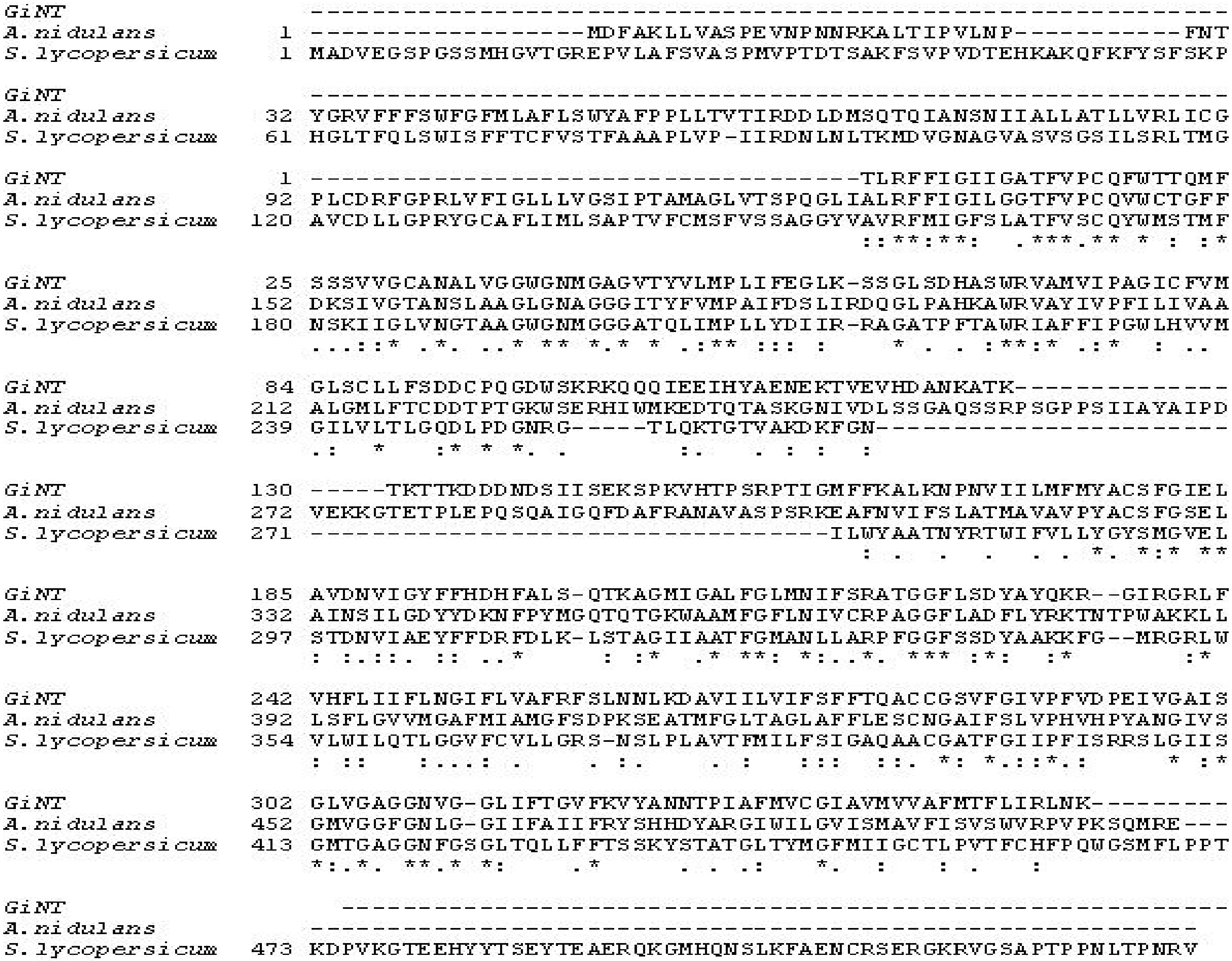
List of supplementary data

Figure S1 Amino acid alignment for *GiNT*
Figure S2 Amino acid alignment and phylogenetic analysis for *GiGS1* and *GiGS2*
Figure S3 Amino acid alignment for *GiGluS*
Figure S4 Amino acid alignment for *GiCPS*
Figure S5 Amino acid alignment for *GiASS*
Figure S6 Amino acid alignment for *GiAL*
Figure S7 Amino acid alignment and phylogenetic analysis for *GiCAR1*
Figure S8 Amino acid alignment for *GiOAT1*
Figure S9 Amino acid alignment for *GiODC*
Figure S10 Amino acid alignment for *GiURE*
Figure S11 Western blot of the expressed proteins in yeast
Table S1 Primers for gene cloning
Table S2 Yeast strains for functional complementation
Table S3 Primers for real time PCR

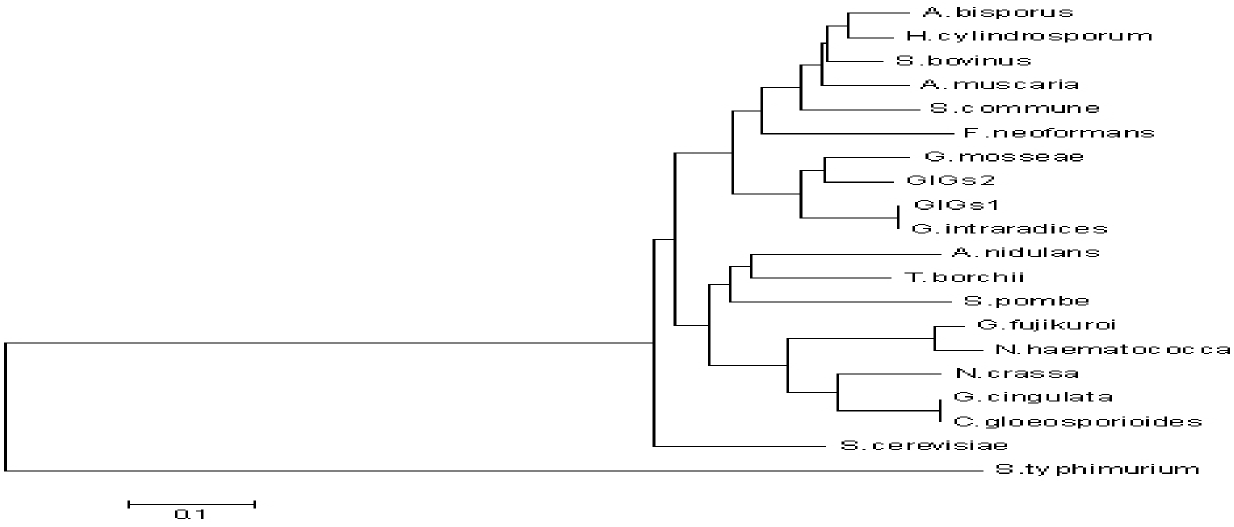


```
GiNT                   ------------------------------------------------------------
A.nidulans           1 ------------------MDFAKLLVASPEVNPNNRKALTIPVLNP-----------FNT
S.lycopersicum       1 MADVEGSPGSSMHGVTGREPVLAFSVASPMVPTDTSAKFSVPVDTEHKAKQFKFYSFSKP

GiNT                   ------------------------------------------------------------
A.nidulans          32 YGRVFFFSWFGFMLAFLSWYAFPPLLTVTIRDDLDMSQTQIANSNIIALLATLLVRLICG
S.lycopersicum      61 HGLTFQLSWISFFTCFVSTFAAAPLVP-IIRDNLNLTKMDVGNAGVASVSGSILSRLTMG

GiNT                 1 ------------------------------------TLRFFIGIIGATFVPCQFWTTQMF
A.nidulans          92 PLCDRFGPRLVFIGLLLVGSIPTAMAGLVTSPQGLIALRFFIGILGGTFVPCQVWCTGFF
S.lycopersicum     120 AVCDLLGPRYGCAFLIMLSAPTVFCMSFVSSAGGYVAVRFMIGFSLATFVSCQYWMSTMF
                                                           ::**:**:  .***.** * : :*

GiNT                25 SSSVVGCANALVGGWGNMGAGVTYVLMPLIFEGLK-SSGLSDHASWRVAMVIPAGICFVM
A.nidulans         152 DKSIVGTANSLAAGLGNAGGGITYFVMPAIFDSLIRDQGLPAHKAWRVAYIVPFILIVAA
S.lycopersicum     180 NSKIIGLVNGTAAGWGNMGGGATQLIMPLLYDIIR-RAGATPFTAWRIAFFIPGWLHVVM
                       ...::* .*. ..* ** *.* * .:** ::: :    * . . :**:* .:*  : ..

GiNT                84 GLSCLLFSDDCPQGDWSKRKQQIEEIHYAENEKTVEVHDANKATK---------------
A.nidulans         212 ALGMLFTCDDTPTGKWSERHIWMKEDTQTASKGNIVDLSSGAQSSRPSGPPSIIAYAIPD
S.lycopersicum     239 GILVLTLGQDLPDGNRG-----TLQKTGTVAKDKFGN-----------------------
                       .:  *   :* * *. .       :.   . : :  :

GiNT               130 -----TKTTKDDDNDSIISEKSPKVHTPSRPTIGMFFKALKNPNVIILMFMYACSFGIEL
A.nidulans         272 VEKKGTETPLEPQSQAIGQFDAFRANAVASPSRKEAFNVIFSLATMAVAVPYACSFGSEL
S.lycopersicum     271 ----------------------------------ILWYAATNYRTWIFVLLYGYSMGVEL
                                                           : .  .  .  . . *. *:* **

GiNT               185 AVDNVIGYFFHDHFALS-QTKAGMIGALFGLMNIFSRATGGFLSDYAYQKR--GIRGRLF
A.nidulans         332 AINSILGDYYDKNFPYMGQTQTGKWAAMFGFLNIVCRPAGGFLADFLYRKTNTPWAKKLL
S.lycopersicum     297 STDNVIAEYFFDRFDLK-LSTAGIIAATFGMANLLARPFGGFSSDYAAKKFG--MRGRLW
                       : :.::. :: ..*     : :*  .* **: *:..*. *** :*:  :*       :*

GiNT               242 VHFLIIFLNGIFLVAFRFSLNNLKDAVIILVIFSFFTQACCGSVFGIVPFVDPEIVGAIS
A.nidulans         392 LSFLGVVMGAFMIAMGFSDPKSEATMFGLTAGLAFFLESCNGAIFSLVPHVHPYANGIVS
S.lycopersicum     354 VLWILQTLGGVFCVLLGRS-NSLPLAVTFMILFSIGAQAACGATFGIIPFISRRSLGIIS
                       : ::   :...: .    . :.    . :   :::  ::. *: *.::*.:     * :*

GiNT               302 GLVGAGGNVG-GLIFTGVFKVYANNTPIAFMVCGIAVMVVAFMTFLIRLNK---------
A.nidulans         452 GMVGGFGNLG-GIIFAIIFRYSHHDYARGIWILGVISMAVFISVSWVRPVPKSQMRE---
S.lycopersicum     413 GMTGAGGNFGSGLTQLLFFTSSKYSTATGLTYMGFMIIGCTLPVTFCHFPQWGSMFLPPT
                       *:.*. **.* *:    .*     . . .:   *.  :   : .   :

GiNT                      ---------------------------------------------------------
A.nidulans             ----------------------------------------------------------
S.lycopersicum     473 KDPVKGTEEHYYTSEYTEAERQKGMHQNSLKFAENCRSERGKRVGSAPTPPNLTPNRV
```

Figure S1. Amino acid alignment of *GiNT* with nitrate transporters from *Aspergillus nidulans* (XP_658612.1) and *Solanum lycopersicum* (AAF00053.1).

