

Supplementary Data

Supplementary experimental procedures

Preparation of the HapC-TNB mixed disulfides

One mg of the respective purified and lyophilized HapC(CSS), HapC(SCS) and HapC(SSC) proteins were dissolved in water and incubated with an excess of DTNB. The final reaction mixture contained 45 μ M of the respective HapC double serine mutant and 2 mM DTNB. The reaction was monitored at 412 nm for the completion of the reaction. The volume of the reaction mixture was then applied to a NAP-5 column (GE Healthcare) to remove the excess of DTNB and released TNB, and for transfer of the mixed disulfides HapC(CSS)-TNB, HapC(SCS)-TNB and HapC(SSC)-TNB into 0.1 M potassium phosphate buffer, 2 mM EDTA, pH 7.5. Then, aliquots of the mixed disulfides HapC-TNB were directly used in a reaction with TrxA(C39S), as described in the following section.

Preparation of the mixed disulfides between HapC(CSS), HapC(SCS), HapC(SSC) and TrxA(C39S)

First, TrxA(C39S) was treated with 100 mM DTT for 30 minutes at room temperature to make sure the remaining nucleophilic active-site thiol (Cys36) was in the reduced state. The excess of DTT was removed by the use of a NAP-5 column. Then, TrxA(C39S) was incubated with equimolar amounts of mixed disulfide HapC(CSS)-TNB, HapC(SCS)-TNB or HapC(SSC)-TNB in 0.1 M potassium phosphate buffer, 2 mM EDTA, pH 7.5 at room temperature. The reaction was monitored at 412 nm for the release of TNB. Upon completion of the reaction the formed product was analyzed on SDS-PAGE.

Subcellular localization of HapC-eGFP and HapC(SSS)-eGFP fusions

The *A. nidulans* strain Δ C-HapCegfp was generated previously (1). A HapC-eGFP fusion in which all cysteines of HapC were exchanged to serines was created by site specific mutagenesis according to the method of Higuchi et al. (2) using the primer pairs HapC5'BamHI/3CS-Backward and HapC3'NcoI/3CS-Forward, and the plasmid pHapC-eGFP as template. A DNA fragment was amplified using the primers HapC5'BamHI and HapC3'NcoI. The resulting PCR fragment was cloned into the *Bam*HI-*Nco*I-digested plasmid p123 and fused to *egfp* under the control of the *otef* promoter. The resulting plasmid was named pHapC(SSS)-eGFP. To use the *pyr-4* gene of *Neurospora crassa* as a selectable

marker for the transformation of *A. nidulans*, the *otef(p)-hapC(SSS)-egfp-nos(t)* cassette, was excised from pHapC(SSS)-eGFP by *ClaI-KpnI* digestion and cloned into the plasmid pKTB1 resulting in the plasmid pKTB1-HapC(SSS)-eGFP. To employ the *A. nidulans pyroA* gene as a selection marker for the transformation of *A. nidulans*, the *pyroA* gene was amplified by PCR using the primers AnPyro-For and AnPyro-Rev and genomic DNA of the AXB4A2 strain as a template. The PCR product was digested with *HindIII* and ligated into the plasmid pHapC-eGFP resulting in the plasmid pHapC-eGFP-pyro. Transformation of the *A. nidulans* $\Delta hapC$ strain Nat24 and the $\Delta trxA$ strain AnTrxAKO with the plasmids pKTB1-HapC(SSS)-eGFP and pHapC-eGFP-pyro resulted in the strains ΔC -HapCegfp-SSS and $\Delta trxA$ -HapCegfp, respectively.

Generation of plasmids and strains for BiFC analysis

To study the interaction between HapC and HapE, the *hapC* gene was PCR amplified, using primers HapC-BiFC-For and HapC-BiFC-Rev to introduce flanking *NcoI* sites. After *NcoI* digestion, the DNA fragment was ligated into the plasmid pEYFPC to yield plasmid pHapC-YC. For employing the *pyr-4* gene of *N. crassa* as a selection marker, the DNA fragment encoding *gpdA(p)-hapC-eyfpC-trpC(t)* was reisolated from plasmid pHapC-YC and cloned into plasmid pKTB1 using *XbaI*. The resulting plasmid was designated pHapC-YC-pyr4. The HapE open reading frame was cloned directly by digesting pHapE-eGFP with *NcoI* and ligated into pEYFPN generating the plasmid pHapE-YN. The recipient strain AXB4A2 was co-transformed with plasmids pHapC-YC-pyr4 and pHapE-YN resulting in the strain yHapC-HapE.

To use pEYFPN as a control for the HapC/HapE interaction experiment, the *gpdA(p)-eyfpN-trpC(t)* cassette was amplified by PCR using the primers EYFP5'EcoRI(C2) and EYFP3'EcoRI(C2) from plasmid pEYFPN as a template. The PCR product was digested with *EcoRI* and ligated into *EcoRI*-digested pKTB1. The generated plasmid was designated pYN-pyr4. Transformation of strain AXB4A2 with the plasmid pYN-pyr4 resulted in the strain yN. To use pEYFPC as a control, the *gpdA(p)-eyfpC-trpC(t)* fragment was excised from plasmid pEYFPC using *XbaI* and cloned into pKTB1. The generated plasmid was designated pYC-pyr4. The plasmid was transformed in the *A. nidulans* strain AXB4A2 resulting in strain yC. The strain yCN was generated by co-transforming strain AXB4A2 with pYC-pyr4 and pEYFPN. The yHapE-C and yHapC-N strains were generated by co-transforming strain AXB4A2 with the plasmid pairs pHapE-YN and pYC-pyr4, and pHapC-YC-pyr4 and pEYFPN, respectively.

