

Colhig3 1 MILPHAAVRSLQALSINAAQPLQSSSIQCPIVLDGRIPQNFTLQVFDTNESPFNPGFTK
Colinc3 1 MMVPHMAVRSLQALSINSAQSLQSSVQCPIVLDGRIPQNFSLQVFDTNESPFNPGFTK
Coltof3 1 MVLPHAAVRSLQALSINSAQSLQLPVQCPIVLDGRIPQNFSLQVFDTNDSPFNPGFTK
Colfio3 1 MILPHAAVRSLQALSINAPRASFDSPVKCPVVDGRIPQNFTLQVFDTYASPFNPGYTK
Colnym3 1 MILPHAAVRSLQALSINAPRASLDSPVKCPVVDGRIPQNFTLQVFDTYASPFNPGYTK
Colsim3 1 MILPHAAVRSLQALSINAPRASLDSPVKCPVVDGRIPQNFTLQVFDTYDSPFNPGYTK
Colsal3 1 MILPHAAVRSLQALALNVPRASLDSPVKCPVVDGRIPQNFTLEVFDTSTSPFNPGFTK
Colchl3 1 MILPHTAVRSLQALSINAPFQSFO-SPVRCPVVDGRIPQNFTLEIFDTNASPFNPGFTK
Colglo3 1 MILPHLAVRAL-S-----ASVQCPIVLDGRISQNFTLENFDTNVSPVNPNGFTK
Colorb3 1 MILPQVAARALLGLS-----SAVKCPIVLDGRIPQNFTLDDFDTNASPFNPGFTK

Colhig3 61 GPAPWSEILLFPAGVTPSRFDVSEPE---TATSEFKPLEVTIDDKSIFD-PSGNNPQLGFR
Colinc3 60 GPAPWSDILLFPTNTTPSRFDVSN-----STGNFKPEVETINDQSIFD-PSGNNPQVIGFR
Coltof3 60 GPAPWSEILLFPALTTPSRFDVSS-----STGDSKPEVETINDQSIFD-PSGNNPQVIGFR
Colfio3 60 GPAKWSEILLFPVPTPSRFDASN-----ATGSEFKPLEVTINDQSIFN-PSGNSPQLGFR
Colnym3 60 GPAKWSEILLFPVPTPSRFDTSN-----ATGNFKPLEVTINDQSIFN-PSGNSPQLGFR
Colsim3 60 GPAKWSEILLFPVPTPSRFDTSN-----ATGNFKPLEVTINDQSIFN-PSGNSPQLGFR
Colsal3 60 GPAPWSEILLFPVPTPSRFDASN-----ATGNFKPLEVTINDQSIFN-PSGNSPQLGFR
Colchl3 60 GPALWSEILLFPNISTTPSRFDVSS-----QTGDFKPLEVTINDQSIFNPPPGNNPQLGFR
Colglo3 48 GSDPWSKIILLFPTVAAPSRFDAAASANATSGSGFKPLEVTINDRSIFNPPPGKNPQLGFR
Colorb3 51 GPDPWSKILLFPFVAQTSRFDVAGR----SPANTKPLEVTINDDSIFNPPPGKNPQLGFR

Colhig3 117 RAGLLLGNGSDASNQGVMTFHWSSRQDPARKLNLTHEYLTSFHETNDSGGAQFSLQVGTGP
Colinc3 114 RAGLLLGNGSDASNQGVMTFHWSSMQDPTRKLNLTHEYLVSFHETNDSGGAQFALQIGTGP
Coltof3 114 RAGLLLGNGSDASNQGVKTFHWSSRQDSTRKLNLTHEYLTFSFHETNDSGGAQFALQIGTGP
Colfio3 114 RAGLLLGNGSDASNQGVMTTHWSSMQDSTRKLNLTHEYLTFSFHETNDSGGAQFALQIGTGP
Colnym3 114 RAGLLLGNGSDASNQGVMTTHWSSMQDSTRKLNLTHEYLTFSFHETNDSGGAQFALQIGTGP
Colsim3 114 RAGLLLGNGSDASNQGVMTTHWSSMQDSTRKLNLTHEYLTFSFHETNDSGGAQFALQIGTGP
Colsal3 114 RAGLLLGNGSDASNQGVMTTHWSSMQDSTRKLNLTHEYLTFSFHETNDSGGAQFALQIGTGP
Colchl3 115 RAGLLLGNGSDASNEGVMTFHWSSRQDSARKLNLTHEYLTAFHETNDSGGAQFALQIGTGP
Colglo3 108 RAGLLLGNGSDASNQGVMTFHWSSAMQDSARKLNLTHEYLTFSFHETNDSGGAQFALQVGLP
Colorb3 107 RAGLLLGNGSDAANECAVTTHWSSRQDPARLNLTHEYATAFHETNDSGGAQFTLQVGLP

