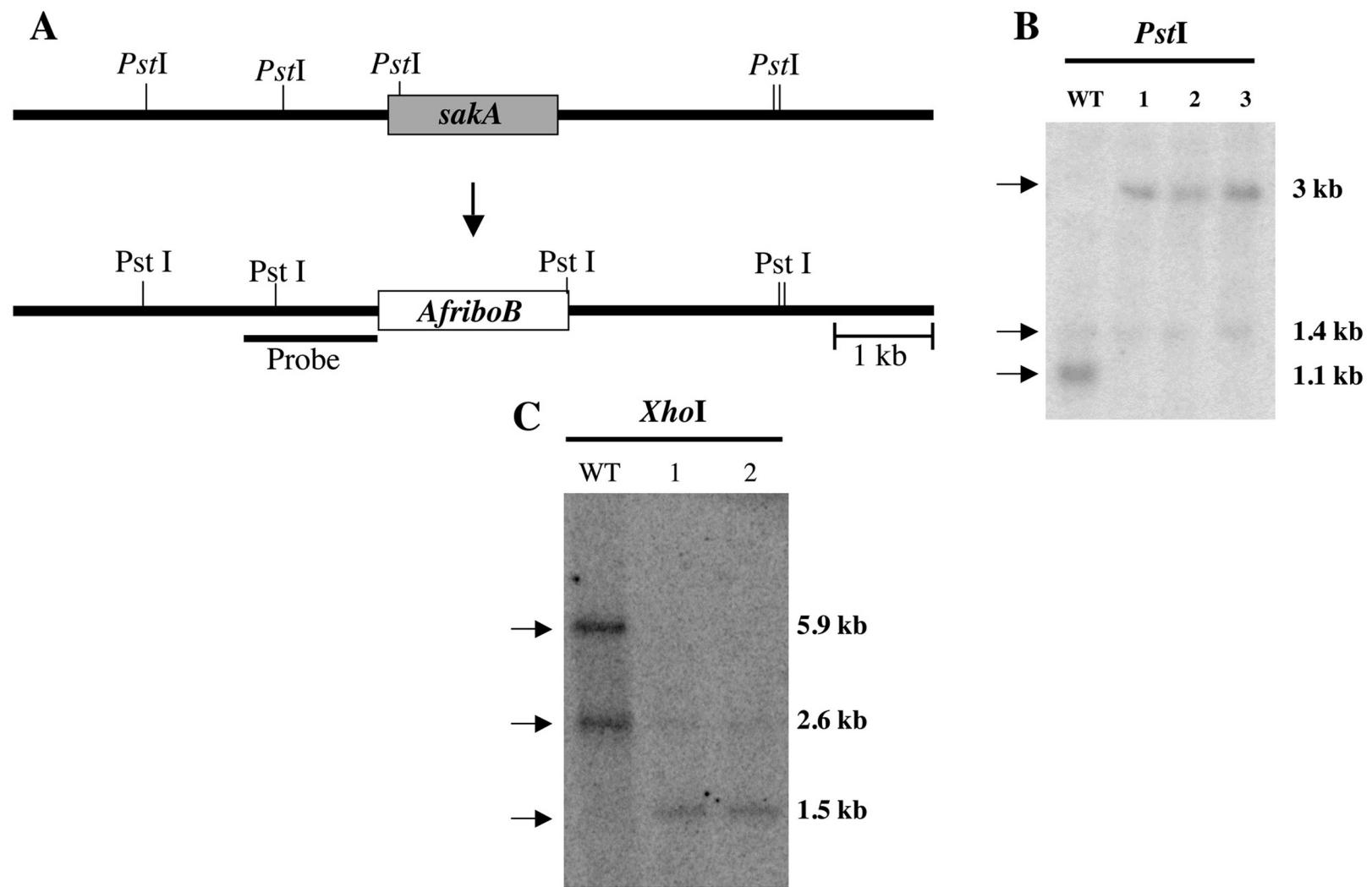


Fig. 3S. Deletion of the *sakA* gene. **(A)** A *sakA* deletion construct containing the *AfriboB* gene as selective marker was used to transform strain 11035. **(B)** To verify *sakA* deletion DNA of WT and RiboB⁺ transformants (1-3) was digested with *PstI* and used for Southern blot analysis with the indicated probe (A). Wild type *PstI* pattern correspond to bands of 1.4 and 1.1 kb; Δ *sakA* pattern corresponds to bands of 1.4 and 1.1 kb. **(C)** Δ *sakA* Δ *atfA* double mutants were generated by transformation of Δ *sakA* strain TFL Δ *sakA*-03 with the *attA*-*Afp*_G construct. *attA* deletion was confirmed in PyrG⁺ transformants 1 and 2, by Southern blot analysis as in Fig. 2S.