To analyse the interaction between HapC and TrxA in the *A. nidulans* wild-type strain AXB4A2, HapC-YC and TrxA-YN fusions were employed. Generation of a plasmid, which contains a HapC-YC fusion, was described above. The plasmid encoding the TrxA-YN fusion was constructed as follows: the *trxA* gene was amplified by PCR using primers TrxNcoI for and TrxNcoI rev and plasmid pET39-AnTrxA(wt)-H6 as a template. The PCR product was digested with *NcoI* and ligated into the *NcoI*-digested pEYFPN. The resulting plasmid was designated pTrxA-YN. Co-transformation of plasmids pHapC-YC-pyr4 and pTrxA-YN into strain AXB4A2 yielded strain yHapC-TrxA. Two control strains were used, yHapC-N and strain yTrxA-C, which was generated by co-transforming strain AXB4A2 with plasmids pTrxA-YN and pYC-pyr4.

Visualization of the interaction between HapC and TrxA in the $\Delta hapE$ strain $\Delta E-89$ was achieved by using HapC-YC and TrxA-YN fusions as described previously. To employ the *pabaA1* gene as a selection marker for the transformation of *A. nidulans*, the *gpdA(p)-hapC-eyfpC-trpC(t)* cassette was amplified by PCR using primers EYFP5'BamHI and EYFP3'BamHI and plasmid pHapC-YC-pyr4 as a template. The PCR product was digested with *BamHI* and ligated into plasmid pabaAnid resulting in plasmid pHapC-YC-paba. To employ the *pyr-4* gene of *N. crassa* as a selection marker, the *gpdA(p)-trxA-eyfpN-trpC(t)* DNA fragment was amplified by PCR using primers EYFP5'KpnI and EYFP3'KpnI and plasmid pTrxA-YN as a template. The PCR product was digested with *Acc65I* (an isoschizomer of *KpnI*) and ligated into *Acc65I*-digested pKTB1. The resulting plasmid was designated pTrxA-YN-pyr4. Co-transformation of the $\Delta E-89$ strain with plasmids pHapC-YC-paba and pTrxA-YN-pyr4 resulted in the strain yHapC-TrxA- ΔE .

Supplementary Figures

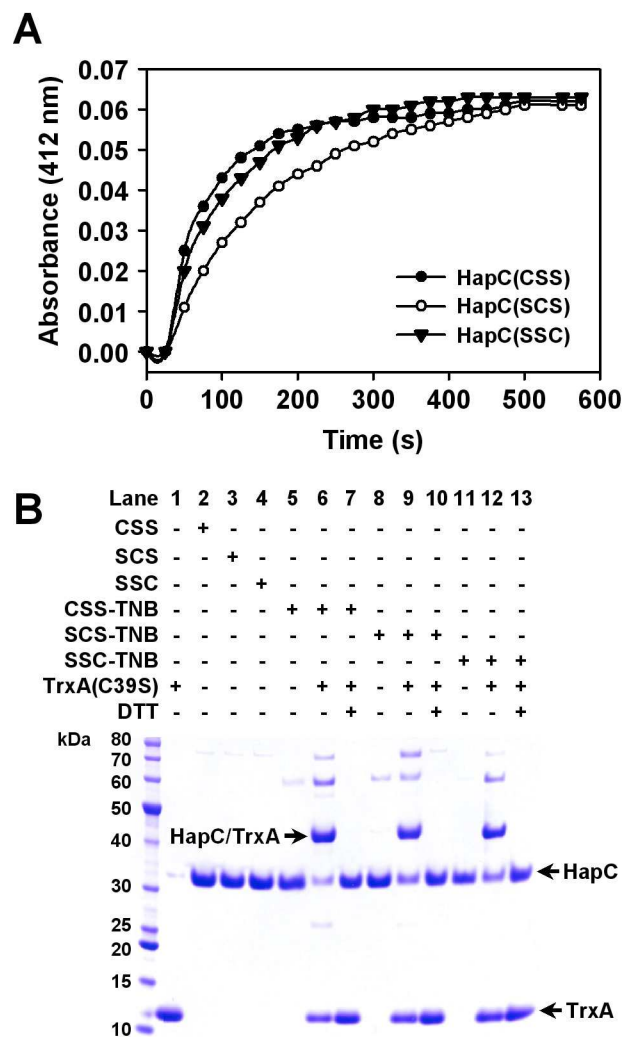


Figure S1. HapC single cysteine mutants CSS, SCS and SSC form stable mixed disulfides with a TrxA(C39S) mutant. Emergence of disulfide linked HapC-TrxA(C39S) heterodimers from HapC-TNB adducts and TrxA(C39S) was monitored at 412 nm by the release of TNB (A) and by SDS-PAGE analysis (B).