Colhig3 177 LGQDGAAGKPAASDWKMLNRNNIVVFSTPLKFDAWQNEAVTLDVPKNTMQVYFSEGNDPL
Colinc3 174 LGQNGTAGRPSPTDWKMLDRNNIVVFSTPLKFDAWQNEAVTLDVPKNTMQVFFSEGNDPL
Coltof3 174 LGQNSTAGRPSLSDWKMLDRNNIVVFSTPLKFDAWQNEAVTLDVPKNTMQVFFSEGNEPL
Colfio3 174 LGQNSTAGKPLSNDWKMLDRNNSVIFSTALKFDVWQNWAVTLDVPKNTIQVFFSEGNDPL
Colnym3 174 LGQNSTAGKPLSTDWKMLDRNNSVIFSTALKFDVWQNWAVTLDVPKNTIQVFFSEGNDPL
Colsim3 174 LGQNSTVGKPLSTDWKMLDRNNSVIFSTALKFDVWQNWAVTLDVPKNTIQVFFSEENDPL
Colsal3 174 LGQNSTTGGPSSADWKMLDRNNSVIFSTALKFDVWQNWAVTLDVPKNTIQVFFSEGNDPL
Colchl3 175 LGQNSTLSGEASSDWKMLDRDNKVVVFSTPLKFDAWQNEGVTLDIPNNTLQVYFSEGDDPL
Colglo3 168 LGQNGT--EPPKENWKMLDRNNSVVFSTPLKFDTWQNWAVTLDVPQNTMQVFFSEGNEPL
Colorb3 167 LGQNKNT--EPPAANWKMLNRNNSIVVFSTPLQFGKWQNEAVTLDVPKNTMQVYYSEGNEPL

Colhig3 237 KPVTEILPNDNSGGGALQIGLLKKPTETKTIVVFDGFGQESNLDEGQILGSI FVEDSANGCV
Colinc3 234 KPVTEVLPNDNSGGGALQIGLLKKPTETKTIVVFDGFGQESNLNEGQILGGIFVENSANGCI
Coltof3 234 KPVTEVEIPNDNSGGGVLOIGLLKKPTETKTAVFDGFGQESNLNEGQILGGIFVEDSANGCV
Colfio3 234 KSVTEVLPNNNAGGGALQIGLLKKPTETKTIVVNDGFGQESNLNEGQILGGIFVENSANGCV
Colnym3 234 KSVTEVLPNNNVGGGALQIGLLKKPTETKTIVVNDGFGQENLNEGQILGGIFVENSANGCV
Colsim3 234 KSVTEVLPNNNVGGGALQIGLLKKPTETKTIVVNDGFGQESNLNEGQILGGIFVENSANGCV
Colsal3 234 KSVTEVLPNNNAGGGALQIGLLKKPTETKTIVVNDGFGQESNLNEGQILGGIFVENSANGCI
Colchl3 235 KPVTEVLPNNNAGGGALQIGLLKKPTETTTAVFDGFGQENLNEGQILGGIFVENSANGCV
Colglo3 226 KAVTKILPNNNAGGGALQIGLLKKPTETKTIVANDGFGQESNLNEGQILGGIFVENSANGCI
Colorb3 225 SAVTGVLPNNNAGGGALQIGLLKKPTETKTIVVFDGFGQESNLNEGQILGGIFLENS SDGCV

Colhig3 297 SL
Colinc3 294 SL
Coltof3 294 SL
Colfio3 294 SL
Colnym3 294 SL
Colsim3 294 SL

