

Supplementary Information

Supplementary Table S1: Strains used in this study: *p_{xyIP}* is the promoter region of the 1,4-endoxylanase (*xyIP*) promoter. The asterisk (*) indicates a second *pyrG* gene copy (endogenous *A. fumigatus pyrG*); both copies are not functional.

Strain	Background	Genotype	Reference
AfS77 (wt)	ATCC46645	<i>ΔakuA::loxP</i>	1
<i>nbp35^{xyIP}</i>	AfS77	<i>p_{nbp35}::hph-p_{xyIP}</i>	this study
<i>nfs1^{xyIP}</i>	AfS77	<i>p_{nfs1}::hph-p_{xyIP}</i>	this study
<i>ΔgshA</i>	AfS77	<i>ΔgshA::ptrA</i>	this study
<i>gshA^{rec}</i>	AfS77	<i>ΔgshA::ptrA, 5'gshA::gshA-hph</i>	this study
A1160	CEA17	<i>ΔakuB::pyrG⁻, pyrG⁻ *</i>	2
A1160P+ (wt)	A1160	<i>ΔpyrG⁻::pyrG⁺</i>	3
<i>ΔmrsA</i>	A1160	<i>ΔmrsA::pyrG⁺</i>	4

Supplementary Table S2: Primers used in this study:

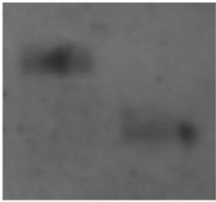
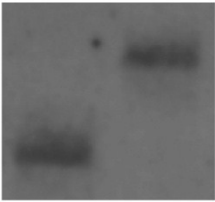
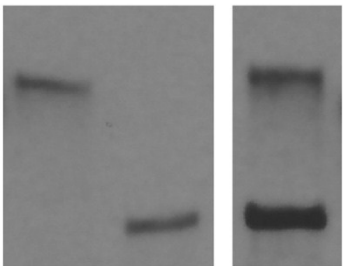
Number	Primer	Sequence
1	nbp35_5'f	AATTCGAGCTCGGTACGTTGCATCTAATACTACATTTTC
2	nbp35_5'r	TACCTAGGTATTATGCGGAAACCAGC
3	hph1_f	GCATAATACCTAGGTACAGAAAGTCC
4	hph1_r	CGCATCAGTGCGGCCTCTAGAAAGAAGGATTACC
5	xyIP1_f	ATCCTTCTTTCTAGAGGCCGCACTGATGCGAGCAACAGTATG
6	xyIP1_r	GGCGCCATGGTTGGTTCTTCGAGT
7	nbp35_3'f	AACCAACCATGGCGCCGTCCGTG
8	nbp35_3'r	GCCAAGCTTGCATGCCGGGCTGTCCGGAAG
9	nfs1_5'f	AATTCGAGCTCGGTACAAGAGTGATATTAGAGCGAA
10	nfs1_5'r	TACCTAGGAATTTAGATGTTGCTCAACC
11	hph2_f	TCTAAATTCCTAGGTACAGAAGTCC
12	xyIP2_r	CTAGACATGGTTGGTTCTTCGAGT
13	nfs1_3'f	AACCAACCATGTCTAGCGTTACGC
14	nfs1_3'r	GCCAAGCTTGCATGCCTGCGTCAACATAGCA
15	gshA_5'f	CACGGTGTGAGTTGGTC
16	gshA_5'r	ATCCTGCAGGACCCAACCAACCCAGTA
17	gshA_3'f	GCTACCTACCTTGACCTC
18	gshA_3'r	CCTCTTGACGGAATCTG
19	gshA_5'nest	TCCGCTTCCTGTCTTTCC
20	gshA_3'nest	TTCTACACCAGCACCTCC
21	gshA_f	AATTCGAGCTCGGTACTTTGTGAGAGACCAGC
22	gshA_r	TACTCGAGGATCAGTTTGGTTACG
23	hph4_f	AACTGATCCTCGAGTACCATTTAATTCT
24	hph4_r	GCCAAGCTTGCATGCCACGCGTTTTATTCTTGTT

Supplementary Table S3: Homologies between *S. cerevisiae*, *A. fumigatus* and *H. sapiens*. *S. cerevisiae* proteins were used as bait to search the *A. fumigatus* and human proteoms (NCBI-BLAST on FungiDB^{5,6}).

