# Supplementary materials

**\*Note:** Full GmPIPs peptide sequences are deposited at the end of the present file.
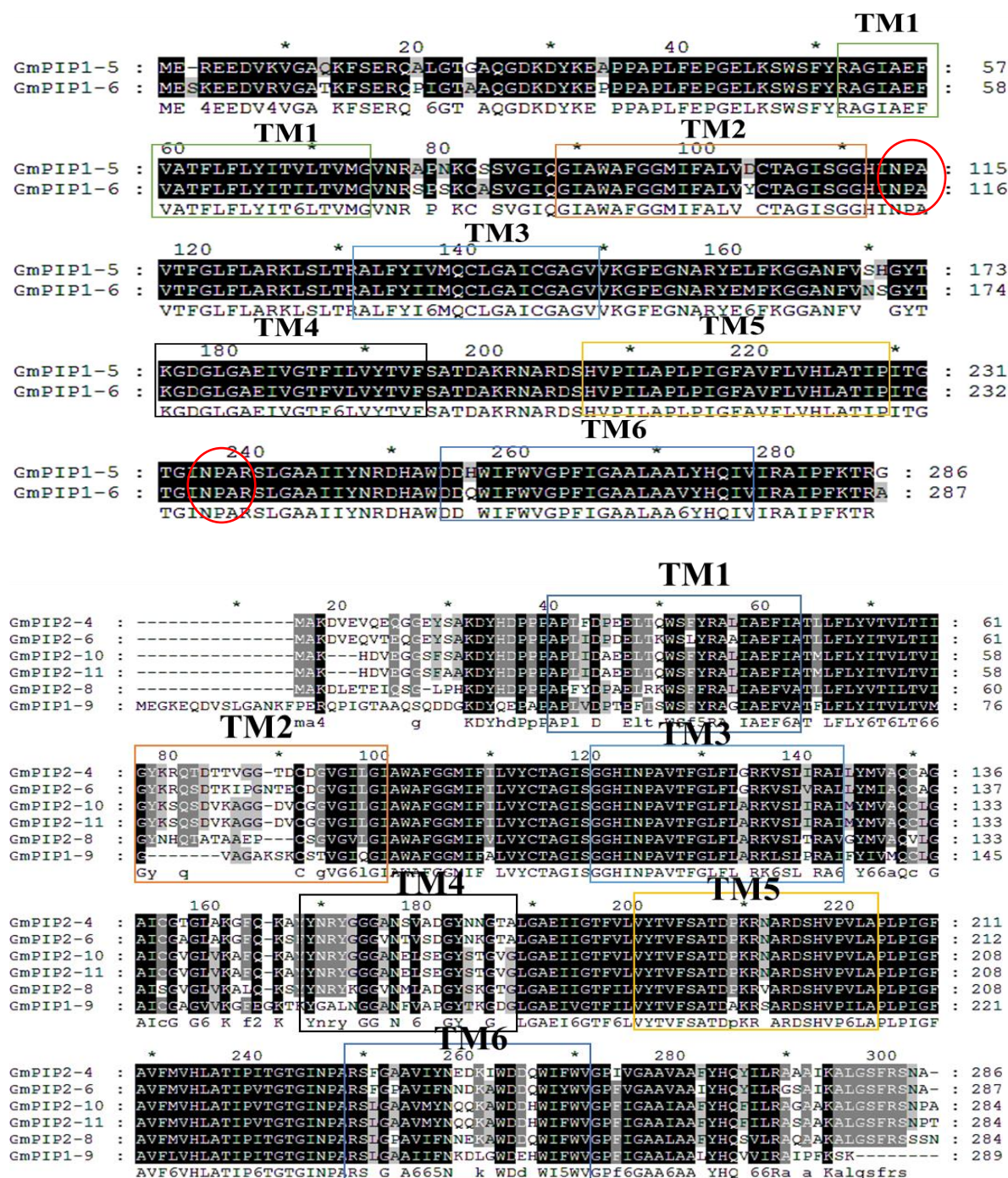
## Supplementary Table S1 The primers used for qRT-PCR.