Colsal3	294	SL
Colchl3	295	SL
Colglo3	286	SL
Colorb3	285	SL

Sordariomycetes

Hypocreomycetidae - Glomerellales (Colletotrichum but not Verticillium)

Colchl3 = OLN81946.1 hypothetical protein CCHL11_08641 [Colletotrichum chlorophyti]
 Colfio3 = EXF72940.1 hypothetical protein CFIO01_04646 [Colletotrichum fiorinae PJ7]
 Colglo3 = KAF3801482.1 hypothetical protein GCG54_00014696 [Colletotrichum gloeosporioides]
 Colletotrichum graminicola - no orthologue
 Colhig3 = CCF43865.1 hypothetical protein CH063_03177 [Colletotrichum higginsianum]
 which is equivalent to CH063_00647 described by Anasontzis et al. (2019)
 Colinc3 = KZL79966.1 hypothetical protein CI238_09667 [Colletotrichum incanum]
 Colnym3 = KXH53449.1 hypothetical protein CNYM01_01194 [Colletotrichum nymphaeae SA-01]
 Colorb3 = TDZ24818.1 hypothetical protein Cob_v002377 [Colletotrichum orbiculare MAFF 240422]
 Colsal3 = KXH68549.1 hypothetical protein CSAL01_02206 [Colletotrichum salicis]
 Colsim3 = KXH27864.1 hypothetical protein CSIM01_06783 [Colletotrichum simmondsii]
 Coltof3 = KZL72700.1 hypothetical protein CT0861_06503 [Colletotrichum tofieldiae]

Hypocreomycetidae - Hypocreales - Clavicipitaceae - no orthologues

Hypocreomycetidae - Hypocreales - Cordycipitaceae - no orthologues

Hypocreomycetidae - Hypocreales - Hypocreaceae - no orthologues

Hypocreomycetidae - Hypocreales - Nectriaceae - no orthologues

Hypocreomycetidae - Hypocreales - Ophiocordycipitaceae - no orthologues

Hypocreomycetidae - Hypocreales - Stachybotryaceae - no orthologues

Hypocreomycetidae - Microascales - no orthologues

Sordariomycetidae - Coniochaetales - no orthologues

Sordariomycetidae - Togniniales - no orthologues

Sordariomycetidae - Diaporthales - no orthologues

Sordariomycetidae - Magnaporthales - no orthologues

Sordariomycetidae - Ophiostomataceae - no orthologues

Sordariomycetidae - Sordariales - Sordariales incertae sedis - no orthologues

Sordariomycetidae - Sordariales - Chaetomiaceae - no orthologues

Sordariomycetidae - Sordariales - Sordariaceae - no orthologues

Xylariomycetidae - Xylariales - no orthologues

Supplementary Figure S8. ChGluc131B alignment. Multiple protein-sequence alignment of ChGluc131B orthologues in the Fol25 clade of Sordariomycete GH131 (glycosyl-hydrolase 131) sequences. All sequences shown were the highest blastP matches to ChGluc131B (= Colhig3 highlighted in green in the alignment) obtained for each genome-sequenced species represented. Sequences were aligned using ClustalW at <https://www.ebi.ac.uk/Tools/msa/mafft/>. Amino-acid identities were highlighted in black and similarities in grey using BoxShade at https://embnet.vital-it.ch/software/BOX_form.html. Conserved cysteines have been highlighted manually in blue, residues conserved with the catalytic residues of PaGluc131A and CcGH131A in red and non-conserved residues in teal. None of these sequences were found to have predicted signal peptide sequences when analysed using SignalP 5.0 (<http://www.cbs.dtu.dk/services/SignalP/>) or SignalP 4.1 (<http://www.cbs.dtu.dk/services/SignalP-4.1/>) but Colorb3 and Colsal3 had predicted signal peptide sequences when analysed using SignalP 3.0 (<http://www.cbs.dtu.dk/services/SignalP-3.0/>) and all were predicted to be secreted proteins when analysed using Secretome P 2.0 (<http://www.cbs.dtu.dk/services/SecretomeP/>).