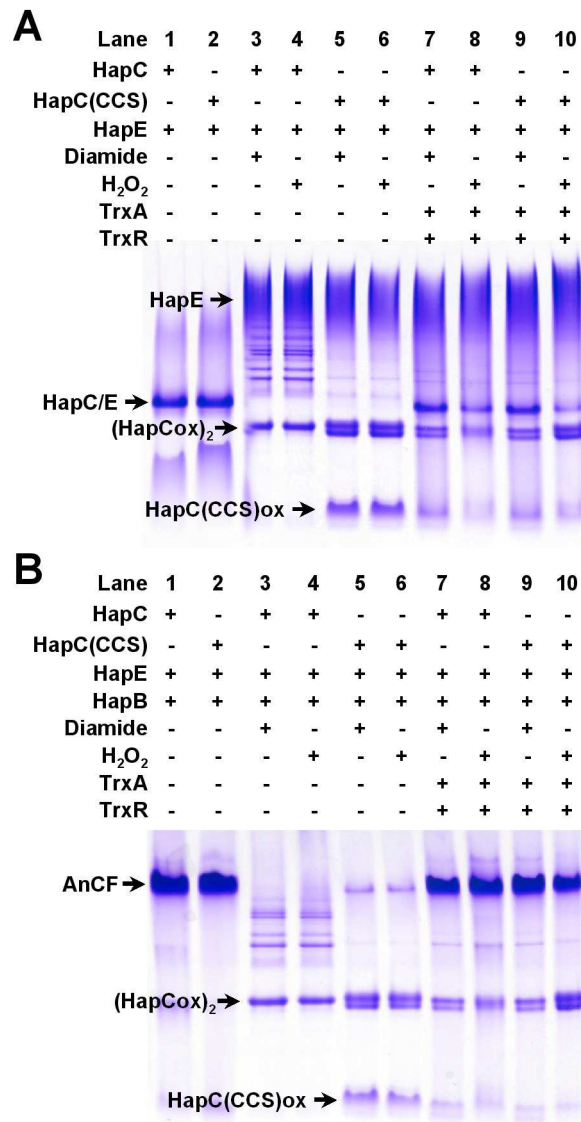


Figure S2. Disruption of the HapC/HapE heterodimer or the AnCF complex by oxidation and the recycling by the *A. nidulans* thioredoxin system *in vitro*. Native PAGE analysis of an oxidation/reduction cycle of HapC/HapE heterodimers (**A**) or the AnCF (**B**) composed of either HapC or HapC(CCS) using diamide or H₂O₂ for oxidation, respectively. Preformed HapC/HapE and AnCF samples (Lanes 1 and 2) contained 10 μ M of each Hap subunit.