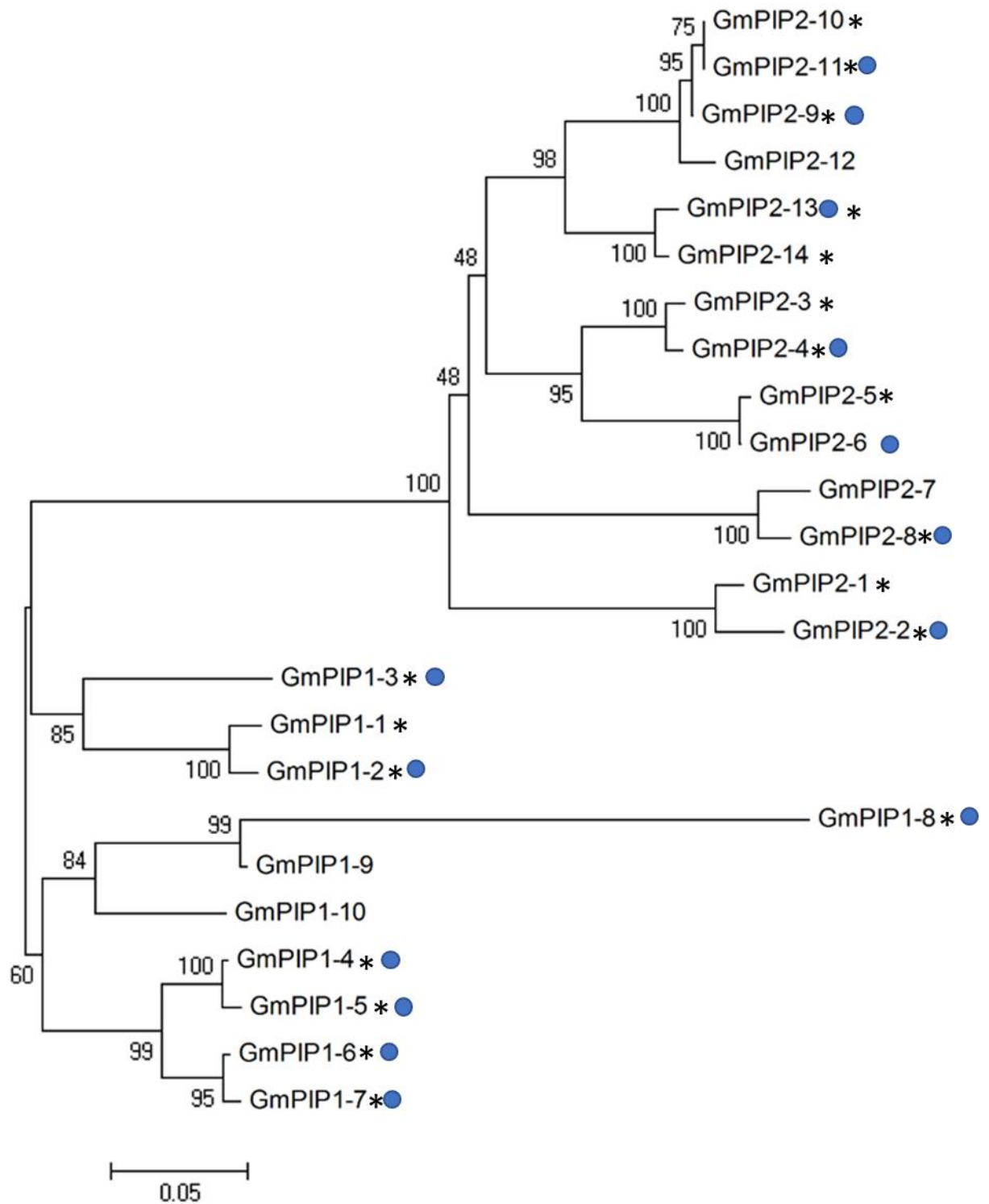| Gene | Forward primers | Reverse primers |
| --- | --- | --- |
| *GmPIP1;1* | GAGGGAAAAGAAGAGGAC | CTTCGACGAGAAGGGGATG |
| *GmPIP1;2* | GAGGGAAGAGATGAGGATG | ACTTGAAAGGAATGGCCCT |
| *GmPIP1;3* | GAGAGGGAGGAAGATGT | CCTTGTCTTGAAAGGAAT |
| *GmPIP1;4* | GAGAGGGAGGAAGATGT | CCTTGTCTTGAATGGAATTG |
| *GmPIP1;5* | GAGAGCAAAGAGGAAGATGT | CACAAAATTGGCTCCACCT |
| *GmPIP1;6* | GAGTAAAGAGGAAGAT | CCTTGTCTTGAAAGGAA |
| *GmPIP1;7* | GAGGGGAAGGAGCAGGAT | GGACTTGAAGGGAATGG |
| *GmPIP1;8* | GAGGGAAAAGAGGAAGAT | GGATTTGAAGGGAATGG |
| *GmPIP1;9* | GAGAGGGAAGAAAGAT | GGATTTGAAGTAATGAA |
| *GmPIP1;10* | GAGCAGGTATATGAATT | CGATTAACTGTCATCCAT |
| *GmPIP2;1* | TCGAAGGAAGTGAGCCAGCA | TGGTAGGGTTGCTCCTGA |
| *GmPIP2;2* | TCGAAGGAAGTGAGCCAAGA | GGTAGGGTTGCTCCTGA |
