SUPPLEMENTAL INFORMATION

Supplemental figure legends

Supplemental Figure 1. Alignment of deduced amino acid sequences of GR from various organisms. Deduced amino acid sequences of GR from *Escherichia coli gorA*, *S. cerevisiae GLR1* (YPL091W), and human (GSHR_HUMAN) were compared with that of AN0932.3 (*glrA*) from *A. nidulans*, and aligned using CLASTAL W software. Motif for active center is highlighted in red font. Other conserved motifs are boxed: yellow and red, Rossmann folds; green, motif for contacting isoalloxazine ring; blue, FAD-binding motif. Met^1 and Met^{86} are shown in blue font.

Supplemental Figure 2. Partial nucleotide and deduced amino acid sequences of *glrA*. Red and blue fonts predicted start sites of transcription and translation (Met^1 and Met^{86}), respectively.

Supplemental Figure 3. Enzymic properties of *A. nidulans* GlrA. *A*, SDS-PAGE of purified enzyme. Purified enzymes (1 μg) were resolved by SDS-PAGE on 10% polyacrylamide gels and stained with Coomassie brilliant blue. Lanes: 1, markers (Precision Protein Standard kit, Bio-Rad); 2, rGlrA. *B*, Superose 6 10/300 GL gel filtration of rGlrA. Circle and squares represent elution volume of rGlrA and molecular weight markers, respectively. Molecular weight markers: 1, bovine serum albumin (67 kDa); 2, ovalbumin (43 kDa); 3, carbonic anhydrase (30 kDa); 4, soybean trypsin inhibitor (20.1 kDa). *C*, Redox titration of rGlrA. rGlrA (38 μM) in 100 mM sodium phosphate buffer (pH 7.2) at 25°C. Visible spectra are representative of those obtained during titration by sodium dithionite. *D*, Spectral changes of rGlrA upon incubation with NADPH. rGlrA (38 μM) in 50 mM sodium phosphate buffer (pH 7.2) was mixed with 0.2 mM NADH under aerobic conditions. Spectra: black line, resting (oxidized); grey line, at 10 s after addition; dotted line, GSSG was added after addition of NADPH.

Supplemental Figure 4. Two-dimensional gel electrophoresis of cell-free extracts of *A. nidulans* WT (A) and DGR1 strains (B). Mycelia of *A. nidulans* WT (FGSC A26) and DGR1 were incubated in MMDN medium for 6 h after pre-cultivating conidia at 30°C for 20 h (WT) and 28 h (DGR1).

Supplemental Figure 5. Two-dimensional gel electrophoresis of cell-free extracts of *A. nidulans* A26 (A) and DGR1 strains (B). Mycelia of *A. nidulans* WT (FGSC A26) and DGR1 were incubated in MMDN medium for 6h after pre-cultivating conidia at 30°C for 20 h (WT) and 28 h (DGR1).

Supplemental Figure 6. Phylogenetic relationship among glutathione-*S*-transferases from various organisms. Multiple alignment and neighbor joining methods were performed by using Molecular Evolutionary Genetics Analysis (MEGA) Software Version 4.0 (http://evolgen.biol.metro-u.ac.jp/MEGA/). Accession numbers were from Broad Institute (http://www.broad.mit.edu/annotation/fgi/) or EMBL/GenBank/DDBJ.

Sato *et al*. Supplemental Fig. 1

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AN0932.3        MLSRSSLRSLSAQASKIRLTRSSLVCSPITQSNRAYTSYGIATRNQKRGVLDSSSRSAIS
S. cerevisiae   MLSATKQTFRSLQIRTMSTN----------------------------------------
Human           MALLPRALSAGAGPSWRRAARAFRG-----------------------------FLLLLP
E. coli         ------------------------------------------------------------

AN0932.3        TPTGLRLASLTRQFSSTSPAANSSNMPPVETKQYDYIVLGGGSGGSGSGRRAAGWYGAKT
S. cerevisiae   ------------------------------TKHYDYLVIGGGSGGVASARRAASYG-AKT
Human           EPAALTRALSRAMACRQEPQPQGPPPAAGAVASYDYLVIGGGSGGLASARRAAELG-ARA
E. coli         -----------------------------MTKHYDYIAIGGGSGGIASINRAAMYG-QKC