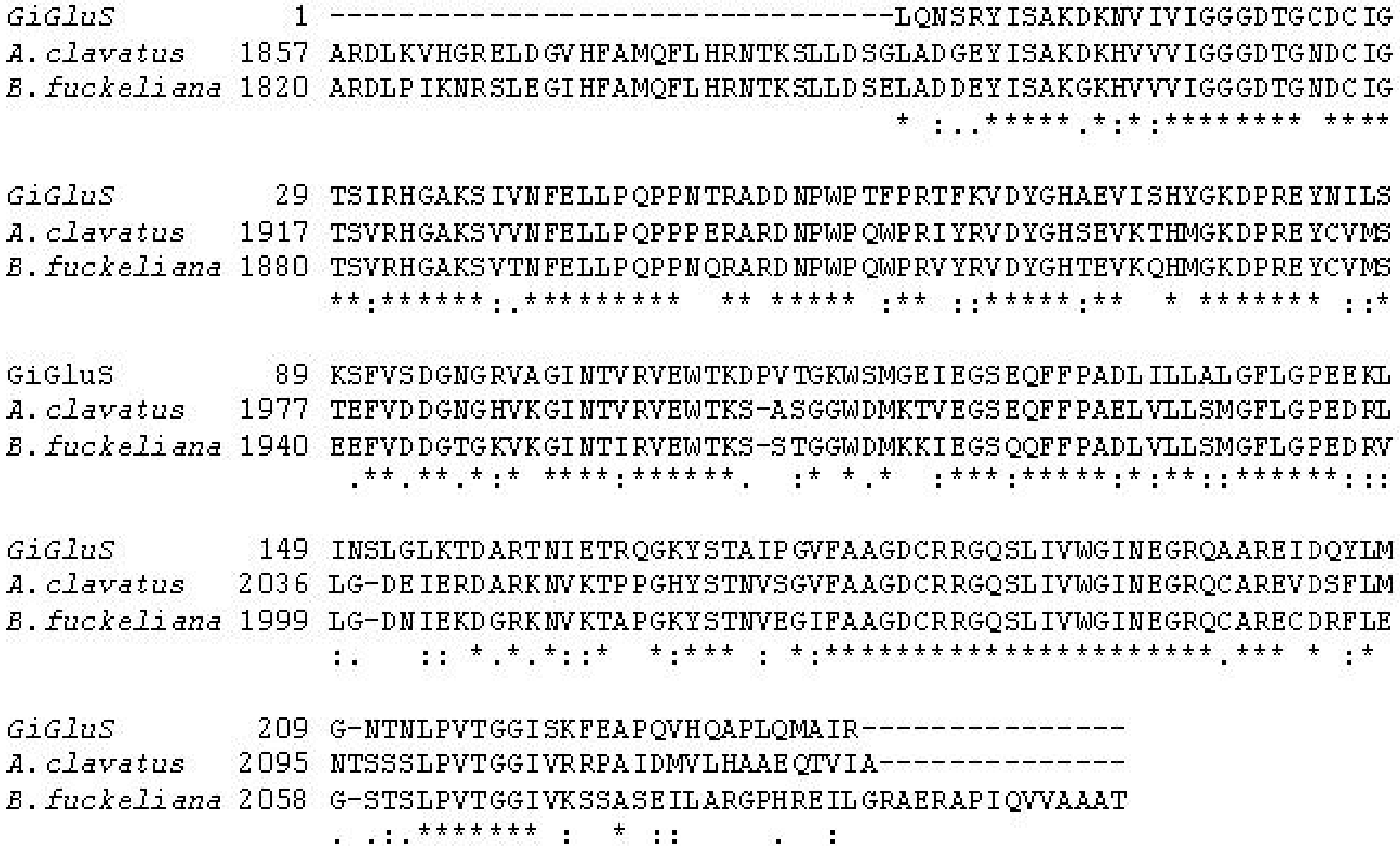
```
F.neoformans        1 ---MSQLIATKRVDLLAPYLALDQGSRVQAEYIWIDAEGGIRSKTMTLDKAPSSVADLKE
S.pombe             1 MSQYDVEPLLSKAAILNKYADLPQNGKVMAEYIWIDGFNHLRSKTMTLDAKPSSIDQLRV
G.mosseae           1 ----MAVANKSNTQTLAKYLSLDQGGKIQAEYIWIDGDGGLRGKTTTLDFKPTDVSELKE
GiGS1               1 ----MYNNNKSNTETLAKYLSLDQGGKVQAEYIWIDGDSGLRSKTRTLDFKPADVSEISE
GiGS2               1 ----MAFINKSNTETLAKYLSLDQCGKIQAEYIWIDGDGGLRGKTTTLDFKPTDVSELKE
G.intraradices      1 ----MYNNNKSNTETLAKYLSLDQGGKVQAEYIWIDGDSGLRSKTRTLDFKPADVSEISE
                                ...  *  *  * * .:: *******. . :*.** ***  *:.: ::
                           GS signature 1
F.neoformans       58 WNFDGSSTNQAPADNSDVFLRPVAIFKDPFRGGANILVLCECYDNDGTPNKSNYRAHCKK
S.pombe            61 WNFDGSSTGQAPGNNSDTLLKPVAMYNDPFRRGDNILVLAACYTADGSPNGFNHRDACAK
G.mosseae          57 WNFDGSSTNQAPGDNSDILLRPCAIFKDPFRGGDNILVLAECYNNDGTPNRTNYRHSCTK
GiGS1              57 WNFDGSSTNQAPGDDSDVLLRPVAIFKDPFRGGDNILVLNECFNNDGTPNRTNHRHSCAK
GiGS2              57 WNFDGSSTNQAPGHDSDILLRPASIFKDPFRGGDNIIVLAECYNNDGTPNRTNYRHSCAK
G.intraradices     57 WNFDGSSTNQAPGDDSDVLLRPVAIFKDPFRGGDNILVLNECFNNDGTPNRTNHRHSCAK
                      ********.***..:** :*:* ::::**** * **:**  *:  **:**  *:*  * *

F.neoformans      117 VMDAAKDTEPWFGLEQEYTLFDADGQVFGWPKNGFPGPQGPYYCGVGAGKVFARDFIEAH
S.pombe           120 LLEKHADKETWFGIEQEYTMLDYYDRPFGWPKGGFPGPQGPFYCGVGTGRVFARDIVEAH
G.mosseae         116 LMSAHSEAKPWFGIEQEYTLFDMDGQVLGWPKGGFPGPQGPYYCSVGANVAFGRDVVEAH
GiGS1             116 VMKTYTDQHPWFGIEQEYTLFESDGSVLGWPKGGFPGPQGPYYCSVGANVAFGRDVVEAH
GiGS2             116 IMETHADAVPWFGIEQEYTLLDADGQVLGWPKGGFPGPQGPYYCSVGANVAFGRDIVEAH
G.intraradices    116 VMKTYTDQHPWFGIEQEYTLFESDGSVLGWPKGGFPGPQGPYYCSVGANVAFGRDVVEAH
                      ::.   :  .***:*****:::  .  :****.********:**.**:. .*.**.:***

F.neoformans      177 YRACLYAGIKISGINAEVMPSQWEFQVGPCTGIEMGDHLWMPRFLLLRIGEEWGITPSLH
S.pombe           180 YKACLYAGINISGINAEVMPSQWEYQVGPCAGIEMGDQLWMSRFLLHRIAEDFGVKISFH
G.mosseae         176 YRACLYSGVNISGVNGEVMPGQWEYQVGPCEGIDMGDHLWVSRYLLQRVAEDFGIVVSFH
GiGS1             176 YRACLYAGVNISGINAEVMPGQWEFQVGPCEGIDMGDHLWIARFLLQRVAEDFGIIVSFH
GiGS2             176 YRACLYAGVNISGVNAEVMPGQWEFQVGPCTGIDMGDHLWVARYLLQRVAEDFGVVVTFH
G.intraradices    176 YRACLYAGVNISGINAEVMPGQWEFQVGPCEGIDMGDHLWIARFLLHRVAEDFGIIVSFH
                      *:****:*::***:*.****.***:***** **:***:**:.*:** *:.*::*:  ::*
                      GSputative ATP-binding region signature
F.neoformans      237 PKPLKGDWNGAGCHSNYSTKDMRTPGKGMAAIEDAIKKLEKKHLEHIAVYGEDNDLRLTG
S.pombe           240 PKPILGDWNGAGCHTNVSTKDTRAEG-GIKAIESYLEKFAKRHKEHIAVYGDDNDLRLTG
G.mosseae         236 PKPIKGDWNGAGCHTNYSTEAMRIEG-GIKAIHDAIDRMSKRHAEHIAVYGEDNDKRLTG
GiGS1             236 PKPIKGDWNGAGCHTNYSTQAMREEG-GIKAIHAAIDKMAARHADHITVYGEDNDQRLTG
GiGS2             236 PKPIKGDWNGAGCHTNYSTKAMREEG-GIKAINEAIERIGKRHAEHIAVYGEDNDQRLTG
G.intraradices    236 PKPIKGDWNGAGCHTNYSTQAMREEG-GIKAIHAAIDKMAARHADHITVYGEDNDQRLTG
                      ***: *********:* **:  *  * *: **.  :.::  :* :**:***:*** ****

F.neoformans      297 KHETASMTTFSAGVANRGASIRIPRHVGAQGYGYLEDRRPASNVDPYRVTAILVETTVLNN
S.pombe           300 RHETGSIDKFTYGVADRGASVRIPRSVAMNGCGYFEDRRPASSIDPYLVTGIITETMFEH-
G.mosseae         296 RHETGHISEFSFGVANRGASIRIPRHVAAQGYGYFEDRRPASNIDPYRVTEVIVESTLG--
GiGS1             296 RHETGHISQFSYGVANRGASIRIPRHVAAEGKGYLEDRRPASNIDPYRVTQIIVESSLA--
GiGS2             296 RHETGHISTFSSGVANRGASIRIPRHVAAEGRGYFEDRRPASNIDPYRVTEIIVESSLG--
G.intraradices    296 RHETGHISQFSYGVANRGASIRIPRHVAAEGKGYLEDRRPASNIDPYRVTQIIVESSL---
                      :***. :  *: ***:****:**** *. :* **:*******.:*** ** ::.*: .
```