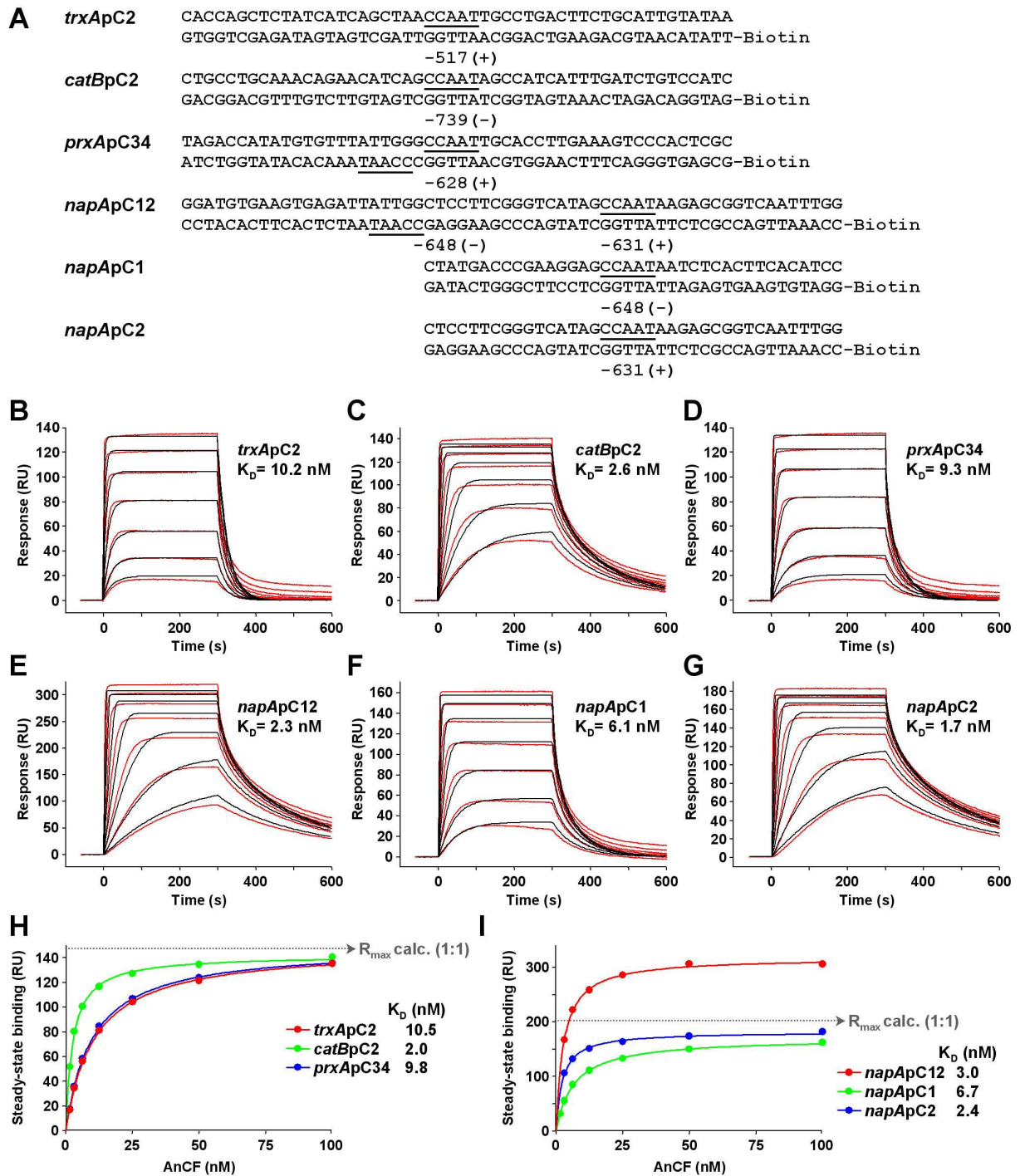


Figure S3. Real-time *in vitro* binding of AnCF to DNA encoding CCAAT boxes of the promoter regions of *A. nidulans* *trxA*, *catB*, *prxA* and *napA*. (A) Sequences of the duplexes used as DNA probes in SPR analyses. Positions of the CCAAT boxes (underlined) with respect to the start codon are indicated. (B-G) Responses of 100, 50, 25, 12.5, 6.25, 3.13 and 1.56 nM AnCF binding (red lines) to DNA duplexes are shown overlaid with the fit derived from a 1:1 interaction model (black lines). (H, I) Fit of the equilibrium data for DNA binding of AnCF. The calculated maximum binding capacities for a 1:1 interaction of AnCF with the immobilized DNA duplexes are marked by dotted arrows.

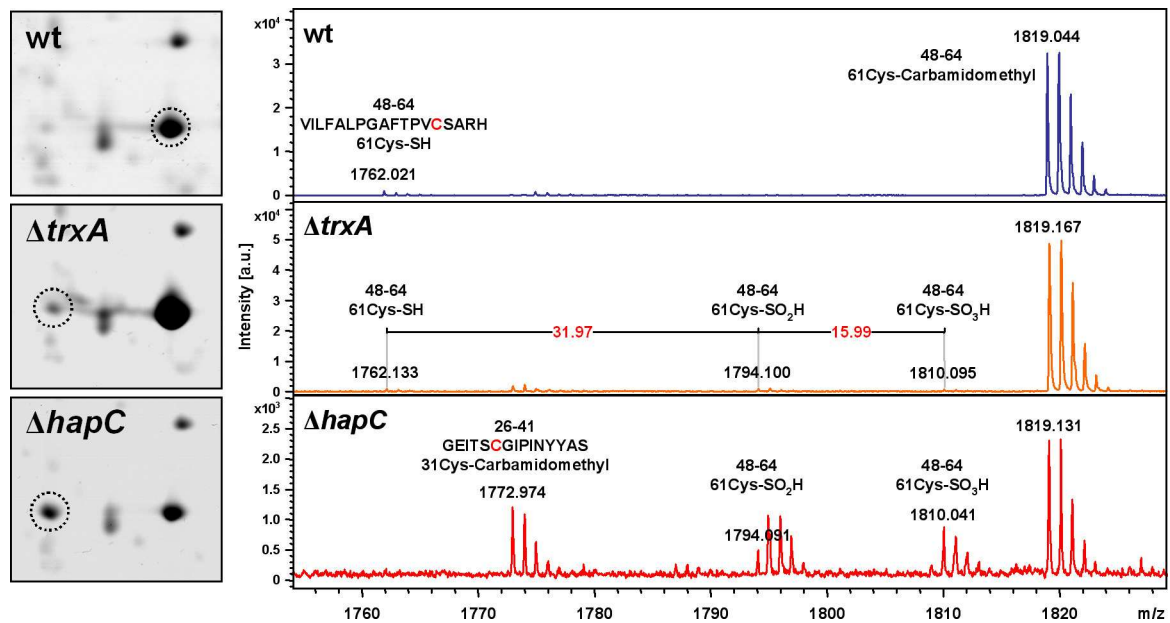


Figure S4. Peroxiredoxin A is hyperoxidized in an *A. nidulans* $\Delta hapC$ strain. The spots corresponding to normal and oxidized PrxA were collected from 2-D gels as indicated by dotted circles (left panel) and analyzed by MALDI MS (right panel). The oxidized spot, picked from a 2-D gel that was loaded with protein extracts from $\Delta hapC$ mycelia, contains both the sulfinic and sulfonic acid forms of peptide 48-64 from PrxA.

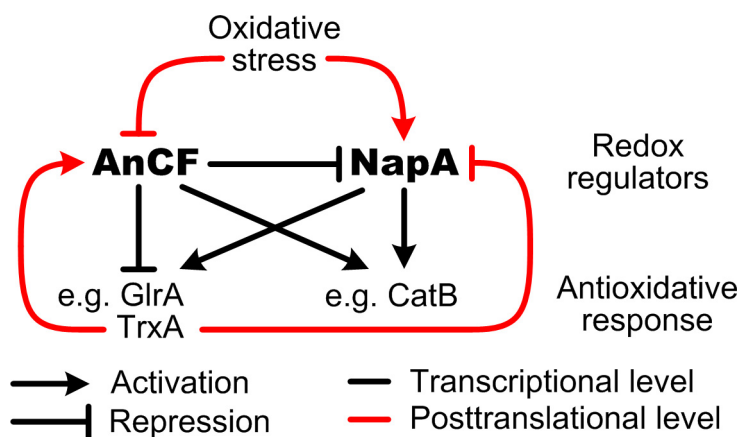


Figure S5. Mechanism for adaptation to oxidative stress situation in *A. nidulans*. The two redox-regulated regulators AnCF and NapA are controlled by interconnected feedback loops. Oxidative stress activates NapA causing nuclear accumulation and inactivates AnCF leading to cytoplasmic retention. The resulting oxidative stress response triggered by NapA activation and AnCF inactivation includes activation of the thioredoxin system, which inactivates NapA and reactivates AnCF. Moreover, AnCF most likely directly transcriptionally represses *napA* and also some NapA target genes, whereas NapA controls AnCF activity indirectly via the cellular redox state. It remains to be elucidated why AnCF affects some genes involved in antioxidative response inversely, e.g., *glrA* and *catB*.