AN0932.3        LIVESGRAGGTCVNVGCVPKKMTWNFASITESIEAGRHYGYDL-----PHNIDVNYTHFK
S. cerevisiae   LLVEAKALGGTCVNVGCVPKKVMWYASDLATRVSHANEYGLYQNLPLDKEHLTFNWPEFK
Human           AVVESHKLGGTCVNVGCVPKKVMWNTAVHSEFMHDH-ADYGFPSCEG-----KFNWRVIK
E. coli         ALIEAKELGGTCVNVGCVPKKVMWHAAQIREAIHMYGPDYGFDTTIN-----KFNWETLI

AN0932.3        KLRDSTIERLNGVYEKNWGNEGIDLVHGRARFVEKKTIEVTNQDGSRTRYTAPHILIATG
S. cerevisiae   QKRDAYVHRLNGIYQKNLEKEKVDVVFGWARFNKDGNVEVQKRDNTTEVYSANHILVATG
Human           EKRDAYVSRLNAIYQNNLTKSHIEIIRGHAAFTSD---PKPTIEVSGKKYTAPHILIATG
E. coli         ASRTAYIDRIHTSYENVLGKNNVDVIKGFARFVD-----AKTLEVNGETITADHILIATG

AN0932.3        GRPSLPD---IKGSEHGISSDGFFEIEELPPKLAVVGAGYIAVELAGVMGTVGVDTHMFI
S. cerevisiae   GKAIFPEN--IPGFELGTDSDGFFRLEEQPKKVVVVGAGYIGIELAGVFHGLGSETHLVI
Human           GMPSTPHESQIPGASLGITSDGFFQLEELPGRSVIVGAGYIAVEMAGILSALGSKTSLMI
E. coli         GRPSHP---DIPGVEYGIDSDGFFALPALPERVAVVGAGYIAVELAGVINGLGAKTHLFV

AN0932.3        RGETFLRKFDPMIQKTMTERYEAVGIHVHKKHPGIKEVQLLR--DGKGKDKLLKLIMNDG
S. cerevisiae   RGETVLRKFDECIQNTITDHYVKEGINVHKLS----KIVKVE--KNVETDKLKIHMNDSK
Human           RHDKVLRSFDSMISTNCTEELENAGVEVLKFSQVKEVKKTLSGLEVSMVTAVPGRLPVMT
E. coli         RKHAPLRSFDPMISETLVEVMNAEGPQLHTNAIPKAVVKNTDGSLTLELEDGR-------

AN0932.3        SEMEVNELLWAIGRVPEVEDLHLEIPGVELNKSGHVVVDEYQNTNVEGIYAIGDVTGQAE
S. cerevisiae   SIDDVDELIWTIGRKSHLG-MGSENVGIKLNSHDQIIADEYQNTNVPNIYSLGDVVGKVE
Human           MIPDVDCLLWAIGRVPNTKDLSLNKLGIQTDDKGHIIVDEFQNTNVKGIYAVGDVCGKAL
E. coli         -SETVDCLIWAIGREPANDNINLEAAGVKTNEKGYIVVDKYQNTNIEGIYAVGDNTGAVE

AN0932.3        LTPVAIAAGRQLGNRLFGGPQFKNAKLSYDNIPTVVFSHPEVGTVGLTEPQARERFGDEN
S. cerevisiae   LTPVAIAAGRKLSNRLFGPEKFRNDKLDYENVPSVIFSHPEAGSIGISEKEAIEKYGKEN
Human           LTPVAIAAGRKLAHRLFEYK--EDSKLDYNNIPTVVFSHPPIGTVGLTEDEAIHKYGIEN
E. coli         LTPVAVAAGRRLSERLFNNK--PDEHLDYSNIPTVVFSHPPIGTVGLTEPQAREQYGDDQ

AN0932.3        VKVYHTRFPAMFYSVFPPEEKAKNPTEFKMVVAGPEEKVVGLHLLGLGVGEMTQGFGVAV
S. cerevisiae    IKVYNSKFTAMYYAMLSE---KSPTRYKIVCAGPNEKVVGLHIVGDSSAEILQGFGVAI
Human           VKTYSTSFTPMYHAVTKR----KTKCVMKMVCANKEEKVVGIHMQGLGCDEMLQGFAVAV
E. coli         VKVYKSSFTAMYTAVTTH----RQPCRMKLVCVGSEEKIVGIHGIGFGMDEMLQGFAVAL

