Isolated polynucleotides and polypeptides encoded thereby are described, together with the use of those products for making transgenic plants with increased tolerance to pH or increased phosphorus efficiency.
FIG. 2
PHOSPHATE USE EFFICIENCY

[0001] This application is a Continuation of co-pending application Ser. No. 11/140,347, filed on May 27, 2005, the entire contents of which are hereby incorporated by reference and for which priority is claimed under 35 U.S.C. § 120.


FIELD OF THE INVENTION

[0003] The present invention relates to isolated polynucleotides, polypeptides encoded thereby, and the use of those sequences for making transgenic plants with modulated pH response and phosphate use efficiency.

BACKGROUND OF THE INVENTION

[0004] Plants are constantly exposed to a variety of biotic (i.e., pathogen infection and insect herbivory) and abiotic (e.g., high pH, low phosphate) stresses. To survive these challenges, plants have developed elaborate mechanisms to perceive external signals and environmental stresses and to manifest adaptive responses with proper physiological and morphological changes (Bohnert et al., 1995). Plants exposed to low or high pH conditions typically have low yields of plant material, seeds, fruit and other edible products. Extreme soil pH conditions have a major influence on nutrient availability resulting in severe agronomic losses. Plants exposed to low pH soil conditions develop deficiencies in nutrients such as copper, molybdate, potassium, sulfur, and nitrogen. Also, plants exposed to high pH soil conditions develop iron, copper, manganese, and zinc deficiencies (FIG. 1). Phosphate deficiency is a problem in both high and low pH soil conditions. Essential mineral nutrients are required in substantial amounts to sustain plant growth and maximize plant yields. 

[0005] Consequently, agricultural and horticultural entities routinely alter the rhizosphere to maximize and maintain crop yields; these frequently result in more pollution and unbalancing of the natural soil mineral balance (National Research Council, 1989 Alternative Agriculture. National Academy Press, Washington D.C.). Excessive over-limiting of acid soils, for instance, has resulted in the induction of iron, manganese, copper, and zinc deficiencies; deficiencies commonly observed in calcareous soil.

[0006] It would, therefore, be of great interest and importance to be able to identify genes that confer improved phosphate efficiency characteristics to thereby enable one to create transformed plants (such as crop plants) with improved phosphate efficiency characteristics to thereby better survive low and high pH conditions. 

[0007] In the field of agriculture and forestry efforts are constantly being made to produce plants with an increased growth potential in order to feed the ever-increasing world population and to guarantee the supply of reproducible raw materials. This is done conventionally through plant breeding. The breeding process is, however, both time-consuming and labor-intensive. Furthermore, appropriate breeding programs must be performed for each relevant plant species.

[0008] Progress has been made in part by the genetic manipulation of plants; that is by introducing and expressing recombinant nucleic acid molecules in plants. Such approaches have the advantage of not usually being limited to one plant species, but instead being transferable among plant species. (Zhang et al. (2004) Plant Physiol. 135:615). There is a need for generally applicable processes that improve forest or agricultural plant growth potential. Therefore, the present invention relates to a process for increasing the abiotic stress tolerance and consequently the growth potential in plants, characterized by expression of recombinant DNA molecules stably integrated into the plant genome.

SUMMARY OF THE INVENTION

[0009] The present invention, therefore, relates to isolated polynucleotides, polypeptides encoded thereby, and the use of those sequences for making transgenic plants with modulated pH tolerance or phosphate use efficiency.

[0010] The present invention also relates to processes for increasing the growth potential in plants under abnormal pH or phosphate conditions, recombinant nucleic acid molecules and polypeptides used for these processes and their uses, as well as to plants themselves.

[0011] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs.

BRIEF DESCRIPTION OF THE FIGURES

[0012] FIG. 1 shows the relationship between soil pH and nutrient uptake.

[0013] FIG. 2 shows pH recovery as measured by volume of seeds collected from a plant containing cDNA 1248777 compared to pH treated and un-treated controls.

DETAILED DESCRIPTION OF THE INVENTION

1. Definitions

[0014] The following terms are utilized throughout this application:

[0015] Constitutive Promoter: Promoters referred to herein as "constitutive promoters" actively promote transcription under most, but not necessarily all, environmental conditions and states of development or cell differentiation. Examples of constitutive promoters include the cauliflower mosaic virus (CaMV) 3S5 transcript initiation region and the 1' or 2' promoter derived from T-DNA of Agrobacterium tumefaciens; and other transcription initiation regions from various plant genes, such as the maize ubiquitin-1 promoter, known to those of skill.

[0016] Domain: Domains are fingerprints or signatures that can be used to characterize protein families and/or parts of proteins. Such fingerprints or signatures can comprise conserved (1) primary sequence, (2) secondary structure, and/or (3) three-dimensional conformation. Generally, each domain has been associated with either a family of proteins or motifs. Typically, these families and/or motifs have been correlated with specific in-vitro and/or in-vivo activities. A domain can be any length, including the entirety of the sequence of a protein. Detailed descriptions of the domains, associated families and motifs, and correlated activities of the polypeptides of the instant invention are described below. Usually, the polypeptides with designated domain(s) can exhibit at least one activity that is exhibited by any polypeptide that comprises the same domain(s).
Endogenous: The term "endogenous," within the context of the current invention refers to any polynucleotide, polypeptide or protein sequence which is a natural part of a cell or organisms regenerated from said cell.

Exogenous: “Exogenous,” as referred to within, is any polynucleotide, polypeptide or protein sequence, whether chimeric or not, that is initially or subsequently introduced into the genome of an individual host cell or the organism regenerated from said host cell by any means other than by a sexual cross. Examples of means by which this can be accomplished are described below, and include Agrobacterium-mediated transformation (of dicots—e.g. Salomon et al. EMBO J. 3:141 (1984); Herrera-Estrella et al. EMBO J. 2:987 (1983); of monocots, representative papers are those by Escudero et al., Plant J. 10:355 (1996), Ishida et al., Nature Biotechnology 14:745 (1996), May et al., Bio/Technology 13:486 (1995), biolistic methods (Armaille et al., Current Genetics 17:97 1990)), electroporation, in planta techniques, and the like. Such a plant containing the exogenous nucleic acid is referred to here as a T0, for the primary transgenic plant and T1, for the first generation. The term “exogenous” as used herein is also intended to encompass inserting a naturally found element into a non-naturally found location.

Functionally Comparable Proteins: This phrase describes those proteins that have at least one characteristic in common. Such characteristics include sequence similarity, biochemical activity, transcriptional pattern similarity and phenotypic activity. Typically, the functionally comparable proteins share some sequence similarity or at least one biochemical and within this definition, homologs, orthologs and analogs are considered to be functionally comparable. In addition, functionally comparable proteins generally share at least one biochemical and/or phenotypic activity.

Functionally comparable proteins will give rise to the same characteristic to a similar, but not necessarily to the same degree. Typically, comparable proteins give the same characteristics where the quantitative measurement due to one of the comparables is at least 20% of the other; more typically, between 30 to 40%; even more typically, between 50-60%; even more typically, 70 to 80%; even more typically between 90 to 100%.

Heterologous sequences: “Heterologous coding sequences” are those that are not operatively linked or are not contiguous to each other in nature. For example, a promoter from corn is considered heterologous to an Arabidopsis coding region sequence. Also, a promoter from a gene encoding a growth factor from corn is considered heterologous to a sequence encoding the corn receptor for the growth factor. Regulatory element sequences, such as UTRs or 3’ end termination sequences that do not originate in nature from the same gene as the coding sequence originates from, are considered heterologous to said coding sequence. Elements operatively linked in nature and contiguous to each other are not heterologous to each other. On the other hand, these same elements remain operatively linked but become heterologous if other filler sequence is placed between them. Thus, the promoter and coding sequences of a corn gene expressing an amino acid transporter are not heterologous to each other, but the promoter and coding sequence of a corn gene operatively linked in a novel manner are heterologous.

High pH: “High pH” can be defined as a non-optimal and terminal alkaline pH value when a given plant can no longer make use of certain essential nutrients, such as phosphate, available in the soil. For instance, if a plant grows optimally at pH of 4.0-5.0, high pH would be any pH greater than 5. If the optimal pH were in the range of 6-6.5, high pH would be a pH greater than pH 6.5. As an example, if a corn crop under optimal pH conditions would yield 134 bushels per acre and all other conditions were held constant, a high pH tolerant variety would produce similar yields at pH 9 or above.

Inducible Promoter: An “inducible promoter” in the context of the current invention refers to a promoter which is regulated under certain conditions, such as light, chemical concentration, protein concentration, conditions in an organism, cell, or organelle, etc. A typical example of an inducible promoter, which can be utilized with the polynucleotides of the present invention, is PARSK1, the promoter from the Arabidopsis gene encoding a serine-threonine kinase enzyme, and which promoter is induced by dehydration, abscisic acid and sodium chloride (Wang and Goodman, Plant J 17:317 1995)). Examples of environmental conditions that may affect transcription by inducible promoters include anaerobic conditions, elevated temperature, or the presence of light.

Low Nitrogen: “Low nitrogen” can be defined as a quantity of nitrogen, whether in the form of ammonium or nitrate, which is insufficient to sustain normal growth and yield for a given plant. The need for nitrogen fertilizers varies considerably among plants. Further, the type of soil and the conditions in the soil have a significant impact on the ability of a plant to take up nitrogen. Supplemental nitrogen fertilizers are often added to soil or applied directly to plants to enhance their growth or appearance. Even with normal fertilizer applications, the amount of nitrogen available to a plant at any given time may be too low to support optimal growth. Hence, low nitrogen must be defined in terms of the specific plant and environment in which the plant is being grown. For example, if under a given set of conditions with a specific corn hybrid the optimal nitrogen level was 160 pounds of nitrogen fertilizer per acre and under such conditions the hybrid were able to achieve a yield of 134 bushels per acre, a low nitrogen tolerant hybrid would grow optimally and produce the same yield with at least 10% less or at least 20% less or at least 30% less or at least 40% less or at least 50% less nitrogen. Further, the low nitrogen hybrid would grow better after much of the initial nitrogen had been depleted and would not require multiple applications of nitrogen.

Low pH: “Low pH” can be defined as that non-optimal and terminal acidic pH value when a given plant can no longer make use of certain essential nutrients, such as potassium, available in the soil. If a plant grows optimally at pH of 4.0-5.0, low pH is any pH less than 4. If the optimal pH is in the range of 6-8, low pH would be a pH less than 6. For example, if a corn crop under optimal pH conditions would yield 134 bushels per acre and all other conditions were held constant, a low pH tolerant variety would produce similar yields at pH 5 or pH 4.

Low Phosphate: “Low phosphate” can be defined as a quantity of phosphate which is insufficient to sustain normal growth and yield for a given plant. The level of phosphate required for optimal plant growth differs among plant species and depends on the condition of the soil and
other environmental conditions. To determine a level of phosphate that is low, comparative experiments are needed. For example, if a corn hybrid in a particular field treated with 40 pounds of phosphate per acre would yield 134 bushels per acre and all other conditions were held constant, a low phosphate tolerant hybrid would produce similar yields at 35 or less pounds of phosphate per acre or 30 or less pounds of phosphate per acre or 25 or less pounds of phosphate per acre or 20 or less pounds of phosphate per acre.

[0027] Masterpool: The “master pools” discussed in these experiments are a pool of seeds from five different transgenic plants transformed with the same exogenous gene.

[0028] Misexpression: The term “misexpression” refers to an increase or a decrease in the transcription of a coding region into a complementary RNA sequence as compared to the wild-type. This term also encompasses expression of a gene or coding region for a different time period as compared to the wild-type and/or from a non-natural location within the plant genome.

[0029] Percentage of sequence identity: “Percentage of sequence identity,” as used herein, is determined by comparing two optimally aligned sequences over a comparison window, where the fragment of the polynucleotide or amino acid sequence in the comparison window may comprise additions or deletions (e.g., gaps or overhangs) as compared to the reference sequence (which does not comprise additions or deletions) for optimal alignment of the two sequences. The percentage is calculated by determining the number of positions at which the identical nucleic acid base or amino acid residue occurs in both sequences to yield the number of matched positions, dividing the number of matched positions by the total number of positions in the window of comparison and multiplying the result by 100 to yield the percentage of sequence identity. Optimal alignment of sequences for comparison may be conducted by the local homology algorithm of Smith and Waterman Add. APL Math. 2:482 (1981), by the homology alignment algorithm of Needleman and Wunsch J. Mol. Biol. 48:443 (1970), by the search for similarity method of Pearson and Lipman Proc. Natl. Acad. Sci. (USA) 85: 2444 (1988), by computerized implementations of these algorithms (GAP, BESTFIT, BLAST, PASTA, and TFASTA) in the Wisconsin Genetics Software Package, Genetics Computer Group (GCC), 575 Science Dr., Madison, Wis.), or by inspection. Given that two sequences have been identified for comparison, GAP and BESTFIT are preferably employed to determine their optimal alignment. Typically, the default values of 5.00 for gap weight and 0.30 for gap weight length are used. The term “substantial sequence identity” between polynucleotide or polypeptide sequences refers to polynucleotide or polypeptide comprising a sequence that has at least 80% sequence identity, preferably at least 85%, more preferably at least 90% and most preferably at least 95%, even more preferably, at least 96%, 97%, 98% or 99% sequence identity compared to a reference sequence using the programs.

[0030] Query nucleic acid and amino acid sequences were searched against subject nucleic acid or amino acid sequences residing in public or proprietary databases. Such searches were done using the Washington University Basic Local Alignment Search Tool Version 1.83 (WU-Blast2) program. The WU-Blast2 program is available on the internet from Washington University, A WU-Blast2 service for Arabidopsis sis can also be found on the internet. Typically the following parameters of WU-Blast2 were used: Filter options were set to “default,” Output format was set to “gapped alignments,” the Comparison Matrix was set to “BLOSUM62,” Cutoff Score (S value) was set to “default,” the Expect (E threshold) was set to “default,” the Number of best alignments to show was set to “100,” and the “Sort output” option was set to sort the output by “p-value.”

[0031] Plant Promoter: A “plant promoter” is a promoter capable of initiating transcription in plant cells and can drive or facilitate transcription of a nucleotide sequence or fragment thereof of the instant invention. Such promoters need not be of plant origin. For example, promoters derived from plant viruses, such as the CaMV 35S promoter or from Agrobacterium tumefaciens such as the T-DNA promoters, can be plant promoters. A typical example of a plant promoter of plant origin is the maize ubiquitin-1 (ubi-1) promoter known to those of skill.

[0032] Specific Promoter: In the context of the current invention, “specific promoters” refers to promoters that have a high preference for being active in a specific tissue or cell and/or at a specific time during development of an organism. By “high preference” is meant at least 3-fold, preferably 5-fold, more preferably at least 10-fold and even more preferably at least 20-fold, 50-fold or 100-fold increase in transcription in the desired tissue over the transcription in any other tissue. Typical examples of temporal and/or tissue specific promoters of plant origin that can be used with the polynucleotides of the present invention, are: SH-EP from Vigna unguiculata (Yamauchi et al. (1996) Plant Mol Biol. 30(2): 321-9); RcC2 and Rcc3, promoters that direct root-specific gene transcription in rice (Xu et al., Plant Mol. Biol. 27:237 (1995) and TohRBB2, a root-specific promoter from tobacco (Yamamoto et al., Plant Cell 3:371 (1991)).

[0033] Stringency: “Stringency” as used herein is a function of probe length, probe composition (G+C content), and salt concentration, organic solvent concentration, and temperature of hybridization or wash conditions. Stringency is typically compared by the parameter $T_m$, which is the temperature at which 50% of the complementary molecules in the hybridization are hybridized, in terms of a temperature differential from $T_m$. High stringency conditions are those providing a condition of $T_m=5^\circ$ C to $T_m=10^\circ$ C. Medium or moderate stringency conditions are those providing $T_m=20^\circ$ C to $T_m=29^\circ$ C. Low stringency conditions are those providing a condition of $T_m=40^\circ$ C to $T_m=48^\circ$ C. The relationship of hybridization conditions to $T_m$ ($\circ$ C) is expressed in the mathematical equation

$$T_m=81.5-16.6 \log ([Na^+]^1/[G+C]\%)(4.14+S)$$

(1)

where N is the length of the probe. This equation works well for probes 14 to 70 nucleotides in length that are identical to the target sequence. The equation below for $T_m$ of DNA-DNA hybrids is useful for probes in the range of 50 to greater than 500 nucleotides, and for conditions that include an organic solvent (formamide).

$$T_m=81.5+16.6 \log ([Na^+]^1/[G+C\%])(4.14+S)$$

(2)

where L is the length of the probe in the hybrid. (P. Tiessen, “Hybridization with Nucleic Acid Probes” in Laboratory Techniques in Biochemistry and Molecular Biology, P. C. van der Vliet, ed., c. 1993 by Elsevier, Amsterdam.) The $T_m$ of equation (2) is affected by the nature of the hybrid; for
DNA-RNA hybrids $T_m$ is 10-15°C higher than calculated, for RNA-RNA hybrids $T_m$ is 20-25°C higher. Because the $T_m$ decreases about 1°C for each 1% decrease in homology when a long probe is used (Bonner et al., J. Mol. Biol. 81:123 (1973)), stringency conditions can be adjusted to favor detection of identical genes or related family members.

