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(54) PROMOTER, PROMOTER CONTROL ELEMENTS, AND COMBINATIONS, AND USES THEREOF

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See application file for complete search history.

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(57) ABSTRACT

The present invention is directed to promoter sequences and promoter control elements, polynucleotide constructs comprising the promoters and control elements, and methods of identifying the promoters, control elements, or fragments thereof. The invention further relates to the use of the present promoters or promoter control elements to modulate transcript levels in plants, and plants containing such promoters or promoter control elements.

25 Claims, 1 Drawing Sheet

Specification includes a Sequence Listing.

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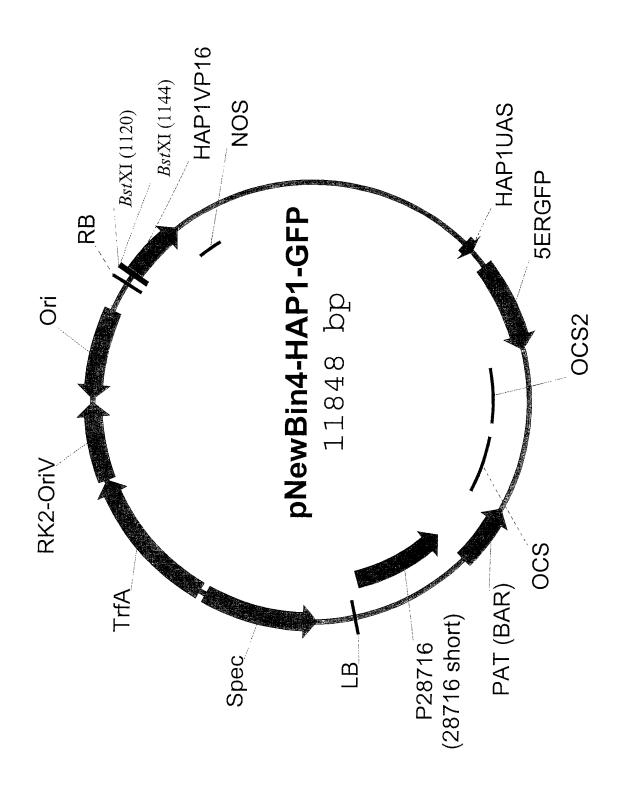
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PROMOTER, PROMOTER CONTROL ELEMENTS, AND COMBINATIONS, AND USES THEREOF

CROSS-REFERENCE TO RELATED APPLICATIONS

This application is a Divisional of application Ser. No. 16/912,222 filed Jun. 25, 2020, now U.S. Pat. No. 11,572, 567, which application is a Divisional of application Ser. No. 15/691,458 filed Aug. 30, 2017, now U.S. Pat. No. 10,815, 490, which is a Divisional of application Ser. No. 14/476, 566 filed on Sep. 3, 2014, now U.S. Pat. No. 9,777,285, which application is a Divisional of application Ser. No. 12/865,719 filed on Apr. 19, 2011 (now abandoned), which is a National Phase of PCT International Application No. PCT/US2009/032485 filed on Jan. 29, 2009, which claims priority to U.S. Provisional Application Ser. No. 61/025,697, filed on Feb. 1, 2008. All of the above applications are hereby expressly incorporated by reference into the present ²⁰ application.

INCORPORATION-BY-REFERENCE OF SEQUENCE LISTING OR TABLE

The material in the accompanying sequence listing is hereby incorporated by reference into this application. The accompanying file, named CRES005USD6_ST26.xml was created on Dec. 6, 2022 and is 153 KB (size as measured in Microsoft Windows® operating system).

FIELD OF THE INVENTION

The present invention relates to promoters and promoter control elements that are useful for modulating transcription of a desired polynucleotide. Such promoters and promoter control elements can be included in polynucleotide constructs, expression cassettes, vectors, or inserted into the chromosome or as an exogenous element, to modulate in vivo and in vitro transcription of a polynucleotide. Host cells, including plant cells, and organisms, such as regenerated plants therefrom, with desired traits or characteristics using polynucleotides comprising the promoters and promoter control elements of the present invention are also a part of the invention.

BACKGROUND OF THE INVENTION

This invention relates to promoter sequences and promoter control element sequences which are useful for the 50 transcription of polynucleotides in a host cell or transformed host organism.

The introduction of genes into plants has resulted in the development of plants having new and useful phenotypes such as pathogen resistance, higher levels of healthier types 55 of oils, novel production of healthful components such as beta-carotene synthesis in rice. An introduced gene is generally a chimeric gene composed of the coding region that confers the desired trait and regulatory sequences. One regulatory sequence is the promoter, which is located 5' to 60 the coding region. This sequence is involved in regulating the pattern of expression of a coding region 3' thereof. The promoter sequence binds RNA polymerase complex as well as one or more transcription factors that are involved in producing the RNA transcript of the coding region.

The promoter region of a gene used in plant transformation is most often derived from a different source than is the 2

coding region. It may be from a different gene of the same species of plant, from a different species of plant, from a plant virus, an algae species, a fungal species, or it may be a composite of different natural and/or synthetic sequences. Properties of the promoter sequence generally determine the pattern of expression for the coding region that is operably linked to the promoter. Promoters with different characteristics of expression have been described. The promoter may confer broad expression as in the case of the widely-used cauliflower mosaic virus (CaMV) 35S promoter. The promoter may confer tissue-specific expression as in the case of the seed-specific phaseolin promoter. The promoter may confer a pattern for developmental changes in expression. The promoter may be induced by an applied chemical compound, or by an environmental condition applied to the plant.

The promoter that is used to regulate a particular coding region is determined by the desired expression pattern for that coding region, which itself is determined by the desired resulting phenotype in the plant. For example, herbicide resistance is desired throughout the plant so the 35S promoter is appropriate for expression of an herbicide-resistance gene. A seed-specific promoter is appropriate for changing the oil content of soybean seed. An endospermspecific promoter is appropriate for changing the starch composition of corn seed. A root-specific promoter can be important for improving water or nutrient up-take in a plant. Control of expression of an introduced gene by the promoter is important because it is sometimes detrimental to have expression of an introduced gene in non-target tissues. For example, a gene which induces cell death can be expressed in male and/or female gamete cells in connection with bioconfinement.

One of the primary goals of biotechnology is to obtain organisms, such as plants, mammals, yeast, and prokaryotes having particular desired characteristics or traits. Examples of these characteristics or traits abound and may include, for example, in plants, virus resistance, insect resistance, herbicide resistance, enhanced stability or additional nutritional value. Recent advances in genetic engineering have enabled researchers in the field to incorporate polynucleotide sequences into host cells to obtain the desired qualities in the 45 organism of choice. This technology permits one or more polynucleotides from a source different than the organism of choice to be transcribed by the organism of choice. If desired, the transcription and/or translation of these new polynucleotides can be modulated in the organism to exhibit a desired characteristic or trait. Alternatively, new patterns of transcription and/or translation of polynucleotides endogenous to the organism can be produced.

SUMMARY OF THE INVENTION

The present invention is directed to isolated polynucleotide sequences that comprise promoters and promoter control elements from plants, especially *Arabidopsis thaliana*, and other promoters and promoter control elements functional in plants.

It is an object of the present invention to provide isolated polynucleotides that are promoter or promoter control sequences. These promoter sequences comprise, for example,

(1) a polynucleotide having a nucleotide sequence according to any one of SEQ. ID. Nos. 1-26 or residues 601-1000 of SEQ ID NO: 26;

- (2) a polynucleotide having a nucleotide sequence having at least 80% sequence identity to a sequence according to SEQ. ID. Nos. 1-26 or residues 601-1000 of SEQ ID NO: 26; and
- (3) a polynucleotide having a nucleotide sequence which hybridizes to a sequence according to SEQ. ID. Nos. 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26 under a condition establishing a Tm-5° C.

Promoter or promoter control element sequences of the present invention are capable of modulating preferential transcription.

In another embodiment, the present promoter control elements are capable of serving as or fulfilling the function, for example, as a core promoter, a TATA box, a polymerase binding site, an initiator site, a transcription binding site, an enhancer, an inverted repeat, a locus control region, and/or a scaffold/matrix attachment region.

It is yet another object of the present invention to provide a polynucleotide that includes at least a first and a second 20 promoter control element. The first promoter control element is a promoter control element sequence as discussed above, and the second promoter control element is heterologous to the first control element; wherein, the first and second control elements are operably linked. Such promoters may modulate transcript levels preferentially in a particular tissue or under particular conditions.

In another embodiment, the present isolated polynucleotide comprises a promoter or a promoter control element as described above, wherein the promoter or promoter control element is operably linked to a polynucleotide to be transcribed.

In another embodiment of the present invention, the promoter and promoter control elements of the instant invention are operably linked to a heterologous polynucleotide that is a regulatory sequence.

It is another object of the present invention to provide a host cell comprising an isolated polynucleotide or vector as described above or fragment thereof. Host cells include, for instance, bacterial, yeast, insect, mammalian, fungus, algae, and plant. The host cell can comprise a promoter or promoter control element exogenous to the genome. Such a promoter can modulate transcription in cis- and in trans-.

In yet another embodiment, the host cell is a plant cell 45 capable of regenerating into a plant.

It is yet another embodiment of the present invention to provide a plant comprising an isolated polynucleotide or vector described above.

It is another object of the present invention to provide a 50 method of modulating transcription in a sample that contains either a cell-free system of transcription or host cell. This method comprises providing a polynucleotide or vector according to the present invention as described above, and contacting the sample of the polynucleotide or vector with 55 conditions that permit transcription.

In another embodiment of the present method, the polynucleotide or vector preferentially modulates, depending upon the function of the particular promoter, constitutive transcription, stress induced transcription, light induced 60 transcription, dark induced transcription, leaf transcription, root transcription, stem or shoot transcription, silique or fruit transcription, callus transcription, rhizome transcription, stem node transcription, gamete tissue transcription, flower transcription, immature bud and inflorescence specific transcription, senescing induced transcription, germination transcription and/or drought transcription.

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One embodiment of the invention is directed to an isolated nucleic acid molecule having promoter activity comprising a nucleotide sequence selected from the group consisting of:

- a. a nucleotide sequence according to any one of SEQ ID NOs. 1-26;
- a nucleotide sequence of nucleic acid residues 601-1000 of SEQ ID NO: 26;
- c. a nucleotide sequence comprising a functional fragment of (a) or (b), wherein said fragment has promoter activity.

and wherein said isolated nucleic acid molecule is not SEQ ID NO: 5.