A



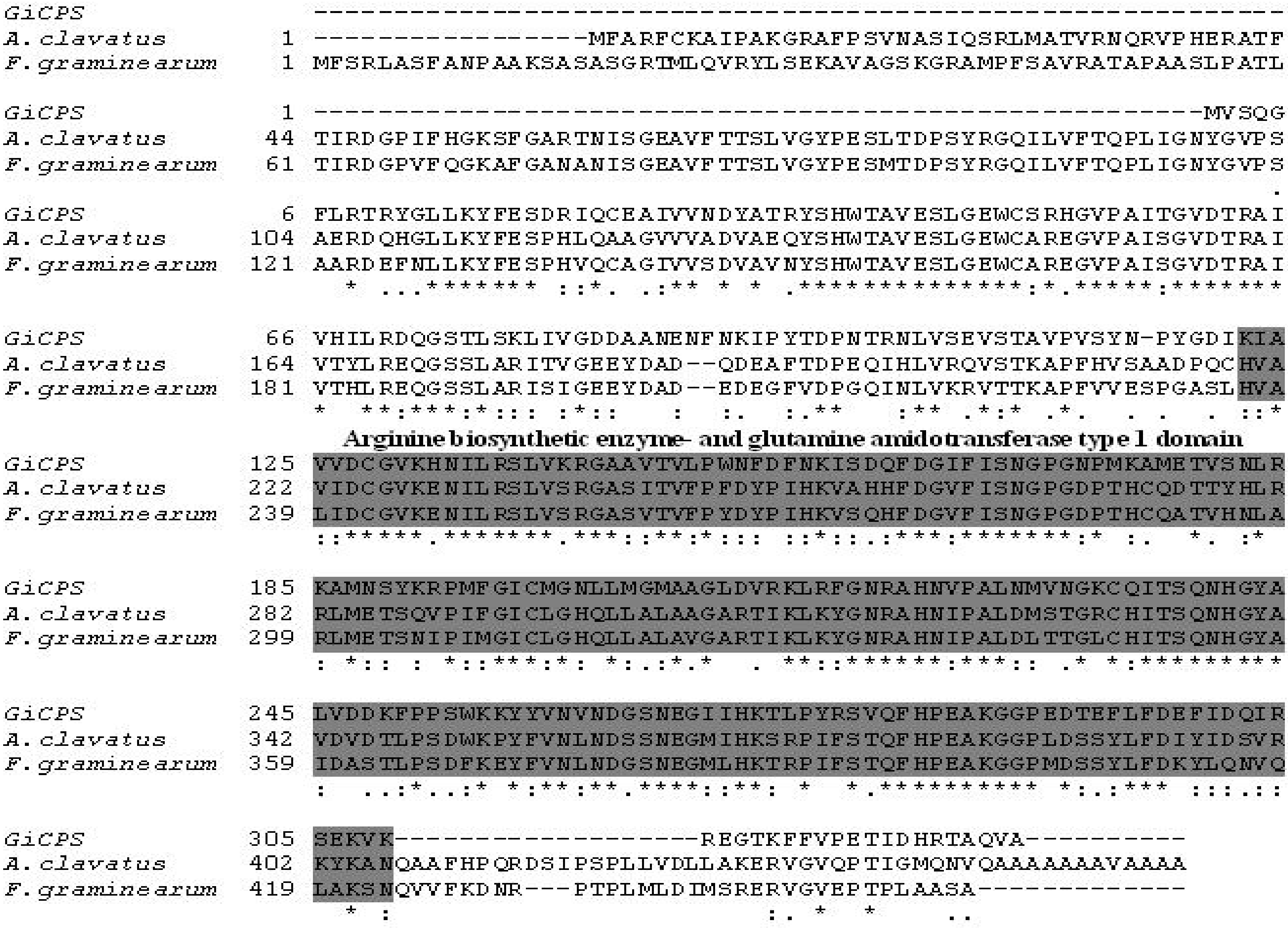
B

Figure S2. Amino acid alignment and phylogenetic analysis for *GiGS1* and *GiGS2*. **A,** The alignment of *GiGS1* and *GiGS2* with *GS* reported from *Glomus intraradices*, *Filobasidiella neoformans, Glomus mosseae*, and *Schizosaccharomyces pombe*. Amino acids shaded in grey represent conserved signature regions. **B,** Phylogenetic relationship among fungal *GS* sequences. The sequence of *Salmonella typhimurium* was used as an outgroup; branches are drawn to scale (the scale bar correspond to 0.1 changes per site). The accession numbers of the sequences used in the alignment are: AF462037 (*Tuber borchii*); O00088 (*Agaricus bisporus*); CAD22045 (*Amanita muscaria*); CAD10037 (*Filobasidiella neoformans*); CAC27836 (*Gibberella fujikuroi*); Q12613 (*Glomerella cingulata*); AAD52617 (*Nectria haematococca*); NCU06724.1 (*Neurospora crassa*); NP_593400 (*Schizosaccharomyces pombe*); AAF27660 (*Schizophyllum commune*); CAD48934 (*Suillus bovinus*); AY360451 (*Glomus mosseae*); DQ063587 (*Glomus intraradices*); M65157 (*Schizosaccharomyces cerevisiae*); AF333968 (*Aspergillus nidulans*); AF411820 (*Hebeloma cylindrosporum*); L78067 (*Colletotrichum gloeosporioides*); 1301273A (*Salmonella typhimurium*).



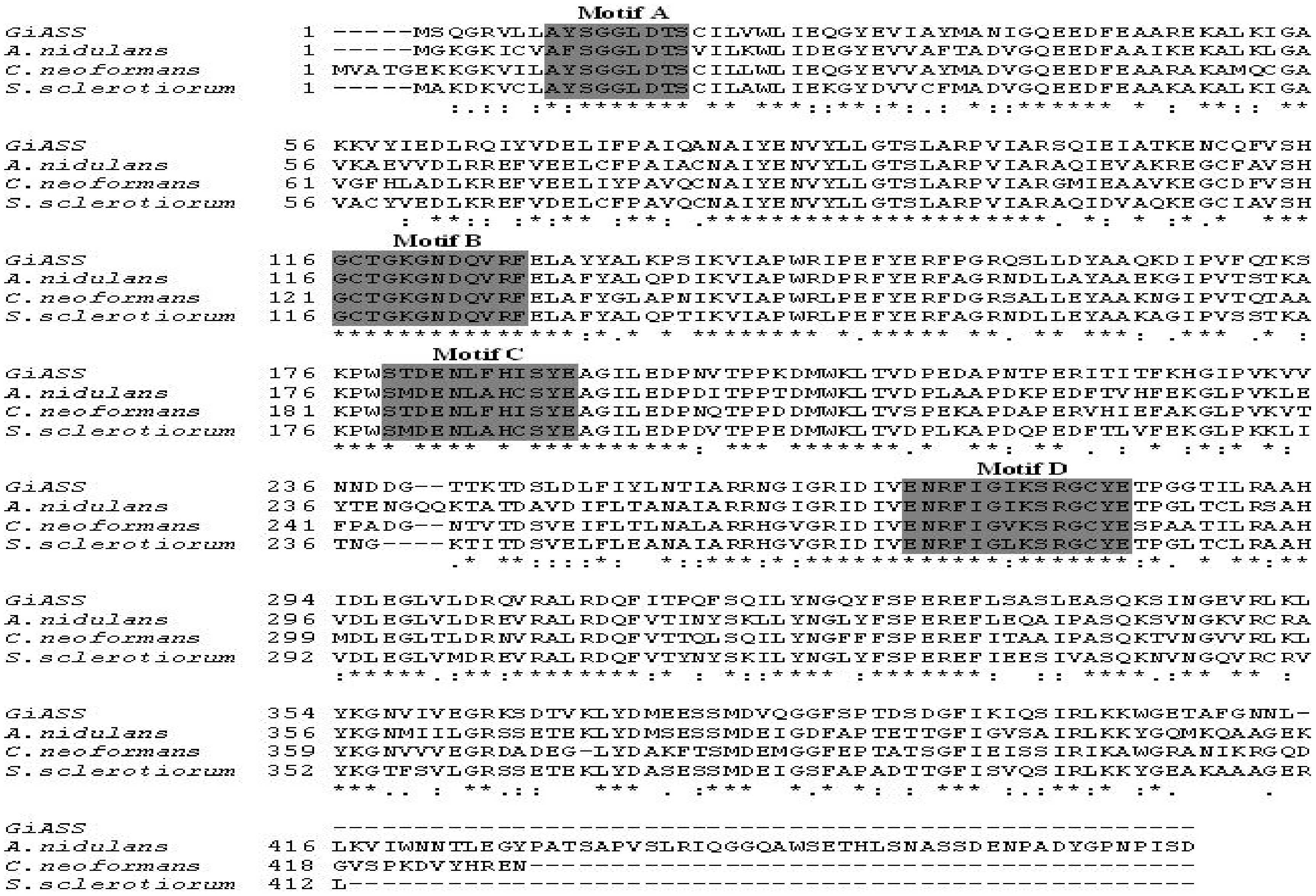
```
GiGluS          1 --------------------------------LQNSRYISAKDKNVIVIGGGDTGCDCIG
A.clavatus   1857 ARDLKVHGRELDGVHFAMQFLHRNTKSLLDSGLADGEYISAKDKHVVVIGGGDTGNDCIG
B.fuckeliana 1820 ARDLPIKNRSLEGIHFAMQFLHRNTKSLLDSELADDEYISAKGKHVVVIGGGDTGNDCIG
                                                  * :..*****.*:*:******** ****

GiGluS         29 TSIRHGAKSIVNFELLPQPPNTRADDNPWPTFPRTFKVDYGHAEVISHYGKDPREYNILS
A.clavatus   1917 TSVRHGAKSVVNFELLPQPPPERARDNPWPQWPRIYRVDYGHSEVKTHMGKDPREYCVMS
B.fuckeliana 1880 TSVRHGAKSVTNFELLPQPPNQRARDNPWPQWPRVYRVDYGHTEVKQHMGKDPREYCVMS
                  **:******:.*********  ** ***** :** ::*****:**  * ******* ::*

GiGluS         89 KSFVSDGNGRVAGINTVRVEWTKDPVTGKWSMGEIEGSEQFFPADLILLALGFLGPEEKL
A.clavatus   1977 TEFVDDGNGHVKGINTVRVEWTKS-ASGGWDMKTVEGSEQFFPAELVLLSMGFLGPEDRL
B.fuckeliana 1940 EEFVDDGTGKVKGINTIRVEWTKS-STGGWDMKKIEGSQQFFPADLVLLSMGFLGPEDRV
                   .**.**.*:* ****:******.  :* *.*  :***:*****:*:**::******:::

GiGluS        149 INSLGLKTDARTNIETRQGKYSTAIPGVFAAGDCRRGQSLIVWGINEGRQAAREIDQYLM
A.clavatus   2036 LG-DEIERDARKNVKTPPGHYSTNVSGVFAAGDCRRGQSLIVWGINEGRQCAREVDSFLM
B.fuckeliana 1999 LG-DNIEKDGRKNVKTAPGKYSTNVEGIFAAGDCRRGQSLIVWGINEGRQCARECDRFLE
                  :.   :: *.*.*::*  *:*** : *:**********************.*** * :* 

GiGluS        209 G-NTNLPVTGGISKFEAPQVHQAPLQMAIR---------------
A.clavatus   2095 NTSSSLPVTGGIVRRPAIDMVLHAAEQTVIA--------------
B.fuckeliana 2058 G-STSLPVTGGIVKSSASEILARGPHREILGRAERAPIQVVAAAT
                  . .:.******* :  * ::     .  :
```

Figure S3. Amino acid alignment of *GiGluS* with NADH dependent glutamate synthases from *Aspergillus clavatus* (XP_001269425.1) and *Botryotinia fuckeliana* (XP_001547433.1). Amino acids shaded in grey represent conserved residues.



```
GiCPS                 ------------------------------------------------------------
A. clavatus        1  -----------------MFARFCKAIPAKGRAFPSVNASIQSRLMATVRNQRVPHERATF
F. graminearum     1  MFSRLASFANPAAKSASASGRTMLQVRYLSEKAVAGSKGRAMPFSAVRATAPAASLPATL

GiCPS              1  -------------------------------------------------------MVSQG
A. clavatus       44  TIRDGPIFHGKSFGARTNISGEAVFTTSLVGYPESLTDPSYRGQILVFTQPLIGNYGVPS
F. graminearum    61  TIRDGPVFQGKAFGANANISGEAVFTTSLVGYPESMTDPSYRGQILVFTQPLIGNYGVPS
                                                                                 .
GiCPS              6  FLRTRYGLLKYFESDRIQCEAIVVNDYATRYSHWTAVESLGEWCSRHGVPAITGVDTRAI
A. clavatus      104  AERDQHGLLKYFESPHLQAAGVVVADVAEQYSHWTAVESLGEWCAREGVPAISGVDTRAI
F. graminearum   121  AARDEFNLLKYFESPHVQCAGIVVSDVAVNYSHWTAVESLGEWCAREGVPAISGVDTRAI
                        * ...******* ::*. .:** * * .**************:*.*****:*******

GiCPS             66  VHILRDQGSTLSKLIVGDDAANENFNKIPYTDPNTRNLVSEVSTAVPVSYN-PYGDIKIA
A. clavatus      164  VTYLREQGSSLARITVGEEYDAD--QDEAFTDPEQIHLVRQVSTKAPFHVSAADPQCHVA
F. graminearum   181  VTHLREQGSSLARISIGEEYDAD--EDEGFVDPGQINLVKRVTTKAPFVVESPGASLHVA
                      *  **:***:*::: :*::   :  :.  :.**   :** .*:* .*.  . .  . ::*
                        Arginine biosynthetic enzyme- and glutamine amidotransferase type 1 domain
GiCPS            125  VVDCGVKHNILRSLVKRGAAVTVLPWNFDFNKISDQFDGIFISNGPGNPMKAMETVSNLR
A. clavatus      222  VIDCGVKENILRSLVSRGASITVFPFDYPIHKVAHHFDGVFISNGPGDPTHCQDTTYHLR
F. graminearum   239  LIDCGVKENILRSLVSRGASVTVFPYDYPIHKVSQHFDGVFISNGPGDPTHCQATVHNLA
                      ::*****.*******.***::**:*::: ::*::.:***:*******:* :.  *. :*

GiCPS            185  KAMNSYKRPMFGICMGNLLMGMAAGLDVRKLRFGNRAHNVPALNMVNGKCQITSQNHGYA
A. clavatus      282  RLMETSQVPIFGICLGHQLLALAAGARTIKLKYGNRAHNIPALDMSTGRCHITSQNHGYA
F. graminearum   299  RLMETSNIPIMGICLGHQLLALAVGARTIKLKYGNRAHNIPALDLTTGLCHITSQNHGYA
                      : *:: : *::***:*: *:.:*.*  . **::******:***:: .* *:*********

GiCPS            245  LVDDKFPPSWKKYYVNVNDGSNEGIIHKTLPYRSVQFHPEAKGGPEDTEFLFDEFIDQIR
A. clavatus      342  VDVDTLPSDWKPYFVNLNDSSNEGMIHKSRPIFSTQFHPEAKGGPLDSSYLFDIYIDSVR
F. graminearum   359  IDASTLPSDFKEYFVNLNDGSNEGMLHKTRPIFSTQFHPEAKGGPMDSSYLFDKYLQNVQ
                      :  ..:*..:* *:**:**.****::**: *  *.********** *:.:*** :::.::

GiCPS            305  SEKVK-------------------REGTKFFVPETIDHRTAQVA---------
A. clavatus      402  KYKANQAAFHPQRDSIPSPLLVDLLAKERVGVQPTIGMQNVQAAAAAAVAAAA
F. graminearum   419  LAKSNQVVFKDNR---PTPLMLDIMSRERVGVEPTPLAASA------------
                        * :                       :. *  *    ..
```