Equation (2) is derived assuming equilibrium and therefore, hybridizations according to the present invention are most preferably performed under conditions of probe excess and for sufficient time to achieve equilibrium. The time required to reach equilibrium can be shortened by inclusion of a hybridization accelerator such as dextran sulfate or another high volume polymer in the hybridization buffer.

Stringency can be controlled during the hybridization reaction or after hybridization has occurred by altering the salt and temperature conditions of the wash solutions used. The formulas shown above are equally valid when used to compute the stringency of a wash solution. Preferred wash solution stringencies lie within the ranges stated above; high stringency is 5-8°C below $T_m$, medium or moderate stringency is 26-29°C below $T_m$, and low stringency is 45-48°C below $T_m$.

Superpool: As used in the context of the current invention, a “superpool” refers to a mixture of seed from 100 different “master pools.” Thus, the superpool contains an equal amount of seed from 500 different events, but only represents 100 transgenic plants with a distinct exogenous nucleotide sequence transformed into them, because the master pools are of 5 different events with the same exogenous nucleotide sequence transformed into them.

$T_c$: As used in the current application, the term “$T_c$” refers to the whole plant, explant, or callous tissue inoculated with the transformation medium.

$T_i$: As used in the current application, the term $T_i$ refers to the either the progeny of the $T_i$ plant, in the case of whole-plant transformation, or the regenerated seedling in the case of explant or callous tissue transformation.

$T_2$: As used in the current application, the term $T_2$ refers to the progeny of the $T_1$ plant. $T_2$ progeny are the result of self-fertilization or cross pollination of a $T_1$ plant.

$T_3$: As used in the current application, the term $T_3$ refers to second generation progeny of the plant that is the direct result of a transformation experiment. $T_3$ progeny are the result of self-fertilization or cross pollination of a $T_2$ plant.

Zero Nitrogen: Nitrogen is not present in any amount.

Zero Phosphorus: Phosphorus is not present in any amount.

2. Important Characteristics of the Polynucleotides and Polypeptides of the Invention

The polynucleotides and polypeptides of the present invention are of interest because they are misexpressed (i.e. when expressed at a non-natural location or in an increased or decreased amount) they produce plants with modified pH tolerance or phosphate use efficiency. “Phosphate use efficiency” is a term that includes various responses to environmental conditions that affect the amount of phosphate available to the plant. For example, under both low and high pH conditions phosphate is bound within the soil, resulting in a decrease of available phosphate for maintaining or initiating physiological processes. As used herein, modulating phosphate use efficiency is intended to encompass all of these situations as well as other environmental situations that affect the plant’s ability to use and/or maintain phosphate effectively (e.g. osmotic stress, etc.).

The polynucleotides and polypeptides of the invention, as discussed above and as evidenced by the results of various experiments, are useful for modulating pH tolerance or phosphate use efficiency. These traits can be used to exploit or maximize plant products for agricultural, ornamental or forestry purposes in different environment conditions of water supply. Modulating the expression of the nucleotides and polypeptides of the present invention leads to transgenic plants that will be less sensitive to variations in pH and that require less phosphate, resulting in better yields under these types of adverse conditions. Both categories of transgenic plants lead to reduced costs for the farmer and better yield in their respective environmental conditions.

3. The Polynucleotides and Polypeptides of the Invention

The polynucleotides of the invention, and the proteins expressed thereby, are set forth in the sequences present in the Sequence Listing. Some of these sequences are functionally comparable proteins.

Functionally comparable proteins are those proteins that have at least one characteristic in common. Such characteristics can include sequence similarity, biochemical activity and phenotypic activity. Typically, the functionally comparable proteins share some sequence similarity and generally share at least one biochemical and/or phenotypic activity. For example, biochemical functionally comparable proteins are those proteins that act on the same reactant to give the same product.

Another class of functionally comparable proteins is phenotypic functionally comparable proteins. The members of this class regulate the same physical characteristic, such as increased drought tolerance. Proteins can be considered phenotypic functionally comparable proteins even if the proteins give rise to the same physical characteristic, but to a different degree.

The polypeptides of the invention also include those comprising the consensus sequences described in Tables 1-5, 2-6 and 3-5. A consensus sequence defines the important conserved amino acids and/or domains within a polypeptide. Thus, all those sequences that conform to the consensus sequence are suitable for the same purpose. Polypeptides comprised of a sequence within and defined by one of the consensus sequences can be utilized for the purposes of the invention namely to make transgenic plants with improved tolerance to heat or high or low water conditions.

4. Use of the Polynucleotides and Polypeptides to Make Transgenic Plants

To use the sequences of the present invention or a combination of them or parts and/or mutants and/or fusions and/or variants of them, recombinant DNA constructs are prepared which comprise the polynucleotide sequences of the invention inserted into a vector, and which are suitable for transformation of plant cells. The construct can be made using standard recombinant DNA techniques (Sambrook et al. 1989) and can be introduced to the species of interest by Agrobacterium-mediated transformation or by other means of transformation as referenced below.
The vector backbone can be any of those typical in the art such as plasmids, viruses, artificial chromosomes, BACs, YACs and PACs and vectors of the sort described by


(b) YAC: Burke et al., Science 236:806-812 (1987);

(c) PAC: Sternberg N. et al., Proc Natl Acad Sci USA. January; 87(1):103-7 (1990);

(d) Bacteria- Yeast Shuttle Vectors: Bradshaw et al., Nucl Acids Res 23: 4850-4856 (1995);


(g) Plasmid vectors: Sambrook et al., infra.

Typically, the construct comprises a vector containing a sequence of the present invention with any desired transcriptional and/or translational regulatory sequences, such as promoters, UTRs, and/or termination sequences. Vectors can also include origins of replication, scaffold attachment regions (SARs), markers, homologous sequences, introns, etc. The vector may also comprise a marker gene that confers a selectable phenotype on plant cells. The marker typically encodes biocide resistance, particularly antibiotic resistance, such as resistance to kanamycin, bleomycin, hygromycin, or herbicide resistance, such as resistance to glyphosate, chlorosulfuron or phosphinotricin.

A plant promoter is used that directs transcription of the gene in all tissues of a regenerant plant, which may be a constitutive promoter, such as p326 or CaMV35S. Alternatively, the plant promoter directs transcription of a sequence of the invention in a specific tissue manner (tissue-specific promoter) or is otherwise under more precise environmental control (inducible promoter). Various plant promoters, including constitutive, tissue-specific and inducible, are known to those skilled in the art and can be utilized in the present invention. Typically, preferred promoters to use in the present invention are those that are induced by heat or low water conditions. Such as the RD29a promoter (Kasuga et al., Plant Cell Physiol. 45:346 (2004) and Yamasaki-Shinozaki and Shinozaki, Mol Gen Genet 236: 331 (1993)) or other DRE-containing (dehydration-responsive elements) promoters (Liu et al, Cell 10: 1391 (1998)). Another preferred embodiment of the present invention is the use of root specific promoters such as those present in the AxTH19, AxTH118, and AxTH200 genes of Arabidopsis Thaliana (Vissenberg et al. (2005) Plant Cell Physiol 46:192) or guard cell specific promoters such as TGG1 or KST1 (Huseby et al. (2002) Plant Physiol 128:1180; Plesch et al. (2001) Planta 237:845). Alternatively, misexpression can be accomplished using a two component system, whereby the first component comprises a transgenic plant comprising a transcriptional activator operatively linked to a promoter and the second component comprises a transgenic plant comprising a sequence of the invention operatively linked to the target binding sequence/region of the transcriptional activator. The two transgenic plants are crossed and the sequence of the invention is expressed in their progeny. In another alternative, the misexpression can be accomplished by transforming the sequences of the two component system into one transgenic plant line.

Any promoter that functions in plants can be used in the first component, such as those discussed above. Suitable transcriptional activator polypeptides include, but are not limited to, those encoding HAP1 and GAL4. The binding sequence recognized and targeted by the selected transcriptional activator protein (e.g. a UAS element) is used in the second component.

Transformation

Nucleotide sequences of the invention are introduced into the genome or the cell of the appropriate host plant by a variety of techniques. These techniques for transforming a wide variety of plant species are well known and described in the technical and scientific literature. See, e.g., Weising et al., Ann. Rev. Gene. 22:421 (1988); and Christon, Euphytica, v. 85, n.1:3-13:27, (1995).

Processes for the transformation and regeneration of monocotyledonous and dicotyledonous plants are known to the person skilled in the art. For the introduction of DNA into a plant host cell a variety of techniques is available. These techniques include transformation of plant cells by injection (e.g. Newell, 2000), microinjection (e.g. Griessbach (1987) Plant Sci. 50 69-77), electroporation of DNA (e.g. Fromm et al. (1985) Proc Natl Acad Sci. USA 82:5824 and War and Lemaux, Plant Physiol. 104 (1994), 37-48, PEFG (e.g. Paszkowski et al. (1984) EMBO J. 3:2717), use of biolistics (e.g. Klein et al. (1987) Nature 327:773), fusion of cells or protoplasts (Willmitzer, L., 1993 Transgenic plants. In: Biotechnology, A Multi-Volume Comprehensive Treatise (H. R. Rehm, G. Reed, A. Phil. P. P. Stadler, eds.), Vol. 2, 627-659, VCH Weinheim-New York-Basel-Cambridge), using T-DNA using Agrobacterium tumefaciens (e.g. Finley et al. (1985) Crit. Rev. Plant Sci. 4: 1-46 and Fromm et al., Biotechnology 8 (1990), 833-844) or Agrobacterium rhizogenes (e.g. Cho et al. (2000) Planta 210:195-204) or other bacterial hosts (e.g. Brouggerts et al. (2005) Nature 433:629-633), as well as further possibilities.

In addition, a number of non-stable transformation methods well known to those skilled in the art may be desirable for the present invention. Such methods include, but are not limited to, transient expression (e.g. Lincoln et al. (1998) Plant Mol. Biol. Rep. 16:14) and viral transfection (e.g. Lacomme et al. (2001) In “Genetically Engineered Viruses” (C. A. Ring and E. D. Blair, Eds). Pp. 59-99, BIOS Scientific Publishers, Ltd. Oxford, UK).

Seeds are obtained from the transformed plants and used for testing stability and inheritance. Generally, two or more generations are cultivated to ensure that the phenotypic feature is stably maintained and transmitted.

One of skill will recognize that after the expression cassette is stably incorporated in transgenic plants and confirmed to be operable, it can be introduced into other plants by sexual crossing. Any of a number of standard breeding techniques can be used, depending upon the species to be crossed.

The nucleic acids of the invention can be used to confer the trait of increased tolerance to heat and/or low water conditions, without reduction in fertility, on essentially any plant.

The nucleotide sequences according to the invention encode appropriate proteins from any organism, in particular from plants, fungi, bacteria or animals.

The process according to the invention can be applied to any plant, preferably higher plants, pertaining to the classes of Angiospermae and Gymnospermae. Plants of the subclasses of the Dicotyledoneae and the Monocotyledoneae are particularly suitable. Dicotyledonous plants belong to the orders of the Magnoliidae, Illiciflorae, Laurales, Piperales Aristhochiales, Nymphaeales, Ranunculales, Papaverales, Saxifragaceae, Trochodendrales, Hamamelidales, Eucomiales, Lecythidales, Myricales, Fagales, Casuarinales, Caryo-

0069 The method of the invention is preferably used with plants that are interesting for agriculture, horticulture, biomass for bioconversion and/or forestry. Examples are tobacco, oilseed rape, sugar beet, potato, tomato, cucumber, pepper, bean, pea, citrus fruit, apple, pear, berries, plum, melon, eggplant, cotton, soybean, sunflower, rose, poinsettia, petunia, guayule, cabbage, spinach, alfalfa, artichoke, corn, wheat, rye, barley, grasses such as switch grass or turf grass, millet, hemp, barbary, poplar, eucalyptus trees, cotyledons.

Homologs Encompassed by the Invention

0070 Agents of the invention include proteins comprising at least about a contiguous 10 amino acid region preferably comprising at least about a contiguous 20 amino acid region, even more preferably comprising at least about a contiguous 25, 35, 50, 75 or 100 amino acid region of a protein of the present invention. In another preferred embodiment, the proteins of the present invention include between about 10 and about 25 contiguous amino acid region, more preferably between about 20 and about 50 contiguous amino acid region, and even more preferably between about 40 and about 80 contiguous amino acid region.

0071 Due to the degeneracy of the genetic code, different nucleotide codons may be used to code for a particular amino acid. A host cell often displays a preferred pattern of codon usage. Nucleic acid sequences are preferably constructed to utilize the codon usage pattern of the particular host cell. This generally enhances the expression of the nucleic acid sequence in a transformed host cell. Any of the above described nucleic acid and amino acid sequences may be modified to reflect the preferred codon usage of a host cell or organism in which they are contained. Modification of a nucleic acid sequence for optimal codon usage in plants is described in U.S. Pat. No. 5,689,052. Additional variations in the nucleic acid sequences may encode proteins having equivalent or superior characteristics when compared to the proteins from which they are engineered.

0072 It is understood that certain amino acids may be substituted for other amino acids in a protein or peptide structure (and the nucleic acid sequence that codes for it) without appreciable change or loss of its biological utility or activity. The amino acid changes may be achieved by changing the codons of the nucleic acid sequence.

0073 It is well known in the art that one or more amino acids in a native sequence can be substituted with other amino acid(s), the charge and polarity of which are similar to that of the native amino acid, i.e., a conservative amino acid substitution, resulting in a silent change. Conservative substitutes for an amino acid within the native polypeptide sequence can be selected from other members of the class to which the amino acid belongs (see below). Amino acids can be divided into the following four groups: (1) acidic (negatively charged) amino acids, such as aspartic acid and glutamic acid; (2) basic (positively charged) amino acids, such as arginine, histidine, and lysine; (3) neutral polar amino acids, such as glycine, serine, threonine, cysteine, cystine, tyrosine, asparagine, and glutamine; and (4) neutral nonpolar (hydrophobic) amino acids such as alanine, leucine, isoleucine, valine, proline, phenylalanine, tryptophan, and methionine.

0074 In a further aspect of the present invention, nucleic acid molecules of the present invention can comprise sequences that differ from those encoding a protein or fragment thereof selected from the group consisting of those sequences present in the Sequence Listing due to the fact that the different nucleic acid sequence encodes a protein having one or more conservative amino acid changes.

0075 In another aspect, biologically functional equivalents of the proteins or fragments thereof of the present invention can have about 10 or fewer conservative amino acid changes, more preferably about 7 or fewer conservative amino acid changes, and most preferably about 5 or fewer conservative amino acid changes. In a preferred embodiment, the protein has between about 5 and about 500 conservative changes, more preferably between about 10 and about 300 conservative changes, even more preferably between about 25 and about 150 conservative changes, and most preferably between about 5 and about 25 conservative changes or between 1 and about 5 conservative changes.

5. Experiments Confirming the Usefulness of the Polynucleotides and Polypeptides of the Invention

0076 5.1 Procedures

0077 The nucleotide sequences of the invention were identified by use of a variety of screens for pH and/or low phosphate and/or low nitrogen conditions. These screens are recognized by those skilled in the art to be predictive of nucleotide sequences that provide plants with improved tolerance to pH and/or low phosphate and/or low nitrogen conditions because they emulate the different environmental conditions that can result from increased pH and/or low phosphate and/or low nitrogen conditions. These screens generally fall into two categories (1) soil screens and (2) in vitro screens.

0078 Soil screens have the advantage of assaying the response of the entire plant to particular conditions, such as high pH or low phosphorus. On the other hand, in vitro screens have the advantage of relying on defined media and so allow more defined manipulation of growth conditions. Each of the screens used is described in more detail below.

0079 In general, the screens used to identify the polynucleotides and polypeptides of the invention were conducted using superpools of Arabidopsis T<sub>2</sub> transformed plants. The T<sub>1</sub> plants were transformed with a Ti plasmid containing a particular SEQ ID NO in the sense orientation relative to a constitutive promoter and harboring the plant-selectable marker gene phosphinotricin acetyltransferase (PAT), which confers herbicide resistance to transformed plants. For in vitro screens, seed from multiple superpools (1,200 T<sub>2</sub> seeds from each superpool) were usually tested. T<sub>2</sub> seed were collected from the resistant plants and retested on one or more in vitro screens. The results of the screens conducted for each SEQ ID NO can be found in the Examples below.

0080 1. High pH

0081 Screens for high pH resistance identify seedlings better able to thrive under nutritional deficiencies (e.g. Phosphate, Manganese, Iron, Boron) imposed by alkaline conditions.
Seeds are sterilized in 50% household bleach for 5 minutes and then washed with double distilled deionized water three times. Sterilized seed is stored in the dark at 4°C for a minimum of 3 days before use.

High pH media is prepared by mixing 0.5 g/l MES hydrate with 1X MS+0.5% Sucrose. Prior to autoclaving, pH is adjusted with 10 N KNH to the following values: pH 5.7 (control), pH 7.03, pH 8.02, pH 9.01 and pH 10.18. The media pH is retested since pH values drop after autoclaving as follows: pH 5.7 → pH 5.66; pH 7.03 → pH 6.50; pH 8.02 → pH 7.50; pH 9.01 → pH 8.91; pH 10.18 → pH 9.91. Generally speaking, pH 9.01 (pH 8.91) allows germination but no growth beyond 2 to 5 mm and no root growth. Germination does not occur at higher pH (e.g. pH 10.81).