Another embodiment of the invention is directed to an isolated nucleic acid molecule comprising a nucleotide sequence that shows at least 80 percent sequence identity to any one of SEQ ID NOs: 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26, wherein said nucleic acid molecule comprises a regulatory region that directs transcription of an operably linked heterologous polynucleotide, and wherein said isolated nucleic acid molecule is not SEQ ID NO: 5.

In another embodiment of the invention the isolated nucleic acid molecule shows at least 85 percent sequence identity to any one of SEQ ID NOs: 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26.

In another embodiment of the invention the isolated nucleic acid molecule has at least 90 percent sequence identity to any one of SEQ ID NOs: 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26.

In another embodiment the isolated nucleic acid molecule comprises at least one member selected from the group consisting of a promoter, an enhancer and an intron.

In a further embodiment of the invention, the isolated nucleic acid molecule consists of any one of SEQ ID NOs: 1-4, 6-26 and the nucleic acid residues 61-1000 of SEQ ID NO: 26

Another embodiment of the invention is directed to a vector construct comprising:

a. a first nucleic acid molecule as described above; and b. a transcribable polynucleotide molecule,

wherein said first nucleic acid molecule and said transcribable polynucleotide molecule are heterologous to each other and are operably linked.

In another embodiment of the invention, the first nucleic acid molecule consists of the nucleic acid molecule set forth in any one of SEQ ID NOs: 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26.

In another embodiment of the invention, the transcribable polynucleotide molecule encodes a polypeptide.

In another embodiment of the invention, the transcribable polynucleotide molecule is operably linked to said first nucleic acid molecule in the sense orientation.

In another embodiment of the invention, the transcribable polynucleotide molecule is transcribed into an RNA molecule that expresses the polypeptide encoded by transcribable polynucleotide molecule.

In another embodiment of the invention, the transcribable polynucleotide molecule is operably linked to said first nucleic acid molecule in the antisense orientation.

In another embodiment of the invention, the transcribable polynucleotide molecule is transcribed into an antisense RNA molecule.

In another embodiment of the invention, the transcribable polynucleotide molecule is transcribed into an interfering RNA against an endogenous gene.

In another embodiment of the invention, the transcribable polynucleotide molecule encodes a polypeptide of agronomic interest.

Another embodiment of the invention is directed to a plant or plant cell comprising:

- a. the nucleic acid molecule described above that is operably linked to a heterologous polynucleotide, or
- b. the vector construct described above.

Another embodiment of the invention is directed to a plant or plant cell stably transformed with the vector construct described above.

Another embodiment of the invention is directed to a seed of a plant as described above.

Another embodiment of the invention is directed to a method of directing transcription by combining, in an environment suitable for transcription:

- a. a first nucleic acid molecule as described above; and
- b. a transcribable polynucleotide molecule;

wherein said first nucleic acid molecule and said transcribable polynucleotide molecule are heterologous to each other 20 — and operably linked.

Another embodiment of the invention is directed to a method of expressing an exogenous coding region in a plant comprising:

- a. transforming a plant cell with a vector as described ²⁵ above.
- b. regenerating a stably transformed plant from the transformed plant cell of step (a); and
- c. selecting plants containing a transformed plant cell, wherein expression of the transcribable polynucleotide molecule results in production of a polypeptide encoded by said transcribable polynucleotide molecule.

Another embodiment of the invention is directed to a method of altering the expression of a gene in a plant comprising:

- a. transforming a plant cell with the nucleic acid molecule as described above that is operably linked to a heterologous polynucleotide, and
- b. regenerating stably transformed plants from said transformed plant cell.

Another embodiment of the invention is directed to a plant prepared according to the method described above.

Another embodiment of the invention is directed to a seed from the plant described above.

Another embodiment of the invention is directed to a 45 method of producing a transgenic plant, said method comprising;

- a. introducing into a plant cell:
 - (i) an isolated polynucleotide comprising the nucleic acid as described above that is operably linked to a 50 heterologous polynucleotide, or
 - (ii) the vector as described above; and
- b. growing a plant from said plant cell.

Other and further objects of the present invention will be made clear or become apparent from the following descrip- 55 tion.

BRIEF DESCRIPTION OF THE TABLES AND FIGURES

The Tables consist of the Expression Reports for some of the promoters of the invention providing the nucleotide sequence for each promoter and details for expression driven by each of the nucleic acid promoter sequences as observed in transgenic plants. The results are presented as summaries 65 of the spatial expression, which provides information as to gross and/or specific expression in various plant organs and 6

tissues. The observed expression pattern is also presented, which gives details of expression during different generations or different developmental stages within a generation. Additional information is provided regarding the source organism of the promoter, and the vector and marker genes used for the construct. The following symbols are used consistently throughout the Tables:

- T1: First generation transformant
- T2: Second generation transformant
- T3: Third generation transformant
- (L): low expression level
- (M): medium expression level
- (H): high expression level

Each row of the table begins with heading of the data to be found in the section. The following provides a description of the data to be found in each section:

	Heading in Tables	Description
1.	Promoter Expression Report #	Identifies the particular promoter by its construct ID.
2.	Promoter tested in:	Identifies the organism in which the promoter-marker vector was tested.
3.	Spatial expression summary:	Identifies the specific parts of the plant where various levels of GFP expression are observed. Expression levels are noted as either low (L), medium (M), or high (H).
4.	Observed expression pattern:	Provides a general explanation of where GFP expression in different generations of plants was observed.
5.	Source promoter organism:	Identifies the plant species from which the promoter was derived.
6.	Vector:	Identifies the vector used into which a promoter was cloned.
7.	Marker type:	Identifies the type of marker linked to the promoter. The marker is used to determine patterns of gene expression in plant tissue.
8.	Generation screened: T1 Mature T2 Seedling T2 Mature T3 Seedling	Identifies the plant generation(s) used in the screening process. T1 plants are those plants subjected to the transformation event while the T2 generation plants are from the seeds collected from the T1 plants and T3 plants are from the seeds of T2 plants.
9.	Inductions completed:	Provides summary of experiment schedule.
10.	T1 Mature Plant Expression:	Identifies plant tissues that were observed for possible expression, and identifies (H, M or L) level of observed expression.
11.	T2 Seedling Expression:	Identifies plant tissues that were observed for possible expression, and identifies (H, M or L) level of observed expression.
12.	T2 Mature Plant Expression:	Identifies plant tissues that were observed for possible expression, and identifies (H, M or L) level of observed expression.
13.	Utility	Provides a description of the utility of the sequence, including a trait area and, in some instances, a sub-trait area.
14.	Construct Promoter Candidate I.D. cDNA I.D.	Identifies the promoter by its construct ID and internal candidate number, and cDNA number
15.	Lines/Events expressing:	Identifies the line/event numbers that expressed under the promoter.

Some promoter reports describe additional experiments and 60 results with the particular promoter.

BRIEF DESCRIPTION OF THE FIGURES

FIG. 1 is a schematic representation of a vector pNewbin4-HAP1-GFP that is useful to insert promoters of the invention into a plant. The definitions of the abbreviations used in the vector map are as follows:

Ori—the origin of replication used by an *E. coli* host RB—sequence for the right border of the T-DNA from pMOG800

BstXI—restriction enzyme cleavage site used for cloning HAP1VP16—coding sequence for a fusion protein of the 5 HAP1 and VP16 activation domains

NOS—terminator region from the nopaline synthase gene HAP1UAS—the upstream activating sequence for HAPI 5ERGFP—the green fluorescent protein gene that has been optimized for localization to the endoplasmic ¹⁰ reticulum

OCS2—the terminator sequence from the octopine synthase 2 gene

OCS—the terminator sequence from the octopine synthase gene

p28716 (a.k.a. 28716 short)—promoter used to drive expression of the PAT (BAR) gene

PAT (BAR)—a marker gene conferring herbicide resistance

LB—sequence for the left border of the T-DNA from 20 pMOG800

Spec—a marker gene conferring spectinomycin resistance

TrfA—transcription repression factor gene

RK2-OriV—origin of replication for Agrobacterium

DETAILED DESCRIPTION OF THE INVENTION

The following definitions and methods are provided to 30 better define the present invention and to guide those of ordinary skill in the art in the practice of the present invention. Unless otherwise noted, terms are to be understood according to conventional usage by those of ordinary skill in the relevant art.

The invention disclosed herein provides promoters capable of driving the expression of an operably linked transgene. The design, construction, and use of these promoters is one object of this invention. The promoter sequences, SEQ ID NOs: 1-26 and residues 601-1000 of 40 SEQ ID NO: 26, are capable of transcribing operably linked nucleic acid molecules in particular plant tissues/organs or during particular plant growth stages, and therefore can selectively regulate expression of transgenes in these tissues/ organs or at these times of plant development.

1. Definitions

Chimeric: The term "chimeric" is used to describe polynucleotides or genes, or constructs wherein at least two of 50 the elements of the polynucleotide or gene or construct, such as the promoter and the polynucleotide to be transcribed and/or other regulatory sequences and/or filler sequences and/or complements thereof, are heterologous to each other.

Broadly Expressing Promoter: Promoters referred to 55 herein as "broadly expressing promoters" actively promote transcription under most, but not necessarily all, environmental conditions and states of development or cell differentiation. Examples of broadly expressing promoters include the cauliflower mosaic virus (CaMV) 35S transcript 60 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.

Domain: Domains are fingerprints or signatures that can 65 be used to characterize protein families and/or parts of proteins. Such fingerprints or signatures can comprise con-

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served (1) primary sequence, (2) secondary structure, and/or (3) three-dimensional conformation. A similar analysis can be applied to polynucleotides. Generally, each domain has been associated with either a conserved primary sequence or a sequence motif. Generally these conserved primary sequence motifs have been correlated with specific in vitro and/or in vivo activities. A domain can be any length, including the entirety of the polynucleotide to be transcribed. Examples of domains include, without limitation, AP2, helicase, homeobox, zinc finger, etc.

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 organism(s) regenerated from said cell. In the context of promoter, the term "endogenous coding region" or "endogenous cDNA" refers to the coding region that is naturally operably linked to the promoter.

Enhancer/Suppressor: An "enhancer" is a DNA regulatory element that can increase the steady state level of a transcript, usually by increasing the rate of transcription initiation. Enhancers usually exert their effect regardless of the distance, upstream or downstream location, or orientation of the enhancer relative to the start site of transcription. In contrast, a "suppressor" is a corresponding DNA regulatory element that decreases the steady state level of a transcript, again usually by affecting the rate of transcription initiation. The essential activity of enhancer and suppressor elements is to bind a protein factor(s). Such binding can be assayed, for example, by methods described below. The binding is typically in a manner that influences the steady state level of a transcript in a cell or in an in vitro transcription extract.