| *GmPIP2;3* | GCTAAAGATGTTGAGGT | CGTTGCTTCTGAAGGATCCA |
| *GmPIP2;4* | GCTAAAGATGTTGAGGT | GCGTTGCTCCTGAAGGATCCA |
| *GmPIP2;5* | GCCAAAGACGTTGAGCA | AGTGTTGCTCCTGAATGATC |
| *GmPIP2;6* | GCGAAAGACGTTGAGCAG | AGCGTTGCTCCTGAAGGATCCA |
| *GmPIP2;7* | GCGAAAGACATCGAAACT | CAGGTTTGAGGAGCTCCTGA |
| *GmPIP2;8* | GCCAAAGACCTCGAAACG | CAGGTTTGAGGAGCTCCTGA |
| *GmPIP2;9* | GCTAAGCATGATGTTGA | AATAGTGGGGTTGCTCCTGA |
| *GmPIP2;10* | GCTAAGCATGATGTTGAG | AATAGCGGGGTTGCTCCTGAAT |
| *GmPIP2;11* | TAAGCATGATGTTGAG | CGGGGTTGCTCCTGAAT |
| *GmPIP2:12* | GCCTAAGAATATCGAGC | ACTTCCCAGTAGATTCCATA |
| *GmPIP2;13* | AGCGTGTTTTGGCAGGA | TTATGGGGGTTACTCCTGAATGA |
| *GmPIP2;14* | GCGAAGGACGTTGAGGTT | ATTATGGGGGTTACTCCTGAA |
| *GmTUBB3* | GGAGGGTGAGTGAGCAGTTC | GCCGCATAACATTGTTCCCA |
| *GmActin* | CGGTGGTTCTATCTTGGCATC | GTCTTTCGCTTCAATAACCCTA |