Figure S4. Amino acid alignment of *GiCPS* with other available *CPS* sequences from *Aspergillus clavatus* (XP_001274012.1) and *Fusarium graminearum* (XP_389730). Amino acids shaded in grey represent signature sequences.

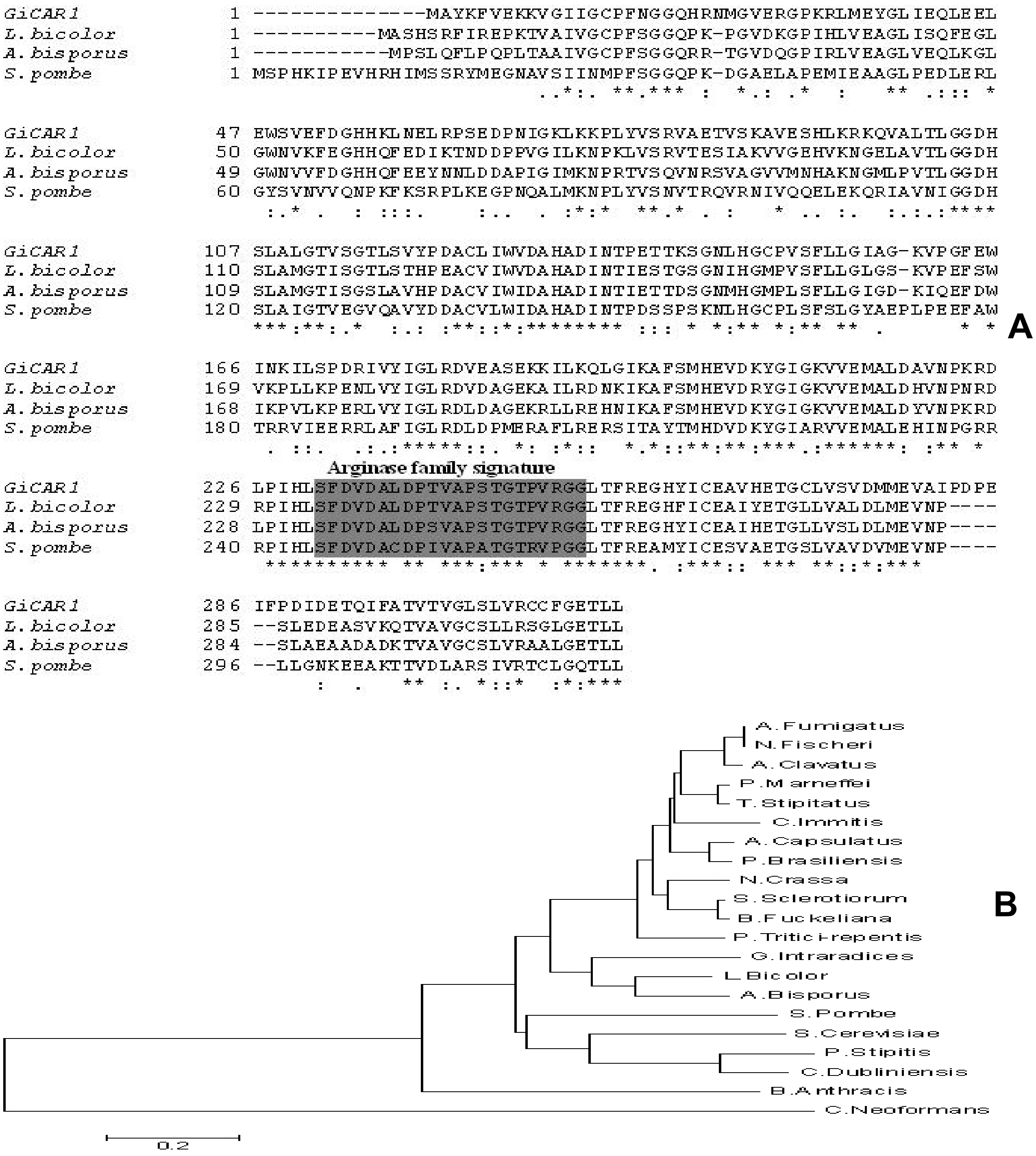


```
                                        Motif A
GiASS                  1 -----MSQGRVLLAYSGGLDTSCILVWLIEQGYEVIAYMANIGQEEDFEAAREKALKIGA
A.nidulans             1 -----MGKGKICVAFSGGLDTSVILKWLIDEGYEVVAFTADVGQEEDFAAIKEKALKLGA
C.neoformans           1 MVATGEKKGKVILAYSGGLDTSCILLWLIEQGYEVVAYMADVGQEEDFEAARAKAMQCGA
S.sclerotiorum         1 -----MAKDKVCLAYSGGLDTSCILAWLIEKGYDVVCFMADVGQEEDFEAAKAKALKIGA
                                :.:: :*:******* ** ***::**:*:.: *::****** * : **:: **

GiASS                 56 KKVYIEDLRQIYVDELIFPAIQANAIYENVYLLGTSLARPVIARSQIEIATKENCQFVSH
A.nidulans            56 VKAEVVDLRREFVEELCFPAIACNAIYENVYLLGTSLARPVIARAQIEVAKREGCFAVSH
C.neoformans          61 VGFHLADLKREFVEELIYPAVQCNAIYENVYLLGTSLARPVIARGMIEAAVKEGCDFVSH
S.sclerotiorum        56 VACYVEDLKREFVDELCFPAVQCNAIYENVYLLGTSLARPVIARAQIDVAQKEGCIAVSH
                             : **:: :*:** :**: .*********************. *: * :*.*  ***
                             Motif B
GiASS                116 GCTGKGNDQVRFELAYYALKPSIKVIAPWRIPEFYERFPGRQSLLDYAAQKDIPVFQTKS
A.nidulans           116 GCTGKGNDQVRFELAFYALQPDIKVIAPWRDPRFYERFAGRNDLLAYAAEKGIPVTSTKA
C.neoformans         121 GCTGKGNDQVRFELAFYGLAPNIKVIAPWRLPEFYERFDGRSALLEYAAKNGIPVTQTAA
S.sclerotiorum       116 GCTGKGNDQVRFELAFYALQPTIKVIAPWRLPEFYERFAGRNDLLEYAAKAGIPVSSTKA
                         ***************:*.* * ******** *.***** **. ** ***: .*** .* :
                               Motif C
GiASS                176 KPWSTDENLFHISYEAGILEDPNVTPPKDMWKLTVDPEDAPNTPERITITFKHGIPVKVV
A.nidulans           176 KPWSMDENLAHCSYEAGILEDPDITPPTDMWKLTVDPLAAPDKPEDFTVHFEKGLPVKLE
C.neoformans         181 KPWSTDENLFHISYEAGILEDPNQTPPDDMWKLTVSPEKAPDAPERVHIEFAKGLPVKVT
S.sclerotiorum       176 KPWSMDENLAHCSYEAGILEDPDVTPPEDMWKLTVDPLKAPDQPEDFTLVFEKGLPKKLI
                         **** **** * **********: *** *******.*  **: ** . : * :*:* *:
                                                                 Motif D
GiASS                236 NNDDG--TTKTDSLDLFIYLNTIARRNGIGRIDIVENRFIGIKSRGCYETPGGTILRAAH
A.nidulans           236 YTENGQQKTATDAVDIFLTANAIARRNGIGRIDIVENRFIGIKSRGCYETPGLTCLRSAH
C.neoformans         241 FPADG--NTVTDSVEIFLTLNALARRHGVGRIDIVENRFIGVKSRGCYESPAATILRAAH
S.sclerotiorum       236 TNG----KTITDSVELFLEANAIARRHGVGRIDIVENRFIGLKSRGCYETPGLTCLRAAH
                                .* **::::*:  *::***:*:************:*******:*. * **:**

GiASS                294 IDLEGLVLDRQVRALRDQFITPQFSQILYNGQYFSPEREFLSASLEASQKSINGEVRLKL
A.nidulans           296 VDLEGLVLDREVRALRDQFVTINYSKLLYNGLYFSPEREFLEQAIPASQKSVNGKVRCRA
C.neoformans         299 MDLEGLTLDRNVRALRDQFVTTQLSQILYNGFFFSPEREFITAAIPASQKTVNGVVRLKL
S.sclerotiorum       292 VDLEGLVMDREVRALRDQFVTYNYSKILYNGLYFSPEREFIEESIVASQKNVNGQVRCRV
                         :*****.:**:********:* : *::**** :*******:  :: ****.:** ** :

GiASS                354 YKGNVIVEGRKSDTVKLYDMEESSMDVQGGFSPTDSDGFIKIQSIRLKKWGETAFGNNL-
A.nidulans           356 YKGNMIILGRSSETEKLYDMSESSMDEIGDFAPTETTGFIGVSAIRLKKYGQMKQAAGEK
C.neoformans         359 YKGNVVVEGRDADEG-LYDAKFTSMDEMGGFEPTATSGFIEISSIRIKAWGRANIKRGQD
S.sclerotiorum       352 YKGTFSVLGRSSETEKLYDASESSMDEIGSFAPADTTGFISVQSIRLKKYGEAKAAAGER
                         ***.. : **.::   *** . :***  *.* *: : ***  :.:**:* :*.     .

GiASS                    -------------------------------------------------------
A.nidulans           416 LKVIWNNTLEGYPATSAPVSLRIQGGQAWSETHLSNASSDENPADYGPNPISD
C.neoformans         418 GVSPKDVYHREN-----------------------------------------
S.sclerotiorum       412 L----------------------------------------------------
```

Figure S5. Amino acid alignment of *GiASS* with argininosuccinate synthases from *Aspergillus nidulans* (CBF85751.1), *Cryptococcus neoformans* (XP_570386.1), and *Sclerotinia sclerotiorum* (XP_001594845.1). Amino acids shaded in grey represent conserved sequences.