Supplementary Tables

Table S1. Plasmids used in this study

Plasmids	Genotype	Reference
p123	Amp ^R ; <i>egfp</i>	(3)
pabaAnid	Amp ^R ; pUC 18, 4.1 kb <i>KpnI</i> fragment containing <i>pabaA1</i> and the flanking regions derived from <i>A. nidulans</i> .	(4)
pET39-AnTrxA(wt)-H6	Kan ^R ; T7 promoter - <i>trxA</i> - His ₆	(5)
pET39H6-AnTrxR	Kan ^R ; T7 promoter-His ₆ - <i>trxR</i>	(5)
pET43.1H6NapA	Amp ^R ; T7 promoter-His ₆ - <i>napA</i>	unpublished
pET43.1H6GpxA	Amp ^R ; T7 promoter-His ₆ - <i>gpxA</i>	unpublished
pEYFPC	Amp ^R ; Hyg ^R ; <i>eyfpC</i> terminal part	(6)
pEYFPN	Amp ^R ; Hyg ^R ; <i>eyfpN</i> terminal part	(6)
pHapC(SSS)-eGFP	Amp ^R ; <i>hapC(SSS)-egfp</i>	This study
pHapC-eGFP	Amp ^R ; <i>hapC-egfp</i>	(1)
pHapC-eGFP-pyro	Amp ^R ; <i>hapC-egfp</i> ; <i>pyroA</i>	This study
pHapC-YC	Amp ^R ; Hyg ^R ; <i>hapC-eyfpC</i>	This study
pHapC-YC-paba	Amp ^R ; <i>pabaA1</i> ; <i>hapC-eyfpC</i>	This study
pHapC-YC-pyr4	Amp ^R ; <i>pyr-4</i> ; <i>hapC-eyfpC</i>	This study
pHapE-eGFP	Amp ^R ; <i>hapE-egfp</i>	(1)
pHapE-YN	Amp ^R ; Hyg ^R ; <i>hapE-eyfpN</i>	This study
pKTB1	Amp ^R ; <i>pyr-4</i>	(7)
pKTB1-HapC(SSS)-eGFP	Amp ^R ; <i>pyr-4</i> ; <i>hapC(SSS)-egfp</i>	This study
pKTB1-HapC-eGFP	Amp ^R ; <i>pyr-4</i> ; <i>hapC-egfp</i>	This study
pTrxA-YN	Amp ^R ; Hyg ^R ; <i>trxA-eyfpN</i>	This study
pTrxA-YN-pyr4	Amp ^R ; <i>pyr-4</i> ; <i>trxA-eyfpN</i>	This study
pYC-pyr4	Amp ^R ; <i>pyr-4</i> ; <i>eyfpC</i> terminal part	This study
pYN-pyr4	Amp ^R ; <i>pyr-4</i> ; <i>eyfpN</i> terminal part	This study

Table S2. Oligonucleotides used in this study**A) BiFC analysis and amplification of hybridization probes**

Oligonucleotide	Sequence (5'→3')
3CS-Backward	AGATTTTTTCGGAAGCCTCGCTAGTAATAAAAGAGATGAATTCGCTCACAG ATTCTTGCATAGATTCTTT AG
3CS-Forward	TCTATGCAAGAATCTGTGAGCGAATTCATCTCTTTTATTACTAGCGAGGCT TCCGAAAAATCTCAACAGGA
AnPyro-For	GAGCAGCTGAAGCTTTGCGCGAAAGCGTAAGGAGA
AnPyro-Rev	GAGCAGCTGAAGCTTTCGCAATCTGACTTGACGC
EYFP3'EcoRI(C2)	GGCCGAATTCCTTACCTCTAAACAAGTGTACCTGTGCATT
EYFP5'EcoRI(C2)	TATAGAATTCGTACAGTGACCGGTGACTCTTTCTGGC
EYFP3'BamHI	GTACACGAGGACTGGATCCAAGAAGGATTACCTCTAAACAA
EYFP5'BamHI	TTGGGCGAGCTCGGATCCGTGACCGGTGACTCTTT
EYFP3'KpnI	GGCCGGTACCTTACCTCTAAACAAGTGTACCTGTGCATT
EYFP5'KpnI	TATAGGTACCGTACAGTGACCGGTGACTCTTTCTGGC
HapC3'NcoI	CAGAAAGCCATGGAAGATTCGCCACCAGC
HapC5'BamHI	CCAACAGCTGGATCCATGTGTCGTCGACC
HapC-BiFC-For	CCAACAGCTCCATGGCAATGTGTCGTCGACC
HapC-BiFC-Rev	AACCATGGGAGGGTATCCATAAGCTGAGGC
TrxNcoI for	GGCGGATCCATGGGTGCCTCTGAACACG
TrxNcoI rev	GATCCCCATGGAAGCAAGCAGAGCCTTG
AnYap1 for ¹	ATGGCCGACTACAATCTTTGTACCAACAC
AnYap1 rev ¹	CTACACGCGGCCAATGATGTCATCCACA
AnTrxA for ¹	ATGGGTGCCTCTGAACACGC
AnTxA rev ¹	CTAAGCAAGCAGAGCCTTGA
AnTrxR for ¹	ATGGTTCACTCCAAAGTAGTTATCATC
AnTrxR rev ¹	TTACAGAGCGGGGTTAGCGTGAGCAG
AnCatB for ²	ATGGTGGCCCGAACTTTGAGCAG
AnCatB rev ²	TCCAGGGCAAAGCGATCCAAGAAC
AnGlrA for ²	GTCCCCAAGAAGATGACCTGGAACCTC
AnGlrA rev ²	GTTAGCTCGGCCTGACCTGTGACA
AnGpxA for ¹	ATGCCGTTACATATTGTGGCTTAGTTCCA
An GpxA rev ¹	TTACGCCAGCTTGGCAGTCTCCCCGTC
AnPrxA for ²	GTCGTCGTTCTTAACACCGTTGG
AnPrxA rev ²	ACATTCCTGGACTGAGGAGAAG

¹ These probe-generating primers were used for amplification of cDNA encoded by the respective plasmids pET39-AnTrxA(wt)-H6, pET39H6-AnTrxR, pET43.1H6NapA and pET43.1H6GpxA.