AN0932.3        KMGATKKDFDSCVAIHPTSAEELVTLR
S. cerevisiae   KMGATKADFDNCVAIHPTSAEELVTMR
Human           KMGATKADFDNTVAIHPTSSEELVTLR
E. coli         KMGATKKDFDNTVAIHPTAAEEFVTMR
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Sato *et al*. Supplemental Fig. 2

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TGCAGATTCCCCCCTCCCCCTCCTTTTATTCCTTCGGGTGCCCGTCAATAATCTCCCTCC
                                     −22

        10        20        30        40        50        60
ATGCTCTCTCGCTCCTCGCTTCGGTCATTAAGTGCCCAAGCGTCGAAGATTCGGCTCACC
M  L  S  R  S  S  L  R  S  L  S  A  Q  A  S  K  I  R  L  T

        70        80        90       100       110       120
CGTTCGTCTCTCGTCTGTTCGCCAATCACTCAATCCAATCGAGCCTACACAAGCTACGGC
R  S  S  L  V  C  S  P  I  T  Q  S  N  R  A  Y  T  S  Y  G

       130       140       150       160       170       180
ATTGCCACTAGAAACCAGAAGAGAGGAGTCCTGGATTCTTCATCTCGCTCAGCTATCTCC
I  A  T  R  N  Q  K  R  G  V  L  D  S  S  S  R  S  A  I  S
                                                  +172
       190       200       210       220       230       240
ACTCCCACTGGGCTGCGTCTGGCCTCGCTCACAAGACAATTTTCATCCACTTCGCCTGCA
T  P  T  G  L  R  L  A  S  L  T  R  Q  F  S  S  T  S  P  A

       250       260       270       280       290       300
GCGAACTCGAGCAACATGCCGCCGGTGGAGACAAAACAGTATGACTATATCGTGTTAGGT
A  N  S  S  N  M  P  P  V  E  T  K  Q  Y  D  Y  I  V  L  G