Exogenous: As referred to within, "exogenous" is any polynucleotide, polypeptide or protein sequence, whether chimeric or not, that is introduced into the genome of a host 35 cell or 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 Agrobacterium-mediated transformation dicots-e.g. Salomon et al. (1984) EMBO J. 3:141; Herrera-Estrella et al. (1983) EMBO J. 2:987; of monocots, representative papers are those by Escudero et al. (1996) *Plant J.* 10:355), Ishida et al. (1996) Nature Biotech 14:745, May et al. (1995) Bio/Technology 13:486), biolistic methods (Armaleo et al. (1990) Current Genetics 17:97), electroporation, in 45 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.

Heterologous sequences: "Heterologous 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, 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.

Homologous: In the current invention, a "homologous" polynucleotide refers to a polynucleotide that shares sequence similarity with the polynucleotide of interest. This 5 similarity may be in only a fragment of the sequence and often represents a functional domain such as, examples including, without limitation, a DNA binding domain or a domain with tyrosine kinase activity. The functional activities of homologous polynucleotides are not necessarily the 10 same.

Inducible Promoter: An "inducible promoter" in the context of the current invention refers to a promoter, the activity of which is influenced by certain conditions, such as light, temperature, chemical concentration, protein concentration, 15 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 an *Arabidopsis* gene encoding a serine-threonine kinase enzyme, and which promoter is 20 induced by dehydration, abscisic acid and sodium chloride (Wang and Goodman (1995) *Plant J.* 8:37). Examples of environmental conditions that may affect transcription by inducible promoters include anaerobic conditions, elevated temperature, the presence or absence of a nutrient or other 25 chemical compound or the presence of light.

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 and/or 30 translation of a gene or coding region or inhibition of such transcription and/or translation for a different time period as compared to the wild-type and/or from a non-natural location within the plant genome, including a gene or coding region from a different plant species or from a non-plant 35 organism.

Modulate Transcription Level: As used herein, the phrase "modulate transcription" describes the biological activity of a promoter sequence or promoter control element. Such modulation includes, without limitation, up- and down- 40 regulation of initiation of transcription, rate of transcription, and/or transcription levels.

Operable Linkage: An "operable linkage" is a linkage in which a promoter sequence or promoter control element is connected to a polynucleotide sequence (or sequences) in 45 such a way as to place transcription of the polynucleotide sequence under the influence or control of the promoter or promoter control element. Two DNA sequences (such as a polynucleotide to be transcribed and a promoter sequence linked to the 5' end of the polynucleotide to be transcribed) 50 are said to be operably linked if induction of promoter function results in the transcription of mRNA encoding the polynucleotide and if the nature of the linkage between the two DNA sequences does not (1) result in the introduction of a frame-shift mutation, (2) interfere with the ability of the 55 promoter sequence to direct the expression of the protein, antisense RNA, RNAi or ribozyme, or (3) interfere with the ability of the DNA template to be transcribed. Thus, a promoter sequence would be operably linked to a polynucleotide sequence if the promoter was capable of effecting 60 transcription of that polynucleotide sequence.

Percentage of sequence identity As used herein, the term "percent sequence identity" refers to the degree of identity between any given query sequence, e.g., SEQ ID NOs:1-26, and a subject sequence. A subject sequence typically has a 65 length that is from about 80 percent to 250 percent of the length of the query sequence, e.g., 82, 85, 87, 89, 90, 93, 95,

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97, 99, 100, 105, 110, 115, or 120, 130, 140, 150, 160, 170, 180, 190, 200, 210, 220, 230, 240, or 250 percent of the length of the query sequence. A query nucleic acid or amino acid sequence is aligned to one or more subject nucleic acid or amino acid sequences using the computer program ClustalW (version 1.83, default parameters), which allows alignments of nucleic acid or protein sequences to be carried out across their entire length (global alignment). Chenna et al. (2003) *Nucleic Acids Res.* 31(13):3497-500.

ClustalW calculates the best match between a query and one or more subject sequences, and aligns them so that identities, similarities and differences can be determined. Gaps of one or more residues can be inserted into a query sequence, a subject sequence, or both, to maximize sequence alignments. For fast pairwise alignment of nucleic acid sequences, the following default parameters are used: word size: 2; window size: 4; scoring method: percentage; number of top diagonals: 4; and gap penalty: 5. For an alignment of multiple nucleic acid sequences, the following parameters are used: gap opening penalty: 10.0; gap extension penalty: 5.0; and weight transitions: yes. For fast pairwise alignment of protein sequences, the following parameters are used: word size: 1; window size: 5; scoring method: percentage; number of top diagonals: 5; gap penalty: 3. For multiple alignment of protein sequences, the following parameters are used: weight matrix: blosum; gap opening penalty: 10.0; gap extension penalty: 0.05; hydrophilic gaps: on; hydrophilic residues: Gly, Pro, Ser, Asn, Asp, Gln, Glu, Arg, and Lys; residue-specific gap penalties: on. The output is a sequence alignment that reflects the relationship between sequences. ClustalW can be run, for example, at the Baylor College of Medicine Search Launcher website and at the European Bioinformatics Institute website on the World Wide Web.

To determine a percent identity of a subject polypeptide or nucleic acid sequence to a query sequence, the sequences are aligned using Clustal W, the number of identical matches in the alignment is divided by the length of the query sequence, and the result is multiplied by 100. The output is the percent identity of the subject sequence with respect to the query sequence. It is noted that the percent identity value can be rounded to the nearest tenth. For example, 78.11, 78.12, 78.13, and 78.14 are rounded down to 78.1, while 78.15, 78.16, 78.17, 78.18, and 78.19 are rounded up to 78.2.

Plant Promoter: A "plant promoter" is a promoter capable of initiating transcription in plant cells and can modulate transcription of a polynucleotide. Such promoters need not be of plant origin. For example, promoters derived from plant viruses, such as the CaMV35S 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 in the art.

Plant Tissue: The term "plant tissue" includes differentiated and undifferentiated tissues or plants, including but not limited to roots, stems, shoots, rhizomes, cotyledons, epicotyl, hypocotyl, leaves, pollen, seeds, gall tissue and various forms of cells in culture such as single cells, protoplast, embryos, and callus tissue. The plant tissue may be in plants or in organ, tissue or cell culture.

Preferential Transcription: "Preferential transcription" is defined as transcription that occurs in a particular pattern of cell types or developmental times or in response to specific stimuli or combination thereof. Non-limitive examples of preferential transcription include: high transcript levels of a desired sequence in root tissues; detectable transcript levels of a desired sequence in certain cell types during embryo-

genesis; and low transcript levels of a desired sequence under drought conditions. Such preferential transcription can be determined by measuring initiation, rate, and/or levels of transcription.

Promoter: A "promoter" is a DNA sequence that directs 5 the transcription of a polynucleotide. Typically a promoter is located in the 5' region of a polynucleotide to be transcribed, proximal to the transcriptional start site of such polynucleotide. More typically, promoters are defined as the region upstream of the first exon; more typically, as a region 10 upstream of the first of multiple transcription start sites; more typically, as the region downstream of the preceding gene and upstream of the first of multiple transcription start sites; more typically, the region downstream of the polyA signal and upstream of the first of multiple transcription start 15 sites; even more typically, about 3,000 nucleotides upstream of the ATG of the first exon; even more typically, 2,000 nucleotides upstream of the first of multiple transcription start sites. The promoters of the invention comprise at least a core promoter as defined above. Frequently promoters are 20 capable of directing transcription of genes located on each of the complementary DNA strands that are 3' to the promoter. Stated differently, many promoters exhibit bidirectionality and can direct transcription of a downstream gene when present in either orientation (i.e. 5' to 3' or 3' to 5' relative to 25 the coding region of the gene). Additionally, the promoter may also include at least one control element such as an upstream element. Such elements include UARs and optionally, other DNA sequences that affect transcription of a polynucleotide such as a synthetic upstream element.

Promoter Control Element: The term "promoter control element" as used herein describes elements that influence the activity of the promoter. Promoter control elements include transcriptional regulatory sequence determinants such as, but not limited to, enhancers, scaffold/matrix attachment regions, TATA boxes, transcription start locus control regions, UARs, URRs, other transcription factor binding sites and inverted repeats.

Public sequence: The term "public sequence," as used in the context of the instant application, refers to any sequence 40 that has been deposited in a publicly accessible database prior to the filing date of the present application. This term encompasses both amino acid and nucleotide sequences. Such sequences are publicly accessible, for example, on the BLAST databases on the NCBI FTP web site (accessible via 45 the internet). The database at the NCBI FTP site utilizes "gi" numbers assigned by NCBI as a unique identifier for each sequence in the databases, thereby providing a non-redundant database for sequence from various databases, including GenBank, EMBL, DBBJ (DNA Database of Japan) and 50 PDB (Brookhaven Protein Data Bank).

Regulatory Regions: The term "regulatory region" refers to nucleotide sequences that, when operably linked to a sequence, influence transcription initiation or translation initiation or transcription termination of said sequence and 55 the rate of said processes, and/or stability and/or mobility of a transcription or translation product. As used herein, the term "operably linked" refers to positioning of a regulatory region and said sequence to enable said influence. Regulatory regions include, without limitation, promoter 60 sequences, enhancer sequences, response elements, protein recognition sites, inducible elements, protein binding sequences, 5' and 3' untranslated regions (UTRs), transcriptional start sites, termination sequences, polyadenylation sequences, and introns.

The nucleic acid sequence set forth in SEQ ID NOs:1-26 are examples of regulatory regions provided herein. How-

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ever, a regulatory region can have a nucleotide sequence that deviates from that set forth in SEQ ID NOs:1-26, while retaining the ability to direct expression of an operably linked nucleic acid. For example, a regulatory region having 80% or greater (e.g. 85% or greater, 90% or greater 91% or greater, 92% or greater, 93% or greater, 94% or greater, 95% or greater, 96% or greater, 97% or greater, 98% or greater, or 99% or greater) sequence identity to the nucleotide sequence set forth in SEQ ID NOs:1-25, or 26 can direct expression of an operably linked nucleic acid.

A regulatory region can also be a fragment of SEQ ID NOs:1-25, or 26, while retaining promoter activity, i.e. the ability to direct expression of an operably linked nucleic acid. Additional examples of regulatory regions are identified in the Sequence Listing.

Regulatory regions can be classified in two categories, promoters and other regulatory regions.

Regulatory Sequence: The term "regulatory sequence," as used in the current invention, refers to any nucleotide sequence that influences transcription or translation initiation and rate, or stability and/or mobility of a transcript or polypeptide product. Regulatory sequences include, but are not limited to, promoter sequences, enhancer sequences, response elements, protein recognition sites, inducible elements, promoter control elements, protein binding sequences, 5' and 3' untranslated regions (UTRs), transcriptional start sites, termination sequences, polyadenylation sequences, introns, certain sequences within amino acid coding sequences such as secretory signals, protease cleavage sites, etc.