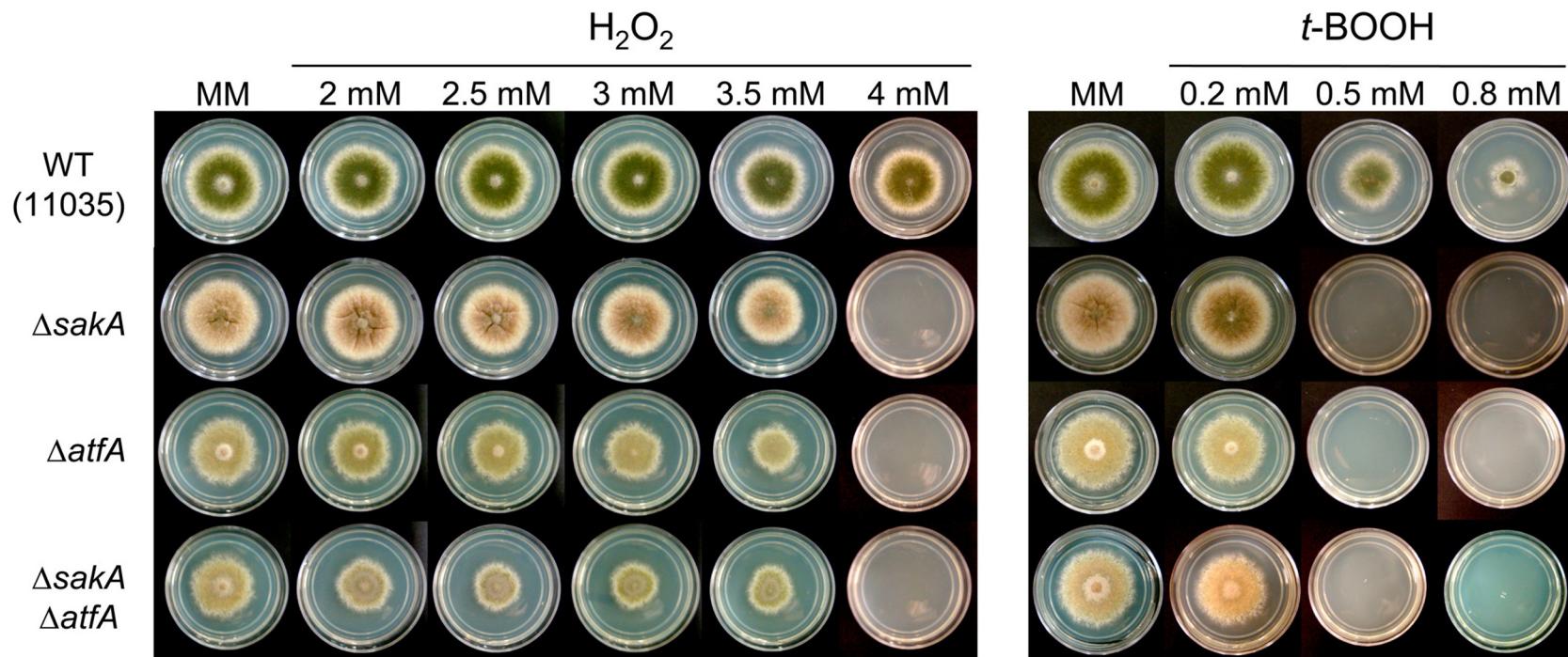


FIG. 4S. Spores from single and double $\Delta s\text{ak}A$ and $\Delta a\text{tf}A$ mutants are equally sensitive to H_2O_2 and $t\text{-BOOH}$. Conidia (10^4) from strains 11035 (WT), TFL $\Delta s\text{ak}A$ -03 ($\Delta s\text{ak}A$), TFL $\Delta a\text{tf}A$ -04 ($\Delta a\text{tf}A$) and TFL4 ($\Delta s\text{ak}A \Delta a\text{tf}A$) were inoculated on plates containing indicated concentrations of H_2O_2 or *tert*-butyl hydroperoxide ($t\text{-BOOH}$) and incubated for 4 days at 37 °C.