       310       320       330       340       350
GGTGGTAGCGGTGGTAGCGGTAGTGGCCGTAGAGCTGCCGGTTGGTAC.....
G  G  S  G  G  S  G  S  G  R  R  A  A  G  W  Y  .....
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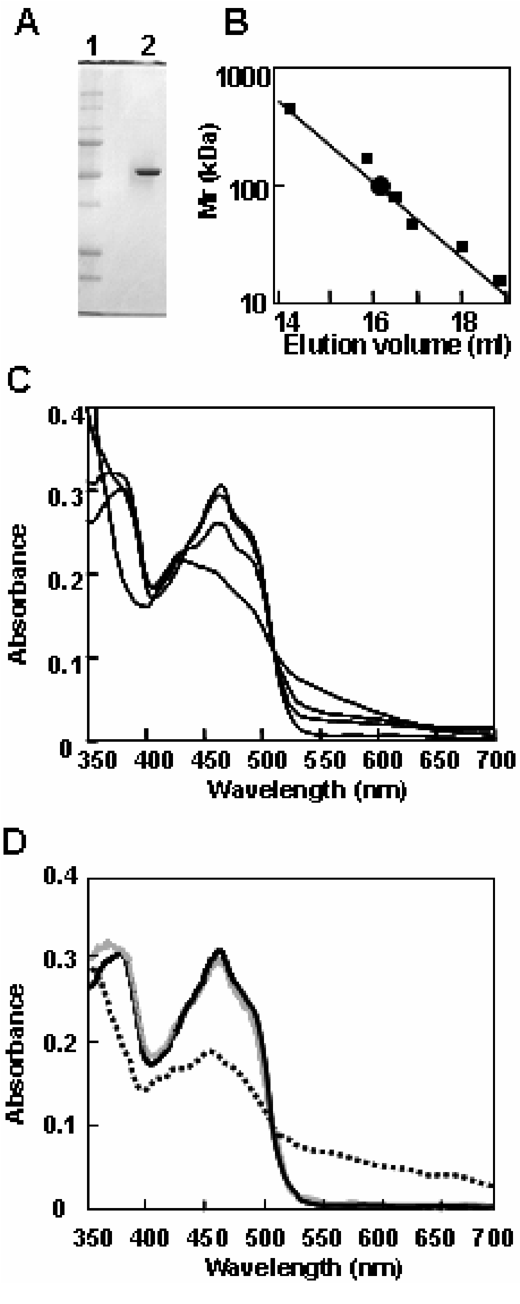
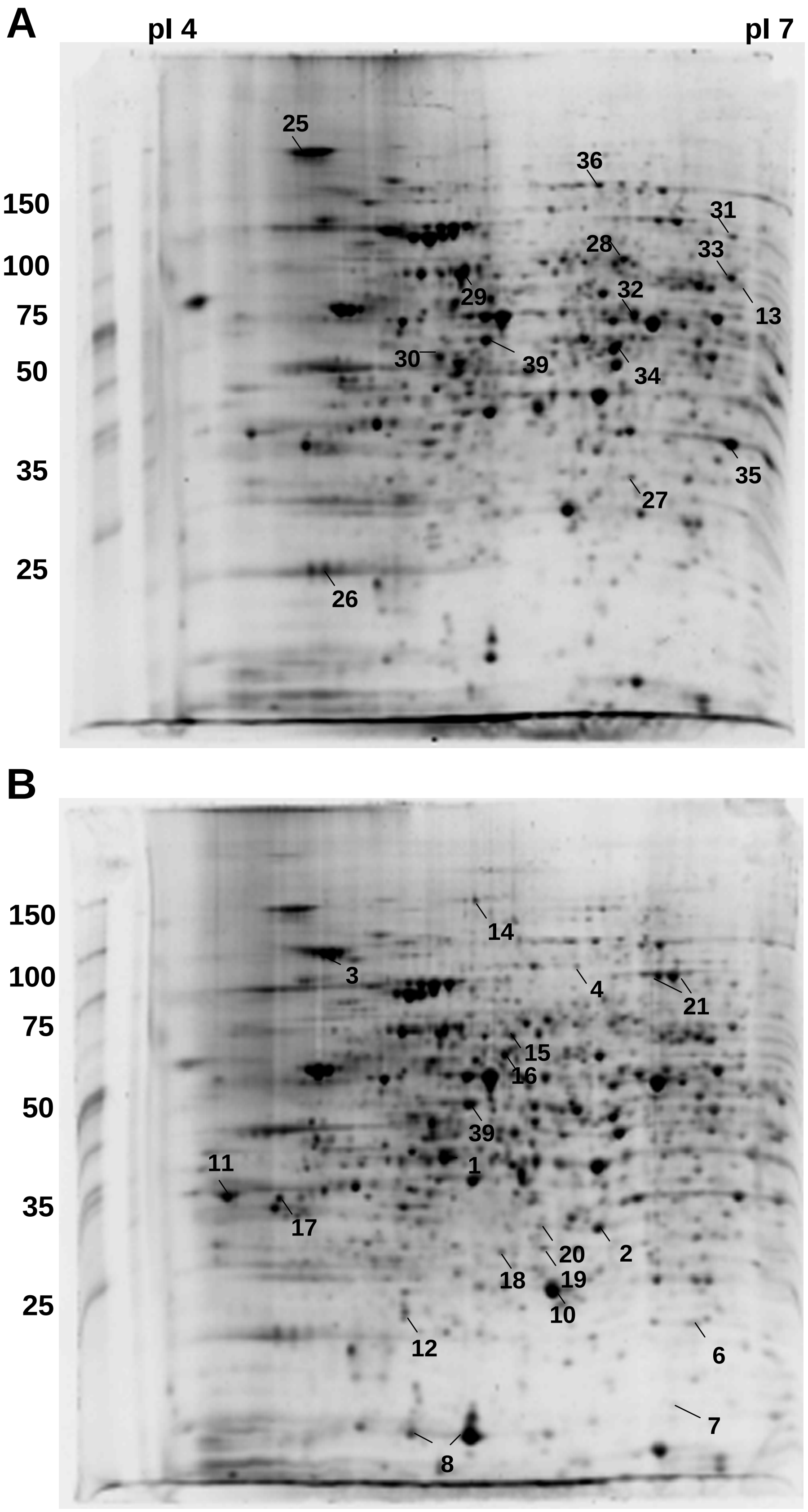
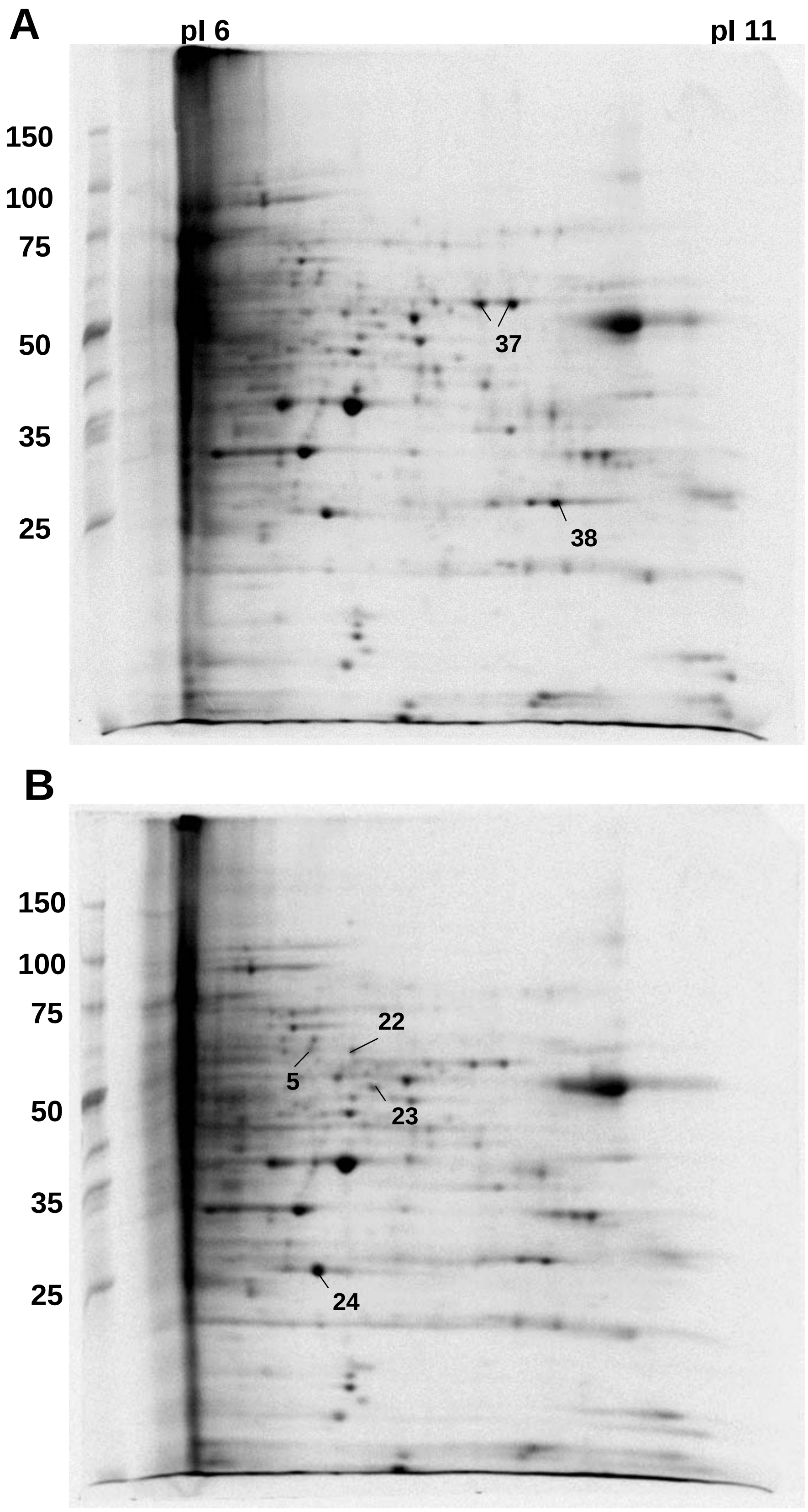
Sato *et al*. Supplemental Fig. 3