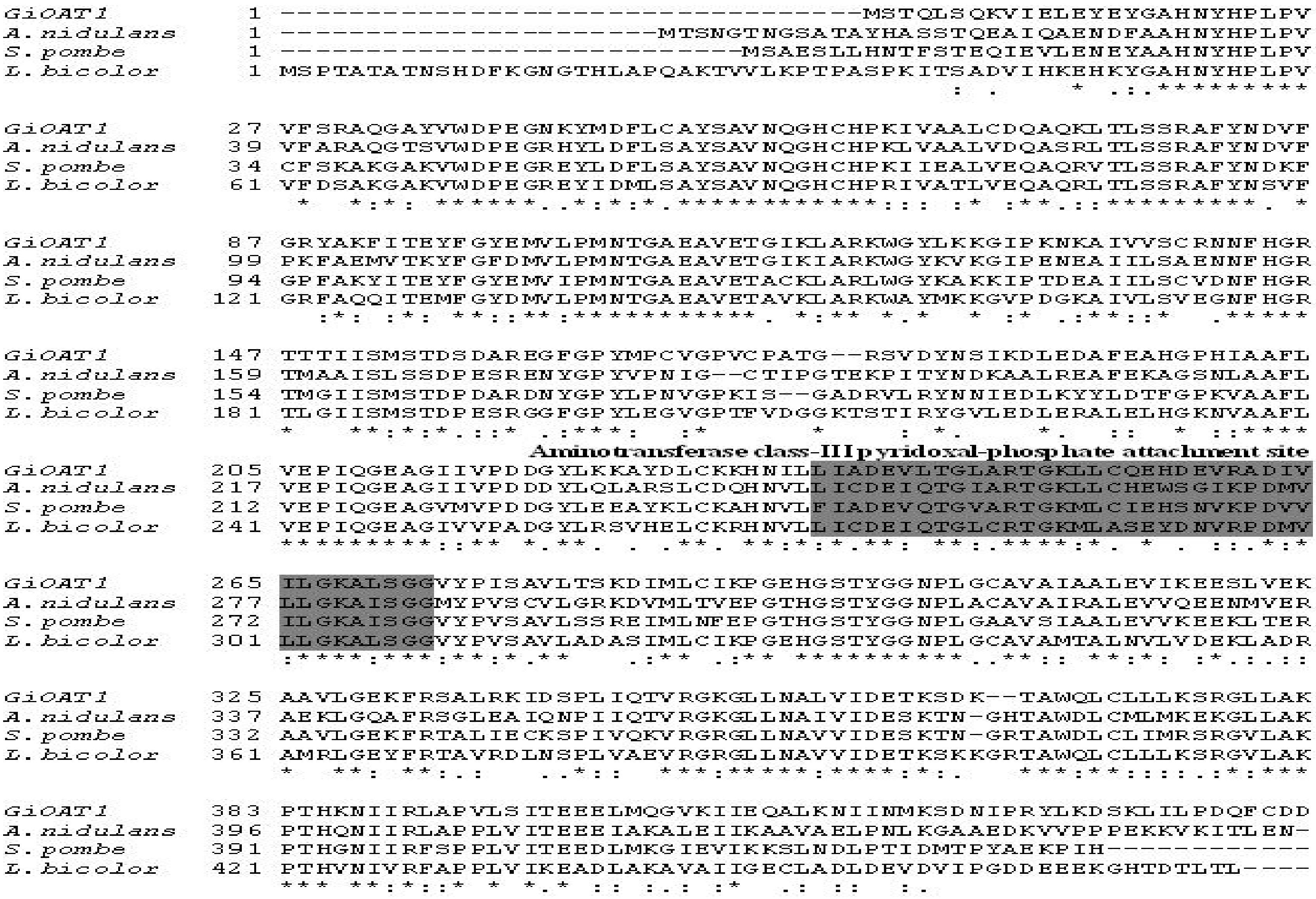
```
GiAL                    ------------------------------------------------------------
S.pombe           1     -----MASSQKLWGGRFTGATDPLMTAYNESIHYDKRMFNADIDGSKAYAKALEQRGIIS
A.clavatus        1     MSASKAVAENMLWGGRFTQGLDPLMVQYNQSLPYDRIFWKQDIAGSIAFARANAKTGILT

GiAL                    ------------------------------------------------------------
S.pombe          56     ADELDKMLDGLEAVRDEWKTNKFTLQPSDEDIHTANERRLGEIIGTGIAGKLHTGRSRND
A.clavatus       61     QQEFTEIERGFKQIADEWSTNTFVVKENDEDIHTANERRLSEIIGKEIGGKLHTGRSRNE

GiAL                    ------------------------------------------------------------
S.pombe         116     QVTTDMRLWLRDELDIIQKSLVGLLQVFVARAESDLDIIMPGYTHLQRAQPIRWSHFLLS
A.clavatus      121     QIATDMRLWLRDELRKLDAVLCDLVKVSIARAESELDFIMPGYTHLQKAQPVRWSHWLLS

GiAL              1     ----MERRWGRLTHIFDRLNVIPLGSGAVAGNPFNIDREFLAKELGFKGVIHNSLLGVSD
S.pombe         176     HAFSILNDLDRLKQIYSRVNRLPLGAGALAGNPFAVDRKFLQKELGFEGVIMNSMNAVAD
A.clavatus      181     HATAFASELQRLREVTKRVNRSPLGTGALAGNPFQIDREAMAQELGFEGLLYNSMNAVAD
                            :     ** .: .*:*  ***:**:***** :**: : :****:*:: **: .*:*
                                                                     C3 region
GiAL             57     RDFVAEILFWSSLTMIHISRFSEDLILYSTSEFGFVTLADAYSTGSSLMPQKKNPDSLEL
S.pombe         236     RDYVIEFMFWASMVMTHISRLAEDLIIYSTSEFNFVTLSDAYSTGSSIMPQKKNPDSLEL
A.clavatus      241     RDFAMETMQWGSSFMLKISRWAEDLIIYSSLEFGFVRLSDAYSTGSSLMPQKKNADSLEL
                        **:. * : *.*  * :*** :****:**: **.** *:********:******.*****

GiAL            117     LRGKSGRVFGQLSGFLMTYKGLPSTYNKDLQEDKEPIFDATDTLSGSLQITTGVLSTLTI
S.pombe         286     LRGKSGRVFGNMMGFMVSVKGIPSTYNKDLQEDKEPLFDSSKTVLDSIQILTGVLSTLTV
A.clavatus      301     LRGKAGRAFGHMAGLMVTIKGLPTTYNKDLQESVEPLLDHIKTVSDSIQIATGVLSTLTV
                        ****:**.**:: *:::: **:*:********. **::*  .*: .*:** ********:

GiAL            177     YPERMKQSLSTDMLATDLAEYLVRKGVPFRETHHIAGAAVRLAEDRNTSMSNLTLEDFKQ
S.pombe         346     NPENIAKSLTADMLATDLAEYLVRKGVPFRETHHISGSAVRMAEERNTTIDKLSVEDFKS
A.clavatus      361     LPEKMTAALAPEMLATEIADYLVRKGVPFREGHHISGRVVALAEKNNVPMDTLSLEQLKT
                         **.:  :*:.:****::*:*********** ***:* .* :**..*..:..*::*::*

GiAL            237     LHQAFEDDVKLIWDYEKSIENRCSLGGTSKQTVKEQIIKLSKWLNE
S.pombe         406     LHPLYEEDVADVFNYESSVEKRCAIGGTAKSCVLEQIQTIKKALE-
A.clavatus      421     VDARFDEDVQACLDYERAVELKDAIGGTSRRAVLEQTAVLKKLL--
                        :.  :::**    :** ::* : ::***::  * **   :.* *
```

Figure S6. Amino acid alignment of *GiAL* with the argininosuccinate lyases (*AL*) from *Aspergillus clavatus* (XP_001270795.1) and from *Schizosaccharomyces pombe* (NP_596817.1). Amino acids shaded in grey represent conserved residues.



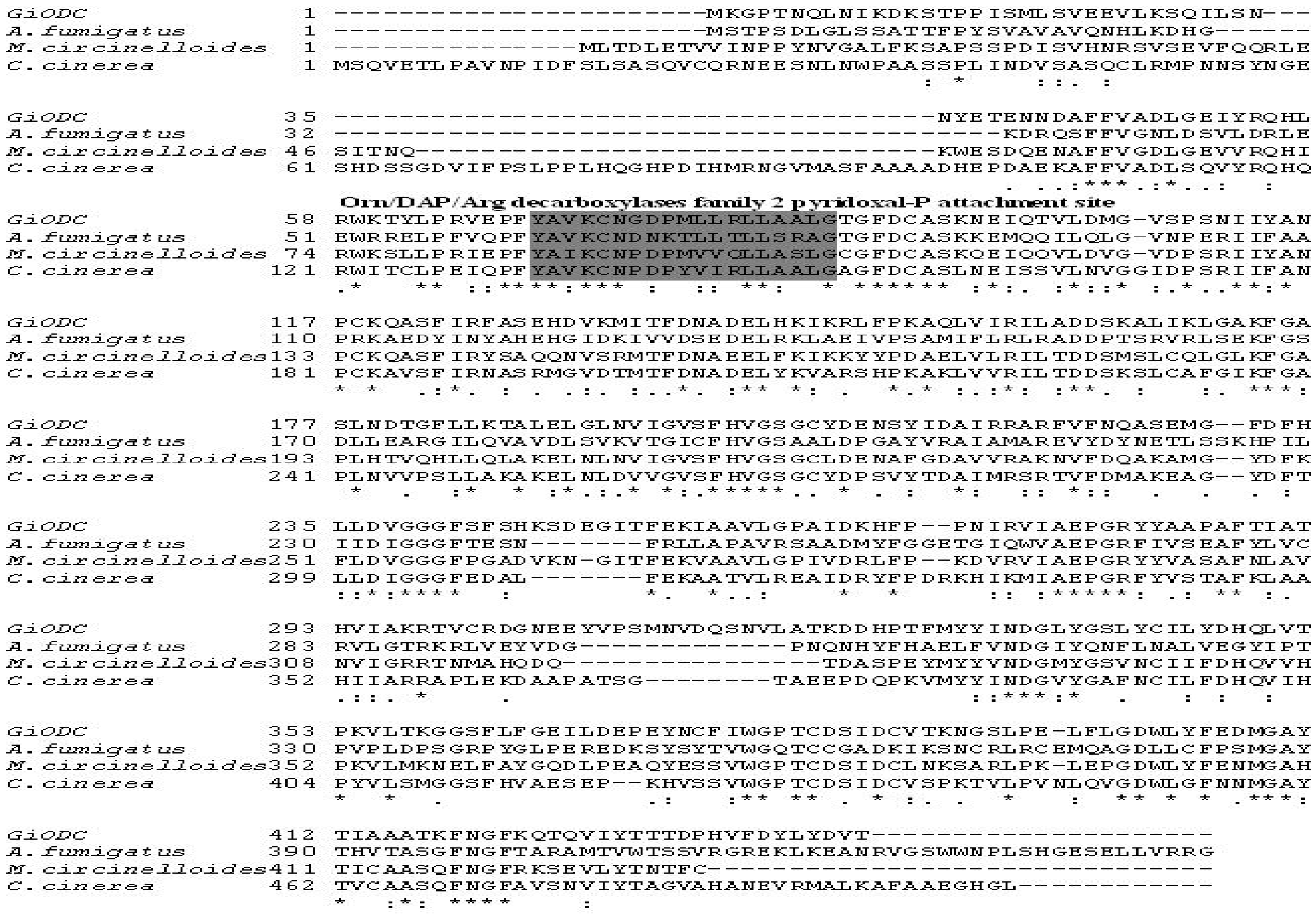
```
GiCAR1         1 --------------MAYKFVEKKVGIIGCPFNGGQHRNMGVERGPKRLMEYGLIEQLEEL
L.bicolor      1 ----------MASHSRFIREPKTVAIVGCPFSGGQPK-PGVDKGPIHLVEAGLISQFEGL
A.bisporus     1 -----------MPSLQFLPQPLTAAIVGCPFSGGQRR-TGVDQGPIRLVEAGLVEQLKGL
S.pombe        1 MSPHKIPEVHRHIMSSRYMEGNAVSIINMPFSGGQPK-DGAELAPEMIEAAGLPEDLERL
                                        ..*:. **.*** :  *.: .*  :   ** .::: *

GiCAR1        47 EWSVEFDGHHKLNELRPSEDPNIGKLKKPLYVSRVAETVSKAVESHLKRKQVALTLGGDH
L.bicolor     50 GWNVKFEGHHQFEDIKTNDDPPVGILKNPKLVSRVTESIAKVVGEHVKNGELAVTLGGDH
A.bisporus    49 GWNVVFDGHHQFEEYNNLDDAPIGIMKNPRTVSQVNRSVAGVVMNHAKNGMLPVTLGGDH
S.pombe       60 GYSVNVVQNPKFKSRPLKEGPNQALMKNPLYVSNVTRQVRNIVQQELEKQRIAVNIGGDH
                  :.* .  : :::.    :..  . :*:*  **.* . :   * .. :.  :.:.:****

GiCAR1       107 SLALGTVSGTLSVYPDACLIWVDAHADINTPETTKSGNLHGCPVSFLLGIAG-KVPGFEW
L.bicolor    110 SLAMGTISGTLSTHPEACVIWVDAHADINTIESTGSGNIHGMPVSFLLGLGS-KVPEFSW
A.bisporus   109 SLAMGTISGSLAVHPDACVIWIDAHADINTIETTDSGNMHGMPLSFLLGIGD-KIQEFDW
S.pombe      120 SLAIGTVEGVQAVYDDACVLWIDAHADINTPDSSPSKNLHGCPLSFSLGYAEPLPEEFAW
                 ***:**:.*  :.: :**::*:******** ::: * *:** *:** ** .      * *

GiCAR1       166 INKILSPDRIVYIGLRDVEASEKKILKQLGIKAFSMHEVDKYGIGKVVEMALDAVNPKRD
L.bicolor    169 VKPLLKPENLVYIGLRDVDAGEKAILRDNKIKAFSMHEVDRYGIGKVVEMALDHVNPNRD
A.bisporus   168 IKPVLKPERLVYIGLRDLDAGEKRLLREHNIKAFSMHEVDKYGIGKVVEMALDYVNPKRD
S.pombe      180 TRRVIEERRLAFIGLRDLDPMERAFLRERSITAYTMHDVDKYGIARVVEMALEHINPGRR
                  .  ::.  .:.:*****::. *: :*::  *.*::**:**:***.:******: :** *
                       Arginase family signature
GiCAR1       226 LPIHLSFDVDALDPTVAPSTGTPVRGGLTFREGHYICEAVHETGCLVSVDMMEVAIPDPE
L.bicolor    229 RPIHLSFDVDALDPTVAPSTGTPVRGGLTFREGHFICEAIYETGLLVALDLMEVNP----
A.bisporus   228 LPIHLSFDVDALDPSVAPSTGTPVRGGLTFREGHYICEAIHETGLLVSLDLMEVNP----
S.pombe      240 RPIHLSFDVDACDPIVAPATGTRVPGGLTFREAMYICESVAETGSLVAVDVMEVNP----
                  ********** ** ***:*** * *******. :***:: *** **::*:***

GiCAR1       286 IFPDIDETQIFATVTVGLSLVRCCFGETLL
L.bicolor    285 --SLEDEASVKQTVAVGCSLLRSGLGETLL
A.bisporus   284 --SLAEAADADKTVAVGCSLVRAALGETLL
S.pombe      296 --LLGNKEEAKTTVDLARSIVRTCLGQTLL
                      :  .   ** :. *::*  :*:***
```

