SUPPLEMENT

Skp1 isoforms are differentially modified by a dual function prolyl 4-hydroxylase/N-acetylglucosaminyltransferase in a plant pathogen

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SUPPLEMENTAL FIGURES

Fig. S1. *P. ultimum* Phgt sequence.

Fig. S2. *P. ultimum* Skp1A sequence.

Fig. S3. *P. ultimum* Skp1B sequence.

Fig. S4. Alignment of predicted Skp1 sequences from select protists.

Fig. S5. Organisms with Phgt-like sequences in their genomes.

Fig. S6. Mass spectrometry of Skp1 modifications.

Fig. S1. *P. ultimum* Phgt sequence. Black nucleotides are from genomic DNA contig ADO01001287 (reverse complement of nt 28981-34080) at the NCBI wgs database; upper case nt represent the predicted ORF, and lower case nucleotides are non-coding; underlined nt represent intron splice signals. Blue nt are codon optimized for expression in *E. coli* and *D. discoideum*, with 5'- and 3'-restriction sites for cloning indicated; green nt encode the N-terminal FLAG-tag from pDM320 plasmid. Native amino acids are in bold black and are from the protein model PYU1_T004073 at www.pythiumdb.org; red amino acids mark notes on the right; green amino acids represent the N-terminal extension that encodes the FLAG tag;

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                            tgccgctccacacagggtgcgtgtggccgtcg
acgaacccgggcaggatgcagaactcgctgccgtcgatctcctcgtcaaagtgtacgggt
tgcggctgcgtctgcagccactgcgccacgtgctcgtccgtgccaacggccgcgatcttg
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ctttatcatgaatactttgctttatcaaagatactcattataaaaggtatgccgactttt
ttctattgaagttctgtttgaggctcgtcacggtctcagactcagatgtcataccgtaca
gagacttagcaaagggccctgatcggatcaggcgtatgctactggatctgataccatcgt  -60

                           M  D  Y  K  D  D  D  D  K  R  S    FLAG-tag
gattggattcgatttgcacacatccaattgaagccacgcgcaccagaagcacgcaggacg    0
                           ATGGATTATAAAGATGATGATGATAAAAGATCT  BglII

M  D  K  K  S  L  L  S  V  L  S  P  Q  L  Q  A  L  V  E  A     20
ATGGACAAGAAATCGCTGCTGAGCGTGCTGTCGCCGCAACTGCAGGCGCTCGTGGAGGCT   60
ATGGATAAAAAGAGCTTGTTGAGCGTTTTGAGCCCGCAATTGCAAGCGTTGGTTGAAGCG


S  L  G  V  P  L  Q  A  N  A  P  A  Q  P  L                    35
TCTCTGGGCGTGCCACTGCAAGCGAACGCTCCAGCACAGCCACgtgcgcctctaaccaat  120
AGCTTGGGTGTGCCGTTGCAAGCGAACGCGCCGGCGCAGCCGT

ccagagagcaacgggaaagtggacacagctggtgacgaagcgacagaagctgcgatagcg  180
gatcagcggacaggtgggagcagcaaagcacccgtggcttcaactacacgcccagcagtg  240
gcaacgggcgtgggtcgtccaaggaaagttgctggtgcgaatgcgaagctgaacccttcc  300
gaattgtttggtacgtacctgaaccagtctgtcatcacgccatatcaaatcgcacgatac  360

                                       L  K  H  P  E  S  P     42
gctttaactttgcgatttgtatgtacgcacttgtcagTTCTGAAGCACCCCGAGTCACCA  420
                                     TGTTGAAGCACCCGGAGAGCCCG

G  F  V  V  K  D  A  F  L  G  S  E  H  A  V  T  V  R  D  A     62 Start of P4H-like
GGCTTTGTCGTGAAAGATGCGTTTCTCGGATCGGAGCATGCTGTCACCGTGCGTGACGCA  480
GGCTTCGTGGTTAAAGATGCGTTTTTGGGTAGCGAACATGCGGTGACCGTTCGTGATGCG

L  L  A  L  T  K  T  E  S  F  H  E  A  K  V  G  H  G  D  H     82
TTGCTCGCGCTCACGAAGACCGAATCGTTTCATGAAGCCAAGGTCGGACACGGCGATCAC  540
TTGTTGGCGTTGACCAAGACCGAGAGCTTCCACGAAGCGAAAGTGGGCCACGGTGATCAC

L  R  N  E  R  A  V  R  G  D  R  I  H  W  V  K  R  P  S  D    102
CTTCGCAACGAGCGTGCCGTGCGCGGAGACCGTATCCACTGGGTCAAGCGTCCCAGTGAT  600
TTGCGTAACGAGCGTGCGGTGCGTGGTGACCGTATTCACTGGGTTAAGCGTCCGAGCGAC

L  N  R  S  D  L  L  H  P  A  I  L  Y  L  M  K  Q  V  E  S    122
CTGAATCGGTCAGATCTGCTGCATCCTGCGATTCTGTACCTGATGAAGCAAGTCGAGAGT  660
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TTGAACCGTAGCGATTTGTTGCACCCGGCGATCTTGTACTTGATGAAGCAGGTTGAAAGC

A  A  Y  A  F  K  Q  T  N  P  D  L  D  L  R  N  V  T  S  T    142
GCGGCGTACGCCTTCAAACAGACCAATCCTGACCTGGATCTTCGGAACGTCACGTCCACG  720
GCGGCGTATGCGTTCAAACAAACCAACCCGGACTTGGATTTGCGTAACGTGACCAGCACC

Q  F  A  I  F                                                 147
CAATTTGCCATTTTTgtatcctttttttcctgcttcatacttgctgcatcgttgtgaccc  780
CAGTTCGCGATTTTT

attggtattggtatcagacattatgctccttgacctttgctgctgtgcttttgtgtgtag  840

P  G  D  G  S  R  F  V  K  H  T  D  T  Y  S  T  A  H  Q  D    167
CCAGGCGATGGGAGTCGATTTGTGAAGCACACAGACACGTACTCGACAGCGCACCAGGAC  900
CCGGGCGATGGTAGCCGTTTTGTTAAACACACCGACACCTACAGCACCGCGCACCAAGAT

E  S  G  T  S  S  S  S  D  G  L  V  R  V  L  T  C  V  Y  Y    187
GAAAGCGGCACTTCGTCTTCTTCCGATGGACTCGTGCGTGTTCTCACGTGTGTGTATTAC  960
GAGAGCGGTACCAGCAGCAGCAGCGATGGCTTGGTGCGTGTTTTGACCTGCGTGTACTAT

L  N  Q  D  W  V  P  E  H  E  G  Q  L  R  V  Y  V  K  G  T    207
CTGAACCAAGATTGGGTACCTGAGCACGAAGGCCAGCTGCGTGTGTACGTGAAGGGAACC 1020
TTGAACCAGGACTGGGTTCCGGAGCACGAAGGCCAATTGCGTGTGTACGTTAAGGGTACC