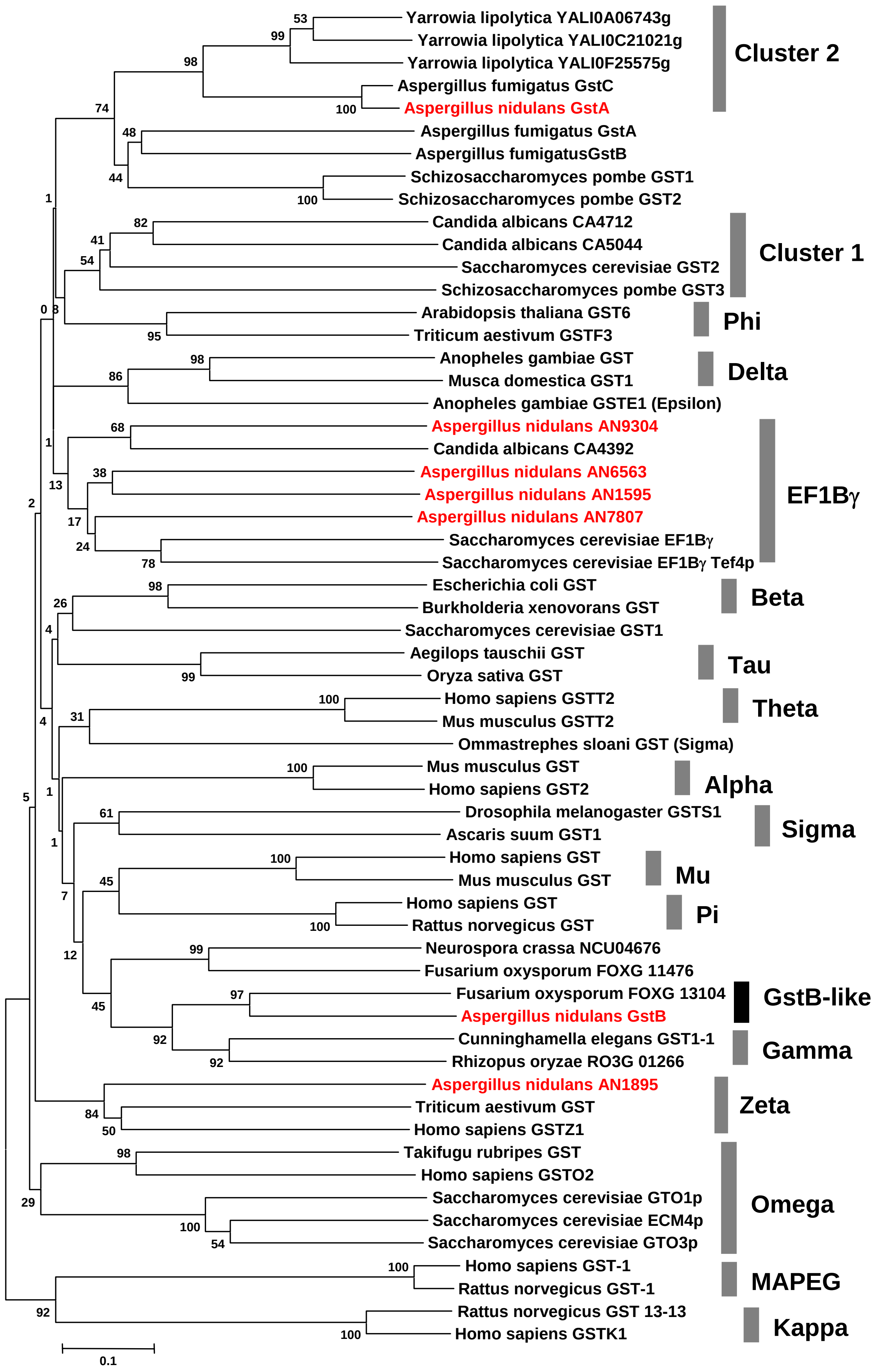
Sato et al. Supplemental Fig. 4



Sato et al. Supplemental Fig. 5



Sato et al. Supplemental Fig. 6



Supplemental Table 1 Distribution of predicted GST proteins among fungi

Predicted GST proteins from *A. nidulans* were compared with fungal genome nucleotide sequences published in Broad Institute (http://www.broad.mit.edu/annotation/fgi/), and more than 30% identical proteins to them are shown. Accession numbers are from Broad Institute. Identities are shown below accession numbers in percents. Blank; corresponding proteins were not found.