² These primers were used for generation of northern hybridization probes based on the amplification of the largest exon of the respective gene.

B) SPR analysis

Gene	Oligo-nucleotide	Sequence ¹ (5'→3')
<i>sreA</i>	SREACB1 B-SREACB1i	CGCCCACCGAGTCTCGCTGCAG <u>CCAAT</u> CACAGCAAGCGTGATGACACTAC Biotin- TGACGAATCAGGTGGCGCGGATTATCATTGGTCAACCCGGTCTTCAAGAC
<i>trxA</i>	<i>trxAC2</i> B- <i>trxAC2i</i>	CACCAGCTCTATCATCAGCTAAC <u>CCAAT</u> TGCCTGACTTCTGCATTGTATAA Biotin- TTATACAATGCAGAAGTCAGGCAATTGGTTAGCTGATGATAGAGCTGGTG
<i>catB</i>	<i>catBC2</i> B- <i>catBC2i</i>	CTGCCTGCAAACAGAACATCAG <u>CCAAT</u> AGCCATCATTGATCTGTCCATC Biotin- GATGGACAGATCAAATGATGGCTATTGGCTGATGTTCTGTTTGCAGGCAG
<i>prxA</i>	<i>prxAC34</i> B- <i>prxAC34i</i>	TAGACCATATGTGTTT <u>ATTGGG</u> <u>CCAAT</u> TGCACCTTGAAAGTCCCACTCGC Biotin- GCGAGTGGGACTTTCAAGGTGCAATTGGCCAATAAACACATATGGTCTA
<i>napA</i>	<i>napAC12</i> B- <i>napAC12i</i>	GGATGTGAAGTGAGATT <u>ATTGG</u> CTCCTTCGGGTCATAG <u>CCAATA</u> AAGAGCG GTCAATTTGG Biotin- CCAAATTGACCGCTCTT <u>ATTGG</u> CTATGACCCGAAGGAG <u>CCAATA</u> AATCTCAC TTCACATCC
<i>napA</i>	<i>napAC1</i> B- <i>napAC1i</i>	CTATGACCCGAAGGAG <u>CCAATA</u> AATCTCACTTCACATCC Biotin-GGATGTGAAGTGAGATT <u>ATTGG</u> CTCCTTCGGGTCATAG
<i>napA</i>	<i>napAC2</i> B- <i>napAC2i</i>	CTCCTTCGGGTCATAG <u>CCAATA</u> AAGAGCGGTCAATTTGG Biotin-CCAAATTGACCGCTCTT <u>ATTGG</u> CTATGACCCGAAGGAG

¹ CCAAT boxes are underlined

Table S3. Comparison of the proteome of the *A. nidulans* wild type and $\Delta hapC$ **A) Proteins with higher levels in $\Delta hapC$ versus wild type**

Putative function	Gene ID	pI/MW	Fold changes	Sequence coverage (%)	Mascot score
Elongation factor 3	AN6700.4	6.2/102.2	2.8	36.2	231.0
pH response regulator PalA	AN4351.4	7.2/94.8	2.8	8.6	56.1
Eukaryotic translation initiation factor subunit eIF2A	AN2775.4	9.6/73.5	2.4	41.1	173.0
ATP citrate lyase, subunit 1	AN2436.4	8.2/71.6	2.4	66.0	275.0
Choline oxidase (CodA)	AN1429.4	6.7/60.0	4.3	47.6	192.0
Translation initiation factor 4B	AN7350.4	9.6/53.6	3.4	28.8	104.0
T-complex protein 1, eta subunit	AN5713.4	6.4/60.8	4.9	39.4	115.0
Nuclear segregation protein (Bfr1)	AN0753.4	9.9/54.3	2.3	21.3	64.9
ATP synthase alpha chain, mitochondrial precursor ²	AN1523.4	9.6/60.1	2.1	36.5	122.0
ThiF domain protein, putative	AN4714.4	7.6/57.3	2.1	37.9	126.0
GDP-mannose pyrophosphorylase A	AN1911.4	7.2/48.5	2.1	18.2	56.9
hypothetical oxidoreductase	AN2682.4	5.9/46.1	2.4	32.0	106.0
Hydroxymethylglutaryl-CoA synthase	AN4923.4	6.2/50.8	2.6	37.0	93.4
Ornithine aminotransferase	AN1810.4	5.9/49.2	2.2	55.9	192.0
hypothetical protein similar to elongation factor EF-Tu	AN1084.4	6.9/48.3	2.1	41.2	113.0
C-3 sterol dehydrogenase/C-4 decarboxylase	AN7575.4	7.1/45.5	2.5	43.2	117.0
60S ribosome biogenesis protein Brx1	AN10055.4	9.7/40.4	2.1	19.8	54.4
conserved expressed oxidoreductase ¹	AN2208.4	7.1/40.1	2.2	59.4	134.0
Glutathione S-transferase ¹	AN10273.4	7.8/40.1	2.1	45.6	153.0
CCCH finger DNA binding protein, putative	AN6922.4	9.4/39.0	6.3	27.6	70.8
conserved hypothetical protein	AN6891.4	10.4/41.5	2.2	45.1	132.0
DnaJ domain protein Psi	AN2238.4	9.9/40.2	4.4	24.9	64.4
Tryptophanyl-tRNA synthetase, putative	AN6488.4	9.8/42.7	3.4	36.5	117.0
translational initiation factor 2 beta	AN2992.4	9.3/33.8	3.5	40.5	79.1
hypothetical protein similar to 60S ribosomal protein L5	AN1013.4	9.1/34.4	2.5	50.5	103.0
Pyridoxine biosynthesis protein PyroA, involved in response to singlet oxygen ¹	AN7725.4	6.3/32.3	3.2	41.1	96.6
Proteasome component Pre8 ²	AN6726.4	5.5/30.1	3.8	21.6	51.8
Proteasome component Pre6, putative	AN8054.4	8.6/27.0	4.2	35.4	51.9
Proteasome core alpha 1 component	AN4869.4	6.7/27.8	2.5	35.8	91.4
hypothetical protein similar to NADP-dependent mannitol dehydrogenase	AN7590.4	6.8/28.3	2.7	47.4	82.3
Formyltetrahydrofolate deformylase, putative	AN0495.4	7.3/31.8	2.0	67.1	154.0
conserved hypothetical protein	AN7208.4	6.5/20.4	2.1	63.8	83.1
Peroxiredoxin/Prx5-like /allergen Asp F3	AN8692.3	5.6/18.5	0.75	32.7	50.1
Peroxiredoxin/Prx5-like /allergen Asp F3 (Sulfinic/Sulfonic acid)	AN8692.3	5.6/18.5	3.2	67.9	99.0
Peroxiredoxin/Prx5-like /allergen Asp F3 (total) ^{1,2}	AN8692.3	5.6/18.5	0.98	-	-
hypothetical protein similar to NADH-ubiquinone oxidoreductase	AN1063.4	8.2/18.2	2.0	53.5	68.9
Thioredoxin ¹	AN0170.4	9.8/20.2	1.4	37.0	48.6