Figure S7. Amino acid alignment and phylogenetic analysis for *GiCAR1***. A,** The alignment of the *GiCAR1* with the reported arginase from *Laccaria bicolor*, *Agaricus bisporus* and *Schizosaccharomyces pombe*. Amino acids shaded in grey represent conserved residues. **B,** Phylogenetic relationship among fungal arginase. The sequence of *Cryptococcus neoformans* was used as an outgroup; branches are drawn to scale (the scale bar corresponds to 0.2 changes per site). The accession numbers of the sequences used in the alignment are: XP_001875445 (*Laccaria bicolor*); CAG26693 (*Agaricus bisporus*); AAW44454 (*Cryptococcus neoformans*); NP_595133 (*Schizosaccharomyces pombe*); XP_001542989 (*Actinobacillus capsulatus*); XP_001941966 (*Pyrenophora tritici-repentis*); XP_001588035 (*Sclerotinia sclerotiorum*); XP_001556008 (*Botryotinia fuckeliana*); XP_002143546 (*Penicillium marneffei*); EED19435 (*Talaromyces stipitatus*); XP_959759 (*Neurospora crassa*); XP_754476 (*Aspergillus fumigatus*); XP_001244242 (*Coccidioides immitis*); XP_001271144 (*Aspergillus clavatus*); XP_001263320 (*Neosartorya fischeri*); XP_001385197 (*Pichia stipitis*); EDV11110 (*Saccharomyces cerevisiae*); CAX41791 (*Candida dubliniensis*); EEH39780 (*Pteronura brasiliensis*); NP_842720 (*Bacillus anthracis str.Ames*).



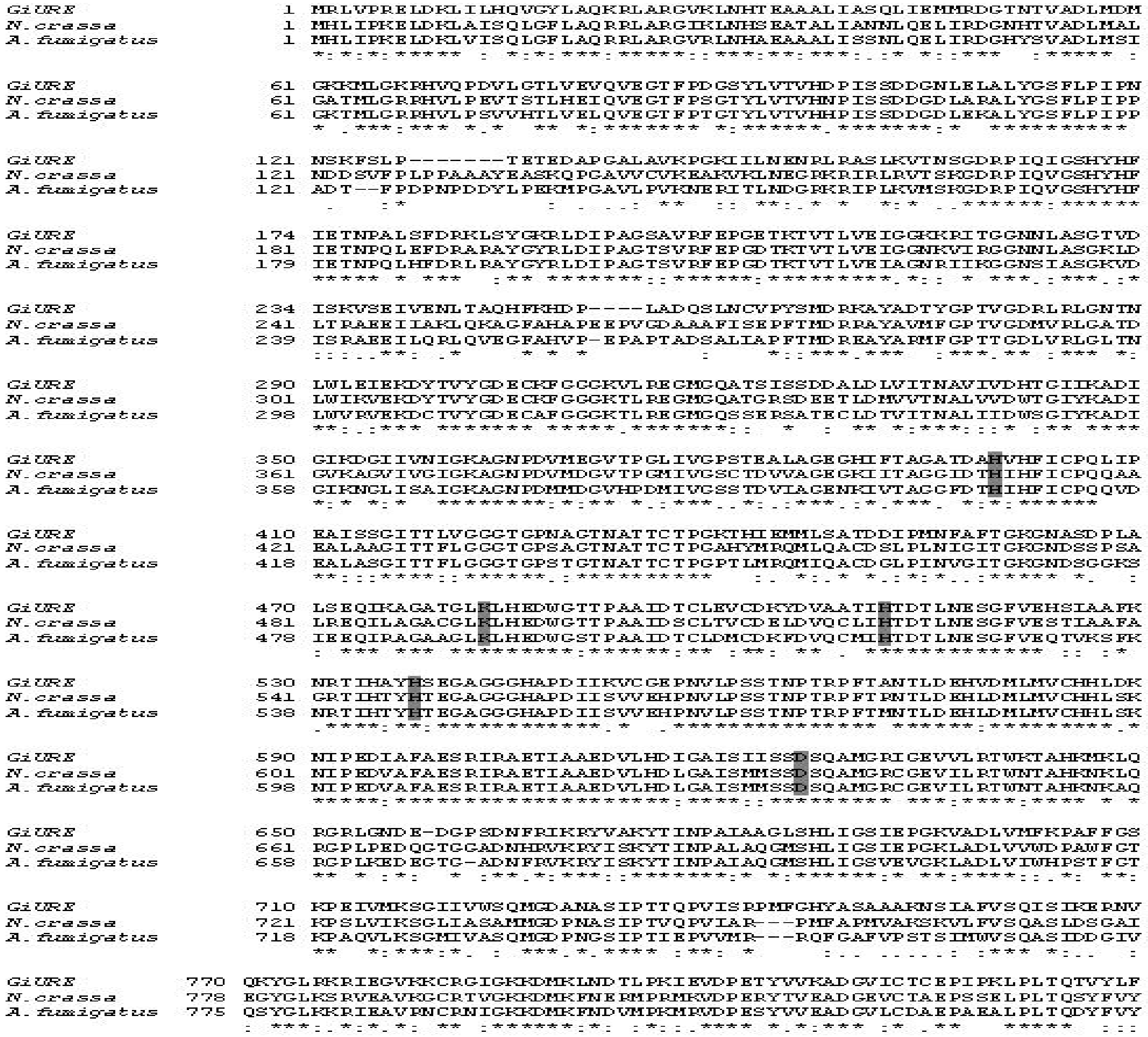
```
GiOAT1           1 ----------------------------------MSTQLSQKVIELEYEYGAHNYHPLPV
A.nidulans       1 ----------------------MTSNGTNGSATAYHASSTQEAIQAENDFAAHNYHPLPV
S.pombe          1 ---------------------------MSAESLLHNTFSTEQIEVLENEYAAHNYHPLPV
L.bicolor        1 MSPTATATNSHDFKGNGTHLAPQAKTVVLKPTPASPKITSADVIHKEHKYGAHNYHPLPV
                                                          : .    * .:.*********

GiOAT1          27 VFSRAQGAYVWDPEGNKYMDFLCAYSAVNQGHCHPKIVAALCDQAQKLTLSSRAFYNDVF
A.nidulans      39 VFARAQGTSVWDPEGRHYLDFLSAYSAVNQGHCHPKLVAALVDQASRLTLSSRAFYNDVF
S.pombe         34 CFSKAKGAKVWDPEGREYLDFLSAYSAVNQGHCHPKIIEALVEQAQRVTLSSRAFYNDKF
L.bicolor       61 VFDSAKGAKVWDPEGREYIDMLSAYSAVNQGHCHPRIVATLVEQAQRLTLSSRAFYNSVF
                    *  *:*: ******..*:*:*.************::: :* :**.::*********. *

GiOAT1          87 GRYAKFITEYFGYEMVLPMNTGAEAVETGIKLARKWGYLKKGIPKNKAIVVSCRNNFHGR
A.nidulans      99 PKFAEMVTKYFGFDMVLPMNTGAEAVETGIKIARKWGYKVKGIPENEAIILSAENNFHGR
S.pombe         94 GPFAKYITEYFGYEMVIPMNTGAEAVETACKLARLWGYKAKKIPTDEAIILSCVDNFHGR
L.bicolor      121 GRFAQQITEMFGYDMVLPMNTGAEAVETAVKLARKWAYMKKGVPDGKAIVLSVEGNFHGR
                     :*: :*: **::**:***********. *:** *.*  * :* .:**::*  .*****

GiOAT1         147 TTTIISMSTDSDAREGFGPYMPCVGPVCPATG--RSVDYNSIKDLEDAFEAHGPHIAAFL
A.nidulans     159 TMAAISLSSDPESRENYGPYVPNIG--CTIPGTEKPITYNDKAALREAFEKAGSNLAAFL
S.pombe        154 TMGIISMSTDPDARDNYGPYLPNVGPKIS--GADRVLRYNNIEDLKYYLDTFGPKVAAFL
L.bicolor      181 TLGIISMSTDPESRGGFGPYLEGVGPTFVDGGKTSTIRYGVLEDLERALELHGKNVAAFL
                   *   **:*:*.::* .:***:  :*      *    : *.    *.  ::  *  :****

                                 Aminotransferase class-III pyridoxal-phosphate attachment site
GiOAT1         205 VEPIQGEAGIIVPDDGYLKKAYDLCKKHNILLIADEVLTGLARTGKLLCQEHDEVRADIV
A.nidulans     217 VEPIQGEAGIIVPDDDYLQLARSLCDQHNVLLICDEIQTGIARTGKLLCHEWSGIKPDMV
S.pombe        212 VEPIQGEAGVMVPDDGYLEEAYKLCKAHNVLFIADEVQTGVARTGKMLCIEHSNVKPDVV
L.bicolor      241 VEPIQGEAGIVVPADGYLRSVHELCKRHNVLLICDEIQTGLCRTGKMLASEYDNVRPDMV
                   *********::** *.**. . .**. **:*:*.**: **:.****:*. * . ::.*:*

GiOAT1         265 ILGKALSGGVYPISAVLTSKDIMLCIKPGEHGSTYGGNPLGCAVAIAALEVIKEESLVEK
A.nidulans     277 LLGKAISGGMYPVSCVLGRKDVMLTVEPGTHGSTYGGNPLACAVAIRALEVVQEENMVER
S.pombe        272 ILGKAISGGVYPVSAVLSSREIMLNFEPGTHGSTYGGNPLGAAVSIAALEVVKEEKLTER
L.bicolor      301 LLGKALSGGVYPVSAVLADASIMLCIKPGEHGSTYGGNPLGCAVAMTALNVLVDEKLADR
                   :****:***:**:*.**   .:** .:** **********..**:: **:*: :*.:.::

GiOAT1         325 AAVLGEKFRSALRKIDSPLIQTVRGKGLLNALVIDETKSDK--TAWQLCLLLKSRGLLAK
A.nidulans     337 AEKLGQAFRSGLEAIQNPIIQTVRGKGLLNAIVIDESKTN-GHTAWDLCMLMKEKGLLAK
S.pombe        332 AAVLGEKFRTALIECKSPIVQKVRGRGLLNAVVIDESKTN-GRTAWDLCLIMRSRGVLAK
L.bicolor      361 AMRLGEYFRTAVRDLNSPLVAEVRGRGLLNAVVIDETKSKKGRTAWQLCLLLKSRGVLAK
                   *  **: **:.:   ..*::  ***:*****:****:*:.   ***:**::::.:*:***

GiOAT1         383 PTHKNIIRLAPVLSITEEELMQGVKIIEQALKNIINMKSDNIPRYLKDSKLILPDQFCDD
A.nidulans     396 PTHQNIIRLAPPLVITEEEIAKALEIIKAAVAELPNLKGAAEDKVVPPPEKKVKITLEN-
S.pombe        391 PTHGNIIRFSPPLVITEEDLMKGIEVIKKSLNDLPTIDMTPYAEKPIH------------
L.bicolor      421 PTHVNIVRFAPPLVIKEADLAKAVAIIGECLADLDEVDVIPGDDEEEKGHTDTLTL----
                   *** **:*::* * *.* :: :.: :*  .: ::  :.
```