**Supplementary Table S2 The primers used for yeast two-hybrid assays (AD and BD).**

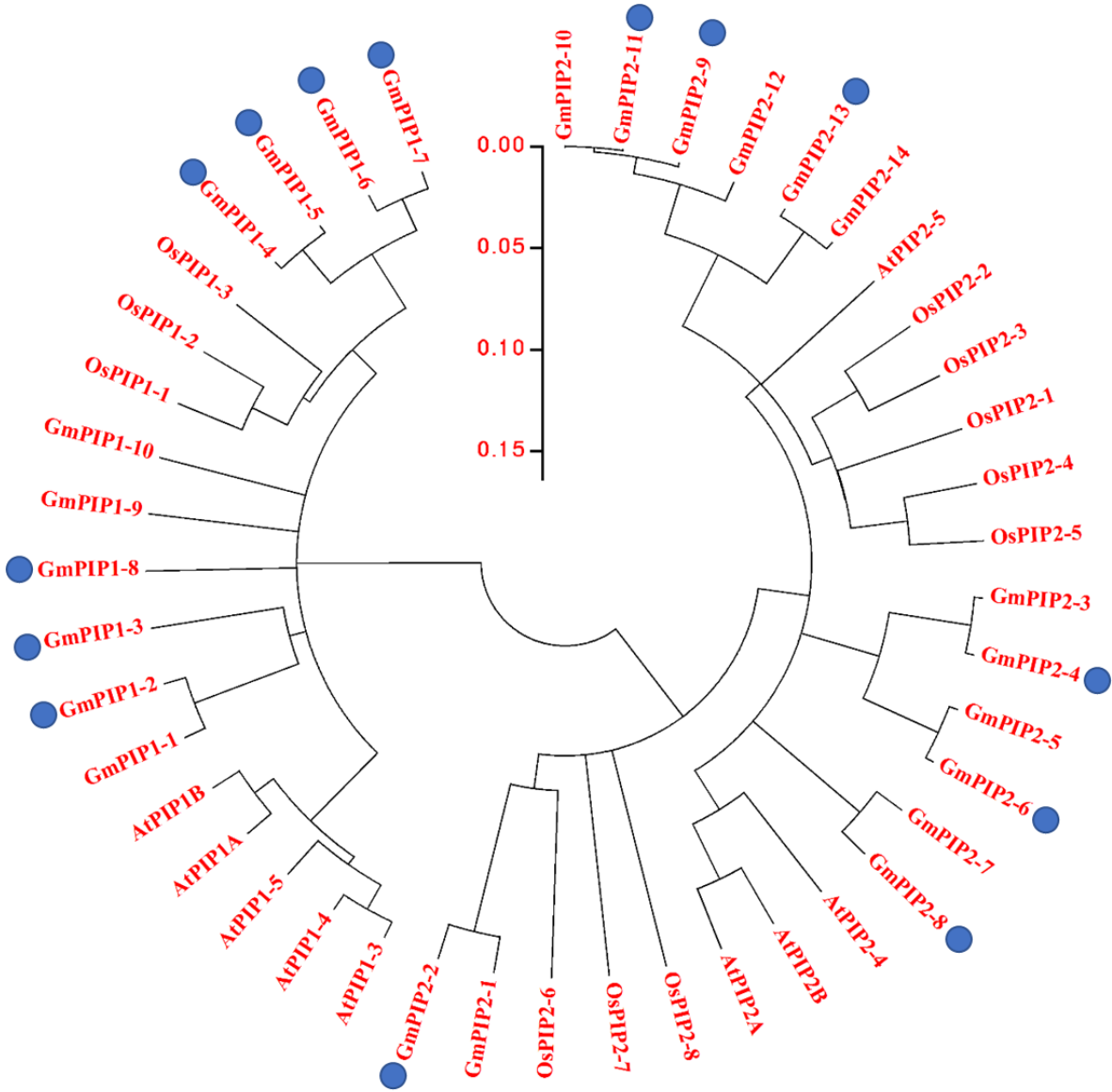| Gene | Forward primers | Reverse primers |
| --- | --- | --- |
| *GmPIP1;2* | cgCCATGGcaATGGAGGGAAGAGATGAGGATG | cGGAATTTCATTTTGACTTGAAAGGAATGGCCCT |
| *GmPIP1;3* | cgCCATGGcaATGGAGAGGGAGGAAGATGT | cGGAATTCTCAACCCCTTGTCTTGAAAGGAA |
| *GmPIP1;4* | cgCCATGGcaATGGAGAGGGAGGAAGATGT | cGGAATTCTCAACCCCTTGTCTTGAATGGAATT |
| *GmPIP1;5* | cgCCATGGcaATGGAGAGCAAAGAGGAAGATGT | cGGAATTCTCAAGCCCTTGTCTTGAAAGGGA |
| *GmPIP1;6* | cgCCATGGcaATGGAGAGTAAAGAGGAAGAT | cGGAATTCTCAAGCCCTTGTCTTGAAAGGA |
| *GmPIP1;7* | cgCCATGGcaATGGAGGGGAAGGAGCAGGAT | cGGAATTCTCACTTGGACTTGAAGGGAAT |
| *GmPIP1;8* | cgCCATGGcaATGGAGGGAAAAGAGGAAGAT | cGGAATTCTCAACTGGATTTGAAGGGAAT |
| *GmPIP2;2* | cgCCATGGcaATGTCGAAGGAAGTGAGCCAAGA | cGGAATTCTCAGTTGGTAGGGTTGCTCCTGA |
| *GmPIP2;4* | cgCCATGGcaATGGCTAAAGATGTTGAGGT | cGGAATTCTCAAGCGTTGCTCCTGAAGGATCCA |
| *GmPIP2;6* | cgCCATGGcaATGGCGAAAGACGTTGAGCAG | cGGAATTCTCAAGCGTTGCTCCTGAAGGATCCA |
| *GmPIP2;8* | cgCCATGGcaATGGCCAAAGACCTCGAAACG | cGGAATTCTCACAGGTTTGAGGAGCTCCTGA |
| *GmPIP2;9* | cgCCATGGcaATGGCTAAGCATGATGTTGA | cGGAATTCTCAAATAGTGGGGTTGCTCCTGA |
| *GmPIP2;11* | cgCCATGGcaATGGCTAAGCATGATGTTGAG | cGGAATTCTCAAATAGCGGGGTTGCTCCTGAAT |
| *GmPIP2;13* | cgCCATGGcaATGAGCGTGTTTTGGCAGGA | cGGAATTCTCAATTATGGGGGTTACTCCTGAATGA |