A 5' untranslated region (5' UTR) of a gene is generally defined as a polynucleotide segment between the transcription start site (TSS) and the coding sequence start site (ATG codon) of a messenger RNA or cDNA. Alternately, 5' UTR can be synthetically produced or manipulated DNA elements. A"plant 5'UTR" can be a native or non-native 5'UTR that is functional in plant cells. A 5' UTR can be used as a 5' regulatory element for modulating expression of an operably linked transcribable polynucleotide molecule. For example, 5' UTRs derived from heat shock protein genes have been demonstrated to enhance gene expression in plants (see for example, U.S. Pat. Nos. 5,659,122 and 5,362,865, all of which are incorporated herein by reference). Examples of 5'UTRs include those shown in SEQ ID NOs: 1-4, 6-10, 13-26.

Specific Promoters: In the context of the current invention, "specific promoters" refers to a subset of promoters that have a high preference for modulating transcript levels in a specific tissue, or organ 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 still more preferably at least 20-fold, 50-fold or 100-fold increase in transcript levels under the specific condition over the transcription under any other reference condition considered. Typical examples of temporal and/or tissue or organ specific promoters of plant origin that can be used with the polynucleotides of the present invention, are: PTA29, a promoter which is capable of driving gene transcription specifically in tapetum and only during anther development (Koltonow et al. (1990) Plant Cell 2:1201; RCc2 and RCc3, promoters that direct root-specific gene transcription in rice (Xu et al. (1995) Plant Mol. Biol. 27:237; TobRB27, a root-specific promoter from tobacco (Yamamoto et al. (1991) Plant Cell 3:371). Examples of tissue-specific promoters under developmental control include promoters that initiate transcription only in certain tissues or organs, such as root, ovule, fruit, seeds, or flowers.

Other specific promoters include those from genes encoding seed storage proteins or the lipid body membrane protein, oleosin. A few root-specific promoters are noted above. See also "Preferential transcription."

A regulatory region can contain conserved regulatory 5 motifs. Such a regulatory region can be any one of the sequences set forth in SEQ ID NOs:1-26, or a regulatory region having a nucleotide sequence that deviates from any one of those set forth in SEQ ID NOs:1-26, while retaining the ability to direct expression of an operably linked nucleic 10 acid. For example, a regulatory region can contain a CAAT box or a TATA box. A CAAT box is a conserved nucleotide sequence involved in initiation of transcription. A CAAT box functions as a recognition and binding site for regulatory proteins called transcription factors. A TATA box is another 15 conserved nucleotide sequence involved in transcription initiation. A TATA box seems to be important in determining accurately the position at which transcription is initiated.

Other conserved regulatory motifs can be identified using methods known in the art. For example, a regulatory region 20 can be analyzed using the PLACE (PLAnt Cis-acting regulatory DNA Elements) Web Signal Scan program on the world wide web at dna.affrc.go.jp/PLACE/signalscan.html. See, Higo et al., *Nucleic Acids Research*, 27(1):297-300 (1999); and Prestridge, *CABIOS*, 7:203-206 (1991). 25 Examples of conserved regulatory motifs can be found in the PLACE database on the world wide web at dna.affrc.go.jp/PLACE/. See, Higo et al., supra.

A regulatory region such as any one of SEQ ID NOs:1-26, or a regulatory region having a nucleotide sequence that 30 deviates from those set forth in SEQ ID NOs:1-26, while retaining the ability to direct expression of an operably linked nucleic acid, can contain one or more conserved regulatory motifs, which can be found in the PLACE database. For example, such a regulatory region can contain a 35 -300CORE motif having the consensus sequence TGTAAAG (SEQ ID NO:27). See, Forde et al., Nucleic Acids Res 13:7327-7339 (1985); Colot et al., EMBO J 6:3559-3564 (1987); Thomas and Flavell, Plant Cell 2:1171-1180 (1990); Thompson et al., Plant Mol Biol 40 15:755-764 (1990); Vicente-Carbajosa et al., Proc Natl Acad Sci USA 94:7685-7690 (1997); Mena et al., Plant J 16:53-62 (1998); Shing, Plant Physiol 118: 1111-1120 (1998). Such a regulatory region can contain an ABREATCONSENSUS motif having the consensus sequence YACGTGGC (SEQ ID 45 NO:28). See, Choi et al., J Biol Chem 275: 1723-1730 (2000); Kang et al., Plant Cell 14: 343-357 (2002); Oh et al., Plant Physiology 138: 341-351 (2005); Choi et al., Plant Physiol 139: 1750-1761(2005). Such a regulatory region can contain an ABREATRD22 motif having the consensus 50 sequence RYACGTGGYR (SEQ ID NO:29). See, Iwasaki et al., Mol Gen Genet 247:391-398 (1995); Bray, Trends in Plant Science 2:48-54 (1997); Busk and Pages, Plant Mol Biol 37:425-435 (1998). A regulatory region can contain an ABRELATERD1 motif having the consensus sequence 55 ACGTG (SEQ ID NO:30). See, Simpson et al., Plant J 33: 259-270 (2003); Nakashima et al., Plant Mol Biol 60:51-68 (2006). A regulatory region can contain an ABREMOTI-FAOSOSEM motif having the consensus sequence TACGTGTC (SEQ ID NO:31). See, Hattori et al., Plant J 7: 60 913-925 (1995); Hobo et al., Proc Natl Acad Sci USA 96: 15348-15353 (1999). A regulatory region can contain an ABRERATCAL motif having the consensus sequence MACGYGB (SEQ ID NO:32). See, Kaplan et al., Plant Cell 18:2733-2748 (2006). A regulatory region can contain an 65 ACGTCBOX motif having the consensus sequence GACGTC (SEQ ID NO:33). See, Foster et al., FASEB J

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8:192-200 (1994); Izawa et al., Plant Cell 6:1277-1287 (1994); Izawa et al., J Mol Biol 230:1131-1144 (1993). A regulatory region can contain an ACGTOSGLUB1 motif having the consensus sequence GTACGTG (SEQ ID NO:34). See, Washida et al., Plant Mol Biol 40:1-12 (1999); Wu et al., Plant J 23: 415-421 (2000). A regulatory region can contain an ACGTTBOX motif having the consensus sequence AACGTT (SEQ ID NO:35). See, Foster et al., FASEB J 8:192-200 (1994). A regulatory region can contain an ACIIPVPAL2 motif having the consensus sequence CCACCAACCCCC (SEQ ID NO:36). See, Patzlaff et al., Plant Mol Biol 53:597-608 (2003); Hatton et al., Plant J 7:859-876 (1995); Gomez-Maldonado et al., Plant J 39:513-526 (2004). A regulatory region can contain an AGL2ATCONSENSUS motif having the consensus sequence NNWNCCAWWWWTRGWWAN (SEQ ID NO:37). See, Huang et al., Plant Cell 8: 81-94 (1996). A regulatory region can contain an AMYBOX2 motif having the consensus sequence TATCCAT (SEQ ID NO:38). See, Huang et al., Plant Mol Biol 14:655-668 (1990); Hwang et al., Plant Mol Biol 36:331-341 (1998). A regulatory region can contain an ANAERO1CONSENSUS motif having the consensus sequence AAACAAA (SEQ ID NO:39). See, Mohanty et al., Ann Bot (Lond).96: 669-681 (2005). A regulatory region can contain an ARE1 motif having the consensus sequence RGTGACNNNGC (SEQ ID NO:40). See, Rushmore et al., J Biol Chem 266:11632-11639 (1991). A regulatory region can contain an ATHB6COREAT motif having the consensus sequence CAATTATTA (SEQ ID NO:41). See, Himmelbach et al., *EMBO J* 21:3029-3038 (2002).A regulatory region can contain an AUXRETGA1GMGH3 motif having the consensus sequence TGACGTAA (SEQ ID NO:42). See, Liu et al., Plant Cell 6:645-657 (1994); Liu et al., Plant Physiol 115:397-407 (1997); Guilfoyle et al., Plant Physiol 118: 341-347 (1998). A regulatory region can contain a BOX-IIPCCHS motif having the consensus sequence ACGTGGC (SEQ ID NO:43). See, Block et al., Proc Natl Acad Sci USA 87:5387-5391(1990); Terzaghi and Cashmore, Annu Rev Plant Physiol Plant Mol Biol 46:445-474 (1995); Nakashima et al., Plant Mol Biol 60: 51-68 (2006). A regulatory region can contain a BOXLCOREDCPAL motif having the consensus sequence ACCWWCC (SEQ ID NO:44). See, Meada et al., Plant Mol Biol 59: 739-752. (2005).Α regulatory region can contain CACGCAATGMGH3 motif having the consensus sequence CACGCAAT (SEO ID NO:45). See, Ulmasov et al., Plant Cell 7: 1611-1623 (1995). A regulatory region can contain a CARGATCONSENSUS motif having the consensus sequence CCWWWWWWGG (SEQ ID NO:46). See, Hepworth et al., EMBO J 21: 4327-4337 (2002); Michaels et al., Plant J 33: 867-874 (2003); Hong et al., Plant Cell 15:1296-1309 (2003); Folter and Angenent, Trends Plant Sci 11:224-231 (2006). A regulatory region can contain a CARGCW8GAT motif having the consensus sequence CWWWWWWWWWG (SEQ ID NO:47). See, Tang and Perry, J Biol Chem 278:28154-28159 (2003); Folter and Angenent, Trends Plant Sci 11:224-231 (2006). A regulatory region can contain a CIACADIANLELHC motif having the consensus sequence CAANNNNATC (SEQ ID NO:48). See, Piechulla et al., Plant Mol Biol 38:655-662 (1998). A regulatory region can contain a DPBFCOREDCDC3 motif having the consensus sequence ACACNNG (SEQ ID NO:49). See, Kim et al., *Plant J* 11: 1237-1251 (1997); Finkelstein and Lynch, Plant Cell 12: 599-609 (2000); Lopez-Molina and Chua, Plant Cell Physiol 41: 541-547 (2000).Α regulatory region can contain