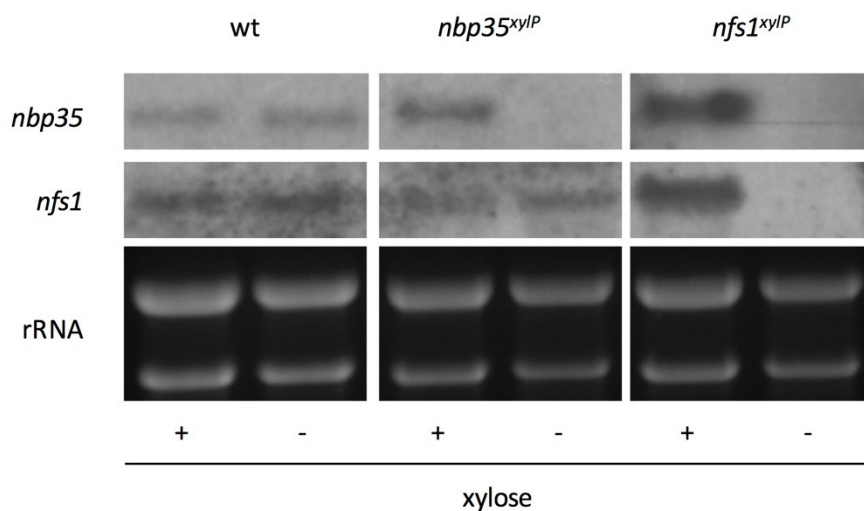
<i>S. cerevisiae</i> , strain S288C		<i>A. fumigatus</i> A1163 homolog	BLASTP <i>A. fumigatus</i>		<i>H. sapiens</i>
Name	ID	ID	E-value	Identity	
MRS3	YJL133W	AFUB_078550	1,00E-92	156/303 (51%)	SLC25A37
MRS4	YKR052C		4,00E-86	149/307 (49%)	
YFH1	YDL120W	AFUB_067610	1,00E-26	50/117 (43%)	FXN
NFS1/SPL1	YCL017C	AFUB_034990	0	311/474 (66%)	NFS1
ISD11	YER048W-A	AFUB_042560	1,00E-16	34/80 (43%)	LYRM4
ARH1	YDR376W	AFUB_028270	3,00E-86	197/533 (37%)	FDXR
YAH1	YPL252C	AFUB_052400	4,00E-61	85/133 (64%)	FDX2
ISU1/NUA1	YPL135W	AFUB_063840	5,00E-63	91/119 (76%)	ISCU
SSQ1/SSH1/SSC2	YLR369W	AFUB_025800	0	326/626 (52%)	HSPA9
JAC1	YGL018C	AFUB_082890	1,00E-27	56/177 (32%)	HSCB
MGE1/YGE1/GRPE	YOR232W	AFUB_028670	1,00E-49	97/198 (49%)	GRPEL1
GRX5	YPL059W	AFUB_063050	3,00E-57	88/144 (61%)	GLRX5
ISA1	YLL027W	AFUB_067770	4,00E-56	81/117 (69%)	ISCA1
ISA2	YPR067W	AFUB_026180	2,00E-22	50/126 (40%)	ISCA2
NFU1/NUB1	YKL040C	AFUB_005020	3,00E-50	97/235 (41%)	NFU1
IBA57/CAF17	YJR122W	AFUB_060100	2,00E-21	127/492 (26%)	IBA57
BOL3/AIM1	YAL046C	AFUB_078500	1,00E-11	24/70 (34%)	BOLA3
CFD1/DRE3	YIL003W	AFUB_052120	3,00E-94	152/306 (50%)	NUBP2
NBP35	YGL091C	AFUB_031640	9,00E-161	210/313 (67%)	NUBP1
DRE2	YKR071C	AFUB_008090	2,00E-28	53/119 (45%)	CIAPIN1
TAH18	YPR048W	AFUB_054840	8,00E-102	227/687 (33%)	NDOR1
GRX3	YDR098C	AFUB_030610	8,00E-68	107/256 (42%)	GLRX3
GRX4	YER174C		5,00E-67	110/259 (42%)	
NAR1	YNL240C	AFUB_068950	2,00E-73	172/490 (35%)	NARFL
CIA1	YDR267C	AFUB_008350	3,00E-64	146/427 (34%)	CIAO1
CIA2	YHR122W	AFUB_083590	5,00E-51	80/131 (61%)	FAM96B
MET18/MMS19	YIL128W	AFUB_082160	1,00E-50	267/1113 (24%)	MMS19