**Supplementary Figure S1:** Multiple alignment of soybean *Gm*PIP1 type and *Gm*PIP2 type aquaporins using Clustal X. GmPIP1s had longer N-terminal ends and GmPIP2s had longer C-terminal ends. The GmPIPs contained six transmembrane domains (rectangles) and two conserved NPA motifs (red circles). The part of the pictures demonstrated below was output directly by the software. In the software ';' was not allowed to appear in the name of a sequence, so we used '-' instead. For example, GmPIP1-5 means *Gm*PIP1;5. (It is the same in supplementary figure 2 & 3)

**Supplementary Figure S2:** The phylogenetic tree of GmPIPs. The aquaporins of which the gene expression levels showed significant variation after salt-stress were marked with asterisks; the dot in blue indicated the GmPIPs selected for yeast two-hybrid assays (In supplementary figure 3 is the same).

**Supplementary Figure S3:** The phylogenetic tree of GmPIPs, AtPIPs and OsPIPs.

**Full peptide sequences of GmPIPs**

>GmPIP1;1

MEGKEEDVRVGANRYGERQPIGTAAQAKDYREPPSAPLFEAGELSS
WSFYRAGIAEFVATFLFLYITVLTVMGVAKSKSKCSTVGIQGIAWAFGGMI
FALVYCTAGISGGHINPAVTFGLFLARKLSMTRAIFYIIMQCLGAICGAGV
VKGFEPHLYERLGGGANTIAKGYTNIAGLGAEIVGTFVLVYTVFSATDAK
RNARDSHVPILAPLPIGFAVFLVHLATIPVTGTGINPARSLGAAIIFNKDQA
WDDH

>GmPIP1;10

MEGKEEDVSLGANKFSERQPIGTAAQSQDDGKDYTEPPPAPLFEPSEL
TSWSFYRAGIAEFVATFLFLYITILTVMGVNRSSSKCATVGIQGIAWAFGG
MIFALVYCTAGISGGHINPAVTFGLFLARKLSLTRALFYMVMQVLGAIVG
AGVVKGFEGKTFYGQHNGGANFVAPGYTKGDGLGAEIVGTFILVYTVFS
ATDAKRSARDSHVPILAPLPIGFAVFLVHLATIPITGTGINPARSLGAAIIFN
KDLGWDDHWIFWVGPFVGAALAALYHQVVIRAIPFKSS*

>GmPIP1;2

MEGKEEDVRVGANRYGERQPIGTAAQAKDYREPPSAPLFEPGELSSW
SFYRAGIAEFVATFLFLYITVLTVMGVFKSKSKCSTVGIQGIAWAFGGMIFA
LVYSTAGISGGHINPAVTFGLFLARKLSLTRAIFYIIMQCLGAICGAGVVKG
FEPHLYERLGGGANTIAKGYTNSAGLGAEIVGTFVLVYTVFSATDAKRNA
RDSHVPILAPLPIGFAVFLVHLATIPVTGTGINPARSLGAAIIFNKDQAWDD
HWIFWVGPFIGAALAALYHQIVIRAIPFSSK*

>GmPIP1;3

LRLPHSHIHIEVKLALKMEGRDEDVRVGANRYGERQPIGTAAQTQDA
KDYREAPPAPLFEPRELTSWSFYRAGIAEFVATFLFLYVTVLTVMGVAKSP
SKCSTVGVQGIAWSFGGMIFALVYCTAGISGGHINPAVTFGLFLARKLSLT
RTVFYMIMQCLGAICGAAVVKGFQSNQYERLGGGANTLSKGYSKGDGL

GAEIVGTFILVYTVFSATDAKRNARDSHVPILAPLPIGFAVFLVHLATIPITG
TGINPARSLGAALVYNKDQAWDNHWIFWVGPFIGAALAALYHQIVLRAIP
FKSK*

>GmPIP1;4

MEREEDVKVGAQKFSERQALGTGAKSDKDYKEAPPAPLFEPGELKS
WSFYRAGIAEFVATFLFLYITVLTVMGVNRAPNKCSSVGIQGIAWAFGGMI
FALVYCTAGISGGHINPAVTFGLFLARKLSLTRAVFYIVMQCLGAICGAGV
VKGFEGNARYELFKGGANFVSHGYTKGDGLGAEIVGTFILVYTVFSATDA
KRNARDSHVPILAPLPIGFAVFLVHLATIPITGTGINPARSLGAAIIYNRDHA
WDDHWIFWVGPFIGAALAAVYHQIVIRAIPFKTRG*