S  T  L  L  M  Q  H  W  D  V  A  P  K  L  D  T  L  V  V  F    227
TCGACTCTTCTAATGCAGCACTGGGACGTTGCCCCAAAGCTGGACACGCTGGTCGTGTTT 1080
AGCACCTTGTTGATGCAGCACTGGGACGTGGCGCCGAAATTGGATACCTTGGTTGTTTTC


R  S  L  D  V  E  H  E  V  L  P  T  F  Y  E  R  M  A  I  T    247
CGAAGTCTTGATGTGGAGCATGAGGTGCTTCCGACTTTTTACGAGCGCATGGCGATAACA 1140
CGTAGCTTGGATGTGGAGCACGAAGTTTTGCCGACCTTTTATGAACGTATGGCGATTACC

V  W  Y  Y  G  H  V  A  K  Q  P  P  D  P  A  A  S  I  E  P    267 End of P4H-like
GTGTGGTACTATGGACACGTTGCAAAGCAACCGCCGGACCCAGCAGCTAGTATCGAGCCT 1200
GTTTGGTACTATGGTCATGTTGCGAAACAACCGCCGGACCCGGCGGCTAGCATTGAGCCG

Q  K  Q  Q  R  Q  N  S  L  E  P  P  P  S  P  S  P  N  I  L    287
CAGAAGCAACAACGACAGAATAGTCTAGAGCCACCGCCATCTCCAAGTCCAAACATTTTA 1260
CAGAAACAGCAACGTCAGAACAGCTTGGAGCCGCCGCCGAGCCCGAGCCCGAACATCTTG

L  S  K  E  A  S  L  G  D  A  R  T  I  F  V  G  I  P  S  Y    307 Start of GlcNAcT-like
TTATCGAAAGAAGCATCTCTGGGGGATGCACGTACCATTTTCGTTGGCATCCCAAGCTAT 1320
TTGAGCAAAGAGGGATCCTTGGGTGACGCGCGTACCATTTTCGTGGGTATCCCGAGCTAC


R  D  P  E  C  R  H  T  V  A  D  L  L  H  K  A  T  F  P  D    327
CGTGATCCGGAATGCAGACACACGGTCGCGGATTTACTGCATAAAGCAACGTTTCCAGAC 1380
CGTGACCCGGAATGCCGTCACACCGTTGCGGATTTGTTGCACAAAGCGACCTTTCCGGAC

R  I  H  I  G  I  Y  L  Q  E  D  E  N  D  D  T  L  R  H  F    347
CGTATTCACATCGGTATTTACTTGCAGGAGGACGAAAATGACGATACGCTCCGCCACTTT 1440
CGTATCCACATTGGCATCTACTTGCAGGAAGACGAAAACGACGATACCTTGCGTCACTTC

E  E  M  Y  P  R  S  Q  V  R  V  Q  F  A  D  Y  R  S  A  A    367
GAAGAGATGTACCCACGCTCTCAAGTGCGCGTGCAGTTTGCGGACTACCGTAGTGCCGCT 1500
GAGGAAATGTACCCGCGTAGCCAGGTTCGTGTTCAATTTGCGGACTATCGTAGCGCGGCG
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G  P  C  V  A  R  A  G  M  Q  K  L  W  D  G  E  D  F  Y  L    387
GGCCCGTGTGTGGCTCGCGCAGGAATGCAGAAGCTGTGGATGGGGAGGACTTCTACCTG 1560
GGTCCGTGCGTGGCGCGTGCGGGTATGCAGAAGTTGTGGACGGCGAGGATTTCTACTTG

Q  I  D  S  H  M  R  F  R  A  G  W  D  V  F  L  L  E  Q  L    407
CAAATCGACAGTCATATGCGATTCCGCGCAGGTTGGGACGTGTTTCTTCTCGAACAACTG 1620
CAAATTGATAGCCACATGCGTTTCCGTGCGGGCTGGGACGTGTTTTTGTTGGAACAGTTG

L  L  C  A  T  P  K  P  I  L  T  T  Y  P  L  G  Y  T  L  P    427
CTACTCTGCGCCACGCCAAAGCCGATCCTGACGACGTATCCGCTTGGGTACACACTGCCG 1680
TTGTTGTGCGCGACCCCGAAACCGATCTTGACCACCTACCCGTTGGGTTATACCTTGCCG

N  N  V  S  S  D  R  R  P  T  L  L  C  A  S  A  F  D  E  N    447
AACAATGTTTCATCCGACCGCCGCCCAACGCTATTATGCGCGTCAGCATTCGATGAGAAC 1740
AACAACGTTAGCAGCGATCGTCGTCCGACCTTGTTATGCGCGAGCGCGTTTGACGAGAAC

G  I  L  R  Q  A  S  K  T  L  A  T  I  S  P  V                463
GGGATACTCCGTCAAGCCAGTAAAACTCTCGCCACGATTTCGCCAGTgtacgtatctgtt 1800
GGCATTTTGCGTCAAGCGAGCAAGACCTTGGCGACCATCAGCCCGGT

gaaagtgcaacgctcttgctattgctgatggtgtgtgtgtgtttatgcattctctctcag 1860

 P  L  P  S  S  F  W  A  A  G  F  A  F  S  S  A  K  V  I  S   483
TCCTCTTCCTTCGAGCTTCTGGGCTGCCGGGTTTGCGTTTTCTTCGGCCAAAGTAATCAG 1920
TCCGTTGCCGAGCAGCTTTTGGGCGGCGGGTTTTGCGTTTAGCAGCGCGAAAGTGATCAG

 E  V  P  Y  D  E  S  L  R  F  L  F  F  G  E  E  A  S  M  S   503
CGAAGTCCCGTACGATGAGTCGTTGCGGTTTTTGTTTTTCGGTGAAGAAGCGTCGATGAG 1980
CGAGGTTCCGTACGATGAAAGCTTGCGTTTTTTGTTCTTTGGCGAGGAAGCGAGCATGAG

 A  R  L  W  T  S  G  W  D  F  F  T  P  G  E  S  I  I  Y  H   523
CGCTCGTCTGTGGACGTCGGGTTGGGATTTCTTTACGCCGGGCGAATCGATTATTTATCA 2040
CGCGCGTTTGTGGACCAGCGGTTGGGACTTCTTTACCCCGGGCGAAAGCATCATTTACCA

 L  W  T  R  S  Y  R  R  V  F  Q  E  I  E  D  N  D  T  A  V   543
CCTGTGGACACGCTCGTATCGCCGTGTGTTTCAGGAGATTGAAGACAACGACACCGCGGT 2100
CTTGTGGACCCGTAGCTATCGTCGTGTGTTCCAGGAGATTGAAGACAACGATACCGCGGT

 W  R  T  A  S  Q  R  Y  V  K  S  L  L  L  G  G  G  N  N  Q   563
GTGGAGAACAGCGTCGCAGCGGTATGTGAAGAGCTTACTTTTGGGTGGTGGCAACAATCA 2160
TTGGCGTACCGCGAGCCAGCGTTATGTTAAGAGCTTATTGTTGGGTGGCGGTAACAACCA

 E  P  T  G  S  I  A  A  G  T  Y  S  L  G  S  E  R  S  L  D   583
AGAGCCAACTGGCAGCATCGCTGCTGGCACTTACTCTCTTGGAAGCGAACGAAGCCTGGA 2220
GGAACCGACCGGCAGCATTGCGGCGGGTACCTACAGCTTGGGCAGCGAACGTAGCTTGGA