Approximately 1200 seeds are evenly spaced per MS-sucrose plate before incubating in the vertical position at 22°C for 14 days. Under these conditions, the plates are exposed to 12,030 LUX from above and 3,190 LUX from the bottom.

Seedlings are scored for root and shoot growth after 7 and 14 days. Putative tolerant seedlings are transferred to MS pH 5.7 for recovery for 14 days prior to transplanting in soil. Finale™ spraying is done after plants are moved to soil to remove non-transgenics from the population.

DNA is isolated from each T2 plant and used in PCR reactions using the following cycling conditions: 95°C for 5 min, 35 cycles of (94°C for 30 sec, then 59°C for 30 sec, then 72°C for 1 min), 72°C for 8 min and 4°C hold. Aliquots of the reaction product are analyzed on a 1.0% agarose gel stained with ethidium bromide. The DNA products are sequenced to determine which insert sequences were in each superpool candidate chosen in the screen.

T3 Seed from those plants containing sequenced PCR products are collected and retested.

3. Zero Phosphate, Zero Nitrogen

Screen for zero phosphate, zero nitrogen tolerance identify seedlings better able to thrive under a phosphate nutritional deficiency.

Seeds are sterilized in 50% household bleach for 5 minutes and then washed with double distilled deionized water three times. Sterilized seed is stored in the dark at 4°C for a minimum of 3 days before use.

Zero phosphate, zero nitrogen media is prepared using commercially available MS media lacking phosphate, pH 5.7.

Approximately 1200 seeds are evenly spaced per MS-P-N plate before incubating in the vertical position at 22°C for 14 days. Under these conditions, the plates are exposed to 12,030 LUX from above and 3,190 LUX from the bottom.

Growth and overall greenness are assayed 10 days post-treatment. Seedling recovery is assessed by adding a thin layer (8.3 ml) of complete MS+P+N media, pH 5.7, softened by the addition of 0.02% agar. Media is added to the edge of the plate and slowly rotated until a thin film of PNP media is present on top of the solidified PNP media. Putative tolerant seedlings are greener and have increased growth compared to controls. Finale™ spraying is done after the plants are moved to soil to remove non-transgenics from the population.

DNA is isolated from each T2 plant and used in PCR reactions using the following cycling conditions: 95°C for 5 min, 35 cycles of (94°C for 30 sec, then 59°C for 30 sec, then 72°C for 1 min), 72°C for 8 min and 4°C hold. Aliquots of the reaction product are analyzed on a 1.0% agarose gel stained with ethidium bromide. The DNA products are sequenced to determine which insert sequences were in each superpool candidate chosen in the screen.

T3 Seed from those plants containing the sequenced PCR products are collected and retested.

5.2 Results

The results of the above experiments are set forth below wherein each individual example relates to all of the experimental results for a particular polynucleotide/polypeptide if the invention.

Example 1

Ceres cDNA 12335629

Clone 40781, Ceres cDNA 12335629, encodes a full-length protein with homology to a ferredoxin thioredoxin reductase from Arabidopsis thaliana.

Ectopic expression of Ceres cDNA 12335629 under the control of the CaMV35S promoter induces the following phenotypes:

Better growth and recovery after exposure to high pH conditions and

Continued growth under high pH induced phosphate and iron deficiencies.

Generation and Phenotypic Evaluation of T1 Lines Containing 35S:cDNA 12335629.

Wild-type Arabidopsis Wassilewskija (WS) plants were transformed with a T1 plasmid containing cDNA 12335629 in the sense orientation relative to the 35S constitutive promoter. The T1 plasmid vector used for this construct, CRS338, contains PAT and confers herbicide resistance to transformed plants. Ten independently transformed events
were selected and evaluated for their qualitative phenotype in the T1 generation. No positive or negative phenotypes were observed in the T1 plants.

Screening of Superpools on High pH Media for pH Tolerance.

Seed from superpools of the 35S:clone 40781 plants on high pH media were evaluated for greenness and size on high pH media as described above. Once cDNA 12335629 was identified in tolerant plants, the five individual T2 events containing this cDNA (ME03527) were screened on high pH media essentially as described above, but where the media pH is 8.5, to identify events with the tolerant phenotype.

Results:

[0112] Qualitative Analysis of the Superpool Containing 35S::clone 40781 Plants on high pH

[0113] The screen resulted in a decrease in germination and/or growth for both wildtype and superpools as compared to seeds on control media. Only one line survived transplantation to soil. The candidate was greener than controls but overall size was comparable to those of wild-type. There was no delay in flowering time or decrease in seed set in comparison to un-treated wild-type but a faster flowering time and greater seed set was apparent when compared to a recovered pH treated wild-type plant (data not shown). These results are consistent with those of the T1 generation which displayed normal flowering time and fertility. Qualitative and Quantitative Analysis of T1 events (T). The experiments were performed on High pH media.

[0114] The plants were treated with Finale™ to eliminate any false-positives or any lines where the Finale™ marker was suppressed. All of the Finale™-resistant candidates flowered and set seed. Finale™ segregation was assessed to identify events containing a single insert segregation in a 3:1 (R:S) ratio as calculated by chi-square test. All of the events segregated for a single functional insert (Table 1-1). The transgenic plants were greener and slightly larger than the control under high pH stress.

<table>
<thead>
<tr>
<th>Event</th>
<th>Generation</th>
<th>Observed</th>
<th>Expected</th>
<th>$\chi^2$</th>
<th>Probability of Chi-Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH Resistant</td>
<td>T3</td>
<td>22</td>
<td>29</td>
<td>0.926</td>
<td>0.35</td>
</tr>
<tr>
<td>pH Sensitive</td>
<td>T3</td>
<td>14</td>
<td>7</td>
<td>2.178</td>
<td>0.35</td>
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<td>N = 36</td>
<td></td>
<td>36</td>
<td>36</td>
<td>3.704</td>
<td>0.35</td>
</tr>
</tbody>
</table>

Qualitative and Quantitative Analysis of Individual T2 Events of cDNA 12335629 on High pH Plate Assay.

[0115] Five individual events of cDNA 12335629 (ME03527) were analyzed for a positive phenotype under high pH conditions. All five T2 events had wild-type germination frequencies on MS pH 5.7 plates (data not shown). All T1 lines and recovered T3 lines showed evidence of a single insert as determined by Chi-square analysis (Table 1-3). Seeds from each of the five independent T2 events were plated on pH 8.5 plates and allowed to germinate and grow for 14 days.

[0116] Four of five T2 events of ME03527 (-02,-03,-04, and -05) had a positive high pH tolerance phenotype as defined by growth and greenness. The phenotype of TV1E03527-01 was too weak to assess as positive compared to the controls (Table 1-4). Phenotype strength varied among the four positive independent events, but all showed better growth than controls. The segregation ratios, determined by a Chi-square test, show that the segregation of the transgene is the same as observed for Finale™ (Table 1-4). ME03527-02,-03,-04, and -05 had the strongest and most consistent pH tolerance phenotypes.

<table>
<thead>
<tr>
<th>Event</th>
<th>Generation</th>
<th>Observed</th>
<th>Expected</th>
<th>$\chi^2$</th>
<th>Probability of Chi-Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>ME03527-01 Finale™</td>
<td>T2</td>
<td>16</td>
<td>18</td>
<td>0.222</td>
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<tr>
<td>Sensitive</td>
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<td>24</td>
<td>0.889</td>
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Qualitative and Quantitative Analysis of cDNA 12335629 Progeny on Media Lacking Phosphate

Before testing independent T3 events, plants containing cDNA 12335629 were re-assayed for phosphate starvation tolerance by growth on media containing no phosphate as described above. After seven days only slightly more tolerance compared to controls is observed, but cDNA 12335629 seedlings are a bit larger and slightly greener than those of the control. Because the slight increase in size was difficult to assess, anything lower or equal to the wild-type average of 0.42 cm was assessed to be sensitive and anything higher was assessed as tolerant. Twenty-four resistant and twelve phosphate starved sensitive seedlings were compared to Finale™ frequencies and found to have a Chi-test probability of 0.49, suggesting a positive fit (Table 1-2).

<table>
<thead>
<tr>
<th>Table 1-2</th>
</tr>
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<tbody>
<tr>
<td>Event</td>
</tr>
<tr>
<td>-----------</td>
</tr>
<tr>
<td>-P Resistant</td>
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<tr>
<td>-P Sensitive</td>
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<tr>
<td>N = 36</td>
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TABLE 1-3-continued

<table>
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<tr>
<th>Event</th>
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<th>Expected</th>
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<th>Probability of Chi-Test</th>
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<tbody>
<tr>
<td>ME03527-02 Finale™ T3</td>
<td>28</td>
<td>27</td>
<td></td>
<td>0.037</td>
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<tr>
<td>ME03527-02 Finale™ R</td>
<td>8</td>
<td>9</td>
<td>0.111</td>
<td>0.30</td>
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<tr>
<td>N = 36</td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME03527-03 Finale™ T3</td>
<td>17</td>
<td>18</td>
<td></td>
<td>0.056</td>
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<tr>
<td>ME03527-03 Finale™ S</td>
<td>7</td>
<td>6</td>
<td>0.167</td>
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<tr>
<td>N = 24</td>
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<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>ME03527-04 Finale™ T3</td>
<td>27</td>
<td>27</td>
<td></td>
<td></td>
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<td>ME03527-04 Finale™ S</td>
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<td>1.0</td>
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<td>N = 36</td>
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<td></td>
</tr>
<tr>
<td>cDNA 12335629 Finale™ T3</td>
<td>22</td>
<td>27</td>
<td></td>
<td>0.926</td>
<td></td>
</tr>
<tr>
<td>cDNA 12335629 Finale™ S</td>
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<td>9</td>
<td>2.778</td>
<td>0.054</td>
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<td>N = 36</td>
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TABLE 1-4

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<th>Event</th>
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<th>Expected</th>
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<th>Probability of Chi-Test</th>
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<tr>
<td>ME03527-01 pH R</td>
<td>15</td>
<td>25.5</td>
<td>4.324</td>
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<td></td>
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<tr>
<td>ME03527-01 pH S</td>
<td>19</td>
<td>85.5</td>
<td>2.970</td>
<td>32E-05</td>
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<tr>
<td>N = 36</td>
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<td></td>
<td></td>
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</tr>
<tr>
<td>ME03527-02 pH R</td>
<td>23</td>
<td>24.75</td>
<td>0.124</td>
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<tr>
<td>ME03527-02 pH S</td>
<td>10</td>
<td>8.25</td>
<td>0.371</td>
<td>0.48</td>
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</tr>
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<td>N = 36</td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME03527-03 pH R</td>
<td>23</td>
<td>23.25</td>
<td>0.003</td>
<td>0.92</td>
<td></td>
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<tr>
<td>ME03527-03 pH S</td>
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<td>7.75</td>
<td>0.008</td>
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<td>N = 36</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>ME03527-04 pH R</td>
<td>24</td>
<td>27</td>
<td>0.333</td>
<td>0.25</td>
<td></td>
</tr>
<tr>
<td>ME03527-04 pH S</td>
<td>12</td>
<td>9</td>
<td>1.000</td>
<td></td>
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</tr>
<tr>
<td>N = 36</td>
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<tr>
<td>ME03527-05 pH R</td>
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<td>27</td>
<td>2.370</td>
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<tr>
<td>ME03527-05 pH S</td>
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<td>9</td>
<td>7.111</td>
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</tr>
<tr>
<td>N = 36</td>
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<tr>
<td>cDNA 12335629 pH T3</td>
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<td>27</td>
<td>2.370</td>
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<td></td>
</tr>
<tr>
<td>cDNA 12335629 pH S</td>
<td>17</td>
<td>9</td>
<td>7.111</td>
<td></td>
<td></td>
</tr>
<tr>
<td>N = 36</td>
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</table>
Table 1-5 provides the results of the consensus sequence analysis based on Ceres cDNA 13487605.

**Table 1-5**

<table>
<thead>
<tr>
<th>CeresClone</th>
<th>g12093884</th>
<th>g14275359</th>
<th>g1505189</th>
<th>Lead clone</th>
<th>CeresClone</th>
<th>1127455</th>
</tr>
</thead>
<tbody>
<tr>
<td>g12093884</td>
<td>TFDGQKLE</td>
<td>CDGQPWTG</td>
<td>KKEFVTNC</td>
<td>KEAKLNC</td>
<td>FGQKT</td>
<td>104</td>
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<tr>
<td>g14275359</td>
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<td>CDGQPWTG</td>
<td>KKEFVTNC</td>
<td>KEAKLNC</td>
<td>FGQKT</td>
<td>104</td>
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<td>g1505189</td>
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<td>CDGQPWTG</td>
<td>KKEFVTNC</td>
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<td>104</td>
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<tr>
<td>Lead clone</td>
<td>TFDGQKLE</td>
<td>CDGQPWTG</td>
<td>KKEFVTNC</td>
<td>KEAKLNC</td>
<td>FGQKT</td>
<td>104</td>
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<td>CeresClone</td>
<td>1127455</td>
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<td>CeresClone</td>
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<td>KKEFVTNC</td>
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<td>Lead clone</td>
<td>TFDGQKLE</td>
<td>CDGQPWTG</td>
<td>KKEFVTNC</td>
<td>KEAKLNC</td>
<td>FGQKT</td>
<td>104</td>
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<tr>
<td>CeresClone</td>
<td>1127455</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Example 2
Ceres cDNA 12330185

0118] Clone 34035, Ceres cDNA 12330185, encodes a 128 amino acid protein of unknown function (DUF423) from Arabidopsis thaliana.

0119] Ectopic expression of Ceres cDNA 12330185 under the control of the 32449 promoter induces the following phenotypes:

0120] Increased size and greenness on nutrient deficiencies incurred by high pH conditions,

0121] Better soil recovery after exposure to high pH stress, and

0122] Better recovery after exposure to conditions lacking both phosphate and nitrogen.

Generation and Phenotypic Evaluation of T1 Lines Containing p32449::cDNA 12330185.

0123] Wild-type Arabidopsis Wassilewskija (WS) plants were transformed with a Ti plasmid containing cDNA 12330185 in the sense orientation relative to the 32449 constitutive promoter. Promoter 32449 has broad expression throughout Arabidopsis, although at much lower expression level than CaMV35S. The T1 plasmid vector used for this construct, CRS331, contains PAT and confers herbicide resistance to transformed plants. Nine independently transformed events were selected and evaluated for their qualitative phenotype in the T1 generation. No positive or negative phenotypes were observed in the T1 plants.

Screens of Superpools on High pH Media for pH Tolerance.

0124] Seed from superpools of the 32449 over-expression lines were evaluated for greenness and size on high pH media as described above. Once cDNA 12330185 was identified in tolerant plants, nine individual T1 events containing this cDNA (ME00077) were screened on high pH media essentially as described above, but where the media pH is 8.5, to identify events with the tolerant phenotype.

Results:

0125] Qualitative Analysis of the Superpool Containing 34449::cDNA 12330185 on High pH

0126] The cDNA 12330185 line displayed a delayed flowering time of ~8 days and decreased seed set in comparison to the un-treated wild-type. However cDNA 12330185 displayed a faster flowering time (~15 days) and greater seed set when compared to the high pH grown wild-type plant.
Qualitative and Quantitative Analysis of the T₃ 32449::cDNA 12330185 on High pH.

**[0127]** The cDNA 12330185 line was tested for Finale™ resistance and re-assayed for continued pH tolerance. The segregation ratio of T₃ seeds from cDNA 12330185 is suggestive of a single insert, as calculated by a Chi-square test (Table 2-1). The cDNA 12330185 line was re-tested on pH 9.0 media as described and found to be tolerant to high pH when compared to controls.

### Table 2-1

<table>
<thead>
<tr>
<th>Event</th>
<th>Observed</th>
<th>Expected</th>
<th>χ²</th>
<th>Probability of Chi-Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Finale™ Resistant</td>
<td>27</td>
<td>27</td>
<td></td>
<td></td>
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<tr>
<td>Finale™ Sensitive</td>
<td>9</td>
<td>9</td>
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<td></td>
</tr>
<tr>
<td>N = 36</td>
<td>36</td>
<td>36</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Qualitative and Quantitative Analysis of Phosphate and Nitrate Starvation of T₃ (cDNA 12330185) Plants.

**[0128]** To ascertain whether the pH tolerant phenotype is related to better survival under nutrient starvation, T₃ seeds were assayed on MS media lacking both phosphate (−P) and nitrate (−N) (pH 5.7) as described above. The cDNA 12330185 line was greener and of equal size compared to wild-type controls. Ten days after the addition of +NP media film, cDNA 12330185 seedlings recovered more quickly than wild type. Twenty-five of 36 seedlings of SP9pH had greater growth when compared to wild type. This increased growth frequency is suggestive of a single insert as determined by Chi-square analysis (Table 2-2).

**[0129]** Seeds from T₃ lines representing nine individual events and containing cDNA 12330185 (ME00077-01, 02, 03, 04, 05, 06, 07, 08, 09) were plated on pH media, pH 8.5 as described above. Plates were evaluated at 7 and 12 days post-plating (Table 2-3). All nine T₃ events had wild-type germination frequencies except for ME00077-04 (Table 2-4). This germination problem however was not observed when seedlings were plated onto high pH plates.