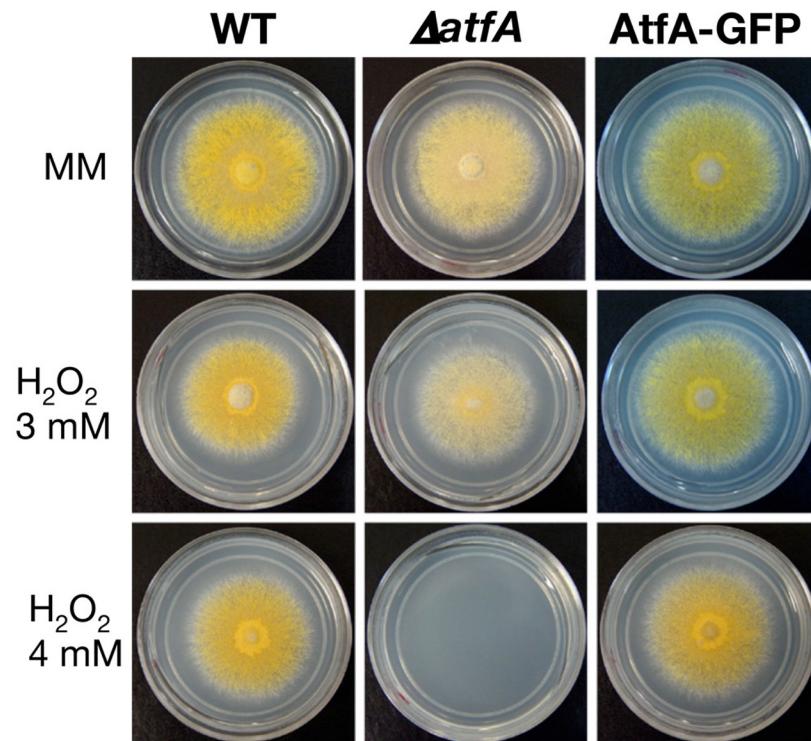
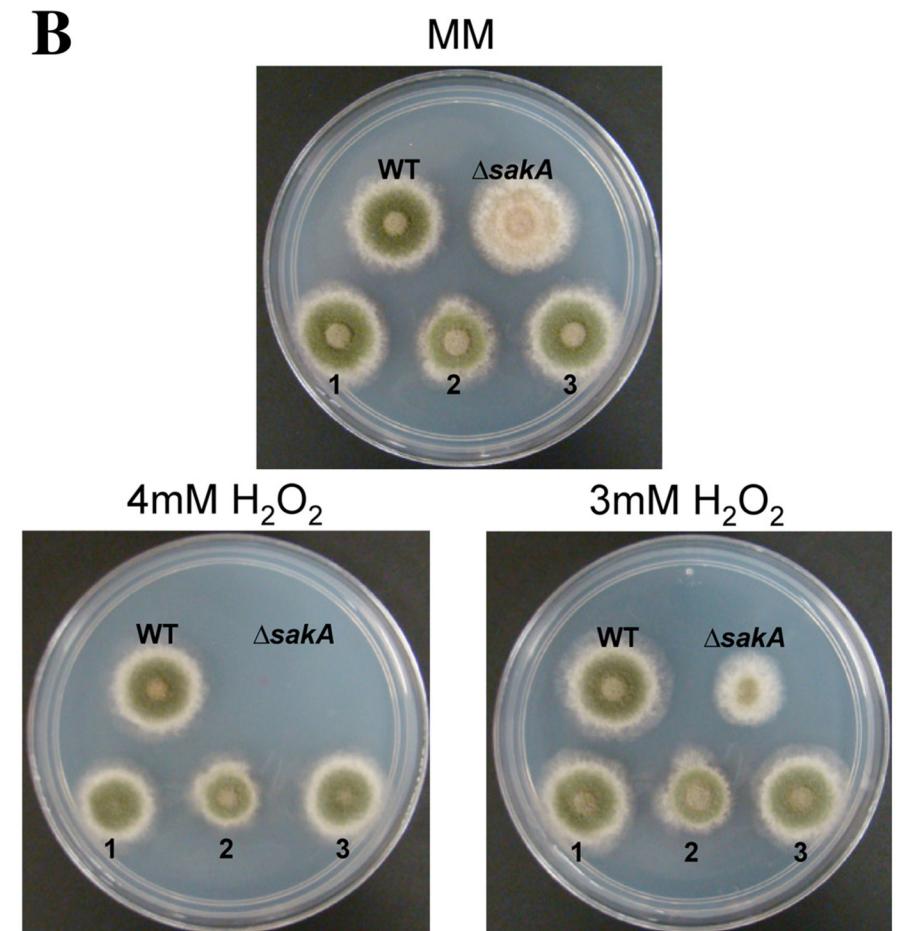
A**B**