DRE2COREZMRAB17 motif having the consensus sequence ACCGAC (SEQ ID NO:50). See, Busk et al., *Plant J* 11: 1285-1295 (1997); Dubouzet et al., *Plant J* 33: 751-763 (2003); Kizis and Pages, Plant J 30:679-689 A regulatory region can contain E2FCONSENSUS motif having the consensus sequence WTTSSCSS (SEQ ID NO:51). See, Vandepoele et al., Plant Physiol 139: 316-328. (2005). A regulatory region can contain an EMHVCHORD motif having the consensus sequence TGTAAAGT (SEQ ID NO:52). See, Muller and 10 Knudsen, Plant J 4: 343-355 (1993). A regulatory region can contain an EVENINGAT motif having the consensus sequence AAAATATCT (SEQ ID NO:53). See, Rawat et al., Plant Mol Biol 57: 629-643 (2005) and Harmer et al., Science 290: 2110-2113 (2000). A regulatory region can 15 contain an GLMHVCHORD motif having the consensus sequence RTGASTCAT (SEQ ID NO:54). See, Albani et al., Plant Cell 9: 171-184 (1997); Muller M Plant J 4: 343-355 (1993); Onate et al., J Biol Chem 274: 9175-9182 (1999). A regulatory region can contain a GT1 Consensus motif hav- 20 ing the consensus sequence GRWAAW (SEQ ID NO:55). See, Terzaghi and Cashmore, supra.; Villain et al., J Biol Chem 271:32593-32598 (1996); Le Gourrierec et al., Plant J 18:663-668 (1999); Buchel et al., Plant Mol Biol 40:387-396 (1999); Zhou, Trends in Plant Science 4:210-214 25 (1999). A regulatory region can contain a GT1GMSCAM4 motif having the consensus sequence GAAAAA (SEQ ID NO:56). See, Park et al., Plant Physiol 135: 2150-2161 regulatory region can contain HDZIP2ATATHB2 motif having the consensus sequence 30 TAATMATTA (SEQ ID NO:57). See, Ohgishi et al., *Plant J* 25: 389-398 (2001). A regulatory region can contain an IBOXCORENT motif having the consensus sequence GATAAGR (SEQ ID NO:58). See, Martinez-Hernandez et al., Plant Physiol 128:1223-1233 (2002). A regulatory 35 region can contain an INRNTPSADB motif having the consensus sequence YTCANTYY (SEQ ID NO:59). See, Nakamura et al., Plant J 29: 1-10 (2002). A regulatory region can contain a LEAFYATAG motif having the consensus sequence CCAATGT (SEQ ID NO:60). See, Kamiya 40 et al., Plant J 35: 429-441 (2003). A regulatory region can contain a LRENPCABE motif having the consensus sequence ACGTGGCA (SEQ ID NO:61). See, Castresana et al., EMBO J 7:1929-1936 (1988). A regulatory region can contain a MARTBOX motif having the consensus sequence 45 TTWTWTTWTT (SEQ ID NO:62). See, Gasser et al., Intnatl Rev Cvto 119:57-96 (1989). A regulatory region can contain a MYBGAHV motif having the consensus sequence TAACAAA (SEQ ID NO:63). See, Gubler et al., Plant Cell 7:1879-1891 (1995); Morita et al., FEBS Lett 423:81-85 50 (1998); Gubler et al., *Plant J* 17:1-9(1999). A regulatory region can contain a MYBPLANT motif having the consensus sequence MACCWAMC (SEQ ID NO:64). See, Sablowski et al., EMBO J 13:128-137 (1994); Tamagnone et al., Plant Cell 10: 135-154 (1998). A regulatory region can 55 contain a NRRBNEXTA motif having the consensus sequence TAGTGGAT (SEQ ID NO:65). See, Elliott and Shirsat, Plant Mol Biol 37:675-687 (1998). A regulatory region can contain an O2F3BE2S1 motif having the consensus sequence TCCACGTACT (SEQ ID NO:66). See, 60 Vincentz et al., Plant Mol Biol 34:879-889 (1997). A regulatory region can contain a P1BS motif having the consensus sequence GNATATNC (SEQ ID NO:67). See, Rubio et al., Genes Dev. 15: 2122-2133.(2001); Shunmann et al., J Exp Bot. 55: 855-865. (2004); Shunmann et al., Plant Physiol 65 136: 4205-4214. (2004). A regulatory region can contain a PRECONSCRHSP70A motif having the consensus

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sequence SCGAYNRNNNNNNNNNNNNNNNNN (SEQ ID NO:68). See, von Gromoff et al., Nucleic Acids Res 34:4767-4779 (2006). A regulatory region can contain a PROXBBN-NAPA motif having the consensus sequence CAAACACC (SEQ ID NO:69). See, Ezcurra et al., Plant Mol Biol 40:699-709 (1999); Busk and Pages, supra.; Ezcurra et al., Plant J 24:57-66 (2000). A regulatory region can contain a PYRIMIDINEBOXHVEPB1 motif having the consensus sequence TTTTTCC (SEQ ID NO:70). See, Cercos et al., Plant J 19: 107-118 (1999). A regulatory region can contain a RBCSCONSENSUS motif having the consensus sequence AATCCAA (SEQ ID NO:71). See, Manzara and Gruissem, Photosynth Res 16:117-139 (1988); Donald and Cashmore, EMBO J 9:1717-1726 (1990). A regulatory region can contain a ROOTMOTIFTAPOX1 motif having the consensus sequence ATATT (SEQ ID NO:72). See, Elmayan and Tepfer, Transgenic Res 4:388-396 (1995). A regulatory region can contain a RYREPEATVFLEB4 motif having the consensus sequence CATGCATG (SEQ ID NO:73). See, Curaba et al., Plant Physiol 136: 3660-3669. (2004); Nag et al., Plant Mol Biol 59: 821-838 (2005). A regulatory region can contain a SEF1MOTIF motif having the consensus sequence ATATTTAWW (SEQ ID NO:74). See, Allen et al., Plant Cell 1:623-631 (1989); Lessard et al., Plant Mol Biol 16:397-413 (1991). A regulatory region can contain a SORLREP3AT motif having the consensus sequence TGTATATAT (SEQ ID NO:75). See, Hudson and Quail, Plant Physiol 133: 1605-1616 (2003). A regulatory region can contain a SURE2STPAT21 motif having the consensus sequence AATACTAAT (SEQ ID NO:76). See, Grierson et al., Plant J 5:815-826 (1994). A regulatory region can contain a SV40COREENHAN motif having the consensus sequence GTGGWWHG (SEQ ID NO:77). See, Weiher et al., Science 219:626-631 (1983); Green et al., EMBO J 6:2543-2549 (1987); Donald and Cashmore, EMBO J 9:1717-1726 (1990). A regulatory region can contain a TATABOX2 motif having the consensus sequence TATAAAT (SEQ ID NO:78). See, Shirsat et al., Mol Gen Genet 215:326-331 (1989); Grace et al., Biol Chem 279: 8102-8110 (2004). A regulatory region can contain a TATA-BOX3 motif having the consensus sequence TATTAAT (SEQ ID NO:79). See, PLACE (PLAnt Cis-acting regulatory DNA Elements) at dna.affrc.go.jp/PLCAE/signalscan.html). A regulatory region can contain a TATABOX4 motif having the consensus sequence TATATAA (SEQ ID NO:80). See, Grace et al., J Biol Chem 279:8102-8110 (2004). A regulatory region can contain a TATABOX5 motif having the consensus sequence TTATTT (SEQ ID NO:81). See, Tjaden et al., *Plant Physiol* 108:1109-1117 (1995). A regulatory region can contain a TATABOXOSPAL motif having the consensus sequence TATTTAA (SEQ ID NO:82). See, Zhu et al., Plant Cell 14: 795-803 (2002). A regulatory region can contain a TELOBOXATEEF1AA1 motif having the consensus sequence AAACCCTAA (SEQ ID NO:83). See, Tremousayque et al., Plant J 20: 553-561 (1999); Axelos et al., Mol Gen Genet 219: 106-112 (1989); Welchen and Gonzalez, Plant Physiol 139: 88-100 (2005). A regulatory region can contain a TL1ATSAR motif having the consensus sequence CTGAAGAAGAA (SEQ ID NO:84). See, Wang et al., Science 308: 1036-1040 (2005). A regulatory region can contain a UP2ATMSD motif having the consensus sequence AAACCCTA (SEQ ID NO:85). See, Tatematsu et al., Plant Physiology 138: 757-766 (2005). A regulatory region can contain a WBBOXPCWRKY1 motif having the consensus sequence TTTGACY (SEQ ID NO:86). See, Ishiguro and Nakamura, Mol Gen Genet 244:563-571 (1994); Rushton et al., Plant Mol Biol 29:691-

702 (1995); Rushon et al., *EMBO J* 15:5690-5700 (1996); de Pater et al., *Nucleic Acids Res* 24:4624-4631 (1996); Eulgem et al., *Trends Plant Sci* 5: 199-206 (2000). A regulatory region can contain a XYLAT motif having the consensus sequence ACAAAGAA (SEQ ID NO:87). See, Ko et al., 5 *Mol Genet Genomics* 276:517-531 (2006).

Stringency: "Stringency," as used herein is a function of nucleic acid molecule probe length, nucleic acid molecule probe composition (G+C content), salt concentration, organic solvent concentration and temperature of hybridization and/or wash conditions. Stringency is typically measured by the parameter T_m , which is the temperature at which 50% of the complementary nucleic acid molecules in the hybridization assay are hybridized, in terms of a temperature differential from T_m . High stringency conditions are those providing a condition of T_m -5° C. to T_m -10° C. Medium or moderate stringency conditions are those providing T_m -20° C. to T_m -29° C. Low stringency conditions are those providing a condition of T_m -40° C. to T_m -48° C. The relationship between hybridization conditions and T_m (in ° C.) is expressed in the mathematical equation:

$$T_m = 81.5 - 16.6(\log_{10}[\text{Na}^+]) + 0.41(\% G + C) - (600/N)$$
 (I)

where N is the number of nucleotides of the nucleic acid $_{25}$ molecule 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 having lengths 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+0.7[Na⁺])}+0.41(%
 $G+C$)-500/L 0.63(% formamide) (II)

where L represents the number of nucleotides in the probe 35 in the hybrid (21). The T_m of Equation II 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 (Frischauf 40 et al. (1983) *J. Mol Biol*, 170: 827-842), stringency conditions can be adjusted to favor detection of identical genes or related family members.

Equation II is derived assuming the reaction is at equilibrium. Therefore, hybridizations according to the present 45 invention are most preferably performed under conditions of probe excess and allowing sufficient time to achieve equilibrium. The time required to reach equilibrium can be shortened by using a hybridization buffer that includes a hybridization accelerator such as dextran sulfate or another 50 high volume polymer.

Stringency can be controlled during the hybridization reaction, or after hybridization has occurred, by altering the salt and temperature conditions of the wash solutions. The formulas shown above are equally valid when used to 55 compute the stringency of a wash solution. Preferred wash solution stringencies lie within the ranges stated above; high stringency is $5\text{-}8^{\circ}$ C. below T_m , medium or moderate stringency is $26\text{-}29^{\circ}$ C. below T_m and low stringency is $45\text{-}48^{\circ}$ C. below T_m .