Ascomycetes

Aspergillus nidulans	Class	*Aspergillus terreus*	*Botrytis cinerea*	*Candida albicans*	*Candida tropicalis*	*Chaetomium globosum*	*Coccidioides immitis*	*Fusarium graminearum*	*Fusarium oxysporum*	*Fusarium verticilloides*	*Histoplasma capsulatum*	*Lodderomyces elongisporus*
AN3299	alpha	ATEG_07534.1 64	BC1G_12605.1 60			CHG06688.1 45		FGSG_00172 45	FOXG_01307 50	FVEG_00186 49		
AN3849	kappa	ATEG_05408.1 60	BC1G_12830.1 42			CHG10295.1 42	CIMG_08781.2 49				HCAG_06266.1 45	
AN5831	omega	ATEG_06899.1 79	BC1G_03433.1 38	CAWG_01559 45	CTRG_00896.3 46	CHG04901.1 35	CIMG_00933.2 67	FGSG_04433 56	FOXG_13780 56	FVEG_11213 56	HCAG_07421.1 67	LELG_00071.1 *48
AN10273	omega	ATEG_02453.1 90	BC1G_03433.1 55	CAWG_01559 56	CTRG_00896.3 56	CHG04901.1 63	CIMG_00293.2 73	FGSG_09688 66	FOXG_08382 67	FVEG_06294 67	HCAG_01006.1 76	LELG_00071.1 55
AN10379	omega	ATEG_02453.1 46	BC1G_09470.1 52	CAWG_01559 49	CTRG_00896.3 48	CHG04901.1 40	CIMG_00293.2 42	FGSG_09688 43	FOXG_16651 63	FVEG_13590 62	HCAG_01006.1 42	LELG_00071.1 47
AN6024 (*gstB*)	GstB-like	ATEG_06528.1 69	BC1G_05767.1 35					*FGSG_03802 58	FOXG_13104 57	*FVEG_12937 61		
AN9299	cluster 2	ATEG_03359.1 71	BC1G_09645.1 38	CAWG_03499 40	CTRG_00284.3 40	CHG07334.1 49	CIMG_03767.2 46	FGSG_02000 47	FOXG_04328 47	FVEG_07456 47		LELG_04375.1 41
AN4905 (*gstA*)	cluster 2	ATEG_04611.1 82	BC1G_09645.1 43	CAWG_03499 45	CTRG_00284.3 49	CHG07334.1 62	CIMG_03767.2 71	FGSG_02000 57	FOXG_04328 57	FVEG_07456 56	HCAG_02355.1 35	LELG_04376.1 47
AN1595	EF1Bγ	ATEG_06992.1 49	BC1G_00939.1 42	CAWG_02910 31		CHG07289.1 44	CIMG_02699.2 44	FGSG_07401 35		FVEG_07856 36	HCAG_08236.1 37	LELG_02811.1 32
AN6563	EF1Bγ	ATEG_06992.1 81	BC1G_00939.1 54	CAWG_02910 45	CTRG_02705.3 46	CHG07289.1 50	CIMG_02699.2 57	FGSG_07401 49	FOXG_01492 50	FVEG_07856 52	HCAG_08236.1 57	LELG_02811.1 46
AN7807	EF1Bγ	ATEG_06992.1 39	BC1G_00939.1 44	CAWG_02910 34	CTRG_02705.3 33	CHG07289.1 42	CIMG_02699.2 40	FGSG_07401 40	FOXG_01492 39	FVEG_07856 42	HCAG_08236.1 42	LELG_02811.1 35
AN9304	EF1Bγ	ATEG_05085.1 77	BC1G_10460.1 44			CHG02356.1 42		FGSG_13223 44		FVEG_11084 45		
AN10038	EF1Bγ	ATEG_06992.1 30	BC1G_00939.1 38			CHG07289.1 37	CIMG_02699.2 38	FGSG_07401 34		FVEG_07856 35	HCAG_08236.1 36	LELG_02811.1 32
AN3255	Ure2p	ATEG_06384.1 44		CAWG_04225 34	CTRG_01617.3 34	CHG09399.1 55	CIMG_00771.2 51	FGSG_01816 40	FOXG_10072 44	FVEG_08420 39	HCAG_02355.1 74	LELG_01948.1 34
AN6612	MAK16	ATEG_07051.1 94	BC1G_08156.1 84	CAWG_04380.1 68	CTRG_01790.3 66	CHGG_07201.1 69	CIMG_10174.3 71	FGSG_05193.3 69	FOXG_07948.2 73	FVEG_04866.3 73	HCAG_05122.1 89	LELG_01895.1 67
AN1895	MAAI	ATEG_05925.1 64					CIMG_01314.2 50	FGSG_02852 49	FOXG_15752 47	FVEG_13215 47	HCAG_02121.1 53	
AN10695	MAPEG	ATEG_03896.1 77	BC1G_03423.1 63			CHG02754.1 35	CIMG_01614.2 68	FGSG_01294 40	FOXG_11417 42	FVEG_10403 48	HCAG_01368.1 36	
AN3298	unclassified	ATEG_05146.1 60	BC1G_04672.1 42				CIMG_03351.2 63	FGSG_07224 33	FOXG_17209 61	FVEG_05362 31	HCAG_02776.1 47	

* Another orthologs were found (FGSG_11062 (53%), FVEG_12433 (56%), RO3G_08558.1 (32%)).