Figure S8. Amino acid alignment of *GiOAT1* with ornithine aminotransferases from *Aspergillus nidulans* (XP_659414.1), *Schizosaccharomyces pombe* (NP_596588.1) and *Laccaria bicolor* (XP_001879943.1). Amino acids shaded in grey represent signature region.



```
GiODC              1 -----------------------MKGPTNQLNIKDKSTPPISMLSVEEVLKSQILSN---
A. fumigatus       1 -----------------------MSTPSDLGLSSATTFPYSVAVAVQNHLKDHG------
M. circinelloides  1 ---------------MLTDLETVVINPPYNVGALFKSAPSSPDISVHNRSVSEVFQQRLE
C. cinerea         1 MSQVETLPAVNPIDFSLSASQVCQRNEESNLNWPAASSPLINDVSASQCLRMPNNSYNGE
                                                         : *    ::. :

GiODC             35 -------------------------------------NYETENNDAFFVADLGEIYRQHL
A. fumigatus      32 -----------------------------------------KDRQSFFVGNLDSVLDRLE
M. circinelloides 46 SITNQ--------------------------------KWESDQENAFFVGDLGEVVRQHI
C. cinerea        61 SHDSSGDVIFPSLPPLHQGHPDIHMRNGVMASFAAAADHEPDAEKAFFVADLSQVYRQHQ
                                                              . ..:***.:*..:  :
                     Orn/DAP/Arg decarboxylases family 2 pyridoxal-P attachment site
GiODC             58 RWKTYLPRVEPFYAVKCNGDPMLLRLLAALGTGFDCASKNEIQTVLDMG-VSPSNIIYAN
A. fumigatus      51 EWRRELPFVQPFYAVKCNDNKTLLTLLSRAGTGFDCASKKEMQQILQLG-VNPERIIFAA
M. circinelloides 74 RWKSLLPRIEPFYAIKCNPDPMVVQLLASLGCGFDCASKQEIQQVLDVG-VDPSRIIYAN
C. cinerea       121 RWITCLPEIQPFYAVKCNPDPYVIRLLAALGAGFDCASLNEISSVLNVGGIDPSRIIFAN
                     .*   ** ::****:*** :  :: **:  * ****** :*:. :*::* :.*..**:*

GiODC            117 PCKQASFIRFASEHDVKMITFDNADELHKIKRLFPKAQLVIRILADDSKALIKLGAKFGA
A. fumigatus     110 PRKAEDYINYAHEHGIDKIVVDSEDELRKLAEIVPSAMIFLRLRADDPTSRVRLSEKFGS
M. circinelloides133 PCKQASFIRYSAQQNVSRMTFDNAEELFKIKKYYPDAELVLRILTDDSMSLCQLGLKFGA
C. cinerea       181 PCKAVSFIRNASRMGVDTMTFDNADELYKVARSHPKAKLVVRILTDDSKSLCAFGIKFGA
                     * *  .:*. : . .:. :..*. :** *: .  *.* :.:*: :**. :   :. ***:

GiODC            177 SLNDTGFLLKTALELGLNVIGVSFHVGSGCYDENSYIDAIRRARFVFNQASEMG--FDFH
A. fumigatus     170 DLLEARGILQVAVDLSVKVTGICFHVGSAALDPGAYVRAIAMAREVYDYNETLSSKHPIL
M. circinelloides193 PLHTVQHLLQLAKELNLNVIGVSFHVGSGCLDENAFGDAVVRAKNVFDQAKAMG--YDFK
C. cinerea       241 PLNVVPSLLAKAKELNLDVVGVSFHVGSGCYDPSVYTDAIMRSRTVFDMAKEAG--YDFT
                      *  .  :*  * :*.:.* *:.*****.. * . :  *:  :: *::  .  .  . :

GiODC            235 LLDVGGGFSFSHKSDEGITFEKIAAVLGPAIDKHFP--PNIRVIAEPGRYYAAPAFTIAT
A. fumigatus     230 IIDIGGGFTESN-------FRLLAPAVRSAADMYFGGETGIQWVAEPGRFIVSEAFYLVC
M. circinelloides251 FLDVGGGFPGADVKN-GITFEKVAAVLGPIVDRLFP--KDVRVIAEPGRYYVASAFNLAV
C. cinerea       299 LLDIGGGFEDAL-------FEKAATVLREAIDRYFPDRKHIKMIAEPGRFYVSTAFKLAA
                     ::*:****  :        *.  *..:    *  *     :: :*****: .: ** :.

GiODC            293 HVIAKRTVCRDGNEEYVPSMNVDQSNVLATKDDHPTFMYYINDGLYGSLYCILYDHQLVT
A. fumigatus     283 RVLGTRKRLVEYVDG-------------PNQNHYFHAELFVNDGIYQNFLNALVEGYIPT
M. circinelloides308 NVIGRRTNMAHQDQ----------------TDASPEYMYYVNDGMYGSVNCIIFDHQVVH
C. cinerea       352 HIIARRAPLEKDAAPATSG--------TAEEPDQPKVMYYINDGVYGAFNCILFDHQVIH
                     .::. *    .                            ::***:*  .   : :  :

GiODC            353 PKVLTKGGSFLFGEILDEPEYNCFIWGPTCDSIDCVTKNGSLPE-LFLGDWLYFEDMGAY
A. fumigatus     330 PVPLDPSGRPYGLPEREDKSYSYTVWGQTCCGADKIKSNCRLRCEMQAGDLLCFPSMGAY
M. circinelloides352 PKVLMKNELFAYGQDLPEAQYESSVWGPTCDSIDCLNKSARLPK-LEPGDWLYFENMGAH
C. cinerea       404 PYVLSMGGSFHVAESEP--KHVSSVWGPTCDSIDCVSPKTVLPVNLQVGDWLGFNNMGAY
                     *  *  .            .:   :** ** . * :. .  *   :  ** * * .***:

GiODC            412 TIAAATKFNGFKQTQVIYTTTDPHVFDYLYDVT---------------------
A. fumigatus     390 THVTASGFNGFTARAMTVWTSSVRGREKLKEANRVGSWWNPLSHGESELLVRRG
M. circinelloides411 TICAASQFNGFRKSEVLYTNTFC-------------------------------
C. cinerea       462 TVCAASQFNGFAVSNVIYTAGVAHANEVRMALKAFAAEGHGL------------
                     *  :*: ****    :
```