- T_{o} : The term " T_{o} " refers to the whole plant, explant or callus tissue, inoculated with the transformation medium.
- T₁: The term T₁ refers to either the progeny of the T₀ plant, in the case of whole-plant transformation, or the 65 regenerated seedling in the case of explant or callous tissue transformation.

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- T_2 : 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.
- T₃: The term T₃ refers to second generation progeny of the plant that is the direct result of a transformation experiment. T₃ progeny are the result of self-fertilization or cross-pollination of a T₂ plant.
- TATA to start: "TATA to start" shall mean the distance, in number of nucleotides, between the primary TATA motif and the start of transcription.
- Transgenic plant: A "transgenic plant" is a plant having one or more plant cells that contain at least one exogenous polynucleotide introduced by recombinant nucleic acid methods.
- Translational start site: In the context of the present invention, a "translational start site" is usually an ATG or AUG in a transcript, often the first ATG or AUG. A single protein encoding transcript, however, may have multiple translational start sites.
- Transcription start site: "Transcription start site" is used in the current invention to describe the point at which transcription is initiated. This point is typically located about 25 nucleotides downstream from a TFIID binding site, such as a TATA box. Transcription can initiate at one or more sites within the gene, and a single polynucleotide to be transcribed may have multiple transcriptional start sites, some of which may be specific for transcription in a particular cell-type or tissue or organ. "+1" is stated relative to the transcription start site and indicates the first nucleotide in a transcript.
- Upstream Activating Region (UAR): An "Upstream Activating Region" or "UAR" is a position or orientation dependent nucleic acid element that primarily directs tissue, organ, cell type, or environmental regulation of transcript level, usually by affecting the rate of transcription initiation. Corresponding DNA elements that have a transcription inhibitory effect are called herein "Upstream Repressor Regions" or "URR"s. The essential activity of these elements is to bind a protein factor. Such binding can be assayed by methods described below. The binding is typically in a manner that influences the steady state level of a transcript in a cell or in vitro transcription extract.
- Untranslated region (UTR): A "UTR" is any contiguous series of nucleotide bases that is transcribed, but is not translated. A 5' UTR lies between the start site of the transcript and the translation initiation codon and includes the +1 nucleotide. A 3' UTR lies between the translation termination codon and the end of the transcript. UTRs can have particular functions such as increasing mRNA message stability or translation attenuation. Examples of 3' UTRs include, but are not limited to polyadenylation signals and transcription termination sequences.

2. Use of the Promoters of the Invention

The promoters and promoter control elements of this invention are capable of modulating transcription. Such promoters and promoter control elements can be used in combination with native or heterologous promoter fragments, control elements or other regulatory sequences to modulate transcription and/or translation.

Specifically, promoters and control elements of the invention can be used to modulate transcription of a desired polynucleotide, which includes without limitation:

(i) antisense;

- (ii) ribozymes;
- (iii) coding sequences; or
- (iv) fragments thereof.

The promoter also can modulate transcription in a host 5 genome in cis- or in trans-.

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In an organism, such as a plant, the promoters and promoter control elements of the instant invention are useful to produce preferential transcription which results in a desired pattern of transcript levels in a particular cells, 10 tissues, or organs, or under particular conditions.

4. Identifying and Isolating Promoter Sequences of the Invention

The promoters and promoter control elements of the present invention are presented in the Promoter Reports of the Tables and were identified from *Arabidopsis thaliana* and *Oryza sativa*. Isolation from genomic libraries of polynucleotides comprising the sequences of the promoters and 20 promoter control elements of the present invention is possible using known techniques. For example, polymerase chain reaction (PCR) can amplify the desired polynucleotides utilizing primers designed from SEQ ID NOs: 1-26 or residues 601-1000 of SEQ ID NO: 26. Polynucleotide 25 libraries comprising genomic sequences can be constructed according to Sambrook et al., *Molecular Cloning: A Laboratory Manual*, 2nd Ed. (1989) Cold Spring Harbor Press, Cold Spring Harbor, NY), for example.

Other procedures for isolating polynucleotides comprising the promoter sequences of the invention include, without limitation, tail-PCR, and 5' rapid amplification of cDNA ends (RACE). See, for tail-PCR, for example, Liu et al. (1995) *Plant J* 8(3): 457-463; Liu et al. (1995) *Genomics* 25: 674-681; Liu et al. (1993) *Nucl. Acids Res.* 21(14): 3333-35334; and Zoe et al. (1999) *BioTechniques* 27(2): 240-248; for RACE, see, for example, *PCR Protocols: A Guide to Methods and Applications*, (1990) Academic Press, Inc.

In addition, the promoters and promoter control elements described in the Promoter Reports in the Tables (SEQ. ID. 40 Nos. 1-26) can be chemically synthesized according to techniques in common use. See, for example, Beaucage et al. (1981) *Tet. Lett.* 22: 1859 and U.S. Pat. No. 4,668,777. Such chemical oligonucleotide synthesis can be carried out using commercially available devices, such as, Biosearch 45 4600 or 8600 DNA synthesizer, by Applied Biosystems, a division of Perkin-Elmer Corp., Foster City, California, USA; and Expedite by Perceptive Biosystems, Framingham, Massachusetts, USA.

Included in the present invention are promoters exhibiting 50 nucleotide sequence identity to SEQ. ID. Nos. 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26 namely that exhibits at least 80% sequence identity, at least 85%, at least 90%, and at least 95%, 96%, 97%, 98% or 99% sequence identity compared to SEQ. ID. Nos. 1-26 or 55 residues 601-1000 of SEQ ID NO: 26. Such sequence identity can be calculated by the algorithms and computers programs described above.

The present invention further encompasses "functional variants" or "function fragments" of the disclosed 60 sequences, particularly fragments of SEQ ID NOs: 1-26 and residues 601-1000 of SEQ ID NO: 5 that retain promoter activity. Functional variants include, for example, regulatory sequences of the invention having one or more nucleotide substitutions, deletions or insertions and wherein the variant 65 retains promoter activity. Functional variants can be created by any of a number of methods available to one skilled in the

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art, such as by site-directed mutagenesis, induced mutation, identified as allelic variants, cleaving through use of restriction enzymes, or the like. Activity can likewise be measured by any variety of techniques, including measurement of reporter activity as is described at U.S. Pat. No. 6,844,484, Northern blot analysis, or similar techniques. The '484 patent describes the identification of functional variants of different promoters.

Functional fragment, that is, a regulatory sequence fragment can be formed by one or more deletions from a larger regulatory element. For example, in some instances, the 5' portion of a promoter up to the TATA box near the transcription start site can be deleted without abolishing promoter activity, as described by Opsahl-Sorteberg, H-G. et 15 al., "Identification of a 49-bp fragment of the HvLTP2 promoter directing aleruone cell specific expression" Gene 341:49-58 (2004). Such fragments should retain promoter activity, particularly the ability to drive expression of operably linked nucleotide sequences. Activity can be measured by Northern blot analysis, reporter activity measurements when using transcriptional fusions, and the like. See, for example, Sambrook et al., Molecular Cloning, A laboratory Manual (1989). Functional fragments can be obtained by use of restriction enzymes to cleave the naturally occurring regulatory element nucleotide sequences disclosed herein; by synthesizing a nucleotide sequence from the naturally occurring DNA sequence; or can be obtained through the use of PCR technology. See particularly, Mullis et al., Methods Enzymol., 155:335-350 (1987) and Erlich, ed., PCR Technology (Stockton Press, New York), (1989).

For example, a routine way to remove part of a DNA sequence is to use an exonuclease in combination with DNA amplification to produce unidirectional nested deletions of double stranded DNA clones. A commercial kit for this purpose is sold under the trade name Exo-SizeTM (New England Biolabs, Beverly, Mass.). Briefly, this procedure entails incubating exonuclease III with DNA to progressively remove nucleotides in the 3' to 5' direction at 5' overhangs, blunt ends or nicks in the DNA template. However, exonuclease III is unable to remove nucleotides at 3', 4-base overhangs. Timed digests of a clone with this enzyme produces unidirectional nested deletions.

5. Testing of Promoters

Promoters of the invention, including functional fragments, are tested for activity by cloning the sequence into an appropriate vector, transforming plants with the construct and assaying for marker gene expression. Recombinant DNA constructs are prepared which comprise the promoter sequences of the invention inserted into a vector 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

- (a) BAC: Shizuya et al. (1992) Proc. Natl. Acad. Sci. USA 89: 8794-8797; Hamilton et al. (1996) Proc. Natl. Acad. Sci. USA 93: 9975-9979;
- (b) YAC: Burke et al. (1987) Science 236:806-812;
- (c) PAC: Sternberg N. et al. (1990) Proc Natl Acad Sci USA. 87(1):103-7;
- (d) Bacteria-Yeast Shuttle Vectors: Bradshaw et al. (1995) Nucl Acids Res 23: 4850-4856;

(e) Lambda Phage Vectors: Replacement Vector, e.g., Frischauf et al. (1983) J. Mol Biol 170: 827-842; or Insertion vector, e.g., Huynh et al. (1985) In: Glover N M (ed) DNA Cloning: A practical Approach, Vol. 1 Oxford: IRL Press; T-DNA gene fusion vectors: Walden et al. 5 (1990) Mol Cell Biol 1: 175-194; and

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

Typically, the construct comprises a vector containing a promoter sequence of the present invention operationally linked to any marker gene. The promoter was identified as a promoter by the expression of the marker gene. Although many marker genes can be used, Green Fluorescent Protein (GFP) is preferred. The vector may also comprise a marker gene that confers a selectable phenotype on plant cells. The $_{15}$ marker may encode biocide resistance, particularly antibiotic resistance, such as resistance to kanamycin, G418, bleomycin, hygromycin, or herbicide resistance, such as resistance to chlorosulfuron or phosphinotricin. Vectors can also include origins of replication, scaffold attachment 20 regions (SARs), markers, homologous sequences, introns, etc.

6. Constructing Promoters with Control Elements

6.1 Combining Promoters and Promoter Control Elements The promoter and promoter control elements of the present invention, both naturally occurring and synthetic, can be used alone or combined with each other to produce the desired preferential transcription. Also, the promoters of the invention can be combined with other known sequences to obtain other useful promoters to modulate, for example, tissue transcription specific or transcription specific to certain conditions. Such preferential transcription can be determined using the techniques or assays described above.