 A  Y  Q  Q  R  I  G  V  N  F  A  K  Q  E  I  Q  W  E  A  E   603
CGCCTATCAGCAGCGTATTGGAGTCAATTTTGCAAAGCAGGAGATTCAGTGGGAAGCAGA 2280
TGCGTATCAGCAACGTATTGGCGTTAACTTTGCGAAACAGGAGATCCAATGGGAGGCGGA

 W  G  N  L  D  P  I  Q  F  E  L  A  T  K  S  A  G  V  H  A   623 End of GlcNAcT-like
ATGGGGTAATCTGGACCCGATCCAATTTGAACTCGCCACCAAATCTGCTGGTGTTCATGC 2340
ATGGGGTAACTTGGACCCGATTCAGTTCGAGTTGGCGACCAAGAGCGCGGGTGTTCATGC

 P  A  *                                                      625
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gtctaggcagcttgcaatcactggaaat
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Fig. S2. *P. ultimum* Skp1A sequence. Black nucleotides are from genomic DNA ADOS01001474.1 at the NCBI wgs database; upper case nucleotides represent the predicted ORF, and lower case nucleotides are non-coding; underlined nt represent intron splice signals; purple nucleotides are EST EL774563.2 (NCBI EST database). Blue nucleotides are codon optimized for expression in *E. coli* and *D. discoideum*, with 5'- and 3'-restriction sites for cloning indicated. Amino acids are in bold black and derived from protein model PYU1_T013675 at www.pythiumdb.org.

```
                          tgtaggggttctctcgcagccagcaaccaataac -480
ttaccaaaggataacgaacccgtaagggagggggagggggcaaaagggggggtacaatt -420
actgttcctgattctgaacggactgacttttgggagggattcgtacttcgggacaaagag -360
atgacagaagaggaatataacgtgtttcctgatctggcacaatttgttttccaaaggcgg -300
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attaaaccaaagaaagcggtcttaagaggacaacccctgccttccctcgctccccttcct  -60

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                 CCAAAAGTCCGCCCACACACGACAGCAGCAGAGCCCCACGACC
                                                          CC   NcoI

M  A  D  T  A  P  A  P  D  A  P  R  Q  E  E  D  T  H  K  V     20
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ATGGCCGACACCGCCCCCGCACCGGACGCCCCTCGCCAGGAGGAGGACACGCACAAGGTA
ATGGCTGATACAGCTCCCGCACCAGATGCACCCAGGCAAGAAGAAGATACTCACAAGGTG

N  L  V  S  M  D  G  D  S  F  E  V  S  R  S  V  A  S  M  S     40
AACCTCGTGTCCATGGACGGCGACAGCTTCGAGGTCTCGCGCAGCGTCGCCTCCATGAGC  120
AACCTCGTGTCCATGGACGGCGACAGCTTCGAGGTCTCGCGCAGCGTCGCCTCCATGAGC
AACCTGGTTAGCATGGATGGTGACAGCTTCGAAGTGAGCCGTAGCGTTGCGAGCATGAGC

E  L  V  K  T  L  I  S  D                                        49
GAGCTCGTCAAGACCCTCATCTCCGgtacgtgcacgcgagagcgaataaagcggcgtttg  180
GAGCTCGTCAAGACCCTCATCTCCG
GAACTGGTTAAAACCCTGATCAGCG

cattattcgtgtgcacgtgtgtggatatgcgtgtatgcgcgcttatgacccgtgaacgtc  240

ctcattctacatagagcgcgtggatttgtggatatgaatgtgatgttagtgagcgcgtgc  300

                                          E  Q  D  D  D  E     55
attgtactaactgtgtttccgtctaattccctatgtgcagATGAACAGGACGACGATGAA  360
                                        ATGAACAGGACGACGATGAA
                                        ACGAGCAGGACGATGACGAA

V  Q  E  I  P  L  P  N  V  K  S  T  V  L  S  K  V  I  E  F     75
GTACAGGAGATCCCGTTGCCGAACGTCAAGAGCACGGTGCTCTCGAAGGTCATCGAGTTC  420
GTACAGGAGATCCCGTTGCCGAACGTCAAGAGCACGGTGCTCTCGAAGGTCATCGAGTTC
GTGCAAGAGATTCCGCTGCCGAACGTGAAGAGCACCGTTCTGAGCAAAGTGATCGAGTTT

C  S  H  H  H  N  N  P  M  R  E  I  E  K                        89
TGCAGCCACCACCACAACAACCCAATGCGTGAGATCGAAAAGgtatgttgactgactgat  480
TGCAGCCACCACCACAACAACCCAATGCGTGAGATCGAAAAG
TGCAGCCACCACCACAACAACCCGATGCGTGAAATTGAGAAG

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         P  L  K  S  S  D  M  H  D  V  V  S  D  W  D  A  N    106
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         CCATTGAAGAGCTCGGACATGCACGACGTGGTCTCGGACTGGGACGCCAAC
         CCGCTGAAAAGCAGCGATATGCACGACGTGGTTAGCGATTGGGACGCGAAC

F  V  D  I  E  Q  D  I  L  F  E  L  I  L  A  A  N  Y  M  D    126
TTTGTGGACATTGAGCAGGACATCCTCTTCGAGCTGATCCTGGCGGCCAACTACATGGAC  660
TTTGTGGACATTGAGCGGGACATCCTCTTCGAGCTGATCCTGGCGGCCAACTACATGGAC
TTCGTTGATATCGAACAAGACATTCTGTTTGAGCTGATCCTGGCGGCGAACTACATGGAT

I  K  S  L  L  D  L  A  C  A  K  V  A  S  M  I  K  G  K  T    146
ATCAAGAGCCTGCTCGATCTGGCGTGCGCGAAGGTCGCGAGCATGATCAAGGGCAAGACC  720
ATCAAGAGCCTGCTCGATCTGGCGTGCGCGAAGGTCGCGAGCATGATCAAGGGCAAGACC
ATTAAGAGCCTGCTGGACCTGGCGTGCGCGAAAGTTGCGAGCATGATCAAGGGCAAAACC

P  Q  E  I  R  E  T  F  N  I  V  N  D  F  T  P  E  E  E  S    166
CCGCAGGAGATCCGCGAGACTTTCAACATTGTGAACGACTTCACACCCGAGGAAGAGAGC  780