¹ Proteins involved in oxidative stress response² Production of these proteins is also impaired in a $\Delta glrA$ strain (8)

B) Proteins with lower levels in *ΔhapC* versus wild type

Putative function	Gene ID	pI/MW	Fold changes	Sequence coverage (%)	Mascot score
Cdc48 involved in response to stress	AN7254.4	4.7/89.4	0.21	26.2	156.0
Phosphoglucomutase	AN2867.4	6.0/60.7	0.35	37.2	151.0
UDP-N-acetylglucosamine pyrophosphorylase ²	AN9094.4	6.4/55.8	0.49	33.7	124.0
Elongation factor 2	AN6330.4	6.6/93.7	0.14	8.4	47.5
Tubulin alpha chain (<i>tubB</i>)	AN7570.4	4.7/50.0	0.21	30.2	91.7
Aminotransferase, classes I and II, putative	AN5591.4	6.0/53.7	0.39	29.7	96.0
4-aminobutyrate aminotransferase	AN2248.4	9.3/55.5	0.46	23.3	69.7
NADP-specific glutamate dehydrogenase	AN4376.4	6.4/49.6	0.24	65.6	202.0
Peptidyl-prolyl cis-trans isomerase (orthologue of <i>S. pombe</i> Wis2, involved in response to stress)	AN4583.4	5.5/41.1	0.32	30.4	96.4
Glutamine synthetase	AN4159.4	5.8/39.7	0.39	31.7	103.0
Elongation factor 1-alpha	AN4218.4	9.7/50.5	0.45	27.6	86.0
S-adenosylmethionine synthetase ²	AN1222.4	5.2/42.2	0.12	21.4	66.7
hypothetical oxidoreductase ¹	AN8815.4	5.5/40.5	0.12	53.2	118.0
Oxidoreductase, 2-nitropropane dioxygenase family	AN6031.4	7.7/37.6	0.49	49.9	105.0
Alcohol dehydrogenase I (<i>alcA</i>) ²	AN8979.4	7.8/37.1	0.03	37.7	79.1
Thiamine biosynthesis protein	AN8009.4	5.8/38.2	0.41	34.6	71.3
Thiazole synthase (<i>thiF</i>)	AN3928.4	5.4/35.6	0.13	28.4	71.6
Inorganic pyrophosphatase	AN2968.4	5.0/32.4	0.08	38.3	97.0
Spermidine synthase, putrescine aminopropyltransferase	AN0687.4	5.1/33.3	0.46	40.8	120.0
NADH-ubiquinone oxidoreductase 24 kDa subunit, mitochondrial	AN6077.4	6.2/30.0	0.48	33.7	70.6
putative 1-Cys peroxiredoxin ^{1,2}	AN10223.4	5.3/23.3	0.17	27.2	65.8
outer mitochondrial membrane protein porin ²	AN4402.4	9.5/29.9	0.45	39.1	73.5
Woronin body major protein ²	AN4695.4	7.6/19.7	0.50	47.5	64.7
Nitroreductase family protein, putative	AN2343.4	5.3/24.5	0.11	29.3	68.7
hypothetical PH domain protein ²	AN3674.4	4.5/58.2	0.24	18.7	83.2
NADH-quinone oxidoreductase Pst2, putative	AN0297.4	6.2/22.0	0.49	53.9	85.6
Acireductone dioxygenase ARD family protein, putative	AN9527.4	5.0/20.9	0.03	59.6	118.0
allergen, putative AhpC/Tsa family protein ¹	AN8080.4	5.7/17.8	0.31	49.7	71.7
Co-chaperone, orthologue of <i>S. cerevisiae</i> AHA1 involved in response to stress	AN5602.4	5.0/36.6	0.13	26.9	59.7
DUF636 domain protein	AN7594.4	6.5/14.8	0.24	64.0	75.5
Methionine-R-sulfoxide reductase MsrB ¹	AN1932.4	6.7/16.6	0.55	45.8	59.2
Ubiquitin-conjugating enzyme	AN8702.4	6.5/16.9	0.43	12.0	27.5
Conserved hypothetical protein	AN5907.4	7.4/16.9	0.43	54.8	85.0
Calponin homology, EB1-like C-terminal motif	AN2862.4	4.7/27.1	0.15	26.0	55.1
RNA binding protein, putative	AN5480.4	6.3/13.1	0.35	64.7	90.5