Promoters can contain any number of control elements. For example, a promoter can contain multiple transcription binding sites or other control elements. One element may confer tissue or organ specificity; another element may limit ers will contain at least a basal or core promoter as described above. Any additional element can be included as desired. For example, a fragment comprising a basal or "core" promoter can be fused with another fragment with any number of additional control elements.

The following are promoters that are induced under stress conditions and can be combined with those of the present invention: ldh1 (oxygen stress; tomato; see Germain and Ricard (1997) Plant Mol Biol 35:949-54), GPx and CAT (oxygen stress; mouse; see Franco et al. (1999) Free Radic 50 Biol Med 27:1122-32), ci7 (cold stress; potato; see Kirch et al. (1997) Plant Mol Biol. 33:897-909), Bz2 (heavy metals; maize; see Marrs and Walbot (1997) Plant Physiol 113:93-102), HSP32 (hyperthermia; rat; see Raju and Maines (1994) Biochim Biophys Acta 1217:273-80), and 55 MAPKAPK-2 (heat shock; Drosophila; see Larochelle and Suter (1995) Gene 163:209-14).

In addition, the following examples of promoters are induced by the presence or absence of light can be used in somerase II (pea; see Reddy et al. (1999) Plant Mol Biol 41:125-37), chalcone synthase (soybean; see Wingender et al. (1989) Mol Gen Genet 218:315-22) mdm2 gene (human tumor; see Saucedo et al. (1998) Cell Growth Differ 9:119-30), Clock and BMAL1 (rat; see Namihira et al. (1999) 65 Neurosci Lett 271:1-4, PHYA (Arabidopsis; see Canton and Quail (1999) Plant Physiol 121:1207-16), PRB-1b (tobacco;

see Sessa et al. (1995) Plant Mol Biol 28:537-47) and Ypr10 (common bean; see Walter et al. (1996) Eur J Biochem 239:281-93).

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The promoters and control elements of the following genes can be used in combination with the present invention to confer tissue specificity: MipB (iceplant; Yamada et al. (1995) Plant Cell 7:1129-42) and SUCS (root nodules; broadbean; Kuster et al. (1993) Mol Plant Microbe Interact 6:507-14) for roots, OsSUTI (rice; Hirose et al. (1997) Plant Cell Physiol 38:1389-96) for leaves, Msg (soybean; Stomvik et al. (1999) Plant Mol Biol 41:217-31) for siliques, cell (Arabidopsis; Shani et al. (1997) Plant Mol Biol 34(6):837-42) and ACT11 (Arabidopsis; Huang et al. (1997) Plant Mol Biol 33:125-39) for inflorescence.

Still other promoters are affected by hormones or participate in specific physiological processes, which can be used in combination with those of present invention. Some examples are the ACC synthase gene that is induced differently by ethylene and brassinosteroids (mung bean; Yi et al. (1999) *Plant Mol Biol* 41:443-54), the TAPG1 gene that is active during abscission (tomato; Kalaitzis et al. (1995) Plant Mol Biol 28:647-56), and the 1-aminocyclopropane-1-carboxylate synthase gene (carnation; Jones et al. (1995) Plant Mol Biol 28:505-12) and the CP-2/cathepsin L gene (rat; Kim and Wright (1997) *Biol Reprod* 57:1467-77), both active during senescence.

Spacing between control elements or the configuration or control elements can be determined or optimized to permit the desired protein-polynucleotide or polynucleotide interactions to occur.

For example, if two transcription factors bind to a promoter simultaneously or relatively close in time, the binding sites are spaced to allow each factor to bind without steric hindrance. The spacing between two such hybridizing con-35 trol elements can be as small as a profile of a protein bound to a control element. In some cases, two protein binding sites can be adjacent to each other when the proteins bind at different times during the transcription process.

Further, when two control elements hybridize the spacing transcription to specific time periods, etc. Typically, promot- 40 between such elements will be sufficient to allow the promoter polynucleotide to hairpin or loop to permit the two elements to bind. The spacing between two such hybridizing control elements can be as small as a t-RNA loop, to as large as 10 kb

> Typically, the spacing is no smaller than 5 bases; more typically, no smaller than 8; more typically, no smaller than 15 bases; more typically, no smaller than 20 bases; more typically, no smaller than 25 bases; even more typically, no smaller than 30, 35, 40 or 50 bases.

> Usually, the fragment size in no larger than 5 kb bases; more usually, no larger than 2 kb; more usually, no larger than 1 kb; more usually, no larger than 800 bases; more usually, no larger than 500 bases; even more usually, no more than 250, 200, 150 or 100 bases. In some embodiments, the nucleic acid of the invention comprises at least one fragment of YP0286 (SEQ ID NO:5), e.g., YP2219 (SEQ ID NO:4), with the proviso that said nucleic acid does not consist of YP0286 (SEQ ID NO:5).

Such spacing between promoter control elements can be combination with those of the present invention: Topoi- 60 determined using the techniques and assays described

6.2 Vectors Used to Transform Cells/Hosts

A plant transformation construct containing a promoter of the present invention may be introduced into plants by any plant transformation method. Methods and materials for transforming plants by introducing a plant expression construct into a plant genome in the practice of this invention

can include any of the well-known and demonstrated methods including electroporation (U.S. Pat. No. 5,384,253); microprojectile bombardment (U.S. Pat. Nos. 5,015,580; 5,550,318; 5,538,880; 6,160,208; 6,399,861; and 6,403, 865); *Agrobacterium*-mediated transformation (U.S. Pat. 5 Nos. 5,824,877; 5,591,616; 5,981,840; and 6,384,301); and protoplast transformation (U.S. Pat. No. 5,508,184).

The present promoters and/or promoter control elements may be delivered to a system such as a cell by way of a vector. For the purposes of this invention, such delivery may 10 range from simply introducing the promoter or promoter control element by itself randomly into a cell to integration of a cloning vector containing the present promoter or promoter control element. Thus, a vector need not be limited to a DNA molecule such as a plasmid, cosmid or bacterial 15 phage that has the capability of replicating autonomously in a host cell. All other manner of delivery of the promoters and promoter control elements of the invention are envisioned. The various T-DNA vector types are a preferred vector for use with the present invention. Many useful vectors are 20 commercially available.

It may also be useful to attach a marker sequence to the present promoter and promoter control element in order to determine activity of such sequences. Marker sequences typically include genes that provide antibiotic resistance, 25 such as tetracycline resistance, hygromycin resistance or ampicillin resistance, or provide herbicide resistance. Specific selectable marker genes may be used to confer resistance to herbicides such as glyphosate, glufosinate or broxynil (Comai et al. (1985) *Nature* 317: 741-744; Gordon-30 Kamm et al. (1990) *Plant Cell* 2: 603-618; and Stalker et al. (1988) *Science* 242: 419-423). Other marker genes exist which provide hormone responsiveness.

The promoter or promoter control element of the present invention may be operably linked to a polynucleotide to be 35 transcribed. In this manner, the promoter or promoter control element may modify transcription by modulating transcript levels of that polynucleotide when inserted into a genome.

However, prior to insertion into a genome, the promoter or promoter control element need not be linked, operably or 40 otherwise, to a polynucleotide to be transcribed. For example, the promoter or promoter control element may be inserted alone into the genome in front of a polynucleotide already present in the genome. In this manner, the promoter or promoter control element may modulate the transcription 45 of a polynucleotide that was already present in the genome. This polynucleotide may be native to the genome or inserted at an earlier time.

Alternatively, the promoter or promoter control element may be inserted into a genome alone to modulate transcription. See, for example, Vaucheret, H et al. (1998) *Plant J* 16: 651-659. Rather, the promoter or promoter control element may be simply inserted into a genome or maintained extrachromosomally as a way to divert transcription resources of the system to itself. This approach may be used to downsequal to the transcript levels of a group of polynucleotide(s).

The nature of the polynucleotide to be transcribed is not limited. Specifically, the polynucleotide may include sequences that will have activity as RNA as well as sequences that result in a polypeptide product. These 60 sequences may include, but are not limited to antisense sequences, RNAi sequences, ribozyme sequences, spliceosomes, amino acid coding sequences, and fragments thereof. Specific coding sequences may include, but are not limited to endogenous proteins or fragments thereof, or 65 heterologous proteins including marker genes or fragments thereof.

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Constructs of the present invention would typically contain a promoter operably linked to a transcribable nucleic acid molecule operably linked to a 3' transcription termination nucleic acid molecule. In addition, constructs may include but are not limited to additional regulatory nucleic acid molecules from the 3'-untranslated region (3' UTR) of plant genes (e.g., a 3' UTR to increase mRNA stability of the mRNA, such as the PI-II termination region of potato or the octopine or nopaline synthase 3' termination regions). Constructs may include but are not limited to the 5' untranslated regions (5' UTR) of an mRNA nucleic acid molecule which can play an important role in translation initiation and can also be a genetic component in a plant expression construct. For example, non-translated 5' leader nucleic acid molecules derived from heat shock protein genes have been demonstrated to enhance gene expression in plants (see for example, U.S. Pat. Nos. 5,659,122 and 5,362,865, all of which are hereby incorporated by reference). These additional upstream and downstream regulatory nucleic acid molecules may be derived from a source that is native or heterologous with respect to the other elements present on the promoter construct.

Thus, one embodiment of the invention is a promoter such as provided in SEQ ID NOs: 1-26 or residues 601-1000 of SEQ ID NO: 26, operably linked to a transcribable nucleic acid molecule so as to direct transcription of said transcribable nucleic acid molecule at a desired level or in a desired tissue or developmental pattern upon introduction of said construct into a plant cell. In some cases, the transcribable nucleic acid molecule comprises a protein-coding region of a gene, and the promoter provides for transcription of a functional mRNA molecule that is translated and expressed as a protein product. Constructs may also be constructed for transcription of antisense RNA molecules or other similar inhibitory RNA in order to inhibit expression of a specific RNA molecule of interest in a target host cell.

Exemplary transcribable nucleic acid molecules for incorporation into constructs of the present invention include, for example, nucleic acid molecules or genes from a species other than the target gene species, or even genes that originate with or are present in the same species, but are incorporated into recipient cells by genetic engineering methods rather than classical reproduction or breeding techniques. Exogenous gene or genetic element is intended to refer to any gene or nucleic acid molecule that is introduced into a recipient cell. The type of nucleic acid molecule included in the exogenous nucleic acid molecule can include a nucleic acid molecule that is already present in the plant cell, a nucleic acid molecule from another plant, a nucleic acid molecule from a different organism, or a nucleic acid molecule generated externally, such as a nucleic acid molecule containing an antisense message of a gene, or a nucleic acid molecule encoding an artificial or modified version of a gene.