CCGCAGGAGATCCGCGAGACTTTCAACATTGTGAACGACTTCACACCCGAGGAAGAGAGC
CCGCAGGAAATCCGTGAGACCTTCAACATTGTGAACGATTTTACCCCGGAGGAAGAGAGC

Q  I  R  E  E  N  K  W  C  E  E  A  *                         178
CAAATCCGCGAGGAGAACAAGTGGTGCGAAGAAGCGtaggcggctagtcagtcaccgaat  840
CAAATCCGCGAGGAGAACAAGTGGTGCGAAGAAGCGTAGGCGGCTAGTCAGTCACCGAAC
CAAATTCGTGAAGAGAACAAGTGGTGCGAAGAGGCGTAAACTAGT                 SpeI


gcagaacggcgacgacccggtccttgggctcgattgccgttgtgcagcagtccgttcacc  900
GCAGAACGGCGACGACCCGGTCCTTGGGCTCGATTGCCGTTGTGCAGCAGTCCGTTCACC

atttgcgctctgcatattagtcagctgattgattgagggagagcagcgagcagtcagtgg  960
ATTTGCGCTCTGCATATTAGTCAGCTGATTGATTGAGGGAGAGCAGCGAGCAGTCAGTGG

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cgtgatctctctgggaggagcaagaacacaaactcaacaggacgggccgctggtataaac 1440
ggtcgctattgcagtcgcttgt                                       1462
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Fig. S3. *P. ultimum* Skp1B sequence. Black nucleotides are from genomic DNA ADOS01001682.1 at the NCBI wgs database; upper case nucleotides represent the predicted ORF, and lower case nucleotides are non-coding; purple nucleotides are ESTs from NCBI EST db (1, EL776046.2; 2, FE995571.1; 3, FF026581.1; 4, FF014550.1; 5, FF017573.1; 6, FF023366.1); blue nucleotides are codon optimized for expression in *E. coli* and *D. discoideum*, with 5'- and 3'-restriction sites for cloning indicated. Amino acids are in bold black and were from protein model PYU1_T011629 at www.pythiumdb.org with the C-terminal region adjusted based on genomic DNA data.

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                                              tccgcagctaacag  -60

cccaagaagcgcctccgccctcccaagaccctcttccgccttcctttctacctgcccagt    0
                                                          TC   BspHI

M  S  D  A  A  T  T  P  L  D  G  R  K  V  K  L  V  S  M  D     20
ATGAGCGACGCTGCTACCACCCCTCTCGACGGCCGCAAGGTCAAGCTGGTCTCCATGGAT   60
                                GGGGGGTTCTAAGCTGGTCTCCATGGAT 4
                                GGGGGGTTC-AAGCTGGTCTCCATGGAT 5
                  CCGGGGTTTACGGTCCGCAAGGTCAAGCTGGTCTCCATGGAT 6
ATGAGCGATGCGGCGACCACCCCGCTGGATGGTCGTAAGGTGAAACTGGTTAGCATGGAC

G  Q  T  Y  E  V  D  A  A  V  A  S  L  S  K  L  V  S  T  L     40
GGCCAGACCTACGAAGTGGACGCCGCTGTGGCCTCGCTCAGCAAGCTGGTGAGCACGCTG  120
GGCCAGACCTACGAAgtggacgccgctgtggcctcgctcagcaagctggtgagcacgctg 4
GGCCAGACCTACGAAgtggacgccgctgtggcctcgctcagcaagctggtgagcacgctg 5
GGCCAGACCTACGAAgtggacgccgctgtggcctcgctcagcaagctggtgagcacgctg 6
GGCCAGACCTACGAAGTGGATGCGGCGGTTGCGAGCCTGAGCAAGCTGGTGTCTACCCTG

V  S  D  N  A  D  E  D  E  V  Q  E  M  P  L  P  N  V  K  S     60
GTGTCGGACAACGCTGACGAGGACGAGGTGCAGGAAATGCCGTTGCCGAACGTCAAGAGC  180
                                        cgttgccgaacgtcaagagc 1
                                  aaatgccgttgccgaacgtcaagagc 2
gtgtcggacaacgctgacgaggacgaggtgcaggaaataccgttgccgaacgtcaagagc 4
gtgtcggacaacgctgacgaggacgaggtgcaggaaataccgttgccgaacgtcaagagc 5
gtgtcggacaacgctgacgaggacgaggtgcaggaaatgccgttgccgaacgtcaagagc 6
GTTAGCGACAACGCGGACGAAGATGAGGTGCAAGAGATGCCGCTGCCGAACGTGAAGAGC

H  V  L  E  K  V  V  A  F  C  E  H  H  H  K  H  P  M  K  E     80
CACGTGCTCGAGAAGGTGGTGGCGTTCTGCGAGCATCACCACAAGCACCCGATGAAGGAG  240
cacgtgctcgagaaggtggtggcgttctgcgagcatcaccacaagcacccgatgaaggag 1
cacgtgctcgagaaggtggtggcgttctgcgagcatcaccacaagcacccgatgaaggag 2
cacgtgctcgagaaggtggtggcgttctgcgagcatcaccacaagcacccgatgaaggag 4
cacgtgctcgagaaggtggtggcgttctgcgagcatcaccacaagcacccgatgaaggag 5
cacgtgctcgagaaggtggtggcgttctgcgagcatcaccacaag                6
CACGTTCTGGAAAAAGTGGTTGCGTTCTGCGAGCACCACCACAAGCACCCGATGAAAGAA

L  E  K  P  L  K  S  N  D  L  R  D  A  V  S  D  W  Y  A  D    100
CTAGAGAAGCCGCTCAAGAGCAACGATCTACGCGATGCGGTCTCGGACTGGTACGCGGAC  300
ctagagaagccgctcaagagcaacgatctacgcgatgcggtctcggactggtacgcggac 1
ctagagaagccgctcaagagcaacgatctacgcgatgcggtctcggactggtacgcggac 2
CTGGAGAAGCCGCTGAAAAGCAACGACCTGCGTGATGCTGTGAGCGACTGGTACGCGGAT

F  V  E  V  K  Q  E  D  Q  E  L  L  F  E  L  I  L  A  A  N    120
TTTGTGGAGGTGAAGCAGGAAGATCAGGAGCTCCTGTTTGAGCTGATCCTGGCCGCGAAC  360
tttgtggaggtgaagcaggaagatcaggagctcctgtttgagctgatcctggccgcgaac 1
tttgtggaggtgaagcaggaagatcaggagctcctgtttgagctgatcctggccgcgaac 2
                    agatcaggagctcctgtttgagctgatcctggccgcgaac 3
TTCGTGGAAGTTAAGCAGGAAGATCAAGAGCTGCTGTTTGAGCTGATCCTGGCGGCGAAC
```