**[0130]** Six of the nine events showed tolerance to high pH as defined by growth and greenness. The strongest tolerance phenotypes were in ME00077-03 and ME00077-05. ME00077-03 and ME00077-05 both had single inserts as determined by Chi-square analysis (Table 2-3).

**[0131]** The pH tolerant phenotype was strongest in the cDNA 12330185 T₃ line recovered from the superpool screen. We did not do a genetic mapping of this line’s insert to determine which event it represented. This line’s phenotype was so strong that it allowed adjacent wild-type quadrants within same plate to grow normally after 14-days. This is most likely due to acidification of surrounding media by the pH tolerant line. ME00077-03, 05 T₃ plants also showed increased recovery during phosphate and nitrogen starvation assays (data not shown). However, the cDNA 12330185 T₃ line recovered from the superpool phenotype was stronger than that observed for lines ME00077-03 and -05 under −NP starvation recovery (as noted above).

### Table 2-2

<table>
<thead>
<tr>
<th>Event</th>
<th>Observed</th>
<th>Expected</th>
<th>χ²</th>
<th>Probability of Chi-Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>NP Resistant</td>
<td>25</td>
<td>27</td>
<td>0.148</td>
<td>0.441</td>
</tr>
<tr>
<td>NP Sensitive</td>
<td>11</td>
<td>9</td>
<td>0.444</td>
<td>0.005</td>
</tr>
<tr>
<td>N = 36</td>
<td>36</td>
<td>36</td>
<td>0.592</td>
<td></td>
</tr>
</tbody>
</table>

Qualitative and Quantitative Analysis of Individual T₃ Events of cDNA 12330185 on High pH.

**[0132]** Observed and expected frequencies assuming a 3:1 (R:S) ratio for Finale™ among progeny of 32449::cDNA 12330185 and T₃ events tested for growth under high pH conditions. α of 0.05. Shading signifies a fit for 3 to 1.

<table>
<thead>
<tr>
<th>Event</th>
<th>Generation</th>
<th>Observed</th>
<th>Expected</th>
<th>χ²</th>
<th>Probability of Chi-Test</th>
<th>pH Phenotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>ME00077-01 Finale™ Resistant</td>
<td>T₂</td>
<td>34</td>
<td>33.75</td>
<td>0.062</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME00077-01 Finale™ Sensitive</td>
<td>T₂</td>
<td>2</td>
<td>2.25</td>
<td>0.028</td>
<td>0.86</td>
<td>No</td>
</tr>
<tr>
<td>ME00077-01 Finale™ Resistant</td>
<td>T₃</td>
<td>34</td>
<td>31.875</td>
<td>0.142</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME00077-01 Finale™ Sensitive</td>
<td>T₃</td>
<td>0</td>
<td>2.25</td>
<td>0.028</td>
<td>0.86</td>
<td>No</td>
</tr>
<tr>
<td>N = 36</td>
<td>34</td>
<td>34</td>
<td>2.267</td>
<td>0.13</td>
<td>0.86</td>
<td>No</td>
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<tr>
<td>ME00077-02 Finale™ Resistant</td>
<td>T₂</td>
<td>32</td>
<td>30.938</td>
<td>0.036</td>
<td></td>
<td></td>
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<tr>
<td>ME00077-02 Finale™ Sensitive</td>
<td>T₂</td>
<td>1</td>
<td>2.062</td>
<td>0.547</td>
<td>0.44</td>
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<td>33</td>
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<td>0.583</td>
<td>0.44</td>
<td>0.86</td>
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<tr>
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<td>36</td>
<td>33.75</td>
<td>0.15</td>
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<td></td>
</tr>
<tr>
<td>ME00077-02 Finale™ Sensitive</td>
<td>T₃</td>
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<td>0.25</td>
<td>0.12</td>
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<td>2.4</td>
<td>0.12</td>
<td>0.86</td>
<td>No</td>
</tr>
<tr>
<td>ME00077-03 Finale™ Resistant</td>
<td>T₂</td>
<td>30</td>
<td>28.25</td>
<td>0.536</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME00077-03 Finale™ Sensitive</td>
<td>T₂</td>
<td>5</td>
<td>8.75</td>
<td>3.607</td>
<td>0.143</td>
<td>Yes</td>
</tr>
<tr>
<td>N = 36</td>
<td>35</td>
<td>35</td>
<td>2.4</td>
<td>0.12</td>
<td>0.86</td>
<td>No</td>
</tr>
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</table>
### TABLE 2-3-continued

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<tr>
<th></th>
<th>T&lt;sub&gt;2&lt;/sub&gt;</th>
<th>% Generation on Finale&lt;sup&gt;TM&lt;/sup&gt; (%)</th>
<th>N = 36</th>
</tr>
</thead>
<tbody>
<tr>
<td>ME00077-04</td>
<td>19</td>
<td>18</td>
<td>0.0556</td>
</tr>
<tr>
<td>ME00077-04</td>
<td>5</td>
<td>6</td>
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</tr>
<tr>
<td>Resistant</td>
<td></td>
<td>0.64</td>
<td>Yes</td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME00077-05</td>
<td>24</td>
<td>24</td>
<td>0.2222</td>
</tr>
<tr>
<td>Resistant</td>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td></td>
<td>3R:1S</td>
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<tr>
<td>Sensitive</td>
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<td></td>
<td>Low</td>
</tr>
<tr>
<td>ME00077-06</td>
<td>10</td>
<td>8.5</td>
<td>0.265</td>
</tr>
<tr>
<td>Resistant</td>
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<td></td>
<td>3R:1S</td>
</tr>
<tr>
<td>Sensitive</td>
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<td></td>
<td>Strong</td>
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<tr>
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<td>0.353</td>
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<td></td>
<td>3R:1S</td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td></td>
<td></td>
</tr>
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<td>26.25</td>
<td>0.536</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Sensitive</td>
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<td></td>
</tr>
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<td>ME00077-09</td>
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<td>2.143</td>
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<tr>
<td>Resistant</td>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td></td>
<td>3R:1S</td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td></td>
<td></td>
</tr>
<tr>
<td>cDNA 12330185</td>
<td>27</td>
<td>27</td>
<td>0</td>
</tr>
<tr>
<td>Resistant</td>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME00077-03</td>
<td>9</td>
<td>9</td>
<td>1</td>
</tr>
<tr>
<td>pH Resistant</td>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td></td>
<td>Yes</td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ME00077-02</td>
<td>36</td>
<td>36</td>
<td>0.0556</td>
</tr>
<tr>
<td>pH Resistant</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td></td>
<td></td>
</tr>
<tr>
<td>cDNA 12330185</td>
<td>36</td>
<td>36</td>
<td>0</td>
</tr>
<tr>
<td>Resistant</td>
<td>T&lt;sub&gt;2&lt;/sub&gt;</td>
<td></td>
<td>3R:1S</td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td></td>
<td>Strong</td>
</tr>
</tbody>
</table>

**Germination reduction in comparison to wild-type control and other ME00077 lines**

### TABLE 2-4

**Observed germination frequencies on Finale<sup>TM</sup> plates among progeny of 32449::cDNA 12330185 T<sub>2</sub> and T<sub>3</sub> events tested for growth under high pH conditions.**

<table>
<thead>
<tr>
<th>Event</th>
<th>Observed pH</th>
<th>Expected pH</th>
<th>% Generation on Finale&lt;sup&gt;TM&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>ME00077-01</td>
<td>26</td>
<td>25.5</td>
<td>0.009</td>
</tr>
<tr>
<td>ME00077-03</td>
<td>8</td>
<td>8.5</td>
<td>0.029</td>
</tr>
<tr>
<td>N = 36</td>
<td>34</td>
<td>34</td>
<td>0.038</td>
</tr>
<tr>
<td>ME00077-05</td>
<td>29</td>
<td>26.25</td>
<td>0.288</td>
</tr>
<tr>
<td>ME00077-07</td>
<td>6</td>
<td>8.75</td>
<td>0.864</td>
</tr>
<tr>
<td>N = 36</td>
<td>35</td>
<td>35</td>
<td>1.152</td>
</tr>
<tr>
<td>cDNA 12330185</td>
<td>31</td>
<td>27</td>
<td>0.592</td>
</tr>
<tr>
<td>Resistant</td>
<td>9</td>
<td>9</td>
<td>0.124</td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td>36</td>
<td>36</td>
</tr>
<tr>
<td>cDNA 12330185</td>
<td>5</td>
<td>9</td>
<td>1.778</td>
</tr>
<tr>
<td>Sensitive</td>
<td>N</td>
<td>36</td>
<td>36</td>
</tr>
<tr>
<td>Probability of Chi-Test</td>
<td>2.370</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table 2-6 provides the results of the consensus sequence analysis based on Ceres cDNA 12330185.

TABLE 2-6

<table>
<thead>
<tr>
<th>CeresClone</th>
<th>Consensus</th>
<th>Gene</th>
<th>MDP</th>
<th>WHKWAA! S GLAACL GTY CAHVFKPON</th>
<th>55</th>
<th>CeresClone:586573</th>
<th>02/10/05</th>
<th>AYLEDRKFST -AP–GGFAF AAWAT LLF 11</th>
</tr>
</thead>
<tbody>
<tr>
<td>CeresClone:586573</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>111</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CeresClone:588155</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>111</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CeresClone:2893889</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>120</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>g</td>
<td>50918749</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>93</td>
<td></td>
<td></td>
</tr>
<tr>
<td>g</td>
<td>7933594</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>115</td>
<td></td>
<td></td>
</tr>
<tr>
<td>g</td>
<td>7933702</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>108</td>
<td></td>
<td></td>
</tr>
<tr>
<td>g</td>
<td>7933702</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>113</td>
<td></td>
<td></td>
</tr>
<tr>
<td>g</td>
<td>7933702</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>129</td>
<td></td>
<td></td>
</tr>
<tr>
<td>g</td>
<td>7933702</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>111</td>
<td></td>
<td></td>
</tr>
<tr>
<td>g</td>
<td>7933702</td>
<td>AYLEDRKFST</td>
<td>MAP</td>
<td>GFFAF</td>
<td>RAWASLLF</td>
<td>129</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

24/05/05 plur=3.000000 -collision -box -nobboxcol collbyconsensus

Example 3

Ceres cDNA 12482777

[0132] Clone 126592, Ceres cDNA 12482777, encodes a full-length protein that has homology to an iron/manganese superoxide dismutase from Arabidopsis thaliana.

[0133] Ectopic expression of Ceres cDNA 12482777 under the control of the CaMV 35S promoter induces the following phenotypes:

[0134] Increased growth under high pH induced stress
[0135] Better recovery after exposure to pH stress
[0136] Reduced height without a reduction in harvest index.

Generation and Phenotypic Evaluation of T1 Lines Containing 35S::cDNA 12482777.

[0137] Wild-type Arabidopsis Wassilewskija (WS) plants were transformed with a Ti plasmid containing cDNA 12482777 in the sense orientation relative to the 35S constitutive promoter. The T1 plasmid vector used for this construct, CRS338, contains PAT and confers herbicide resistance to transformed plants. Seven independently transformed events were selected and evaluated for their qualitative phenotype in the T1 generation. No negative phenotypes were observed in the T1 plants, although an increase in the number of branches was observed one of the events.

Screens of Superpools on High pH Media for pH Tolerance.

[0138] Seed from superpools of the 35S over-expression lines were evaluated for greenness and size on high pH media as described above. T1 seed were also assayed for total seed yield, total tissue dry weight and harvest index as described above.

Results:

[0139] Qualitative Analysis of the Superpool Containing 35S::cDNA 12482777 Plants on High pH

[0140] The screen identified a single event that was greener and the overall size was comparable to the controls. There was no delay in flowering time or decrease in seed set compared to un-treated wild-type. After recovery, the plant con-
containing cDNA 12482777 had significantly better seed yield, as determined by seed volume, than controls (FIG. 2).

Qualitative and Quantitative Analysis of T₃-cDNA 12482777 on High pH.

[0141] The plants were treated with Finale™ to eliminate any false-positives or any lines where the Finale™ marker was suppressed. All of the Finale™-resistant candidates flowering and set seed. Finale™ resistance segregation in the T₃ line suggested a segregation ratio of 1:1 (R:S) as calculated by chi-square test (Table 3-1).

[0142] The plants were greener than the pre-pH treated control. There was no tolerant effect found under low phosphate conditions (data not shown), suggesting that the tolerant response is not to the nutrient deficiencies imposed by the high pH but rather to oxidative stress induced by alkalinity.

Table 3-1

<table>
<thead>
<tr>
<th>Event</th>
<th>Generation</th>
<th>Observed</th>
<th>Expected</th>
<th>χ²</th>
<th>Probability of Chi-Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>cDNA 12482777</td>
<td></td>
<td>23</td>
<td>27</td>
<td>0.593</td>
<td></td>
</tr>
<tr>
<td>pH Resistant</td>
<td></td>
<td>13</td>
<td>9</td>
<td>1.778</td>
<td></td>
</tr>
<tr>
<td>cDNA 12482777</td>
<td></td>
<td>36</td>
<td>36</td>
<td>2.371</td>
<td></td>
</tr>
<tr>
<td>pH Sensitive</td>
<td></td>
<td>36</td>
<td>36</td>
<td>2.371</td>
<td></td>
</tr>
</tbody>
</table>

Qualitative and Quantitative Analysis of Harvest Index, Seed Yield, and Plant Height of T₃ Progeny of 35S:: cDNA 12482777.

[0143] A segregating population of 17 plants containing cDNA 12482777 was analyzed for harvest index and seed yield compared to wild-type populations. Based upon stem height measurements, the transgenic population of 35S:: cDNA 12482777 (10 plants) was significantly smaller than both internal (6 plants) and external wild-type/control populations. Internal wild-types/controls were those plants segregating from the T₃ population of the 35S::cDNA 12482777 line which did not contain the insert (segregating non-transgensics). External wild-types were non-transgenic plants from an outside source which shared no lineage with the line being tested. External wild-types are added to the experiment as a process control to ensure the quality of the growth conditions. Average height for transgenic plants of cDNA 12482777 was 33.44 cm±0.78 versus 44.65 cm±0.70 for the internal wild-type controls. Despite this decrease in plant height, harvest index, as measured by seed weight/total plant weight remained unaffected, i.e., these transgenic plants still produced the same ratio of total seed weight/total plant weight (biomass) as non-transgenic controls. This result means that although the total seed yield is decreased in cDNA 12482777 lines, it still has the same seed proportionally as controls. The cDNA 12482777 plants had a harvest index of 56.96±2.99 compared to the wild-type population’s harvest index of 44.92±2.67 (Table 3-2A). This increase in harvest index was significant at a P-value of 0.009 (Table 3-3A).

[0144] It is important to note that seed weight of cDNA 12482777 plants with a larger harvest index was 0.309777g±0.025 while the wild-type population had an average seed weight of 0.37155g±0.027 (Table 3-3B). cDNA 12482777 has a slightly smaller seed weight than the wild-type population but not statistically different at a P-value of 0.12 (Table 3-3B), suggesting that the harvest index of 35S:: cDNA 12482777 is comparable to, if not greater than, wild-type plants. This increase in harvest index is not due to an increase in number of branches (data not shown) as observed in the T₃ generation. Instead, the internode length between siliques is reduced compared to the internal wild-type control, suggesting that cDNA 12482777 plants have more siliques per stem length.

Table 3-2A

<table>
<thead>
<tr>
<th>Harvest Index: cDNA 12482777 small stature</th>
<th>Transgenic Population</th>
<th>Harvest Index of cDNA 12482777 Wild-type stature</th>
<th>Internal Wild-type Population</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>56.9582619</td>
<td>Mean</td>
<td>44.91972222</td>
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<tr>
<td>Standard Error</td>
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<td>Standard Error</td>
<td>2.667294601</td>
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<tr>
<td>Median</td>
<td>56.6889324</td>
<td>Median</td>
<td>45.56319444</td>
</tr>
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<td>Standard Deviation</td>
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<td>Standard Deviation</td>
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<td>Sample Variance</td>
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</table>

(95.0%)
### TABLE 3-2B

Descriptive statistical comparison of total seed weight (g) at time of harvest between segregating T₂ populations containing cDNA 12482777.

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<th>Total Seed Weight (g)</th>
<th>Transgenic Population</th>
<th>Wild-type Stature Population</th>
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### TABLE 3-4A

Statistical comparison of harvest index between transgenic populations of clone 126592 and internal wild-type populations using a t-test on two samples assuming unequal variances. cDNA 12482777 Wt stature (internal wild-type population) and cDNA 12482777 small stature (transgenic population).

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### TABLE 3-4B

Statistical comparison of seed weight between transgenic population of clone 126592 and internal wild-type populations using a t-test on two samples assuming unequal variances. cDNA 12482777 Wt stature (internal wild-type population) and cDNA 12482777 small stature (transgenic population).

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Table 3-5 provides the results of the consensus sequence analysis based on Ceres cDNA 12482777.

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**Example 4**

Ceres cDNA 12333678

[0145] Clone 26006, Ceres cDNA 12333678, encodes a full-length glycosyl hydrolase. Ectopic expression of Ceres cDNA 12333678 under the control of the CaMV35S promoter induces the following phenotypes:

[0146] Germination on high concentrations of polyethylene glycol (PEG), mannitol and abscisic acid (ABA).