Supplemental Table 1 Distribution of predicted GST proteins among fungi (continued)

								Basidiomycetes			**Chytridiomycetes**	**Zygomycetes**
Aspergillus nidulans	Class	*Magnaporthe grisea*	*Neurospora crassa*	*Sclerotinia sclerotiorum*	*Saccharomyces cerevisiae RM11-1a*	*Schizo-saccharomyces japonicus*	*Uncinocarpus reesii*	*Coprinus cinerea*	*Cryptococcus neoformans Serotype A*	*Ustilago maydis*	*Batrachochytrium dendrobatidis*	*Rhizopus oryzae*
AN3299	alpha			SS1G_01922.1 41				CC1G_10591.1 35				
AN3849	kappa		NCU01636 49	SS1G_09579.1 42			UREG_02764.1 47			UM05678.1 31		
AN5831	omega	MGG_01410.5 47	NCU04368 46	SS1G_08258.1 *50	SCRG_02146 38		UREG_00366.1 37	CC1G_11081.1 42	CNAG_01889.1 46	UM01515.1 47		RO3G_13689.1 *53
AN10273	omega	MGG_01410.5 68	NCU04368 64	SS1G_08258.1 59	SCRG_04091 48		UREG_00366.1 71	CC1G_11081.1 55	CNAG_01893.1 55	UM01515.1 48		RO3G_13689.1 56
AN10379	omega	MGG_02244.5 62	NCU04368 45	SS1G_05554.1 58	SCRG_04091 39			CC1G_11081.1 46	CNAG_01893.1 46	UM01515.1 39		RO3G_13689.1 44
AN6024 (*gstB*)	GstB-like											*RO3G_01266.1 33
AN9299	cluster 2	MGG_06747.5 44	NCU04109 43	SS1G_07195.1 47			UREG_07517.1 47	CC1G_00666.1 40	CNAG_04110.1 39		BDEG_08491 43	
AN4905 (*gstA*)	cluster 2	MGG_06747.5 57	NCU05780 39	SS1G_07195.1 65	SCRG_03353	SJAG_00238 38	UREG_07517.1 70	CC1G_00666.1 44	CNAG_04110.1 44	UM06325.1 39	BDEG_08491 44	RO3G_09910.1
AN1595	EF1Bγ	MGG_06936.5 41	NCU03826 42	SS1G_00220.1 62			UREG_03068.1 39					
AN6563	EF1Bγ	MGG_06936.5 50	NCU03826 52	SS1G_00220.1 54	SCRG_02444 43	SJAG_02017 37	UREG_03068.1 52	CC1G_01604.1 39	CNAG_00417.1 37	UM02442.1 37	BDEG_06207 36	RO3G_13129.1 37
AN7807	EF1Bγ	MGG_06936.5 40	NCU03826 42	SS1G_00220.1 44	SCRG_02444 31		UREG_03068.1 41				BDEG_06207 32	RO3G_09910.1 31
AN9304	EF1Bγ	MGG_12569.5 38	NCU10521 39					CC1G_01604.1 36			BDEG_06207 35	RO3G_13129.1 39
AN10038	EF1Bγ	MGG_06936.5 32	NCU03826 34	SS1G_00220.1 38			UREG_03068.1 32					
AN3255	Ure2p	MGG_09138.5 44	NCU05780 44	SS1G_10108.1 52	SCRG_03353 36	SJAG_00179 41	UREG_00745.1 51					
AN6612	MAK16	MGG_04069 59	NCU04150 62	SS1G_08782 70	SCRG_05659.1 66	SJAG_00689.2 64	UREG_03547.1 86			UM05387.1 56	BDEG_01326.1 54	RO3G_16892.1 65
AN1895	MAAI						UREG_01240.1 51				BDEG_01302 38	RO3G_11518.1 33
AN10695	MAPEG	MGG_05367.5 43	NCU00549 39	SS1G_06623.1 65			UREG_03256.1 39	CC1G_06890.1 44	CNAG_06238.1 37	UM01870.1 48		
AN3298	unclassified	MGG_06383.5 35	NCU06494 35	SS1G_12640.1 43			UREG_07177.1 60				BDEG_08674 35	