Figure S9. Amino acid alignment of *GiODC* with ornithine decarboxylases from *Aspergillus fumigatus* (XP_748770.1), *Caprinopsis cinerea* (XP_001836715.1) and *Mucor circinelloides* (CAB61758.1). The amino acids shaded in grey represent a conserved signature region.



```
GiURE             1 MRLVPRELDKLILHQVGYLAQKRLARGVKLNHTEAAALIASQLIEMMRDGTNTVADLMDM
N.crassa          1 MHLIPKELDKLAISQLGFLAQRRLARGIKLNHSEATALIANNLQELIRDGNHTVADLMAL
A.fumigatus       1 MHLIPKELDKLVISQLGFLAQRRLARGVRLNHAEAAALISSNLQELIRDGHYSVADLMSI
                    *:*:*:***** : *:*:***:*****::***:**:***:.:* *::***  :***** :

GiURE            61 GKKMLGKRHVQPDVLGTLVEVQVEGTFPDGSYLVTVHDPISSDDGNLELALYGSFLPIPN
N.crassa         61 GATMLGRRHVLPEVTSTLHEIQVEGTFPSGTYLVTVHNPISSDDGDLARALYGSFLPIPP
A.fumigatus      61 GKTMLGRRHVLPSVVHTLVELQVEGTFPTGTYLVTVHHPISSDDGDLEKALYGSFLPIPP
                    * .***:*** *.*  ** *:******* *:******.*******:*  **********

GiURE           121 NSKFSLP-------TETEDAPGALAVKPGKIILNENRLRASLKVTNSGDRPIQIGSHYHF
N.crassa        121 NDDSVFPLPPAAAYEASKQPGAVVCVKEAKVKLNEGRKRIRLRVTSKGDRPIQVGSHYHF
A.fumigatus     121 ADT--FPDPNPDDYLPEKMPGAVLPVKNERITLNDGRKRIPLKVMSKGDRPIQVGSHYHF
                     .   :*          : . ..: **  :: **:.* *  *:* ..******:******

GiURE           174 IETNPALSFDRKLSYGKRLDIPAGSAVRFEPGETKTVTLVEIGGKKRITGGNNLASGTVD
N.crassa        181 IETNPQLEFDRARAYGYRLDIPAGTSVRFEPGDTKTVTLVEIGGNKVIRGGNNLASGKLD
A.fumigatus     179 IETNPQLHFDRLRAYGYRLDIPAGTSVRFEPGDTKTVTLVEIAGNRIIKGGNSIASGKVD
                    ***** * ***  :** ****::******:*********.*:: * ***.:***.:*

GiURE           234 ISKVSEIVENLTAQHFKHDP----LADQSLNCVPYSMDRKAYADTYGPTVGDRLRLGNTN
N.crassa        241 LTRAEEIIAKLQKAGFAHAPEEPVGDAAAFISEPFTMDRRAYAVMFGPTVGDMVRLGATD
A.fumigatus     239 ISRAEEILQRLQVEGFAHVP-EPAPTADSALIAPFTMDREAYARMFGPTTGDLVRLGLTN
                    :::..**: .*    * * *        :    *::***.***  :***.** :*** *:

GiURE           290 LWLEIEKDYTVYGDECKFGGGKVLREGMGQATSISSDDALDLVITNAVIVDHTGIIKADI
N.crassa        301 LWIKVEKDYTVYGDECKFGGGKTLREGMGQATGRSDEETLDMVVTNALVVDWTGIYKADI
A.fumigatus     298 LWVRVEKDCTVYGDECAFGGGKTLREGMGQSSERSATECLDTVITNALIIDWSGIYKADI
                    **:.:*** ****** *****.*******::  *  : ** *:***:::* :** ****

GiURE           350 GIKDGIIVNIGKAGNPDVMEGVTPGLIVGPSTEALAGEGHIFTAGATDAHVHFICPQLIP
N.crassa        361 GVKAGVIVGIGKAGNPDVMDGVTPGMIVGSCTDVVAGEGKIITAGGIDTHIHFICPQQAA
A.fumigatus     358 GIKNGLISAIGKAGNPDMMDGVHPDMIVGSSTDVIAGENKIVTAGGFDTHIHFICPQQVD
                    *:* *:*  ********:*:** *.:***..*:.:***.:*.***. *:*:******

GiURE           410 EAISSGITTLVGGGTGPNAGTNATTCTPGKTHIEMMLSATDDIPMNFAFTGKGNASDPLA
N.crassa        421 EALAAGITTFLGGGTGPSAGTNATTCTPGAHYMRQMLQACDSLPLNIGITGKGNDSSPSA
A.fumigatus     418 EALASGITTFLGGGTGPSTGTNATTCTPGPTLMRQMIQACDGLPINVGITGKGNDSGGKS
                    **:::****::******.:*********   :. *: * *.:*:*..:***** *.  :

GiURE           470 LSEQIKAGATGLKLHEDWGTTPAAIDTCLEVCDKYDVAATIHTDTLNESGFVEHSIAAFK
N.crassa        481 LREQILAGACGLKLHEDWGTTPAAIDSCLTVCDELDVQCLIHTDTLNESGFVESTIAAFA
A.fumigatus     478 IEEQIRAGAAGLKLHEDWGSTPAAIDTCLDMCDKFDVQCMIHTDTLNESGFVEQTVKSFK
                    : *** *** *********:******:** :**: ** . ************* :: :*

GiURE           530 NRTIHAYHSEGAGGGHAPDIIKVCGEPNVLPSSTNPTRPFTANTLDEHVDMLMVCHHLDK
N.crassa        541 GRTIHTYHTEGAGGGHAPDIISVVEHPNVLPSSTNPTRPFTPNTLDEHLDMLMVCHHLSK
A.fumigatus     538 NRTIHTYHTEGAGGGHAPDIISVVEHPNVLPSSTNPTRPFTMNTLDEHLDMLMVCHHLSK
                    .****:**:************.*  .*************** ******:*********.*

GiURE           590 NIPEDIAFAESRIRAETIAAEDVLHDIGAISIISSDSQAMGRIGEVVLRTWKTAHKMKLQ
N.crassa        601 NIPEDVAFAESRIRAETIAAEDVLHDLGAISMMSSDSQAMGRCGEVILRTWNTAHKNKLQ
A.fumigatus     598 NIPEDVAFAESRIRAETIAAEDVLHDLGAISMMSSDSQAMGRCGEVILRTWNTAHKNKAQ
                    *****:********************:****::********* ***:****:**** * *

GiURE           650 RGRLGNDE-DGPSDNFRIKRYVAKYTINPAIAAGLSHLIGSIEPGKVADLVMFKPAFFGS
N.crassa        661 RGPLPEDQGTGGADNHRVKRYISKYTINPALAQGMSHLIGSIEPGKLADLVVWDPAWFGT
A.fumigatus     658 RGPLKEDEGTG-ADNFRVKRYISKYTINPAIAQGMSHLIGSVEVGKLADLVIWHPSTFGT
                    ** * :*:  * :**.*:***::*******:* *:******:* **:****::.*: **:

GiURE           710 KPEIVMKSGIIVWSQMGDANASIPTTQPVISRPMFGHYASAAAKNSIAFVSQISIKERNV
N.crassa        721 KPSLVIKSGLIASAMMGDPNASIPTVQPVIAR---PMFAPMVAKSKVLFVSQASLDSGAI
A.fumigatus     718 KPAQVLKSGMIVASQMGDPNGSIPTIEPVVMR---RQFGAFVPSTSIMWVSQASIDDGIV
                    **  *:***:*. : ***.*.**** :**: *     :.. .....: :*** *:..  :

GiURE           770 QKYGLRKRIEGVKKCRGIGKKDMKLNDTLPKIEVDPETYVVKADGVICTCEPIPKLPLTQTVYLF
N.crassa        778 EGYGLKSRVEAVKGCRTVGKKDMKFNERMPRMKVDPERYTVEADGEVCTAEPSSELPLTQSYFVY
A.fumigatus     775 QSYGLKKRIEAVPNCRNIGKKDMKFNDVMPKMPVDPESYVVEADGVLCDAEPAEALPLTQDYFVY
                    : ***:.*:*.*  ** :******:*: :*:: **** *.*:*** :* .**   *****  :::
```

Figure S10. Amino acid alignment of *GiURE* with ureases from *Neurospora crassa* (XP_964986.1) and *Aspergillus fumigatus* (CAE17672.1). Amino acids shaded in grey represent conserved signature residues.

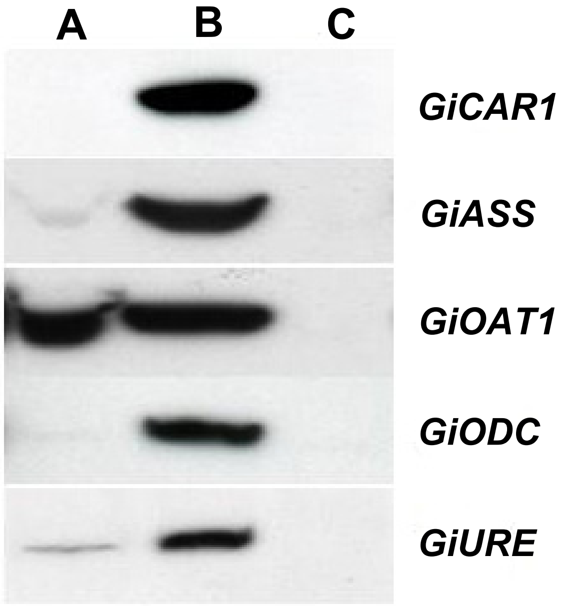


Figure S11. Western blot of the expressed proteins in yeast. Whole-cell protein extracts from the yeast transformed with Gi genes were fractionated on a denaturing 12% polyacrylamide gel, transferred to polyvinylidene fluoride (PVDF) membranes and detected with Anti-His(C-term)-HRP antibody. The yeast knockout mutants were independently transformed with the empty pYES2.1 vector or the vector fused with *GiCAR1*, *GiASS*, *GiOAT1*, *GiODC* and *GiURE*. **a**, The expression of the Gi genes without the induction of galactose; **b**, The expression of the Gi genes with the induction of galactose; **c**, The expression of the empty vector with the induction of galactose (as the negative control).

Supplementary Table S1. Primers for gene cloning

Gene	**Forward (5’- 3’)**	**Reverse (5’- 3’)**
GiASS	ATGTCACAAGGTCGCGTTCTTCTCG	TAAATTGTTGCCAAAAGCAGTTTCTCC
GiCAR1	GTAACTAACTCTATAAAGATGGCC	TAACAAAGTTTCACCGAAACAACA
GiGS1	TTAAATCTGAAAGATGTATAACAA	TGCAAGTGAAGACTCAACAATAAT
GiGS2	AAACATTAATAACGAAAATGGCTTTTAT	ACCAAGAGAAGATTCAACGATGATC
GiOAT1	ATGTCAACTCAACTTAGTCAAAAAG	ATCATCGCAAAACTGATCA
GiODC	ATGAAGGGGCCTACGAATCAGC	TGTAACATCATAGAGATAATCAAAAACATGAGG
GiURE	ATGAGGTTAGTACCGAGAGAATTA	AAACAAGTAAACCGTTTGAGTTAAAGG

Supplementary Table S2. Yeast strains for functional complementation

Yeast knockout mutant	Corresponding parent line	Source of the strain
Haploid ΔGLN1 (MATa, his3D1, leu2D0, met15D0, ura3D0, YPR035w::kanMX4)	YPR035W (MATa, his3D1, leu2D0, ura3D0, ΔGLN1::kanMX4)	Made from diploid YPR035W by sporulation
YPR035W (MATa, his3D1, leu2D0, ura3D0, ΔGLN1::kanMX4)	BY4743 (MATa, his3, leu2, ura3)	Open Biosystem, USA
YOL058W (MATa, his3, leu2, lys2, ura3, ARG1 Δ ::kanMX4))	BY4742 (MATa, his3, leu2, lys2, ura3)	Open Biosystem, USA
YKL184W (MATa, his3, leu2, lys2, ura3, SPE1 Δ ::kanMX4)	BY4742 (MATa, his3, leu2, lys2, ura3)	Open Biosystem, USA
YBR208C (MATa, his3, leu2, lys2, ura3, Dur1,2 Δ ::kanMX4)	BY4742 (MATa, his3, leu2, lys2, ura3)	Open Biosystem, USA
O2463d ΔARG81 (leu2, ura3)	O2463d	Evelyne Dubois, Université Libre de Bruxelles
12T7cI ura3, ΔCAR1	2T7cI ura3	Evelyne Dubois, Université Libre de Bruxelles

Supplementary Table 3 Primers for real time PCR

Gene	**Forward (5’- 3’)**	**Reverse (5’- 3’)**
DcGS	GGATGGCCAAATGGAGGATT	TGTCGGCACCAATACCACAGT
DcEAF	AGGATGATCCCGCCAAAGAG	CCAGGGTGGTTCATGATGATAA
GiAL	ACGGACTTGGCTGAATATTTGGT	GCCCCTGCAATATGATGAGTTT
GiASS	GCATTGGTCGTATTGATATTGTTGA	TTGTTCCCCCAGGAGTTTCA
GiCAR1	TGATGCGGTGAATCCTAAGAGA	GATCAAGTGCATCAACGTCAAAG
GiCPS	GATCGCCGTCGTTGACTGT	CGCGCCGCGTTTAACTAA
GiGS1	AGTGGCCTTCGTTCAAAGACTAG	CATCACCAGGTGCTTGATTAGTA
GiGS2	CCAACATTGATCCTTATCGTGTC	CTCCTAAATTAGAGAAAGAAAAAAAGGG
GiGluS	TTCGCAGCCGGTGATTG	CGACCCTCATTTATACCCCATACA
GiNT	GGCTTGCTGCGGTTCAGT	CGACTAATCCTGATATTGCACCAA
GiOAT1	GGTTCGAGCGGATATTGTCATAC	AGGACTGCTGATATTGGGTAAACG
GiOAT2	CGGGTAAGATGCTTTGTCAAGA	GCCTGAAAGTGCTTTACCAAGTATAAC
GiODC	TTGATTGCGTTACCAAAAATGG	TCGAAATACAACCAGTCACCAAGA
GiSR4	AAGCCGCCTACGTGTCGTT	AACAGGTGGTAGAAATATGGGAAG
GiURE	CGCAAATGGGAGATGCAAAT	CAGCACTTGCATAATGACCAAAC