Fig. 5S. GFP tagging does not interfere with *atfA* or *sakA* function. **(A)** Conidiospores (10^4) from strains CLK43 (WT), CFL Δ atfA-02 (Δ atfA) and TFL3 (*atfA*-GFP) were inoculated on plates containing 3 or 4 mM H_2O_2 and incubated at $37^\circ C$ for 4 days. **(B)** Conidiospores from strains A1155 (WT), TFL Δ sakA-03 (Δ sakA) and three *sakA*-GFP transformants were point inoculated on plates containing 3 or 4 mM H_2O_2 and incubated at $37^\circ C$ for 3 days. Transformant 3 was named TFL6 and selected for further experiments.

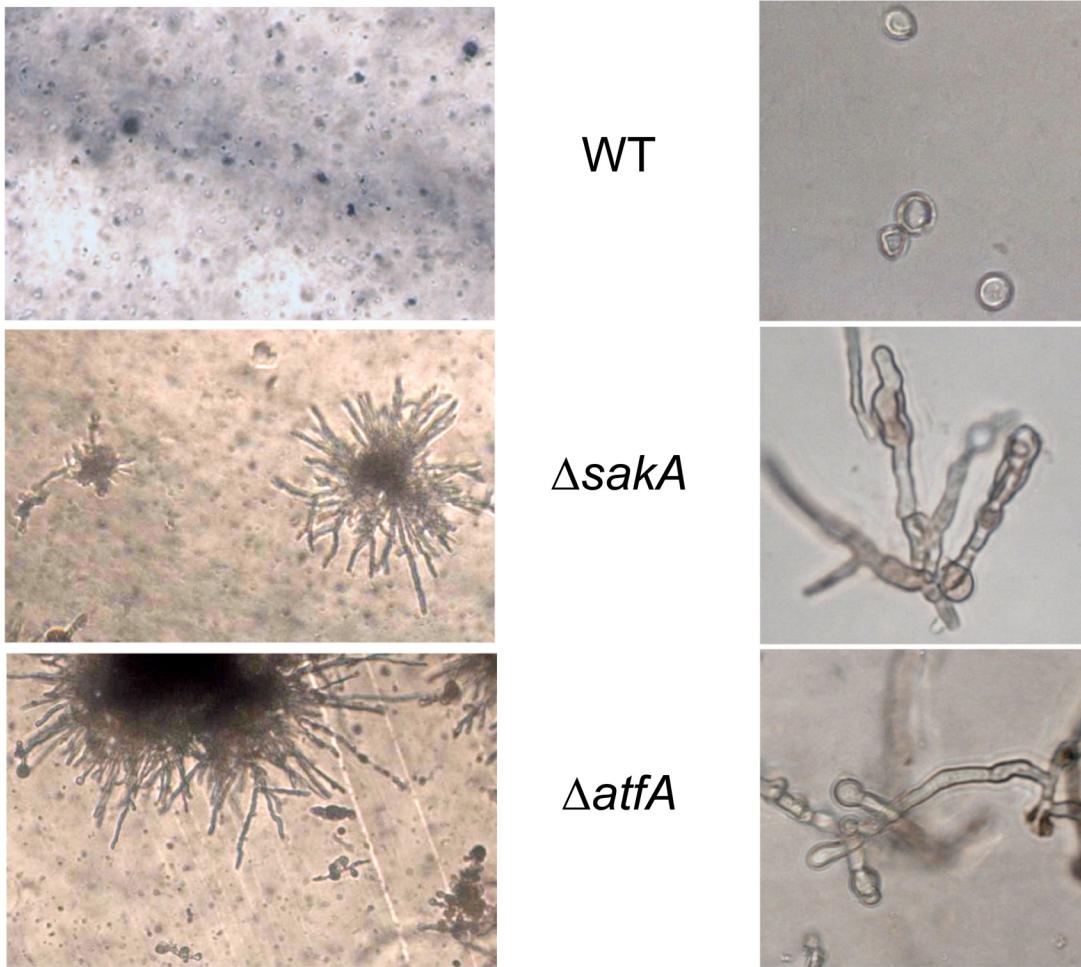


Fig. 6S. Germination of WT, Δ *sakA* and Δ *atfA* conidia in the presence of fludioxonil on solid medium.
Conidia from indicated strains were streaked on plates containing minimal solid medium plus 2 $\mu\text{g}/\text{ml}$ of fludioxonil and incubated at 37 °C for 4 days. Pictures were taken under the microscope directly from inverted plates (left, 10X objective) or from samples scraped and transferred to a glass slide (right, 40X objective).