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Y  L  D  I  K  N  L  L  D  L  A  C  A  K  V  A  T  M  I  K     140
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tacttagatatcaagaatctactcgatttggcgtgcgcaaaggttgcgacaatgatcaaa 1
tacttagatatcaagaatctac                                       2
tacttagatatcaagaatctactcgatttggcgtgcgcaaaggttgcgacaatgatcaaa 3
TATCTGGACATTAAGAACCTGCTGGATCTGGCGTGCGCGAAAGTTGCGACCATGATCAAG

G  K  T  P  E  E  I  R  A  T  F  G  I  T  E  E  F  T  P  E     160
GGCAAGACCCCTGAAGAGATCCGTGCGACGTTTGGCATTACTGAGGAGTTCACTCCCGAA  480
ggcaagacccctgaagagatccgtgcgacgtttggcattactgaggagttcactcccgaa 1
ggcaagacccctgaagagatccgtgcgacgtttggcattactgaggagttcactcccgaa 3
GGTAAAACCCCGGAGGAAATCCGTGCGACCTTCGGCATTACCGAGGAATTTACCCCGGAG

E  E  Q  R  I  R  E  E  N  K  W  C  E  D  I  *                175
GAAGAACAGCGCATTCGTGAAGAGAACAAGTGGTGCGAAGATATTtaagcgcgagttcac  540
gaagaacagcgcattcgtgaagagaacaagtggtgcgaagatatttaagcgcgagttcac 1
gaagaacagcgcattcgtgaagagaacaagtggtgcg                        3
GAAGAGCAGCGTATCCGTGAAGAGAACAAATGGTGCGAAGACATTTAAACTAGT        SpeI

gagcgaatggaacaggtaagttgcgtgtggatgtatttctgtatgtgcctttctctcatg  600
gagcgaatggaacaggtaagttgcgtgtggatgtatttctgtatgtgcctttctctcatg 1

cctactagaggtgctggattcagataatggtcatgaatgaaggctcagtttgtattgcac  660
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gagcgcgtgtttttatgccgtcaccatccccttgcgcgtttatgttgagcgacgaaacga  720
cagaccatgccgcgcgagagtgtggaagcaatggacagcgacaagcgatgcttttgtttt  780
ggcaggtcagctcgcttcaagcgcaagtgagcattgccccaattgtaccagcaacctcaa  840
cgagcagatactgagatgcaggctctagttcaatatgcatcaactgcagcctgccatgtt  900
acatacacgtgtgcatttgaagctccttgttattggcacggaggtc                960
```

Fig. S4. Predicted Skp1 sequences from select protists. The modified Pro residue is marked with an asterisk. Uncertain amino acids are replaced with a question mark. To facilitate visualization of relatedness, acidic residues are in blue, basic in dark red, Gly and Pro in red, and hydrophobic in green. Positions possessing a consensus chemical characteristic are highlighted, in yellow (hydrophobic), gray (acidic), dark grey (basic), or teal (small).

Dd Skp1A	*Dictyostelium discoideum*	amoebozoa/mycetozoa	NCBI, AAA67888.1
Tg Skp1	*Toxoplasma gondii*	alveolate/apicomplexa	ToxoDB, TGARI_207680
Ng Skp1A	*Naegleria gruberi*	excavate/percolozoa	DOE, version 1.0, 2/2009: in scaffold 5: 131842-132718
Ng Skp1B	*N. gruberi*		NCBI, XP_002681295.1
Tp Skp1A	*Thalassiosira pseudonana* 38460	stramenopile/diatom	NCBI, XP_002294707.1
Tp Skp1B	*T. pseudonana* 10077		NCBI, XP_002294049.1
Fc Skp1A	*Fragilariopsis cylindrus* 212070	stramenopile/ diatom	NCBI, OEU09899.1
Fc Skp1B	*F. cylindrus* 213790		NCBI, OEU16705.1
Ps Skp1A	*Phytophthora sojae* 108319	stramenopile/oomycete	NCBI, XP_009526241.1
Ps Skp1B	*P. sojae* 108223		NCBI, XP_009532045.1
Pr Skp1A	*Phytophthora ramorum* 71411	stramenopile/oomycete	DOE, 71411|fgenesh1_pm.C_scaffold_18000002, 6/2007
Pr Skp1B	*P. ramorum* 71622		DOE, 71622|fgenesh1_pm.C_scaffold_32000004, 6/2007
Pi Skp1A	*Phytophthora infestans* 01260.1	stramenopile/oomycete	NCBI, XP_002907933.1
Pi Skp1B	*P. infestans* 15216.1		NCBI, XP_002898672.1
Pu Skp1A	*Pythium ultimum*	stramenopile/oomycete	PYU1_T013675, www.pythiumdb.org
Pu Skp1B	*P. ultimum*		PYU1_T011629, www.pythiumdb.org, EST data NCBI
Es Skp1A	*Ectocarpus siliculosus*	heterokontophyta/brown alga	NCBI, CBJ48672.1
Es Skp1B	*E. siliculosus*		NCBI, CBJ48849.1
Gt Skp1A	*Guillardia theta CCMP2712*	cryptophyta/cryptomonad	NCBI, XP_005838866.1
Gt Skp1B	*G. theta CCMP2712*		NCBI, XP_005840806.1
Al Skp1	*Albugo laibachii*	stramenopile/oomycete	NCBI, CCA15417.1
Eh Skp1	*Emiliania huxleyi*	haptophyte/phytoplankton	NCBI, XP_005781634.1
Vc Skp1	*Volvox carteri*	cryptophyta/green alga	NCBI, XP_002947872.1
Cv Skp1	*Chlorella virus* PBCV-1	virus	NCBI, NP_048387.1
Sc Skp1	*Saccharomyces cerevisiae*	yeast	NCBI, NP_010615.3
Hs Skp1	Homo sapiens OCP2	human	NCBI, NP_008861.2