[0147] Continued growth on high PEG, mannitol and ABA.

Generation and Phenotypic Evaluation of T1 Lines Containing 35S::cDNA 12333678.

[0148] Wild-type Arabidopsis Wassilewskija (WS) plants were transformed with a Ti plasmid containing cDNA 12333678 in the sense orientation relative to the CaMV35S constitutive promoter. The T1 plasmid vector used for this construct, CRS338, contains PAT and confers herbicide resistance to transformed plants. Ten independently transformed events were selected and evaluated for their qualitative phenotype in the T1 generation. No positive or negative phenotypes were observed in the T2 plants.
Screens of Superpools on High PEG, Mannitol and ABA as Surrogate Screens for Drought Tolerance.

[0149] Seeds from 13 superpools (1,200 T2 seeds from each superpool) from the CaMV 35S or 32449 over-expression lines were tested on high pH media as described above. T2 seeds were collected from the tolerant plants and analyzed for tolerance on all additional high pH screens.

[0150] Once cDNA 12333678 was identified in tolerant plants, the individual T2 events containing this cDNA (ME01334) were screened on high PEG, mannitol and ABA to identify events with the resistance phenotype.

[0151] Superpools (SP) are referred to as SP1, SP2 and so on. The letter following the hyphen refers to the screen (P=PEG, M=mannitol, and A=ABA) and the number following the letter refers to a number assigned to each plant obtained from that screen on that superpool. For example, SP1-M18 is the 18th plant isolated from a mannitol screen of Superpool 1.

Results:

[0152] Qualitative Assessment of ME01334 on high pH.

[0153] Superpool 1 was screened on high pH media as described above. PCR analyses identified ME01334 as one of the ME lines showing high pH resistance. Testing of the second generation confirmed the inheritance of the pH resistance (data not shown).

[0154] ME01334 plants that recovered after high pH produced an exceptionally large number of seeds compared to wild-type controls. Additional testing confirmed that these plants statistically produce 30-80% more seeds than either wild-type or transgenic control plants that are recovered from this screen or transferred from regular MS media.

[0155] Table 4-1 provides the results of the consensus sequence analysis based on Ceres cDNA 12333678.

---

**Table 4-1**

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23/05/05 plur=9.500000 ~col=1/12 boxed=1/12 colbyconsensus
The invention being thus described, it will be apparent to one of ordinary skill in the art that various modifications of the materials and methods for practicing the invention can be made. Such modifications are to be considered within the scope of the invention as defined by the following claims.

Each of the references from the patent and periodical literature cited herein is hereby expressly incorporated in its entirety by such citation.
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His Thr Ala Ala Leu Val Ala Ala Pro Ile Thr Lys His Pro Asn Val
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50  55  60

Phe Gly Gly Leu Leu Thr Ala Gly Ile Leu Ala Phe Ser Gly Thr Cys
65  70  75  80

Tyr Thr Val Ala Phe Leu Glu Asp Arg Lys Tyr Ser Thr Met Ala Pro
85  90  95

Phe Gly Gly Phe Ala Phe Ile Ala Ala Trp Gly Ser Leu Phe Phe
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<210> SEQ ID NO 7
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<212> TYPE: PRT
<213> ORGANISM: Triticum aestivum
<220> FEATURE:
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<223> OTHER INFORMATION: CeresClone:678257

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Met Val Met Pro Thr Asp Pro Met Leu Trp His Lys Val Ala Ala Val
1   5   10   15

Ser Gly Val Val Ala Leu Gly Leu Gly Thr Tyr Gly Ala His Met Phe
20  25  30
Arg Pro Gln Asn Pro Arg Tyr Lys Ile Trp Gln Thr Ala Ser Leu 35 40 45
Tyr His Leu Val His Thr Ala Ala Leu Leu Gly Ala Pro Met Thr Lys 50 55 60
Arg Pro Asn Ile Phe Gly Gly Leu Thr Thr Gly Ile Val Leu Phe 65 70 75 80
Ser Gly Thr Cys Tyr Thr Val Ala Tyr Leu Glu Asp Arg Lys Phe Ser 85 90 95
Ser Pro Ala Pro Ile Gly Gly Phe Ala Phe Ile Ala Ala Trp Ala Ser 100 105 110
Leu Leu Phe 115

<210> SEQ ID NO 8
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<213> ORGANISM: Zea mays
<220> FEATURE:
<221> NAME/KEY: misc
<223> OTHER INFORMATION: CeresClone:289088

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Ser Gly Val Ala Ala Leu Gly Leu Gly Thr Tyr Gly Ala His Met Phe 20 25 30
Arg Pro Lys Asn Pro Ala Tyr Lys Val Leu Trp His Thr Ala Ser Leu 35 40 45
Tyr His Leu Val His Thr Ala Ala Leu Leu Gly Ala Pro Ile Thr Lys 50 55 60
Arg Pro Asn Val Phe Gly Gly Leu Thr Ala Gly Ile Val Leu Phe 65 70 75 80
Ser Gly Thr Cys Tyr Thr Val Ala Tyr Leu Glu Asp Arg Lys Phe Ser 85 90 95
Ser Pro Ala Pro Leu Gly Gly Phe Ala Phe Ile Ala Ala Trp Ala Ser 100 105 110
Leu Leu Phe 115

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<212> TYPE: PRT
<213> ORGANISM: Psathyrostachys juncea
<220> FEATURE:
<221> NAME/KEY: misc
<223> OTHER INFORMATION: gi[79]63694

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Leu Gly Thr Tyr Gly Ala His Met Phe Arg Pro Gin Asn Pro Lys Tyr 20 25 30
Lys Glu Ile Trp Gin Thr Ala Phe Leu Tyr His Leu Val His Thr Ala 35 40 46
Ala Leu Leu Gly Ala Pro Met Thr Lys Arg Pro Asn Ile Phe Gly Gly 50 55 60
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<211> LENGTH: 106
<212> TYPE: PRT
<213> ORGANISM: Agropyron cristatum
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi(79)63702

<400> SEQUENCE: 10

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Ser Gly Val Ala Ala Leu Gly Leu Gly Thr Tyr Gly Ala His Met Phe
20  25  30
Arg Pro Gln Arg Pro Arg Tyr Glu Ile Trp Glu Thr Ala Ser Leu
35  40  45
Tyr His Leu Val His Thr Ala Ala Leu Leu Gly Ala Pro Met Thr Lys
50  55  60
Arg Pro Asn Ile Phe Gly Gly Leu Leu Thr Gly Ile Val Leu Phe
65  70  75  80
Ser Gly Thr Cys Tyr Thr Val Ala Tyr Leu Glu Asp Arg Lys Phe Ser
85  90  95
Ser Pro Ala Pro Ile Gly Gly Phe Ala Phe
100 105

<210> SEQ ID NO 11
<211> LENGTH: 120
<212> TYPE: PRT
<213> ORGANISM: Oryza sativa subsp. japonica
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi(50)919749

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Lys Val Ala Ala Ile Ser Gly Val Ala Ala Leu Gly Leu Gly Thr Tyr
20  25  30
Gly Ala His Met Phe Arg Pro Lys Asn Pro Ala Tyr Lys Glu Val Trp
35  40  45
His Thr Ala Ser Leu Tyr His Leu Val His Thr Ala Ala Leu Leu Gly
50  55  60
Ala Pro Ile Thr Lys Arg Pro Asp Val Phe Gly Gly Leu Leu Thr Ala
65  70  75  80
Gly Ile Val Leu Phe Ser Gly Thr Cys Tyr Thr Val Ala Tyr Leu Glu
85  90  95
Asp Arg Lys Tyr Ser Ser Thr Ala Pro Leu Gly Gly Phe Ala Phe Ile
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Ala Ala Trp Ala Ser Leu Leu Phe
115 120

<210> SEQ ID NO 12
<211> LENGTH: 689
<212> TYPE: DNA
<213> ORGANISM: Arabidopsis thaliana
<220> FEATURE:
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<223> OTHER INFORMATION: clone40781_planta.experimental_L43

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ttcctctta gataatttc tttagtagtg 689

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<212> TYPE: PPT
<213> ORGANISM: Arabidopsis thaliana
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<223> OTHER INFORMATION: peptide_clone40781_planta.experimental_L43

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Leu Gly Val Thr Pro Arg Thr Ser Phe Arg Arg Phe Val Ile Arg Ala 20 25 30
Lys Thr Glu Pro Ser Glu Lys Ser Val Glu Ile Met Arg Lys Phe Ser 35 40 45
Glu Gln Tyr Ala Arg Arg Ser Gly Thr Tyr Phe Cys Val Asp Lys Gly 50 55 60
Val Thr Ser Val Ile Lys Gly Leu Ala Glu His Lys Asp Ser Tyr 65 70 75 80
Gly Ala Pro Leu Cys Pro Cys Arg His Tyr Asp Asp Lys Ala Ala Glu 85 90 95
Val Gly Gln Gly Phe Trp Asn Cys Pro Cys Val Pro Met Arg Glu Arg 100 105 110
Lys Glu Cys His Cys Met Leu Phe Leu Thr Pro Asp Asn Phe Ala 115 120 125
Gly Lys Asp Gln Thr Ile Ser Asp Glu Ile Lys Glu Thr Thr Ala 130 135 140
Asn Met 145

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<212> TYPE: PRT
<213> ORGANISM: Spinacia oleracea
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<400> SEQUENCE: 14

Met Lys Ala Leu Gln Ala Ser Thr Ser Tyr Ser Phe Ser Ser Lys Ser
1 5   10 15

Ser Ser Ala Thr Leu Gln Arg Arg Thr His Arg Pro Gln Cys Val Ile
20   25   30

Leu Ser Lys Val Glu Pro Ser Asp Lys Ser Val Glu Ile Met Arg Lys
35   40   45

Phe Ser Glu Gln Tyr Ala Arg Arg Ser Gly Thr Tyr Phe Cys Val Asp
50   55   60

Lys Gly Val Thr Ser Val Ile Lys Gly Leu Ala Glu His Lys Asp
65   70   75   80

Ser Leu Gly Ala Pro Leu Cys Pro Cys Arg Tyr Tyr Asp Ser Lys Ala
85   90   95

Ala Glu Ala Thr Gln Gly Phe Trp Asn Cys Pro Cys Val Pro Met Arg
100 105 110

Glu Arg Lys Glu Cys His Cys Met Leu Phe Thr Pro Glu Asn Asp
115 120 125

Phe Ala Gly Lys Asp Gln Thr Ile Gly Leu Asp Glu Ile Arg Glu Val
130 135 140

Thr Ala Asn Met
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<221> NAME/KEY: misc.feature
<223> OTHER INFORMATION: CeresClone:1127455
<220> FEATURE:
<221> NAME/KEY: misc.feature
<223> OTHER INFORMATION: Xaa can be any naturally occurring amino acid
<220> FEATURE:
<221> NAME/KEY: misc.feature
<223> OTHER INFORMATION: Xaa can be any naturally occurring amino acid

<400> SEQUENCE: 15

Met Asn Pro Gln Ala Val Ser Cys Ser Phe Gly Phe Val Ser Ala Pro
1 5 10 15

Leu Val Ser Pro Arg Thr Ser Arg Phe Val Val Gln Ala Lys Ser Glu
20 25   30

Pro Ser Glu Xaa Ser Val Glu Ile Met Arg Lys Phe Ser Glu Gln Tyr
35   40   45

Ala Arg Arg Ser Gly Thr Phe Cys Val Asp Lys Gly Val Xaa Ser
50   55   60

Val Val Ile Lys Gly Leu Ala Glu His Lys Asp Ser Tyr Gly Ala Pro
65   70   75   80

Leu Cys Pro Cys Arg His Tyr Asp Asp Lys Ala Ala Glu Val Gly Gln
85   90   95
-continued

| Gly Phe Trp Asn Cys Pro Cys Val Pro Met Arg Glu Arg Lys Glu Cys | 100 105 110 |
| His Cys Met Leu Phe Leu Thr Pro Asp Asn Asp Phe Ala Gly Lys Asp | 115 120 125 |
| Gln Thr Ile Thr Ser Asp Glu Ile Lys Glu Thr Thr Ala His Met | 130 135 140 |

<210> SEQ ID NO 16
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<212> TYPE: PRT
<213> ORGANISM: Glycine max
<220> FEATURE:
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Met Thr Thr Gln Ala Ser Thr Phe Ala Val Ala Val Pro Ser Val Ala
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Thr Pro Phe Arg Arg His Arg Asn Pro Phe Val Arg Ala Gln Ala
20  25  30
Glu Pro Ser Asp Lys Ser Val Glu Ile Met Arg Lys Phe Ser Glu Gln
35  40  45
Tyr Ala Arg Lys Ser Gly Thr Tyr Phe Cys Val Asp Lys Gly Val Thr
50  55  60
Ser Val Val Ile Lys Gly Leu Ala Asp His Lys Asp Thr Leu Gly Ala
65  70  75  80
Pro Leu Cys Pro Cys Arg His Tyr Asp Lys Ala Ala Gln Val Ala
85  90  95
Gln Gly Phe Trp Asn Cys Pro Cys Val Pro Met Arg Glu Arg Lys Glu
100 105 110
Cys His Cys Met Leu Phe Leu Thr Pro Asp Asn Asp Phe Ala Gly Asn
115 120 125
Glu Glu Thr Ile Thr Leu Asp Glu Ile Lys Glu Ser Thr Ala Asn Met
130 135 140

<210> SEQ ID NO 17
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<212> TYPE: PRT
<213> ORGANISM: Solanum tuberosum
<220> FEATURE:
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Met Arg Thr Leu Gln Ala Ser Thr Ser Tyr Ser Val Gly Phe Gly Ile
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Ser Ser Phe Ala Thr Arg Phe Pro Ser Thr His Arg Cys Leu Thr
20  25  30
Val Ala Lys Met Gly Pro Ser Gly Lys Ser Val Glu Ile Met Arg Lys
35  40  45
Phe Ser Glu Gln Tyr Ala Arg Arg Ser Glu Thr Tyr Phe Cys Met Asp
50  55  60
Lys Gly Val Thr Ser Val Val Ile Lys Gly Leu Ala Ala Glu His Lys Asp
65  70  75  80
Thr Leu Gly Ala Pro Leu Cys Pro Cys Arg His Tyr Asp Lys Ala
85  90  95
 Ala Gln Ala Glu Gln Gly Phe Trp Asn Cys Pro Cys Val Pro Met Arg
Glu Arg Lys Glu Cys His Cys Met Leu Phe Leu Thr Pro Asn Asp
Phe Ala Gly Glu Glu Gln Thr Ile Ser Met Glu Glu Ile Lys Glu Thr
Thr Ala Asn Met

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<223> DESCRIPTION: ORGANISM: Oryza sativa subsp. japonica

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<223> OTHER INFORMATION: clone126592_expected_L44

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180
tatatcttg gtgatgt gtaacgacc tccatatcct tctgtacgcct tggaacggca
240
tgatctgc gaaaccttgg attacactg gggcaccacct cacaacactt atgttagaa
300
tctctctcag caaatctttag gcacgatct agatgctatct ctctctctct ctctctctct cggcttgag 420
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<213> ORGANISM: Arabidopsis thaliana
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<223> OTHER INFORMATION: peptide_clone126592_expected_L44

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Arg Asn Gly Lys Arg Arg Leu Gly Thr Lys Val Ala Val Ser Gly Val
35 40 45
Ile Thr Ala Gly Phe Glu Leu Lys Pro Pro Pro Tyr Pro Leu Asp Ala
50 55 60
Leu Glu Pro His Met Ser Arg Glu Thr Leu Asp Tyr His Thr Gly Lys
65 70 75 80
His His Lys Thr Tyr Val Glu Asn Leu Asn Lys Gln Ile Leu Gly Thr
85 90 95
Asp Leu Asp Ala Leu Ser Leu Glu Val Val Leu Leu Ser Tyr Asn
100 105 110
Lys Gly Asn Met Leu Pro Ala Phe Asn Asn Ala Ala Gln Ala Trp Asn
115 120 125
His Glu Phe Phe Trp Glu Ser Ile Gln Pro Gly Gly Gly Gly Lys Pro
130 135 140
Thr Gly Glu Leu Leu Leu Arg Leu Ile Glu Arg Asp Phe Gly Ser Phe Glu
145 150 155 160
Glu Phe Leu Glu Arg Phe Lys Ser Ala Ala Ala Ser Asp Phe Gly Ser
165 170 175
Gly Thr Thr Thr Leu Ala Tyr Lys Ala Asn Arg Leu Asp Val Ala Asn
180 185 190
Ala Val Asn Pro Leu Pro Lys Glu Asp Lys Leu Val Ile Val
195 200 205
Lys Thr Pro Asn Ala Val Asn Pro Leu Val Asp Tyr Ser Pro Leu
210 215 220
Leu Thr Ile Asp Thr Trp Glu His Ala Tyr Tyr Leu Asp Phe Glu Asn
225 230 235 240
Arg Arg Ala Glu Tyr Ile Asn Thr Phe Met Glu Lys Leu Val Ser Trp
245 250 255
Glut Val Ser Thr Arg Leu Glu Ser Ala Ile Ala Arg Ala Val Gln
260 265 270
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Glu Val Pro Glu Val Tyr Leu Ser Asp Ser Ile Asp Val Ser Glu Val
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Asp
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<212> TYPE: DNA
<213> ORGANISM: Arabidopsis thaliana
<220> FEATURE: misc_feature
<223> OTHER INFORMATION: clone126592.in planta.experimental.L44