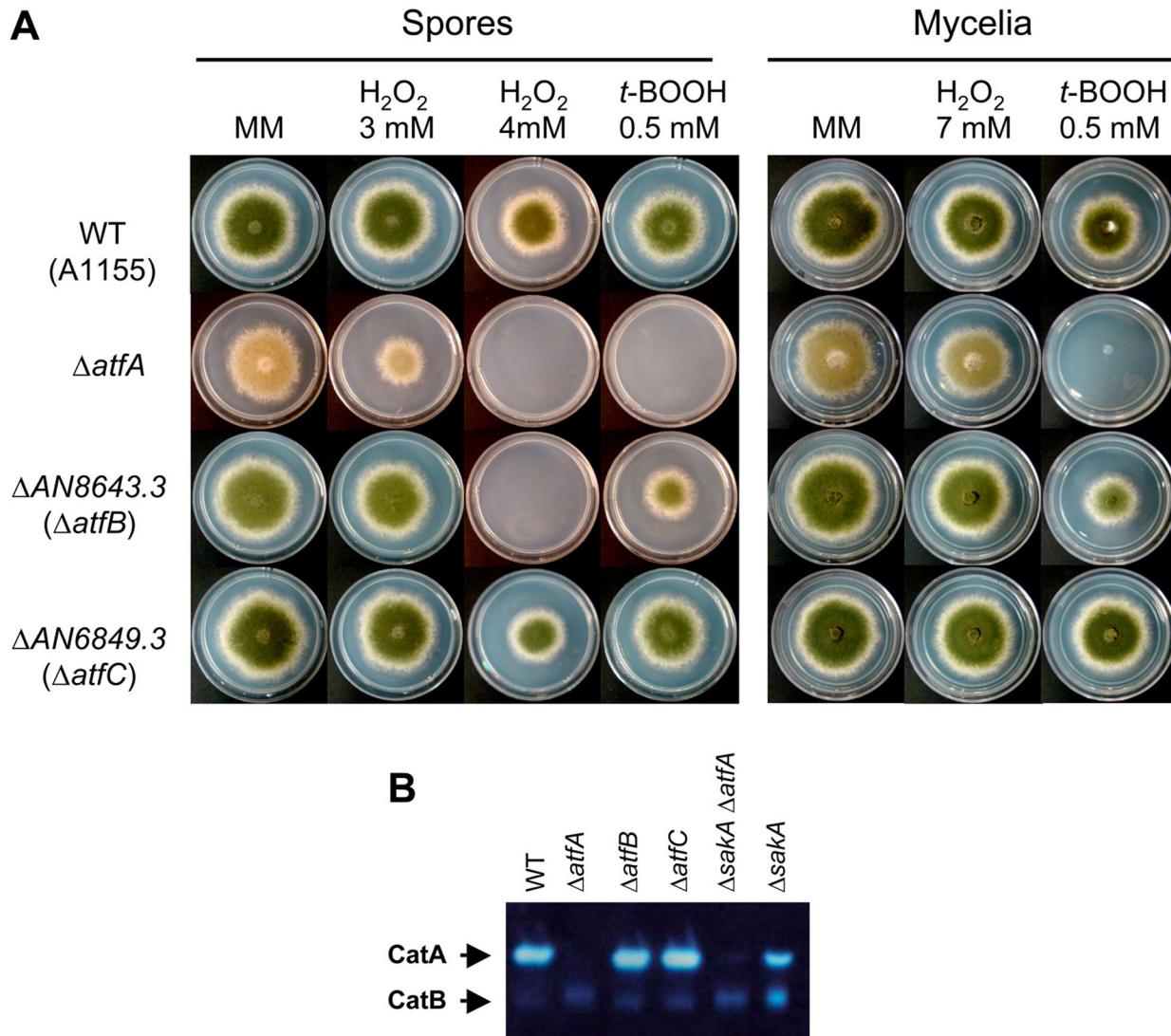


Fig 7S. Oxidative stress sensitivity and spore catalase activity in mutants lacking putative transcription factors AtfA, AtfB and AtfC. (A) Genes AN8643.3 (*atfB*) and AN6849.3 (*atfC*) were deleted in strain A1155, confirmed by Southern blot analysis and tested as in Fig. 1. (B) 30 µg of protein extract prepared from spores of the indicated strains were processed as in Fig. 2, to determine catalase activity.