```
Dd Skp1A                                     MSLVKLESSDEKVFEIEKEIACMSVTIKNMIEDIGESDS--------------
Tg Skp1                              MSKERMGDARKVTLVSQEGDEFDVDIEVASMSALIKTMVEEDSDCQE--------------
Ng Skp1A
Ng Skp1B                                  MSETKQVELTSKDKVSFKVDRDVILMSGLVKDMLEEG--DEDETP-----------
Tp Skp1A                                         ?--SKEGDTFPVDIEVARMSELVKGMLEDDADDDEEAT-----------
Tp Skp1B                               MDGDDAHGTVKLVSKEGDTFEVPIEVAKLSNLVVTTLGEEDDYDDDDDNMV--------
Fc Skp1A                  MDVEDSKETAASTADTSSSGVTIQLISKEGDKFPVPITVSNMSELVKSMMDDKDDDDDDDDDEDDDGKEKIT
Fc Skp1B                                 MDEENGIVKLVSKEGDVHEVPINVAKMSSLILSTLDDDEEDDDDDEETKKHL-----
Ps Skp1A                    MSDAAAAPAQDVPKEEDSRKVNLVSMDGDSFEVSRSVAAMSELVKTLISDD-ADDDEVQ-----------
Ps Skp1B                             MAPQEPAADMKVKLVSMDGEAFEVDAKVAVMSQLVQTLVADEQEQGDEVQ-----------
Pr Skp1A                     MSDAAAPAQDAPAQEDLRKVNLVSMDGDSFEVSRSVAAMSELVKTLISDD-ADDDEVQ-----------
Pr Skp1B                                 MAPHETKVKLVSMDGEAFEVDASVAVMSQLVQTLVAD---EGDEVQ-----------
Pi Skp1A                    MSDAAAPAQDTPRQEEDSRKVNLVSMDGDSFEVSRGVAAMSELVKTLISDD-ADDDEVQ-----------
Pi Skp1B                                MAPSTKMKVKLVSMDGEAFEVDTSVAVMSELVKTLVADDQEEGGEVQ-----------
Pu Skp1A                     MADTAPAPDAPRQEEDTHKVNLVSMDGDSFEVSRSVASMSELVKTLISDE-QDDDEVQ-----------
Pu Skp1B                           MSDAATTPLDGRKVKLVSMDGQTYEVDAAVASLSKLVSTLVSDN-ADEDEVQ-----------
Es Skp1A                 MSALEEKSQAADAPADETEDTRSVHLVSQEGESFDIKVSVAKMSNLVKTMIDEDAEEDGDAQ-----------
Es Skp1B  MATESGSSSRVQKTTSSDGAVGSARTTRTDTGASSSGAVTLVSMDGDGFVVEASAIEVSKLLKAMVDGP--TENAAK-----------
Gt Skp1A                                 MSEADPEITLETYDKHQIKIPKSIATRSAIINMMIEDT-GDVNEVV-----------
Gt Skp1B                                 MSSSKRMVTLLSPDKQTFQVPWDVAMRSTVIKQMLEDL-PEPEEGAEDQTDPV----
Al Skp1                  MEEAATAGGMQPETAAPHEDGRKVNLVSMDGDSFEVSRSVASMSELVKTLIADG-TDDQEIQ-----------
Eh Skp1                                MADDSEATAVKLKSKQEEIFEVEKEVACRSVTVKNMVEDT---GLDT------------
Vc Skp1                                    MSGSKVKLMSSDTQMFEVDEEAAFQSQTVKNLVEDA---GTDD------------
Cv Skp1                                       MSIVFITSDFHRIEADKNVLDQSTVINDIVEDV---GDDE------------
Sc Skp1                                    MVTSNVVLVSGEGERFTVDKKIAERSLLLKNYLNDMHDSNLQNNSDSESDSDSET
Hs Skp1                                      MPSIKLQSSDGEIFEVDVEIAKQSVTIKTMLEDL-GMDDEGDDD---------
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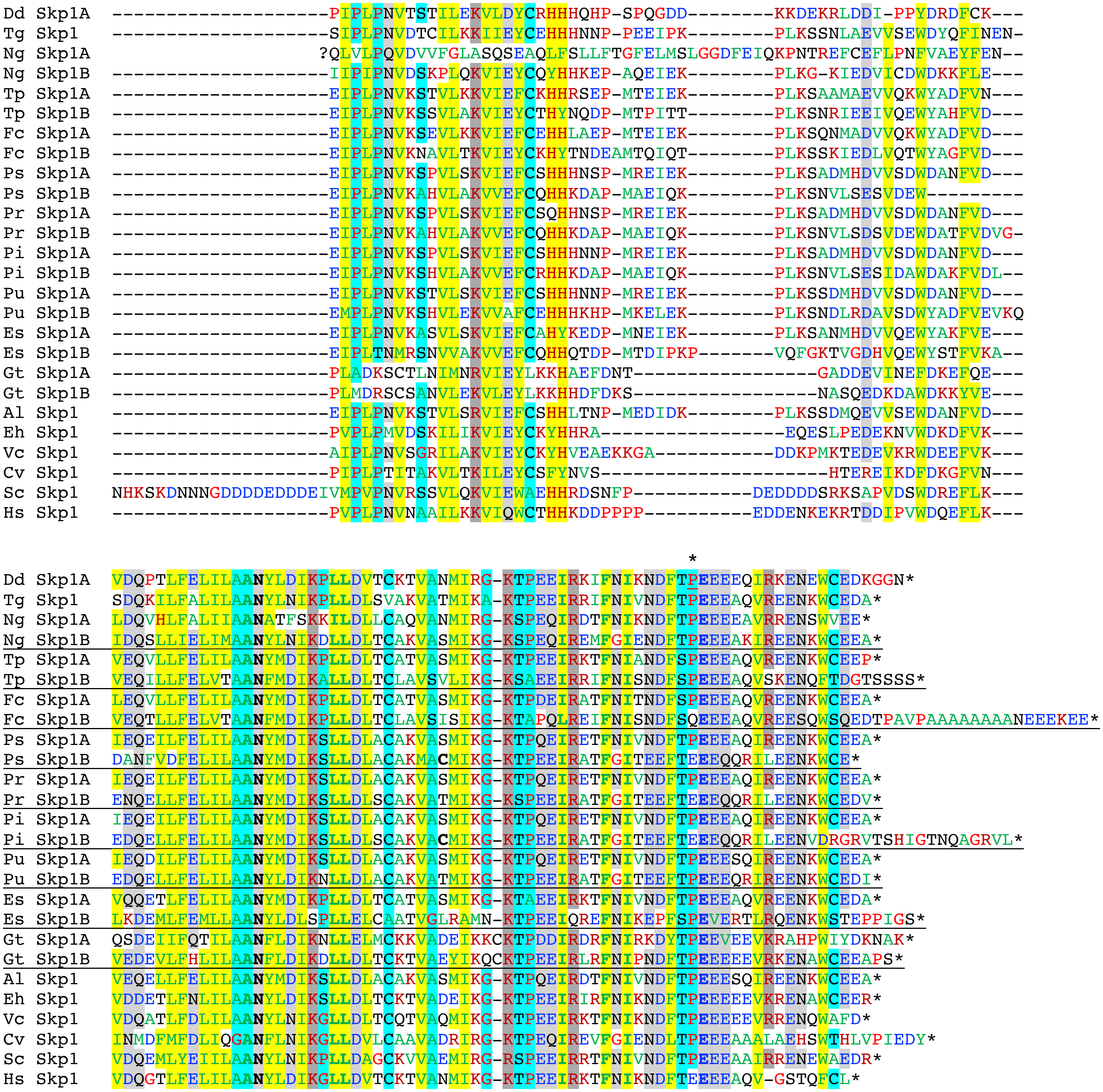