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aatctgaatgg aatgaagacag gaaagagaat gttgaggatc aagtagaggt tttctggtgt 180
tatccagct gggatggagc gtaagccacc ttcatactct cttgatgctc tggacgac 240
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tatgacgccg gaaacctttg aggcaaacat cacaaccatt aatgtagaa 300
cctgaccaag caaatcttg agaacgatct atatcattta tctttggaag aagcttgctt 360
tttttcata cacaagagca aatatgttcc tgcttttcaac aaagctgcac aggaggtgaa 420
cacgcaggtct tgtgggagt cttcaccacc tcggagcttgga ggaagagccaa tgtggagct 480
cocagattata atagaaagagt attttgggtc tttggaaggt tttttggaa ggttcaatgc 540
ggtgcagct tcgaatcttg gcagcgggtg acaatcgcttc gatataagg cgaatagact 600
tgcggtgcga aatgacgtaa atctctcccc aagggggga gacaagaaaattgtaggtg 660
gagagcgccc aatgcagtaa atctgctcgct atggattatt ttcaccacttc acacgattga 720	acgccgggag aagcgttcat aatctgccaggt ttgaaacagaga agagctgatt acataaatac 780
ttcctgagaa aagaggttgct caggggaaac tctaaacgca aagttgaggat ccgcaatttc 840
tcggagctcg caagagaaac gagaagagac agacagcaga gatgaagaga atccgattga 900
tgctcrgcag accatttgat tagattgtga ccagcgtgtc tcgaggtttg acataaacc 960
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<213> ORGANISM: Arabidopsis thaliana
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<222> OTHER INFORMATION: peptide_clone126592_inplanta_experimental_L44
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Arg Asn Gly Lys Arg Arg Leu Gly Thr Lys Val Ala Val Ser Gly Val
35       40      45
Ile Thr Ala Gly Phe Glu Leu Lys Pro Pro Pro Tyr Pro Leu Asp Ala
50       55      60
Leu Glu Pro His Met Ser Arg Glu Thr Leu Asp Tyr His Trp Gly Lye
65       70      75      80
His His Lys Thr Tyr Val Glu Asn Leu Asn Lys Glu Ile Leu Gly Thr
85       90      95
Asp Leu Asp Ala Leu Ser Leu Glu Val Val Leu Leu Ser Tyr Asn
100      105      110
Lys Gly Asn Met Leu Pro Ala Phe Asn Ala Ala Glu Ala Trp Asn
115      120      125
His Glu Phe Phe Trp Glu Ser Ile Gln Pro Gly Gly Gly Gly Gly Lys Pro
130      135      140
Thr Gly Leu Leu Arg Leu Ile Glu Arg Asp Phe Gly Ser Phe Glu
145      150      155      160
Glu Phe Leu Glu Arg Phe Lys Ser Ala Ala Ala Ser Asn Phe Gly Ser
165      170      175
Gly Trp Thr Trp Leu Ala Tyr Lys Ala Asn Arg Leu Asp Val Ala Asn
180      185      190
Ala Val Asn Pro Leu Pro Lys Glu Asp Lys Lye Val Val Ile Val
195      200      205
Lys Thr Pro Asn Ala Val Asn Pro Leu Val Trp Asp Tyr Ser Pro Leu 210 215 220
Leu Thr Ile Asp Thr Trp Glu His Ala Tyr Tyr Leu Asp Phe Glu Asn 225 230 235 240
Arg Arg Ala Glu Tyr Ile Asn Thr Phe Met Glu Lys Leu Val Ser Trp 245 250 255
Glu Thr Val Ser Thr Arg Leu Glu Ser Ala Ile Ala Arg Ala Val Gln 260 265 270
Arg Glu Gln Glu Arg Thr Glu Thr Glu Asp Glu Glu Asn Pro Asp Asp 275 280 285
Glu Val Pro Glu Val Tyr Leu Asp Ser Asp Ile Asp Val Ser Glu Val 290 295 300
Asp 305

<210> SEQ ID NO 24
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<213> ORGANISM: Brassica napus
<220> FEATURE:
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<223> OTHER INFORMATION: CeresClone:970125

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Phe Glu Leu Lys Pro Pro Pro Tyr Pro Leu Asp Ala Leu Glu Pro His 50 55 60
Met Ser Arg Glu Thr Met Asp Tyr His Trp Gly Lys His His Arg Thr 65 70 75 80
Tyr Val Glu Asn Leu Asn Lys Glu Ile Leu Gly Thr Asp Leu Asp Gly 85 90 95
Leu Ser Leu Glu Glu Val Val Leu Ser Tyr Arg Val Gly Asn Met 100 105 110
Leu Pro Val Phe Asn Asn Ala Ala Glu Asn Trp Asn His Glu Phe Phe 115 120 125
Trp Glu Ser Ile Gln Pro Gly Gly Gly Lys Pro Ser Gly Asp Leu 130 135 140
Leu Arg Leu Ile Glu Arg Asp Phe Gly Ser Phe Asp Asp Phe 145 150 155

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<212> TYPE: PRT
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35 40 45
Gly Arg Thr Lys Ile Thr Ala Lys Phe Glu Leu Lys Pro Pro Tyr
50 55 60
Pro Leu Ser Ala Leu Glu Pro Ile Met Ser Gln Glu Thr Leu Glu Tyr
65 70 75 80
His Trp Gly Lys His His Arg Thr Tyr Val Asp Asn Leu Asn Arg Gln
85 90 95
Ile Asp Gly Thr Asp Leu Asp Gly Asn Ser Leu Glu Asn Thr Ile Val
100 105 110
Ile Thr Tyr Asn Lys Gly Asp Ile Leu Pro Ala Phe Asn Asn Ala Ala
115 120 125
Gln Ala Trp Asn His Asp Phe Phe Glu Trp Glu Ser Met Lys Pro Gly Gly
130 135 140
Gly Gly Arg Pro Ser Gly Asp Leu Leu Asn Ile Glu Arg Asp Phe
145 150 155 160
Gly Ser Phe Glu Lys Phe Leu Asp Glu Phe Lys Thr Ala Ala Ser Thr
165 170 175
Gln Phe Gly Ser Gly Trp Ala Trp Leu Ala Tyr Lys Glu Ser Arg Leu
180 185 190
Asp Val Glu Asn Ala Val Asn Pro Leu Glu Ser Asp Glu Asp Lys Lys
195 200 205
Leu Val Val Val Lys Thr Pro Asn Ala Val Asn Pro Leu Val Trp Asn
210 215 220
Tyr Tyr His Pro Leu Leu Thr Ile Asp Val Trp Glu His Ala Tyr Phe
225 230 235 240
Ile Asp Phe Glu Asn Glu Arg Arg Tyr Ile Ser Val Phe Met Asp
245 250 255
Lys Leu Val Ser Trp Asp Ala Val Ser Ser Arg Leu Glu Glu Ala Lys
260 265 270
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Glu Glu Glu Lys Arg Thr Ser Ser Glu Ala Ile Pro Glu Ile Tyr Ser
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Asp Gly Asp Ala Asp Leu Asp Ala Glu
305 310

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Ser Val Thr Thr Phe Lys Phe Ser Lys Lys Gin Gly Arg Cys Ile Arg
Arg Ala Gly Gly Thr Gln Ile Thr Ala Lys Phe Glu Leu Lys Pro Pro
Pro Tyr Pro Leu Asn Ala Ser Glu Pro Ile Met Ser Glu Asn Thr Phe
Glu Tyr His Trp Gly Lys His His Arg Ala Tyr Val Asp Asn Leu Asn
Lys Gln Ile Glu Gly Thr Asp Leu Asp Gly Lys Ser Leu Glu Glu Thr
Ile Ile Met Ser Tyr Asn Asn Gly Asp Ile Leu Pro Ala Phe Asn Asn
Ala Ala Gln Val Trp Asn His Asp Phe Phe Trp Glu Ser Met Lys Pro
Gly Gly Gly Lys Pro Ser Gly Glu Leu Leu Lys Leu Leu Ile Glu Arg
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Trp Asn His His Pro Leu Thr Ile Asp Val Trp Glu His Ala
Tyr Tyr Leu Asp Tyr Gln Asn Arg Asp Glu Tyr Ile Ser Val Phe
Met Asp Lys Leu Val Ser Trp Glu Ala Val Ser Ser Arg Leu Glu Lys
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Arg Glu Glu Glu Lys Ser Thr Thr Gly Glu Asp Thr Pro Ala Pro
Glu Ile Phe Ala Asp Ser Asp Thr Asp

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Phe Tyr Trp Arg Ser Met Lys Pro Gly Gly Gly Lys Pro Pro Glu
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<400> SEQUENCE: 28

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coaaaat gcacccatgg ctctatctgga gttggtttta atttatcaca aatotttatg 1260
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gacaacaca taataataatc tactcccttt ataaagagttt taatgtaaatt tctgataatt 1440
aagatatatttt ttacaacacac aacccaaattt attatatattt tttctcttattt acacaacca 1500
gaaagaaaaa ctttttttttt tggcaagaaag aggggagatt aatgaaacag aaaaaacag 1560
gaaatataat aaccgacact tcttaattaa cacttctcaaa taaggaatttt tattgacggc 1620
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cggcgcctag atcattaaaac tctcatggac caattttttg acgccagatc gaaactctctg 1860
cacaacacaa awaatatat taagaagatc cttttctctgg tttggttctt caaaaaactac 1920
acccggagc ccttacctgt ctctccgtt ctctagtattt ttcctcagct tcgctcttta 1980
gatctcatgt ggtttccaaa tttctcogagata agggcctt 2016

<210> SEQ ID NO 30
<211> LENGTH: 870
<212> TYPE: DNA
<213> ORGANISM: Arabidopsis thaliana
<220> FEATURE: 
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: Ceres cDNA 12333678

<400> SEQUENCE: 30

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gcgcacgcgct cgcctgctct atccggctct cctccgctat aagcgcacc aaccccccatca 180
tctgagcatc tcacatggaa ccaatctcttg aagcatttgaag agatcaatgc acatcttgcc 240
gcaagatgca gaagaggtgta ctctctgcgtg ttagtctggg aagttttgag ttaggtctttg 300
cctgggataa gttcccctga aaaaatctctg tttctgcttt ctgggactca ttcattgccc 360
acacacaca ctcacccatac ctgctcgaggg aaaaatgtagc aagcagcaagct acacacggg 420
agaggtctg cccctagctc gagacataag ttgctagata ataaggtctctg ctgctctctctc 480
tcagaccaag ccatctaatc cccacactctt cccctgctcttg gatagtgaccc 540
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acatttggag gaaagaggtgct gagtctctgta ctctctcctctt ccctgctcttg 660
acatattgatc ggaagagcatc gaagagatgtg cggcctctaa ttgctcgccgct aattctatga 720
ttgagaggtgaa agagagaggtc cttcttgccaa atctccagta ctaagttgaccc 780
atcattttggtc aatactctgct ctttaattt aataatattt tattgagaaaa ttatgtttgga 840
gtgtagataa taaaatgttt tccaataggg 870
**Continued**

<210> SEQ ID NO 31
<211> LENGTH: 263
<212> TYPE: PRT
<213> ORGANISM: Arabidopsis thaliana
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: Peptide Cerec cDNA 12333670

<400> SEQUENCE: 31

Met Ser Glu Glu Lys Arg Lys Glu His Phe Val Leu Val His Gly Ala
1   5   10  15
Cys His Gly Ala Trp Cys Trp Tyr Lys Val Lys Pro Leu Leu Glu Ala
20  25  30
Leu Gly His Arg Val Thr Ala Leu Asp Leu Ala Ala Ser Gly Ile Asp
35  40  45
Thr Thr Arg Ser Ile Thr Asp Ile Ser Thr Cys Glu Glu Tyr Ser Glu
50  55  60
Pro Leu Met Glu Leu Met Thr Ser Leu Pro Asp Asp Glu Lys Val Val
65  70  75  80
Leu Val Gly His Ser Phe Gly Gly Leu Ser Leu Ala Leu Ala Met Asp
85  90  95
Lys Phe Pro Asp Lys Ile Ser Val Ser Val Phe Val Thr Ala Phe Met
100 105 110
Pro Asp Thr Lys His Ser Pro Ser Phe Val Glu Glu Lys Phe Ala Ser
115 120 125
Ser Met Thr Pro Glu Gly Trp Met Gly Ser Leu Glu Thr Tyr Gly
130 135 140
Ser Asp Asn Ser Gly Leu Ser Val Phe Ser Thr Asp Phe Met Lys
145 150 155 160
His Arg Leu Tyr Glu Leu Ser Pro Val Glu Asp Leu Leu Gly Leu
165 170 175
Leu Leu Lys Arg Pro Ser Ser Leu Phe Ile Asn Glu Leu Ser Lys Met
180 185 190
Glu Asn Phe Ser Glu Lys Gly Tyr Gly Ser Val Pro Arg Ala Tyr Ile
195 200 205
Val Cys Lys Glu Asp Asn Ile Ser Glu Asp His Glu Arg Trp Met
210 215 220
Ile His Asn Tyr Pro Ala Asn Leu Val Ile Glu Met Glu Thr Asp
225 230 235 240
His Met Pro Met Phe Cys Lys Pro Glu Val Leu Ser Asp His Leu Leu
245 250 255
Ala Ile Ala Asp Asn Phe Ser
260

<210> SEQ ID NO 32
<211> LENGTH: 267
<212> TYPE: PRT
<213> ORGANISM: Citrus sinensis
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi[14]279437

<400> SEQUENCE: 32

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1   5   10  15
Gly Val Asn His Gly Ala Trp Cys Trp Tyr Lys Leu Lys Ala Arg Leu
Val Ala Gly Gly His Arg Val Thr Ala Val Asp Leu Ala Ala Ser Gly
6 Val Met Asn His Thr Leu Thr Ala Asp Ala Gly Ala Ser Thr Ala
11 Val Met Leu Met Gly Val Leu Ala Ser Leu Pro Ala Glu Glu Lys Val
16 Val Leu Val Gly His Ser Leu Gly Gly Val Thr Leu Ala Leu Ala Gly
21 Asp Lys Phe Pro His Lys Ile Ser Val Ala Val Phe Val Thr Ala Phe
26 Met Pro Asp Thr Thr His Arg Pro Ser Phe Val Leu Glu Glu Tyr Ser
31 Glu Lys Met Gly Lys Asp Asp Ser Trp Leu Asp Thr Glu Phe Ser
36 Glu Cys Asp Ala Ser Asn Pro Ser His Ile Ser Met Leu Phe Gly Arg
41 Glu Phe Leu Thr Ile Lys Ile Tyr Glu Leu Cys Pro Pro Glu Asp Leu
46 Glu Leu Ala Lys Met Leu Val Arg Pro Gly Ser Met Phe Ile Asp Asn
51 Leu Ser Lys Glu Ser Lys Phe Ser Asp Glu Gly Tyr Gly Ser Val Lys
56 Arg Val Tyr Leu Val Cys Glu Glu Asp Ile Gly Leu Pro Lys Glu Phe
61 Gin His Trp Met Ile Gin Asn Tyr Pro Val Asn Glu Glu Val Met Glu Ile
66 Lys Gly Gly Asp His Met Ala Met Leu Ser Asp Pro Gin Lys Leu Cys
71 Asp Cys Leu Ser Gin Ile Ser Leu Lys Tyr Ala
76

<210> SEQ ID NO 33
<211> LENGTH: 263
<212> TYPE: PRT
<213> ORGANISM: Arabidopsis thaliana
<220> FEATURE: misc_feature
<223> OTHER INFORMATION: CeresClone:1010900
<400> SEQUENCE: 33

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Cys His Gly Ala Trp Cys Trp Tyr Lys Val Lys Pro Leu Leu Glu Ala
20 25 30
Val Gly His Arg Val Thr Ala Val Asp Leu Ala Ala Ser Gly Ile Asp
35 40 45
Thr Thr Arg Ser Ile Thr Asp Ile Pro Thr Cys Glu Gin Tyr Ser Glu
50 55 60
Pro Leu Thr Lys Leu Leu Thr Ser Leu Pro Asn Asp Glu Lys Val Val
65 70 75 80
Leu Val Gly His Ser Phe Glu Glu Leu Asn Leu Ala Ile Ala Met Glu
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<210> SEQ ID NO 34
<211> LENGTH: 263
<212> TYPE: PRT
<213> ORGANISM: Arabidopsis thaliana
<220> FRAGMENT:
<221> NAME/KEY: misc.feature
<222> OTHER INFORMATION: gi[20]146998