>GmPIP1;5

MEREEDVKVGAQKFSERQALGTGAQGDKDYKEAPPAPLFEPGELKS
WSFYRAGIAEFVATFLFLYITVLTVMGVNRAPNKCSSVGIQGIAWAFGGMI
FALVDCTAGISGGHINPAVTFGLFLARKLSLTRALFYIVMQCLGAICGAGV
VKGFEGNARYELFKGGANFVSHGYTKGDGLGAEIVGTFILVYTVFSATDA
KRNARDSHVPILAPLPIGFAVFLVHLATIPITGTGINPARSLGAAIIYNRDHA
WDDHWIFWVGPFIGAALAALYHQIVIRAIPFKTRG*

>GmPIP1;6

MESKEEDVRVGATKFSERQPIGTAAQGDKDYKEPPPAPLFEPGELKS
WSFYRAGIAEFVATFLFLYITILTVMGVNRSPSKCASVGIQGIAWAFGGMIF
ALVYCTAGISGGHINPAVTFGLFLARKLSLTRALFYIIMQCLGAICGAGVV
KGFEGNARYEMFKGGANFVNSGYTKGDGLGAEIVGTFVLVYTVFSATDA
KRNARDSHVPILAPLPIGFAVFLVHLATIPITGTGINPARSLGAAIIYNRDHA
WDDQWIFWVGPFIGAALAAVYHQIVIRAIPFKTRA*

>GmPIP1;7

MESKEEDVNVGANKFSERQPIGTAAQGGGDKDYKEAPPAPLFEPGEL
KSWSFYRAGIAEFVATFLFLYITILTVMGVNRSPSKCASVGIQGIAWAFGG

MIFALVYCTAGISGGHINPAVTFGLFLARKLSLTRALFYIIMQCLGAICGAG

VVKGFEGNANYELFKGGANFVNSGYTKGDGLGAEIVGTFVLVYTVFSAT

DAKRNARDSHVPILAPLPIGFAVFLVHLATIPITGTGINPARSLGAAIIYNRD

HAWDDQWIFWVGPFIGAALAAVYHQIVIRAIPFKTRA*

>GmPIP1;8

MEGKEQDVSLGANKFPERQPIGTAAQSQDDGKDYQEPAPAPLVDPTE

FTSWSFYRAGIAEFVATFLFLYITVLTVMGVAGAKSKCSTVGIQGIAWAFG

GMIFALVYCTAGISGGHINPAVTFGLFLARKLSLPRAIFYIVMQCLGAICG

AGVVKGFEGKTKYGTLNGGANFVAPGYTKGDGLGAEIVGTFVLVYTVFS

ATDAKRNARDSHVPVSPPPSVENLFNCFFSLKKKYYFCGLNLIVWVEYMS

RFWHLCLLGSRCSWFTWQPSLSPALVSTLLVVLVLLSSSTRTLVGMITGSS

GWDLSLVLHLQHSTTRS*

>GmPIP1;9

MEGKEQDVSLGANKFPERQPIGTAAQSQDDGKDYQEPAPAPLVDPTE

FTSWSFYRAGIAEFVATFLFLYITVLTVMGVAGAKSKCSTVGIQGIAWAFG

GMIFALVYCTAGISGGHINPAVTFGLFLARKLSLPRAIFYIVMQCLGAICG

AGVVKGFEGKTKYGALNGGANFVAPGYTKGDGLGAEIVGTFILVYTVFS

ATDAKRSARDSHVPILAPLPIGFAVFLVHLATIPITGTGINPARSLGAAIIFN

KDLGWDEHWIFWVGPFIGAALAALYHQVVIRAIPFKSK*

>GmPIP2;1

MSKEVSQQRKDYVDPPPAPLIDLAEIKLWSFYRALIAEFIATLLFLYVT

VATVIGHKKQTGPCDGVGLLGIAWAFGGMIFVLVYCTAGISGGHINPAVTF

GLFLARKVSLIRALFYMVAQCLGAICGVGLVKAFMKHSYNSLGGGANSV

SAGYNKGSALGAEIIGTFVLVYTVFSATDPKRSARDSHIPVLAPLPIGFAVF

MVHLATIPITGTGINPARSFGAAVIYNNGKVWDDHWIFWVGPFVGALAA

AAYHQYILRAAAIKALGSFRSNPTN*

>GmPIP2;10

MAKHDVEGGSFSAKDYHDPPPAPLIDAEELTQWSFYRALIAEFIATML
FLYITVLTVIGYKSQSDVKAGGDVCGGVGILGIAWAFGGMIFILVYCTAGI
SGGHINPAVTFGLFLARKVSLIRAIMYMVAQCLGAICGVGLVKAFQKAYY
NRYGGGANELSEGYSTGVGLGAEIIGTFVLVYTVFSATDPKRNARDSHVP
VLAPLPIGFAVFMVHLATIPVTGTGINPARSLGAAVMYNQQKAWDDHWIF
WVGPFIGAAIAAFYHQFILRAGAAKALGSFRSNPAI*