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Dd Skp1A    --------------------PIPLPNVTSTILEKVLDYCRHHHQHP-SPQGDD--------KKDEKRLDDI-PPYDRDFCK---
Tg Skp1     --------------------SIPLPNVDTCILKKIIEYCEHHHNNP-PEEIPK--------PLKSSNLAEVVSEWDYQFINEN-
Ng Skp1A                       ?QLVLPQVDVVFGLASQSEAQLFSLLFTGFELMSLGGDFEIQKPNTREFCEFLPNFVAEYFEN--
Ng Skp1B    --------------------IIPIPNVDSKPLQKVIEYCQYHHKEP-AQEIEK--------PLKG-KIEDVICDWDKKFLE---
Tp Skp1A    --------------------EIPLPNVKSTVLKKVIEFCKHHRSEP-MTEIEK--------PLKSAAMAEVVQKWYADFVN---
Tp Skp1B    --------------------EIPLPNVKSSVLAKVIEYCTHYNQDP-MTPITT--------PLKSNRIEEIVQEWYAHFVD---
Fc Skp1A    --------------------EIPLPNVKSEVLKKVIEFCEHHLAEP-MTEIEK--------PLKSQNMADVVQKWYADFVD---
Fc Skp1B    --------------------EIPLPNVKNAVLTKVIEYCKHYTNDEAMTQIQT--------PLKSSKIEDLVQTWYAGFVD---
Ps Skp1A    --------------------EIPLPNVKSPVLSKVIEFCSHHHNSP-MREIEK--------PLKSADMHDVVSDWDANFVD---
Ps Skp1B    --------------------EIPLPNVKAHVLAKVVEFCQHHKDAP-MAEIQK--------PLKSNVLSESVDEW---------
Pr Skp1A    --------------------EIPLPNVKSPVLSKVIEFCSQHHNSP-MREIEK--------PLKSADMHDVVSDWDANFVD---
Pr Skp1B    --------------------EIPLPNVKAHVLAKVVEFCQHHKDAP-MAEIQK--------PLKSNVLSDSVDEWDATFVDVG-
Pi Skp1A    --------------------EIPLPNVKSPVLSKVIEFCSHHHNNP-MREIEK--------PLKSADMHDVVSDWDANFVD---
Pi Skp1B    --------------------EIPLPNVKSHVLAKVVEFCRHHKDAP-MAEIQK--------PLKSNVLSESIDAWDAKFVDL--
Pu Skp1A    --------------------EIPLPNVKSTVLSKVIEFCSHHHNNP-MREIEK--------PLKSSDMHDVVSDWDANFVD---
Pu Skp1B    --------------------EMPLPNVKSHVLEKVVAFCEHHHKHP-MKELEK--------PLKSNDLRDAVSDWYADFVEVKQ
Es Skp1A    --------------------EIPLPNVKASVLSKVIEFCAHYKEDP-MNEIEK--------PLKSANMHDVVQEWYAKFVE---
Es Skp1B    --------------------EIPLTNMRSNVVAKVVEFCQHHQTDP-MTDIPKP-------VQFGKTVGDHVQEWYSTFVKA--
Gt Skp1A    --------------------PLADKSCTLNIMNRVIEYLKKHAEFDNT-----------------GADDEVINEFDKEFQE---
Gt Skp1B    --------------------PLMDRSCSANVLEKVLEYLKKHHDFDKS-----------------NASQEDKDAWDKKYVE---
Al Skp1     --------------------EIPLPNVKSTVLSRVIEFCSHHLTNP-MEDIDK--------PLKSSDMQEVVSEWDANFVD---
Eh Skp1     --------------------PVPLPMVDSKILIKVIEYCKYHHRA-----------------EQESLPEDEKNVWDKDFVK---
Vc Skp1     --------------------AIPLPNVSGRILAKVIEYCKYHVEAEKKGA-----------DDKPMKTEDEVKRWDEEFVK---
Cv Skp1     --------------------PIPLPTITAKVLTKILEYCSFYNVS---------------------HTEREIKDFDKGFVN---
Sc Skp1     NHKSKDNNNGDDDDEDDDEIVMPVPNVRSSVLQKVIEWAEHHRDSNFP-----------DEDDDDSRKSAPVDSWDREFLK---
Hs Skp1     --------------------PVPLPNVNAAILKKVIQWCTHHKDDPPPP----------EDDENKEKRTDDIPVWDQEFLK---