<400> SEQUENCE: 34

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Cys  His  Gly  Ala  Trp  Cys  Trp  Tyr  Lys  Val  Lys  Pro  Leu  Leu  Glu  Ala
20   25    30  
Val  Gly  His  Arg  Val  Thr  Ala  Val  Asp  Leu  Ala  Ala  Ser  Gly  Ile  Asp
35   40    45  
Thr  Thr  Arg  Ser  Ile  Thr  Asp  Ile  Pro  Thr  Cys  Glu  Glu  Tyr  Ser  Glu
50   55    60  
Pro  Leu  Thr  Lys  Leu  Leu  Thr  Ser  Leu  Pro  Asn  Asp  Glu  Lys  Val  Val
65   70    75   80  
Leu  Val  Gly  His  Ser  Phe  Gly  Leu  Asn  Leu  Ala  Ala  Met  Glu 85   90   95  
Lys  Phe  Pro  Glu  Lys  Ile  Ser  Val  Ala  Val  Phe  Thr  Ala  Phe  Met
100  105   110  
Pro  Asp  Thr  Glu  His  Ser  Pro  Ser  Phe  Val  Leu  Asp  Lys  Phe  Gly  Ser
115  120   125  
Asn  Met  Pro  Gln  Glu  Ala  Trp  Met  Gly  Thr  Glu  Phe  Glu  Pro  Tyr  Gly
130  135   140  
Ser  Asp  Asn  Ser  Gly  Leu  Ser  Met  Phe  Phe  Ser  Pro  Asp  Phe  Met  Lys
145  150   155  160  
Leu  Gly  Leu  Tyr  Gln  Leu  Ser  Pro  Val  Glu  Asp  Leu  Glu  Leu  Gly  Leu
165  170   175  

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Leu Leu Met Arg Pro Gly Ser Leu Phe Ile Asn Asp Leu Ser Lys Met

Lys Asn Phe Ser Asp Glu Gly Tyr Gly Ser Val Pro Arg Val Phe Ile

Val Cys Lys Glu Asp Lys Ala Ile Pro Glu Gly Arg Glu Arg Trp Met

Ile Asp Asn Phe Pro Val Asn Leu Val Met Glu Met Glu Glu Thr Asp

His Met Pro Met Phe Cys Lys Pro Glu Gln Leu Ser Asp Tyr Phe Leu

Lys Ile Ala Arg Lys Phe Val

<210> SEQ ID NO 35
<211> LENGTH: 263
<212> TYPE: PRT
<213> ORGANISM: Arabidopsis thaliana
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi[27]754457

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Val Gly His Arg Val Thr Ala Val Asp Leu Ala Ala Ser Gly Ile Asp
35 40 45
Thr Thr Arg Ser Ile Thr Asp Ile Pro Thr Cys Glu Gln Tyr Ser Glu
50 55 60
Pro Leu Thr Lys Leu Leu Thr Ser Leu Pro Asn Asp Glu Lys Val Val
65 70 75 80
Leu Val Gly His Ser Phe Gly Gln Met Leu Asn Leu Ala Ala Ser Gly Met
85 90 95
Lys Phe Pro Lys Lys Ile Ser Val Ala Val Phe Leu Thr Ala Phe Leu
100 105 110
Pro Asp Thr Glu His Ser Pro Ser Phe Val Leu Asp Lys Phe Gly Ser
115 120 125
Asn Met Pro Glu Ala Trp Met Gly Thr Gly Phe Glu Pro Tyr Gyl
130 135 140
Ser Asp Asn Ser Gly Leu Ser Met Phe Phe Ser Pro Asp Phe Met Lys
145 150 155 160
Leu Gly Leu Tyr Gln Leu Ser Pro Val Glu Asp Leu Glu Leu Gly Leu
165 170 175
Leu Leu Met Arg Pro Gly Ser Leu Phe Ile Asn Asp Leu Ser Lys Met
180 185 190
Lys Asn Phe Ser Asp Glu Gly Tyr Gly Ser Val Pro Arg Val Phe Ile
195 200 205
Val Cys Lys Glu Asp Lys Ala Ile Pro Glu Gly Arg Glu Arg Trp Met
210 215 220
Ile Asp Asn Phe Pro Val Asn Leu Val Met Glu Met Glu Glu Thr Asp
225 230 235 240
His Met Pro Met Phe Cys Lys Pro Glu Gln Leu Ser Asp Tyr Phe Leu
Lys Ile Ala Asp Lys Phe Val
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<210> SEQ ID NO 36
<211> LENGTH: 257
<212> TYPE: PRT
<213> ORGANISM: Hevea brasiliensis
<220> FEATURE:
<221> NAME KEY: misc feature
<223> OTHER INFORMATION: gi[64]35646
<400> SEQUENCE: 36

Met Ala Phe Ala His Phe Val Leu Ile His Thr Ile Cys His Gly Ala
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Trp Ile Trp His Lys Leu Lys Pro Leu Leu Glu Ala Leu Gly His Lys
20 25 30
Val Thr Ala Leu Asp Leu Ala Ala Ser Gly Val Asp Pro Arg Gln Ile
35 40 45
Glu Glu Ile Gly Ser Phe Asp Glu Tyr Ser Glu Pro Leu Leu Thr Phe
50 55 60
Leu Glu Ala Leu Pro Pro Gly Glu Lys Val Ile Leu Val Gly Glu Ser
65 70 75 80
Cys Gly Gly Leu Asn Ile Ala Ala Ala Asp Tyr Cys Glu Lys
85 90 95
Ile Ala Ala Ala Val Phe His Asn Ser Val Leu Pro Asp Thr Glu His
100 105 110
Cys Pro Ser Tyr Val Val Asp Leu Met Glu Val Phe Pro Asp Trp
115 120 125
Lys Asp Thr Thr Tyr Phe Thr Tyr Thr Tyr Lys Asp Gly Lys Glu Ile Thr
130 135 140
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145 150 155 160
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165 170 175
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180 185 190
Gly Tyr Gly Ser Ile Lys Lys Ile Tyr Val Trp Thr Asp Glu Asp Glu
195 200 205
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210 215 220
Asp Lys Val Tyr Lys Val Glu Gly Gly Asp His Leu Leu Gln Leu Thr
225 230 235 240
Lys Thr Lys Glu Ile Ala Glu Ile Leu Gln Glu Val Ala Asp Thr Tyr
245 250 255
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Val Thr Ala Leu Asp Leu Ala Ala Ser Gly Val Asp Pro Arg Gin Ile
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Glu Gin Ile Gly Ser Phe Asp Glu Tyr Ser Glu Pro Leu Leu Thr Phe
50  55  60
Leu Glu Ala Leu Pro Pro Gly Glu Lys Val Ile Leu Val Gly Glu Ser
65  70  75  80
Cys Gly Gly Leu Asn Ile Ala Ala Ala Asp Lys Tyr Cys Glu Lys
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Lys Asp Thr Thr Tyr Phe Thr Tyr Thr Lys Asp Gly Lys Glu Ile Thr
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Gly Leu Lys Leu Gly Phe Thr Leu Leu Arg Glu Asn Leu Tyr Thr Lys
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Cys Gly Pro Glu Glu Tyr Glu Leu Ala Lys Met Leu Thr Arg Lys Gly
165  170  175
Ser Leu Phe Gin Asn Ile Ala Ala Lys Arg Pro Phe Phe Thr Lys Glu
180  185  190
Gly Tyr Gly Ser Ile Lys Lys Ile Tyr Val Thr Asp Gin Asp Glu
195  200  205
Ile Phe Leu Pro Glu Phe Gin Leu Trp Gin Ile Gin Asn Tyr Thr Pro
210  215  220
Asp Lys Val Tyr Lys Val Glu Gly Gly Asp His Lys Leu Gin Leu Thr
225  230  235  240
Lys Thr Lys Gin Ile Ala Glu Ile Leu Gin Glu Val Ala Asp Thr Tyr
245  250  255
Asn
-continued

Cys Gly Gly Leu Asn Ile Ala Ile Ala Ala Asp Lys Tyr Pro Glu Lys
  95  90 95
Ile Ala Ala Ala Val Phe Gln Asn Ser Leu Leu Pro Asp Thr Lys His
  100 105 110
Lys Pro Ser Tyr Val Val Asp Tyr Leu Met Glu Val Phe Pro Asp Trp
  115 120 125
Lys Asp Thr Glu Tyr Phe Glu Phe Ser Asn Ser Asn Gly Glu Thr Ile
  130 135 140
Thr Gly Met Val Leu Gly Leu Leu Met Arg Glu Asn Leu Tyr Thr
  145 150 155 160
Ile Cys Pro Pro Glu Asp Tyr Glu Leu Ala Lys Met Leu Thr Arg Arg
  165 170 175
Gly Ser Leu Phe Gln Ser Ile Leu Ala Gin Arg Glu Lys Phe Thr Glu
  180 185 190
Lys Gly Tyr Gly Ser Ile Lys Lys Ile Tyr Val Trp Thr Gly Asp Asp
  195 200 205
Lys Ile Phe Leu Pro Glu Phe Gin Leu Trp Gin Ile Glu Asn Tyr Lys
  210 215 220
Pro Asp Leu Val Phe Arg Val Met Gly Gly Asp His Leu Gin Leu
  225 230 235 240
Thr Lys Thr Asn Glu Ile Ala Gly Ile Leu Gin Lys Val Ala Asp Ile
  245 250 255 260

Tyr Ala
-continued

Leu Ser Pro Ile Glu Asp His Ala Leu Gly Lys Ile Leu Val Arg Pro 165 170 175
Gly Ser Leu Phe Ile Glu Asp Leu Leu Lys Ala Glu Lys Phe Thr Glu 180 185 190
Glu Gly Phe Gly Ser Val Pro Arg Val Tyr Val Ile Ala Ala Glu Asp 195 200 205
Lys Thr Ile Pro Pro Glu Phe Gln Arg Trp Met Ile Glu Asn Asn Pro 210 215 220
Val Lys Glu Val Lys Glu Ile Gly Ala Asp His Met Pro Met Phe 225 230 235 240
Ser Lys Pro Asp Glu Leu Ser Glu Cys Leu Leu Asp Ile Ala Lys Lys 245 250 255 260

His Ala

<210> SEQ ID NO 40
<211> LENGTH: 264
<212> TYPE: PRT
<213> ORGANISM: Rauvolfia serpentina
<220> FEATURE:
<221> NAME/KEY: misc.feature
<222> OTHER INFORMATION: gi|5681393

<400> SEQUENCE: 40
Met His Ser Ala Ala Asn Ala Lys Gln Gln Lys His Phe Val Leu Val 1 5 10 15
His Gly Gly Cys Leu Gly Ala Trp Ile Trp Tyr Lys Leu Lys Pro Leu 20 25 30
Leu Glu Ser Ala Gly His Lys Val Thr Ala Val Asp Leu Ser Ala Ala 35 40 45
Gly Ile Asn Pro Arg Arg Leu Asp Glu Ile His Thr Phe Arg Asp Tyr 50 55 60
Ser Glu Pro Leu Met Glu Val Met Ala Ser Ile Pro Pro Asp Glu Lys 65 70 75 80
Val Val Leu Leu Gly His Ser Phe Gly Gly Met Ser Leu Gly Leu Ala 85 90 95
Met Glu Thr Tyr Pro Glu Lys Ile Ser Val Ala Val Phe Met Ser Ala 100 105 110
Met Met Pro Asp Pro Asn His Ser Leu Thr Tyr Pro Phe Glu Lys Tyr 115 120 125
Asn Glu Lys Cys Pro Ala Asp Met Met Leu Asp Ser Glu Phe Ser Thr 130 135 140
Tyr Gly Asn Pro Glu Asn Pro Gly Met Ser Met Ile Leu Gly Pro Gln 145 150 155 160
Phe Met Ala Leu Lys Met Phe Gln Asn Cys Ser Val Glu Asp Leu Glu 165 170 175
Leu Ala Lys Met Leu Thr Arg Pro Gly Ser Leu Phe Phe Gln Asp Leu 180 185 190
Ala Lys Ala Lys Lys Phe Ser Thr Glu Arg Tyr Glu Ser Val Lys Arg 195 200 205
Ala Tyr Ile Phe Cys Asn Glu Asp Lys Ser Phe Pro Val Glu Phe Gln 210 215 220
Lys Trp Phe Val Glu Ser Val Gly Ala Asp Lys Val Lys Glu Ile Lys 225 230 235 240
Glu Ala Asp His Met Gly Met Leu Ser Gln Pro Arg Glu Val Cys Lys
245
250
Cys Leu Leu Asp Ile Ser Asp Ser
260

<210> SEQ ID NO 41
<211> LENGTH: 262
<212> TYPE: PRT
<213> ORGANISM: Lycopersicon esculentum
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi[41]814856

<400> SEQUENCE: 41

Met Glu Lys Gly Asp Lys Asn His Phe Val Leu Val His Gly Ala Cys
1   5   10   15
His Gly Ala Trp Cys Trp Tyr Lys Val Val Thr Ile Leu Arg Ser Glu
20  25  30
Gly His Lys Val Ser Val Leu Asp Met Ala Ala Ser Gly Ile Asn Pro
35  40
45
Lys His Val Asp Asp Leu Asn Ser Met Ala Asp Tyr Asn Pro Glu Pro Leu
50  55  60
Met Glu Phe Met Asn Ser Leu Pro Glu Leu Glu Arg Val Val Leu Val
65  70  75  80
Gly His Ser Met Gly Gly Ile Asn Ile Ser Leu Ala Met Gly Lys Phe
85  90  95
Pro Gln Lys Ile Val Val Ala Val Phe Val Thr Ala Phe Met Pro Gly
100 105 110
Pro Asp Leu Asn Leu Val Ala Leu Gly Gln Gln Tyr Asn Gln Gln Val
115 120 125
Glu Ser His Met Asp Thr Glu Phe Val Tyr Asn Asn Gly Glu Asp Lys
130 135 140
Ala Pro Thr Ser Leu Val Leu Gly Pro Glu Val Leu Ala Thr Asn Phe
145 150 155 160
Tyr Glu Leu Ser Pro Pro Glu Asp Leu Thr Leu Ala Thr Tyr Leu Val
165 170 175
Arg Pro Val Pro Leu Phe Asp Glu Ser Ile Leu Leu Ala Asn Thr Thr
180 185 190
Leu Ser Lys Glu Lys Tyr Gly Ser Val His Arg Val Tyr Val Val Cys
195 200 205
Asp Lys Asp Asn Val Leu Lys Glu Gin Gin Phe Gin Lys Trp Leu Ile
210 215 220
Asn Asn Pro Pro Asp Glu Val Gin Ile Ile His Asn Ala Asp His
225 230 235 240
Met Val Met Phe Ser Lys Pro Arg Asp Leu Ser Ser Cys Leu Val Met
245 250 255
Ile Ser Gin Lys Tyr Tyr
260

<210> SEQ ID NO 42
<211> LENGTH: 260
<212> TYPE: PRT
<213> ORGANISM: Nicotiana tabacum
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi[40]549303

<400> SEQUENCE: 42

Met Lys Glu Gly Lys His Phe Val Leu Val His Gly Ala Cys His Gly
1  5 10 15
Gly Trp Ser Trp Tyr Lys Leu Lys Pro Leu Leu Glu Ala Ala Gly His
20 25 30
Lys Val Thr Ala Leu Asp Leu Ala Ala Ser Gly Thr Asp Leu Arg Lys
35 40 45
Ile Glu Glu Leu Arg Thr Leu Tyr Asp Tyr Thr Pro Leu Met Glu
50 55 60
Leu Met Glu Ser Leu Ser Ala Asp Glu Val Ile Leu Val Gly His
65 70 75 80
Ser Leu Gly Glu Met Asn Leu Gly Leu Ala Met Glu Lys Tyr Pro Gln
85 90 95
Lys Ile Tyr Ala Ala Val Phe Leu Ala Ala Phe Met Pro Asp Ser Val
100 105 110
His Asn Ser Ser Phe Val Leu Glu Gln Tyr Asn Glu Arg Thr Pro Ala
115 120 125
Glu Asn Trp Leu Asp Thr Gln Phe Leu Pro Tyr Ser Pro Glu Glu
130 135 140
Pro Leu Thr Ser Met Phe Phe Gly Pro Lys Phe Leu Ala His Lys Leu
145 150 155 160
Tyr Gln Leu Cys Ser Pro Glu Asp Ala Leu Ala Ser Ser Leu Val
165 170 175
Arg Pro Ser Ser Leu Phe Met Glu Asp Leu Ser Lys Ala Lys Tyr Phe
180 185 190
Thr Asp Glu Arg Phe Gly Ser Val Lys Arg Val Tyr Ile Val Cys Thr
195 200 205
Glu Asp Lys Gly Ile Pro Glu Phe Gln Arg Trp Gln Ile Asp Asn
210 215 220
Ile Gly Val Thr Glu Ala Ile Glu Ile Lys Gly Ala Asp His Met Ala
225 230 235 240
Met Leu Cys Glu Pro Gln Lys Leu Cys Ala Ser Leu Leu Glu Ile Ala
245 250 255
His Lys Tyr Asn
260

<210> SEQ ID NO 43
<211> LENGTH: 262
<212> TYPE: PRT
<213> ORGANISM: Solanum tuberosum
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi[56]392765