Supplementary Table S4: Numerical values [$\mu\text{mol/g}$ dry weight] for iron uptake, FC- and chelatable iron content

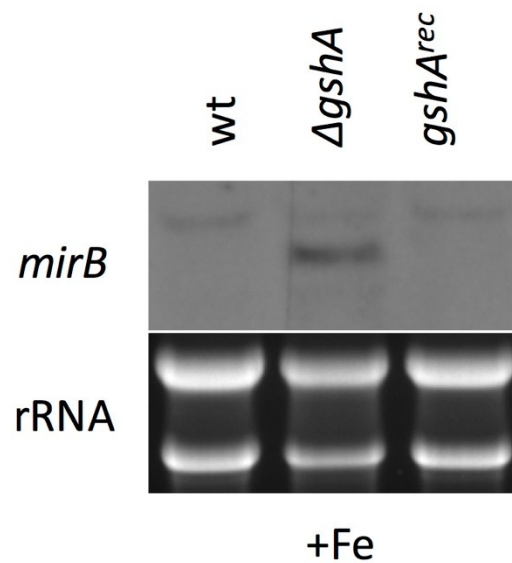
	TAFC uptake		FC content		chelatable iron	
	mean	standard deviation	mean	standard deviation	mean	standard deviation
AfS77	1.10	0.22	0.45	0.11	0.39	0.05
<i>nbp35^{xyIP}</i>	0.82	0.10	0.30	0.09	0.80	0.09
<i>nfs1^{xyIP}</i>	29.87	8.38	4.27	1.07	19.27	6.72
<i>ΔgshA</i>	32.63	9.15	4.07	1.02	20.63	7.07
A1160P+	1.20	0.46	0.44	0.05	0.37	0.15
<i>ΔmrsA</i>	20.67	2.29	2.50	0.40	7.40	0.72

Gene	<i>nbp35</i>		<i>nfs1</i>		<i>gshA</i>		
	MT	WT	MT	WT	MT	WT	REC
							
Enzyme	BamHI/EcoRI		PstI		SmaI/XhoI		
Probe	3'		5'		5'		
Signal	3419 or 2341 bp		3374 or 5540 bp		9500 or 4529 bp		

Supplemental Fig. S1: Southern blot confirmed correct gene manipulation. Target genes are indicated at the top. WT is the recipient strain; MT is the manipulated mutant strain. Enzyme indicates the restriction enzyme(s) used to digest genomic DNA. Probe indicates if the hybridization probe targeted 5'- or 3' homologous region used for homologous recombination. Signal specifies the length of the expected fragments for WT/MT.



Supplemental Fig. S2: Xylose-dependent transcription of p_{xyIP} -targeted genes. Strains were cultivated as described in the experimental part for 20 hours in MM +Fe 0.1% xylose 25°C. After washing the germlings one half from each strain was added to MM +Fe 0.1% xylose, the other half to MM +Fe and no xylose for 20 hours at 37°C. Expression levels of *nbp35* and *nfs1* were compared at different conditions by performing Northern analysis.



Supplemental Fig. S3: GSH depletion derepresses *mirB* during iron sufficiency. In contrast to Fig. 5, the film for detection of signals was exposed longer. Ribosomal RNA (rRNA; same as in Fig. 5) is shown as control for RNA quality and loading.

HapX	277	...	CGFCSDGTPCIC	...	288
Yap5	178	...	CGFCNDNTTCVC	...	290

Supplemental Fig. S4: Alignment of the cysteine-rich regions of HapX and Yap5.

References:

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