¹ Proteins involved in oxidative stress response

² Production of these proteins is also impaired in a *ΔglrA* strain (8)

Table S4. Comparison of the proteome of the *A. nidulans* wild type and Δ *trxA*

Putative function	Gene ID	pI/MW	Fold changes	Sequence coverage (%)	Mascot score
Proteins with higher levels in Δ<i>trxA</i> versus wild type					
Elongation factor 3*	AN6700.4	6.2/102.2	2.1	36.2	231.0
Phenylalanyl-tRNA synthetase, beta subunit	AN4086.4	5.3/67.7	2.2	23.8	108.0
Thioredoxin reductase	AN3581.4	5.0/36.1	5.5	67.8	160.0
DnaJ domain protein Psi*	AN2238.4	9.9/40.2	2.2	30.0	73.8
Tryptophanyl-tRNA synthetase, putative	AN6488.4	9.8/42.7	2.5	36.5	117.0
Proteasome component Pre6, putative*	AN8054.4	8.6/27.0	2.4	35.4	51.9
Peroxiredoxin/Prx5-like /allergen Asp F3	AN8692.3	5.6/18.5	2.8	32.7	50.1
Peroxiredoxin/Prx5-like /allergen Asp F3 (Sulfinic/Sulfonic acid)	AN8692.3	5.6/18.5	2.4	57.7	91.9
Peroxiredoxin/Prx5-like /allergen Asp F3 (total)*	AN8692.3	5.6/18.5	2.8	-	-
Proteins with lower levels in Δ<i>trxA</i> versus wild type					
Heat shock protein	AN10202.4	5.3/42.7	0.38	50.3	132.0
Heat shock 70 kDa protein	AN11227.4	5.4/40.8	0.35	57.3	151.0
Mannitol dehydrogenase, putative	AN2815.4	6.6/56.6	0.49	31.5	116.0
Elongation factor 2*	AN6330.4	6.6/93.7	0.40	8.4	47.5
Tubulin alpha chain (<i>tubB</i>)*	AN7570.4	4.7/50.0	0.32	30.2	91.7
eukaryotic initiation factor 4A	AN2932.4	4.8/44.9	0.28	53.3	162.0
Peptidyl-prolyl cis-trans isomerase (orthologue of <i>S. pombe</i> Wis2, involved in response to stress)*	AN4583.4	5.5/41.1	0.42	30.4	96.4
Aspartate transaminase	AN6048.4	7.2/48.8	0.42	62.0	196.0
Septin CDC11, putative	AN4667.4	4.7/43.1	0.30	31.9	88.7
Elongation factor 1-alpha*	AN4218.4	9.7/50.5	0.32	27.6	86.0
S-adenosylmethionine synthetase*	AN1222.4	5.2/42.2	0.35	60.8	176.0
Thioredoxin, putative	AN1639.4	4.7/36.3	0.28	26.7	61.4
Alcohol dehydrogenase I (<i>alcA</i>)*	AN8979.4	7.8/37.1	0.26	37.7	79.1
Translation initiation factor 3 subunit EifCj, putative	AN5745.4	4.6/29.4	0.45	34.0	50.5
Cofactor for methionyl- and glutamyl-tRNA synthetases, putative	AN10474.4	7.1/ 46.5	0.28	25.6	66.5
Thiazole synthase (<i>thiF</i>)*	AN3928.4	5.4/35.6	0.29	41.7	103.0
Electron transfer flavoprotein alpha subunit, putative	AN6699.4	5.5/36.4	0.15	52.7	142.0
Inorganic pyrophosphatase*	AN2968.4	5.0/32.4	0.18	38.3	97.0
Heat-shock protein	AN5781.4	5.2/23.4	0.17	45.9	93.8
NADH-ubiquinone oxidoreductase 24 kDa subunit, mitochondrial	AN6077.4	6.2/30.0	0.41	33.7	70.6
40S ribosomal protein S3Ae	AN8870.4	10.8/29.1	0.27	25.4	52.2
hypothetical protein similar to NADP-dependent mannitol dehydrogenase	AN7590.4	6.8/28.3	0.40	47.4	100.0
Cysteine dioxygenase Cdo1, putative	AN4081.4	6.2/23.9	0.45	52.8	94.4
Cytochrome c subunit Vb, putative	AN4525.4	6.2/22.0	0.43	43.3	59.2
ATP synthase D chain, mitochondrial	AN6631.4	8.9/19.5	0.09	49.1	84.3
40S ribosomal protein S19	AN4060.4	9.4/16.4	0.18	37.2	61.1
Methionine-R-sulfoxide reductase MsrB*	AN1932.4	6.7/16.6	0.40	45.8	59.2
Chaperonin, putative	AN2432.4	9.1/11.2	0.45	52.4	92.5

* Proteins with higher and lower abundance in both Δ *trxA* and Δ *hapC* strains, respectively

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