<400> SEQUENCE: 43

Met Glu Lys Gly Asn Lys Asn His Phe Val Leu Val His Gly Ala Cys
1  5 10 15
His Gly Ala Trp Cys Trp Tyr Lys Val Val Thr Ile Leu Arg Ser Glu
20 25 30
Gly His Lys Val Ser Val Leu Asp Met Ala Ala Ser Gly Ile Asn Pro
35 40 45
Lys His Val Glu Asp Leu Asn Ser Met Ala Asp Tyr Asn Glu Pro Leu
Met Glu Phe Met Asn Ser Leu Pro Gln Gln Glu Arg Val Val Leu Val
50 70 75 80
Gly His Ser Met Gly Gly Ile Asn Ile Ser Leu Ala Met Glu Lys Phe
95 90 95
Pro His Lys Ile Ala Val Ala Val Phe Val Ser Ala Ser Met Pro Gly
100 105 110
Pro Asp Leu Asn Leu Val Ala Thr Gin Gin Tyr Ser Gin Gin Gin Val
115 120 125
Glu Thr Pro Met Asp Thr Glu Val Tyr Asn Asn Gly Leu Asp Lys
130 135 140
Gly Pro Thr Ser Val Val Leu Gly Pro Lys Val Leu Ala Thr Ile Tyr
145 150 155 160
Tyr Gin Phe Ser Pro Pro Glu Asp Leu Thr Leu Ala Thr Tyr Leu Val
165 170 175
Arg Pro Val Pro Leu Phe Asp Glu Ser Val Leu Thr Asn Thr Thr
190 195 190
Leu Ser Lys Glu Lys Tyr Gly Ser Val His Arg Val Tyr Val Val Cys
195 200 205
Asp Lys Asp Lys Val Leu Lys Glu Glu Glu Gin Arg Trp Leu Ile
210 215 220
Lys Asn Asn Pro Pro Asn Glu Val Gin Met Ile His Asp Ala Gly His
225 230 235 240
Met Val Met Phe Ser Lys Pro Arg Glu Leu Cys Ser Cys Leu Val Met
245 250 255
Ile Ser Gin Lys Tyr His
260

<210> SEQ ID NO 44
<211> LENGTH: 266
<212> TYPE: PRT
<213> ORGANISM: Triticum aestivum
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> OTHER INFORMATION: CeresClone:644331

<400> SEQUENCE: 44
Met Glu Ala Cys Ala Gly Gln Ala Ser Ser Ala His Ile Val Leu Val
1 5 10 15
His Gly Ala Cys Leu Gly Gly Trp Ser Trp Phe Lys Val Ala Thr Arg
20 25 30
Leu Arg Ser Ala Gly His Arg Val Ser Thr Pro Asp Leu Ala Ala Ser
35 40 45
Gly Val Asp Pro Arg Pro Leu Arg Glu Val Pro Thr Phe Arg Asp Tyr
50 55 60
Thr Lys Pro Leu Leu Asp Leu Leu Glu Ser Leu Pro Ser Gly Glu Lys
65 70 75 80
Val Val Leu Val Gly His Ser Leu Gly Val Asn Val Ala Leu Ala
85 90 95
Cys Glu Leu Phe Pro Glu Lys Ile Ala Ala Ala Val Phe Val Ala Ala
100 105 110
Phe Met Pro Asp His Arg Ser Pro Pro Ser Tyr Val Leu Glu Lys Phe
115 120 125
<210> SEQ ID NO 45
<211> LENGTH: 265
<212> TYPE: PRT
<213> ORGANISM: Triticum aestivum
<220> FEATURE: 
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: CeresClone:936068

<400> SEQUENCE: 45
Met Glu Gly Ser Ser Ser Gly His Phe Ile Leu Ile His Gly Leu 1 5 10 15
Cys His Gly Ala Trp Cys Trp Tyr Lys Leu Val Pro Met Leu Arg Ala 20 25 30
Ala Gly His Arg Val Thr Ala Leu Asp Met Ala Ala Ser Gly Ala His 35 40 45
Pro Ala Arg Met Asp Glu Val Pro Ser Phe Glu Asp Tyr Ser Trp Pro 50 55 60
Leu Leu Asp Ala Val Ala Ala Val Gly Glu Arg Leu Val Leu 65 70 75 80
Val Gly His Ser Leu Gly Leu Asn Ile Ala Leu Ala Met Glu Arg 95 100 105 110
Phe Pro Arg Lys Val Ala Ala Val Phe Leu Ala Ala Cys Met Pro 115 120 125
Cys Val Gly Arg His Met Gly Ala Thr Thr Glu Glu Ile Met Arg Arg 130 135 140
Ile Lys Pro Asp Phe Phe Met Asp Met Lys Arg Met Val Leu Asn Thr 145 150 155 160
Ser Glu Gly Pro Arg Pro Ala Leu Val Phe Gly Pro Lys Ile Leu Ala 165 170 175
Ala Lys Leu Tyr Asp Arg Ser Ser Gly Glu Asp Gin Thr Leu Ala Thr 180 185 190
Met Leu Val Arg Pro Gly Cys Gin Phe Leu Asp Asp Pro Thr Met Lys 195 200 205
Asp Glu Ala Leu Leu Thr Glu Ala Lys Tyr Gly Ser Val Lys Lys Val
Tyr Val Val Met Ala Asp Ala Ser Asn Ser Glu Glu Met Gln Arg
210 215 220

Trp Met Val Asp Met Ser Pro Gly Thr Glu Ala Glu Glu Ile Ala Gly
225 230 235 240

Ala Asp His Met Ala Met Cys Ser Lys Pro Arg Glu Lys Asp Val
245 250 255

Leu Leu Arg Ile Ala Asp Lys Tyr Glu
260 265

<210> SEQ ID NO 46
<211> LENGTH: 268
<212> TYPE: PRT
<213> ORGANISM: Oryza sativa subsp. japonica
<220> FEATURE:
<221> ... Met Ala Met Cys Ser Lys Pro Arg Glu Lys Asp Val
245 250 255
<400> SEQUENCE: 46

Met Glu Ile Ser Ser Ser Ser Lys His Phe Ile Leu Val His Gly
1  5  10  15

Leu Cys His Gly Ala Trp Cys Trp Tyr Arg Val Val Ala Ala Leu Arg
20 25 30

Ala Ala Gly His Arg Ala Thr Ala Leu Asp Met Ala Ala Ser Gly Ala
35 40 45

His Pro Ala Arg Val Asp Glu Val Gly Thr Phe Glu Glu Tyr Ser Arg
50 55 60

Pro Leu Leu Asp Ala Val Ala Ala Ala Ala Pro Gly Glu Arg Leu
65 70 75 80

Val Leu Val Gly His Ser His Gly Leu Ser Val Ala Leu Ala Met
85 90 95

Glu Arg Phe Pro Asp Lys Val Ala Ala Val Phe Val Ala Ala Ala
100 105 110

Met Pro Cys Val Gly Lys His Met Gly Val Pro Thr Glu Glu Phe Met
115 120 125

Arg Arg Thr Ala Pro Glu Leu Leu Met Asp Cys Met Met Val Ala
130 135 140

Ile Asn Asn Ser Glu Gly Ser Gly Val Ala Ile Asn Leu Gly Pro Thr
145 150 155 160

Phe Leu Ala Gln Lys Tyr Tyr Gln Glu Ser Pro Ala Glu Asp Leu Ala
165 170 175

Leu Ala Lys Met Leu Val Arg Pro Gly Asn Gin Phe Met Asp Arg Pro
180 185 190

Val Met Lys Asp Glu Ser Leu Thr Asn Gly Tyr Gly Ser Val
195 200 205

Lys Lys Val Tyr Val Ile Ala Lys Ala Asp Ser Ser Ser Thr Glu Glu
210 215 220

Met Gln Arg Trp Met Val Ala Met Ser Pro Gly Thr Asp Val Glu Glu
225 230 235 240

Ile Ala Gly Ala Asp His Ala Val Met Asn Ser Lys Pro Arg Glu Leu
245 250 255

Cys Asp Ile Leu Ile Lys Ala Asn Lys Tyr Glu
260 265
<210> SEQ ID NO 47
<211> LENGTH: 262
<212> TYPE: PRT
<213> ORGANISM: Oryza sativa (japonica cultivar-group)
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi[57]899620

<400> SEQUENCE: 47

Met Glu Gly Ser Ser Ser Ser Ser Ser Ser Ser Ser Ser Ser Ser Leu Val Val Leu Val His Gly
1   5   10   15
Leu Cys His Gly Ala Trp Cys Trp Tyr Lys Val Val Thr Met Leu Arg
20  25  30
Ser Glu Gly His Arg Val Thr Ala Leu Gln Leu Leu Ala Ser Gly Val
35  40  45
His Pro Ala Arg Val Asp Glu Val His Ser Phe Glu Glu Tyr Ser Gln
50  55  60
Pro Leu Leu Asp Ala Val Ala Glu Ala Pro Ala Gly Glu Arg Leu Ile
65  70  75  80
Leu Val Gly His Ser Phe Gly Leu Ser Ser Ile Ala Leu Ala Ala Met Glu
85  90  95
Arg Phe Pro Glu Lys Ile Ala Val Ala Val Phe Val Ala Ala Ala Val
100 105 110
Pro Cys Val Gly Lys Arg Ile Ile Pro Glu Leu Ile Arg Glu Lys Ala
115 120 125
Pro Lys Asp Met Leu Leu Asp Ser Lys Met Ile Pro Ile Asn Asn Lys
130 135 140
Gln Gly Pro Gly Thr Ala Ile Leu Leu Gly Pro Asn Phe Leu Ala Glu
145 150 155 160
Lys Gly Tyr Pro Leu Ser Pro Ala Glu Asp Leu Thr Leu Ala Lys Leu
165 170 175
Leu Val Arg Pro Thr Ser Glu Phe Val Asp Asp Pro Thr Met Lys Asp
180 185 190
Asp Arg Leu Leu Thr Ser Ala Asn Tyr Gly Ser Val Lys Arg Val Cys
195 200 205
Leu Met Ala Met Glu Asp Leu Lys Glu Val His Arg Tyr Met Ile
210 215 220
Thr Leu Ser Pro Gly Val Glu Val Glu Ile Ala Gly Ala Asp His
225 230 235 240
Ala Val Met Cys Ser Arg Pro Arg Glu Leu Ser Asp Leu Leu Ala Lys
245 250 255
Ile Gly Ser Lys Tyr Asp
260

<210> SEQ ID NO 48
<211> LENGTH: 265
<212> TYPE: PRT
<213> ORGANISM: Capsella rubella
<220> FEATURE:
<221> NAME/KEY: misc_feature
<223> OTHER INFORMATION: gi[15]866583

<400> SEQUENCE: 48

Met Gly Gly Asp Gly Gly Ala Glu Gln Pro Val Ile His Phe Val Phe
1   5   10   15
Val His Gly Ala Ser His Gly Ala Trp Cys Trp Tyr Lys Leu Thr Ser
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<210> SEQ ID NO 49
<211> LENGTH: 265
<212> TYPE: PRT
<213> ORGANISM: Lycopersicon hirsutum f. glabratum
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> OTHER INFORMATION: gi:[56]393011
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (46)...(46)
<223> OTHER INFORMATION: Xaa can be any naturally occurring amino acid

<400> SEQUENCE: 49
Met Glu Lys Ser Met Ser Pro Phe Val Lys Lys His Phe Val Leu Val
His Thr Ala Phe His Gly Ala Trp Cys Trp Tyr Lys Ile Val Ala Leu
Met Arg Ser Ser Gly His Asn Val Thr Ala Leu Asp Leu Xaa Ala Ser
Gly Ile Asn Pro Lys Gln Ala Leu Gln Ile Pro Asn Phe Ser Asp Tyr
Leu Ser Pro Leu Met Glu Phe Met Ala Ser Leu Pro Ala Asn Glu Lys
What is claimed is:

1. An isolated nucleic acid molecule comprising:
   a) a nucleic acid having a nucleotide sequence which encodes an amino acid sequence exhibiting at least 85% sequence identity to any one of those sequences present in the Sequence Listing;
   b) a nucleic acid which is a complement of a nucleotide sequence according to paragraph (a);
   c) a nucleic acid which is the reverse of the nucleotide sequence according to subparagraph (a), such that the reverse nucleotide sequence has a sequence order which is the reverse of the sequence order of the nucleotide sequence according to subparagraph (a); or
   d) a nucleic acid capable of hybridizing to a nucleic acid according to any one of paragraphs (a)-(c), under conditions that permit formation of a nucleic acid duplex at a temperature from about 40°C and 48°C below the melting temperature of the nucleic acid duplex.

2. The isolated nucleic acid molecule according to claim 1, which has the nucleotide sequence according to any one of those sequences present in the Sequence Listing.

3. The isolated nucleic acid molecule according to claim 1, wherein said amino acid sequence comprises a polypeptide according to any one of the consensus sequences set forth in Tables 1-5, 2-6, 3-5 or 4-1.

4. The isolated nucleic acid molecule according to claim 1, wherein said amino acid sequence has a sequence according to any one of those sequences present in the Sequence Listing.

5. A vector construct comprising:
   a) a first nucleic acid having a regulatory sequence capable of causing transcription and/or translation in a plant; and
   b) a second nucleic acid having the sequence of the isolated nucleic acid molecule according to claim 1;
   wherein said first and second nucleic acids are operably linked and
   wherein said second nucleic acid is heterologous to any element in said vector construct.

6. The vector construct according to claim 5, wherein said first nucleic acid is native to said second nucleic acid.

7. The vector construct according to claim 5, wherein said first nucleic acid is heterologous to said second nucleic acid.

8. A host cell comprising an isolated nucleic acid molecule according to claim 1 wherein said nucleic acid molecule is flanked by exogenous sequence.

9. A host cell comprising a vector construct according to claim 5.

10. An isolated polypeptide comprising an amino acid sequence exhibiting at least 85% sequence identity to any of those sequences present in the Sequence Listing.

11. A method of introducing an isolated nucleic acid into a host cell comprising:
   a) providing an isolated nucleic acid molecule according to claim 1; and
   b) contacting said isolated nucleic acid with said host cell under conditions that permit insertion of said nucleic acid into said host cell.

12. A method of transforming a host cell that comprises contacting a host cell with a vector construct according to claim 5.
13. A method for detecting a nucleic acid in a sample which comprises:
   a) providing an isolated nucleic acid molecule according to claim 1;
   b) contacting said isolated nucleic acid molecule with a sample under conditions which permit a comparison of
      the sequence of said isolated nucleic acid molecule with the sequence of DNA in said sample; and
   c) analyzing the result of said comparison.
14. A plant, plant cell, plant material or seed of a plant which comprises a nucleic acid molecule according to claim
    1 which is exogenous or heterologous to said plant or plant cell.
15. A plant, plant cell, plant material or seed of a plant which comprises a vector construct according to claim 5.
16. A plant that has been regenerated from a plant cell or seed according to claim 14.
17. A plant, plant cell, plant material or seed of a plant which comprises a nucleic acid molecule according to claim
    1, wherein said plant has improved pH tolerance or phosphate use efficiency characteristics as compared to a wild-type plant
    cultivated under the same conditions.
18. A method for increasing pH tolerance or phosphate use efficiency in a plant comprising transforming a plant with a
    nucleic acid sequence according to claim 1.
19. A transgenic plant having a gene construct comprising a nucleic acid encoding a pH tolerance or phosphate use
    efficiency component operably linked to a plant promoter so that the pH tolerance or phosphate use efficiency component
    is ectopically overexpressed in the transgenic plant, and the transgenic plant exhibits:
   i) faster rate of growth,
   ii) greater fresh or dry weight at maturation,
   iii) greater fruit or seed yield,
   iv) higher tolerance to pH,
   v) higher tolerance to low phosphate concentration, or
   vi) higher tolerance to low nitrogen concentration
    than a progenitor plant which does not contain the polynucleotide construct, when the transgenic plant and the
    progenitor plant are cultivated under identical environmental conditions, wherein the pH or phosphate use
    efficiency component is any one of the polypeptides set forth in the Sequence Listing, or any one of the consensus
    sequences in claim 3.
20. A method for pH tolerance or phosphate use efficiency in a plant which comprises transforming a plant with a nucleic
    acid sequence that encodes a polypeptide that comprises at least one of the following:
   (a) an amino acid sequence that comprises the residues at positions 29-154 of the consensus sequence of Table
       1-5,
   (b) an amino acid sequence that comprises the residues at positions 18-128 of the consensus sequence of Table
       2-6,
   (c) an amino acid sequence that comprises the residues at positions 57-230 of the consensus sequence of Table
       3-5,
   (d) an amino acid sequence that comprises the residues at positions 234-248 of the consensus sequence of Table
       3-5, and
   (e) an amino acid sequence that comprises the residues at positions 10-276 of the consensus sequence of Table
       4-1.
21. A plant, plant cell, plant material of a plant with improved pH tolerance or phosphate use efficiency characteristics as
    compared to a wild-type plant cultivated under the same conditions which comprises a nucleic acid sequence
    that encodes a polypeptide that comprises at least one of the following:
   (a) an amino acid sequence that comprises the residues at positions 29-154 of the consensus sequence of Table
       1-5,
   (b) an amino acid sequence that comprises the residues at positions 18-128 of the consensus sequence of Table
       2-6,
   (c) an amino acid sequence that comprises the residues at positions 57-230 of the consensus sequence of Table
       3-5,
   (d) an amino acid sequence that comprises the residues at positions 234-248 of the consensus sequence of Table
       3-5, and
   (e) an amino acid sequence that comprises the residues at positions 10-276 of the consensus sequence of Table
       4-1.