                                                                 *
Dd Skp1A    VDQPTLFELILAANYLDIKPLLDVTCKTVANMIRG-KTPEEIRKIFNIKNDFTPEEEEQIRKENEWCEDKGGN*
Tg Skp1     SDQKILFALILAANYLNIKPLLDLSVAKVATMIKA-KTPEEIRRIFNIVNDFTPEEEAQVREENKWCEDA*
Ng Skp1A    LDQVHLFALIIAANATFSKKILDLLCAQVANMIRG-KSPEQIRDTFNIKNDFTPEEEEAVRRENSWVEE*
Ng Skp1B    IDQSLLIELIMAANYLNIKDLLDLTCAKVASMIKG-KSPEQIREMFGIENDFTPEEEAKIREENKWCEEA*
Tp Skp1A    VEQVLLFELILAANYMDIKPLLDLTCATVASMIKG-KTPEEIRKTFNIANDFSPEEEAQVREENKWCEEP*
Tp Skp1B    VEQILLFELVTAANFMDIKALLDLTCLAVSVLIKG-KSAEEIRRIFNISNDFSPEEEAQVSKENQFTDGTSSSS*
Fc Skp1A    LEQVLLFELILAANYMDIKPLLDLTCATVASMIKG-KTPDEIRATFNITNDFSPEEEAQVREENKWCEEA*
Fc Skp1B    VEQTLLFELVTAANFMDIKPLLDLTCLAVSISIKG-KTAPQLREIFNISNDFSQEEEAQVREESQWSQEDTPAVPAAAAAAAANEEEKEE*
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Ps Skp1B    DANFVDFELILAANYMDIKSLLDLACAKMACMIKG-KTPEEIRATFGITEEFTEEEQQRILEENKWCE*
Pr Skp1A    IEQEILFELILAANYMDIKSLLDLACAKVASMIKG-KTPQEIRETFNIVNDFTPEEEAQIREENKWCEEA*
Pr Skp1B    ENQELLFELILAANYMDIKSLLDLSCAKVATMIKG-KSPEEIRATFGITEEFTEEEQQRILEENKWCEDV*
Pi Skp1A    IEQEILFELILAANYMDIKSLLDLACAKVASMIKG-KTPQEIRETFNIVNDFTPEEEAQIREENKWCEEA*
Pi Skp1B    EDQELLFELILAANYMDIKSLLDLSCAKVACMIKG-KTPEEIRATFGITEEFTEEEQQRILEENVDRGRVTSHIGTNQAGRVL*
Pu Skp1A    IEQDILFELILAANYMDIKSLLDLACAKVASMIKG-KTPQEIRETFNIVNDFTPEEESQIREENKWCEEA*
Pu Skp1B    EDQELLFELILAANYLDIKNLLDLACAKVATMIKG-KTPEEIRATFGITEEFTPEEEQRIREENKWCEDI*
Es Skp1A    VQQETLFELILAANYMDIKPLLDLTCATVASMIKG-KTAEEIRKTFNIVNDFTPEEEAQVREENKWCEDA*
Es Skp1B    LKDEMLFEMLLAANYLDLSPLLELCAATVGLRAMN-KTPEEIQREFNIKEPFSPEVERTLRQENKWSTEPPIGS*
Gt Skp1A    QSDEIIFQTILAANFLDIKNLLELMCKKVADEIKKCKTPDDIRDRFNIRKDYTPEEVEEVKRAHPWIYDKNAK*
Gt Skp1B    VEDEVLFHLILAANFLDIKDLLDLTCKTVAEYIKQCKTPEEIRLRFNIPNDFTPEEEEEVRKENAWCEEAPS*
Al Skp1     VEQELLFELILAANYMDIKSLLDLACAKVASMIKG-KTPQEIRDTFNIVNDFTPEEESQIREENKWCEEA*
Eh Skp1     VDDETLFNLILAANYLDIKSLLDLTCKTVADEIKG-KTPEEIRIRFNIKNDFTPEEEEEVKRENAWCEER*
Vc Skp1     VDQATLFDLILAANYLNIKGLLDLTCQTVAQMIKG-KTPEEIRKTFNIKNDFTPEEEEEVRRENQWAFD*
Cv Skp1     INMDFMFDLIQGANFLNIKGLLDVLCAAVADRIRG-KTPEQIREVFGIENDLTPEEEAAALAEHSWTHLVPIEDY*
Sc Skp1     VDQEMLYEIILAANYLNIKPLLDAGCKVVAEMIRG-RSPEEIRRTFNIVNDFTPEEEAAIRRENEWAEDR*
Hs Skp1     VDQGTLFELILAANYLDIKGLLDVTCKTVANMIKG-KTPEEIRKTFNIKNDFTEEEEAQV-GSTQFCL*
```

Fig. S5. Organisms with Phgt-like sequences in their genomes.

Green algae

Charophytes

Kn	*Klebsormidium nitens* (green algae)

Chlorophytes

Vc	*Volvox carteri* (chromalveolata – chlorophyta)
Chlorella	*Chlorella variabilis*
Coccomyxa	*Coccomyxa subellipsoidea*

Stramenopiles

Oomycetes

Pi	*Phytophtora infestans*
Ps	*Phytophthora sojae*
Pr	*Phytophtora ramorum* (heterokont)
Pu	*Pythium ultimum* (parasitic oomycete, stramenopile)(heterokont)
Pa	*Pythium aphanidermatum* (parasitic oomycete, stramenopile)
Al	*Albugo laibachii* Nc14, white rust oomycete
Spar	*Saprolegnia parasitica* (heterokontophyta; fish pathogen) (Heterokonta – Oomycete)
Ae	*Aphanomyces euteiches*, strain 109
Ai	*Aphanomyces invadans*

Diatoms

Tp	*Thalassiosira pseudonana* (DOE diatom db; jgi|Thaps3|4441|fgenesh1_pg.C_chr_4000391)
To	*Thalassiosira oceanica*
Fc	*Fragilariopsis cylindrus* (coldwater (arctic) diatom, stramenopile, from DOE)

Brown algae

Es	E*ctocarpus siliculosus* (brown algae, stramenopile) (Heterokonta – Phaeophyceae)

Alveolates

Ciliates

Sl	*Stylonychia lemnae*
Ot	*Oxytricha trifallax* (ciliated protozoan)

Dinoflagellate

Sm	*Symbiodinium microadriaticum* (Alveolata - Dinophyceae)

Cryptophytes

Gt	*Guillardia theta* CCMP2712 (GI:428176508) (Chromalveolata – cryptophyta)

Haptophytes

Eh	*Emiliania huxleyi* (see E.huxleyi doc in “worldwide”) (Chromalveolata – Haptophyta)

Excavates

Ng	*Naegleria gruberi* (amoeba-flagellate) (excavate – percolozoa)

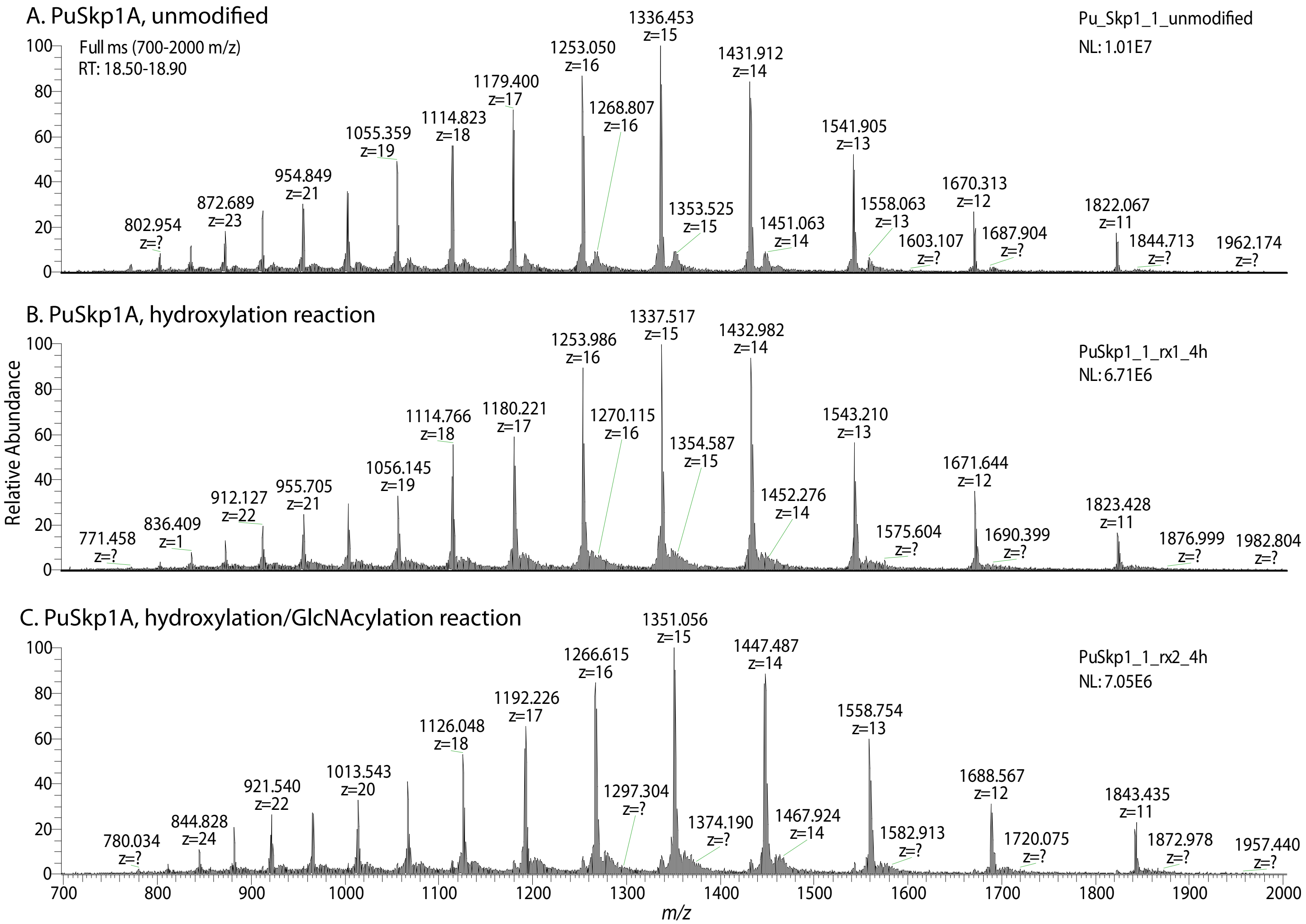
Fungi

Chytridiomycota

Rhizo	*Rhizoclosmatium globosum* (fungi)
Gona	*Gonapodya proliferia* (Fungi)
Spun	*Spizellomyces punctatus* DAOM BR117 (Fungi, Chytridiomycota, zoospore-producing soil fungi)

Fig. S6. Mass spectrometry of Skp1 modifications. Reactions were conducted at a PuSkp1A:PuPhgt ratio of 1:10 for 4 hrs. **(A)** Unreacted PuSkp1A. **(B)** The hydroxylation reaction included αKG and O_2. **(C)** The combined hydroxylation/GlcNAcylation reaction also included UDP-GlcNAc. Samples were analyzed by separation on a C4-column mounted on an Ultimate 3000 nLC in-line with a Q-Exactive+ Orbitrap. Data from these and other samples are summarized in Table II.

Figure S6



A. PuSkp1A, unmodified

B. PuSkp1A, hydroxylation reaction

C. PuSkp1A, hydroxylation/GlcNAcylation reaction