>GmPIP2;11

MAKHDVEGGSFAAKDYHDPPPAPLIDAEELTQWSFYRALIAEFIATML
FLYITVLTVIGYKSQSDVKAGGDVCGGVGILGIAWAFGGMIFILVYCTAGI
SGGHINPAVTFGLFLARKVSLIRAIMYMVAQCLGAICGVGLVKAFQKAYY
NRYGGGANELSEGYSTGVGLGAEIIGTFVLVYTVFSATDPKRNARDSHVP
VLAPLPIGFAVFMVHLATIPVTGTGINPARSLGAAVMYNQQKAWDDHWIF
WVGPFIGAAIAAFYHQFILRASAAKALGSFRSNPTI*

>GmPIP2;12

MAKHDVEGGSFSAKDYHDPPPAPLIDAEELTQWSFYRALIAEFIATLL
FLYITVLTVIGYKSQSDVKAGGDVCGGVGILGIAWAFGGMIFILVYCTAGI
SGGHINPAVTFGLFLARKVSLIRAIMYMVAQCLGAMCGVGLVKAFQKAY
YNRYGGGANELSEGYSTGVGLGAEIIGTFVLVYTVFSATDPKRNARDSHV
PVLAPLPIGFAVFMVHLATIPVTGTGINPARSFGAAVMYNQKKAWDDQWI
FWVGPFIGAAIAAFYHQFILRASAAKAVGSFRSNPTI*

>GmPIP2;13

MSVFWQEGGMAKDVEVAERGSFSGKDYQDPPPAPLIDAEELTKWSF
YRALIAEFIATLLFLYITVLTVIGYNHQTDLKENGEICGGVGILGIAWAFGG
MIFILVYCTAGISGGHINPAVTFGLFLARKVSLIRAIMYMVAQCLGAICGV
GLVKAFQKSYFNKYGGGANSLAAGYSTGTGLGAEIIGTFVLVYTVFSATD
PKRNARDSHVPVLAPLPIGFAVFMVHLATIPVTGTGINPARSLGAAVIYNQ
DKPWDDHWIFWVGPFIGAAIAAFYHQFILRAGAAKALGSFRSNPHN*

**>GmPIP2;14**

MAKDVEVAERGSFSGKDYQDPPPAPLIDAEELTKWSFYRALIAEFIAT
LLFLYITVLTVIGYKHQTDHADACGGVGILGIAWAFGGMIFILVYCTAGIS
GGHINPAVTFGLFLARKVSLIRAIMYMVAQCLGAICGVGLVKAFQKSYFN
KYGGGANSLADGYSTGTGLGAEIIGTFVLVYTVFSATDPKRNARDSHVPV
LAPLPIGFAVFMVHLATIPVTGTGINPARSLGAAVIYNQDKPWDDHWIFW
VGPFIGAAIAAFYHQFILRAGAAKALGSFRSNPHN*

**>GmPIP2;2**

MSKEVSQEGLQRKDYVDPPPAPLFDLAEIKLWSFYRALIAEFIASLLFL
YVTVATIIGHKKQTGPCDGVGLLGIAWSFGGMIFVLVYCTAGISGGHINPA
VTFGLFLARKVSLIRAVFYMVAQCLGAICGVGLVKAFMKHSYNSLGGGA
NSVSAGYNKGSALGAEIIGTFVLVYTVFSATDPKRSARDSHVPVLAPLPIG
FAVFMVHLATIPITGTGINPARSLGAAVIYNNGKVWDEHWIFWVGPLVGA
LAAAAYHQYILRAGAIKALGSFRSNPTN*

**>GmPIP2;3**

MAKDVEVQEQGGEYSAKDYHDPPPAPLFDPEELTQWSFYRALIAEFI
ATLLFLYVTVLTIIGYKRQTDATLGGTECDGVGILGIAWAFGGMIFILVYC
TAGISGGHINPAVTFGLFLGRKVSLIRALLYMVAQCAGAICGTGLAKGFQ
KSYYNRYGGGANSVADGYNNGTALGAEIIGTFVLVYTVFSATDPKRNARD
SHVPVLAPLPIGFAVFMVHLATIPITGTGINPARSFGAAVIYNKDKIWDDQ
WIFWVGPIVGAAVAAFYHQYILRAAAIKALGSFRSNA*

**>GmPIP2;4**

MAKDVEVQEQGGEYSAKDYHDPPPAPLFDPEELTQWSFYRALIAEFI
ATLLFLYVTVLTIIGYKRQTDTTVGGTDCDGVGILGIAWAFGGMIFILVYC
TAGISGGHINPAVTFGLFLGRKVSLIRALLYMVAQCAGAICGTGLAKGFQ
KAYYNRYGGGANSVADGYNNGTALGAEIIGTFVLVYTVFSATDPKRNARD
SHVPVLAPLPIGFAVFMVHLATIPITGTGINPARSFGAAVIYNEDKIWDDQ

WIFWVGPIVGAAVAAFYHQYILRAAAIKALGSFRSNA*

>GmPIP2;5

MAKDVEQVTEQGEYSAKDYHDPPPAPLIDPDELTKWSLYRAAIAEFIA
TLLFLYITVLTIIGYKRQSDTKIPGNTECDGVGILGIAWAFGGMIFILVYCT
AGISGGHINPAVTFGLFLGRKVSLVRALLYMIAQCAGAICGAGLAKGFQK
SYYNRYGGGVNTVSDGYNKGTALGAEIIGTFVLVYTVFSATDPKRSARDS
HVPVLAPLPIGFAVFMVHLATIPVTGTGINPARSFGPAVIFNNDKAWDDQW
IYWVGPFVGAAVAAFYHQYILRAAAIKALGSFRSNT*

>GmPIP2;6

MAKDVEQVTEQGEYSAKDYHDPPPAPLIDPDELTKWSLYRAAIAEFIA
TLLFLYITVLTIIGYKRQSDTKIPGNTECDGVGILGIAWAFGGMIFILVYCT
AGISGGHINPAVTFGLFLGRKVSLVRALLYMIAQCAGAICGAGLAKGFQK
SFYNRYGGGVNTVSDGYNKGTALGAEIIGTFVLVYTVFSATDPKRNARDS
HVPVLAPLPIGFAVFMVHLATIPVTGTGINPARSFGPAVIFNNDKAWDDQW
IYWVGPFVGAAVAAIYHQYILRGSAIKALGSFRSNA*

>GmPIP2;7

MAKDIETEVQSGLPHKDYHDPPAAAFYDPAELRKWSFYRALIAEFVA
TLLFLYVTILTVIGYNHQTATGSPDLCNGVGVLGIAWAFGGMIFVLVYCTA
GISGGHINPAVTFGLFLARKVSLIRAVGYMVAQVLGAISGVGLVKALQKSY
YNRYNGGVNMLADGYSKGTGLGAEIIGTFILVYTVFSATDPKRVARDSHV
PVLAPLPIGFAVFIVHLATIPITGTGINPARSLGPAVIFNNEKAWDDQWIFW
VGPFIGAAIAAFYHQSVLRAQAAKALGSFRSSSNL*

>GmPIP2;8

MAKDLETEIQSGLPHKDYHDPPPAPFYDPAELRKWSFFRALIAEFVAT
LLFLYVTILTVIGYNHQTATAAEPCSGVGVLGIAWAFGGMIFVLVYCTAGI
SGGHINPAVTFGLFLARKVSLTRAVGYMVAQVLGAISGVGLVKALQKSYY
NRYKGGVNMLADGYSKGTGLGAEIIGTFILVYTVFSATDPKRVARDSHVP

VLAPLPIGFAVFMVHLATIPITGTGINPARSLGPAVIFNNEKAWDDQWIFW

VGPFIGAALAAFYHQSVLRAQAAKALGSFRSSSNL*

>GmPIP2;9

　　MAKHDVEGGSFSAKDYHDPPPAPLIDAEELTQWSFYRALIAEFIATLL

FLYITVLTVIGYKSQSDVKAGGDVCGGVGILGIAWAFGGMIFILVYCTAGI

SGGHINPAVTFGLFLARKVSLIRAIMYMVAQCLGAICGVGLVKAFQKAYY

NRYGGGANELSEGYSTGVGLGAEIIGTFVLVYTVFSATDPKRNARDSHVP

VLAPLPIGFAVFMVHLATIPVTGTGINPARSLGAAVMYNQQKAWDDHWIF

WVGPFIGAAIAAFYHQFILRAGAAKALGSFRSNPTI*