The promoters of the present invention can be incorporated into a construct using marker genes as described, and tested in transient analyses that provide an indication of gene expression in stable plant systems. As used herein the term "marker gene" refers to any transcribable nucleic acid molecule whose expression can be screened for or scored in some way. Methods of testing for marker gene expression in transient assays are known to those of skill in the art. Transient expression of marker genes has been reported using a variety of plants, tissues, plant cell(s), and DNA delivery systems. For example, types of transient analyses can include but are not limited to direct gene delivery via electroporation or particle bombardment of tissues in any

transient plant assay using any plant species of interest. Such transient systems would include, but are not limited to, electroporation of protoplasts from a variety of tissue sources or particle bombardment of specific tissues of interest. The present invention encompasses the use of any 5 transient expression system to evaluate promoters or promoter fragments operably linked to any transcribable nucleic acid molecules, including but not limited to selected reporter genes, marker genes, or genes of agronomic interest. Examples of plant tissues envisioned to test in transients via 10 an appropriate delivery system would include, but are not limited to, leaf base tissues, callus, cotyledons, roots, endosperm, embryos, floral tissue, pollen, and epidermal tissue.

Promoters and control elements of the present invention 15 are useful for modulating metabolic or catabolic processes. Such processes include, but are not limited to, secondary product metabolism, amino acid synthesis, seed protein storage, increased biomass, oil development, pest defense and nitrogen usage. Some examples of genes, transcripts and 20 peptides or polypeptides participating in these processes, which can be modulated by the present invention: are tryptophan decarboxylase (tdc) and strictosidine synthase (str1), dihydrodipicolinate synthase (DHDPS) and aspartate kinase (AK), 2S albumin and alpha-, beta-, and gamma- 25 zeins, ricinoleate and 3-ketoacyl-ACP synthase (KAS), Bacillus thuringiensis (Bt) insecticidal protein, cowpea trypsin inhibitor (CpTI), asparagine synthetase and nitrite reductase. Alternatively, expression constructs can be used to inhibit expression of these peptides and polypeptides by 30 incorporating the promoters in constructs for antisense use, co-suppression use or for the production of dominant negative mutations.

As explained above, several types of regulatory elements exist concerning transcription regulation. Each of these 35 regulatory elements may be combined with the present vector if desired. Translation of eukaryotic mRNA is often initiated at the codon that encodes the first methionine. Thus, when constructing a recombinant polynucleotide according to the present invention for expressing a protein product, it 40 is preferable to ensure that the linkage between the 3' portion, preferably including the TATA box, of the promoter and the polynucleotide to be transcribed, or a functional derivative thereof, does not contain any intervening codons which are capable of encoding a methionine.

The vector of the present invention may contain additional components. For example, an origin of replication allows for replication of the vector in a host cell. Additionally, homologous sequences flanking a specific sequence allow for specific recombination of the specific sequence at 50 a desired location in the target genome. T-DNA sequences also allow for insertion of a specific sequence randomly into a target genome.

The vector may also be provided with a plurality of restriction sites for insertion of a polynucleotide to be 55 transcribed as well as the promoter and/or promoter control elements of the present invention. The vector may additionally contain selectable marker genes. The vector may also contain a transcriptional and translational initiation region, and a transcriptional and translational termination region may be native with the host cell. The termination region, may be native with the polynucleotide to be transcribed, or may be derived from another source. Convenient termination regions are available from the Ti-plasmid of *A. tumefaciens*, 65 such as the octopine synthase and nopaline synthase termination regions. See also, Guerineau et al. (1991) *Mol. Gen.*

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Genet. 262:141-144; Proudfoot (1991) Cell 64:671-674; Sanfacon et al. (1991) Genes Dev. 5:141-149; Mogen et al. (1990) Plant Cell 2:1261-1272; Munroe et al. (1990) Gene 91:151-158; Ballas et al. (1989) Nucleic Acids Res. 17:7891-7903; Joshi et al. (1987) Nucleic Acid Res. 15:9627-9639.

Where appropriate, the polynucleotide to be transcribed may be optimized for increased expression in a certain host cell. For example, the polynucleotide can be synthesized using preferred codons for improved transcription and translation. See U.S. Pat. Nos. 5,380,831, 5,436,391; see also and Murray et al. (1989) *Nucleic Acids Res.* 17:477-498.

Additional sequence modifications include elimination of sequences encoding spurious polyadenylation signals, exon intron splice site signals, transposon-like repeats, and other such sequences well characterized as deleterious to expression. The G-C content of the polynucleotide may be adjusted to levels average for a given cellular host, as calculated by reference to known genes expressed in the host cell. The polynucleotide sequence may be modified to avoid hairpin secondary mRNA structures.

A general description of expression vectors and reporter genes can be found in Gruber, et al. (1993) "Vectors for Plant Transformation" In *Methods in Plant Molecular Biology & Biotechnology*, Glich et al. Eds. pp. 89-119, CRC Press. Moreover GUS expression vectors and GUS gene cassettes are available from Clonetech Laboratories, Inc., Palo Alto, California while luciferase expression vectors and luciferase gene cassettes are available from Promega Corp. (Madison, Wisconsin). GFP vectors are available from Aurora Biosciences.

6.3 Polynucleotide Insertion into a Host Cell

The promoters according to the present invention can be inserted into a host cell. A host cell includes but is not limited to a plant, mammalian, insect, yeast, and prokaryotic cell, preferably a plant cell.

The method of insertion into the host cell genome is chosen based on convenience. For example, the insertion into the host cell genome may either be accomplished by vectors that integrate into the host cell genome or by vectors which exist independent of the host cell genome.

The promoters of the present invention can exist autonomously or independent of the host cell genome. Vectors of these types are known in the art and include, for example, certain type of non-integrating viral vectors, autonomously replicating plasmids, artificial chromosomes, and the like.

Additionally, in some cases transient expression of a promoter may be desired.

The promoter sequences, promoter control elements or vectors of the present invention may be transformed into host cells. These transformations may be into protoplasts or intact tissues or isolated cells. Preferably expression vectors are introduced into intact tissue. General methods of culturing plant tissues are provided for example by Maki et al. (1993) "Procedures for Introducing Foreign DNA into Plants" In *Methods in Plant Molecular Biology & Biotechnology*, Glich et al. Eds. pp. 67-88 CRC Press; and by Phillips et al. (1988) "Cell-Tissue Culture and In-Vitro Manipulation" In *Corn & Corn Improvement*, 3rd Edition Sprague et al. eds., pp. 345-387, American Society of Agronomy Inc. et al.

Methods of introducing polynucleotides into plant tissue include the direct infection or co-cultivation of plant cell with *Agrobacterium tumefaciens*, Horsch et al. (1985) *Science*, 227:1229. Descriptions of *Agrobacterium* vector systems and methods for *Agrobacterium*-mediated gene transfer provided by Gruber et al. supra.

Alternatively, polynucleotides are introduced into plant cells or other plant tissues using a direct gene transfer method such as microprojectile-mediated delivery, DNA injection, electroporation and the like. More preferably polynucleotides are introduced into plant tissues using the 5 microprojectile media delivery with the biolistic device. See, for example, Tomes et al., "Direct DNA transfer into intact plant cells via microprojectile bombardment" In: Gamborg and Phillips (Eds.) Plant Cell, Tissue and Organ Culture: Fundamental Methods, Springer Verlag, Berlin (1995).

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Methods for specifically transforming dicots are well known to those skilled in the art. Transformation and plant regeneration using these methods have been described for a number of crops including, but not limited to, cotton (*Gossypium hirsutum*), soybean (*Glycine max*), peanut (*Arachis* 15 hypogaea), and members of the genus *Brassica*.

Methods for transforming monocots are well known to those skilled in the art. Transformation and plant regeneration using these methods have been described for a number of crops including, but not limited to, barley (Hordeum 20 vulgarae); maize (Zea mays); oats (Avena sativa); orchard grass (Dactylis glomerata); rice (Oryza sativa, including indica and japonica varieties); sorghum (Sorghum bicolor); sugar cane (Saccharum sp); tall fescue (Festuca arundinacea); turfgrass species (e.g. species: Agrostis stolonifera, 25 Poa pratensis, Stenotaphrum secundatum); wheat (Triticum aestivum), switchgrass (Panicum vigatum) and alfalfa (Medicago sativa). It is apparent to those of skill in the art that a number of transformation methodologies can be used and modified for production of stable transgenic plants from any 30 number of target plants of interest.

The polynucleotides and vectors described herein can be used to transform a number of monocotyledonous and dicotyledonous plants and plant cell systems, including species from one of the following families: Acanthaceae, 35 Alliaceae, Alstroemeriaceae, Amaryllidaceae, Apocynaceae, Arecaceae, Asteraceae, Berberidaceae, Bixaceae, Brassicaceae, Bromeliaceae, Cannabaceae, Caryophyllaceae, Cephalotaxaceae, Chenopodiaceae, Colchicaceae, Cucurbitaceae, Dioscoreaceae, Ephedraceae, Erythroxylaceae, Euphorbiaceae, Fabaceae, Lamiaceae, Linaceae, Lycopodiaceae, Malvaceae, Melanthiaceae, Musaceae, Myrtaceae, Nyssaceae, Papaveraceae, Pinaceae, Plantaginaceae, Poaceae, Rosaceae, Rubiaceae, Salicaceae, Sapindaceae, Solanaceae, Taxaceae, Theaceae, or Vitaceae.

Suitable species may include members of the genus Abelmoschus, Abies, Acer, Agrostis, Allium, Alstroemeria, Ananas, Andrographis, Andropogon, Artemisia, Arundo, Atropa, Berberis, Beta, Bixa, Brassica, Calendula, Camellia, Camptotheca, Cannabis, Capsicum, Carthamus, Cath- 50 aranthus, Cephalotaxus, Chrysanthemum, Cinchona, Citrullus, Coffea, Colchicum, Coleus, Cucumis, Cucurbita, Cynodon, Datura, Dianthus, Digitalis, Dioscorea, Elaeis, Ephedra, Erianthus, Erythroxylum, Eucalyptus, Festuca, Fragaria, Galanthus, Glycine, Gossypium, Helianthus, 55 Hevea, Hordeum, Hvoscvamus, Jatropha, Lactuca, Linum, Lolium, Lupinus, Lycopersicon, Lycopodium, Manihot, Medicago, Mentha, Miscanthus, Musa, Nicotiana, Oryza, Panicum, Papaver, Parthenium, Pennisetum, Petunia, Phalaris, Phleum, Pinus, Poa, Poinsettia, Populus, Rauwol- 60 fia, Ricinus, Rosa, Saccharum, Salix, Sanguinaria, Scopolia, Secale, Solanum, Sorghum, Spartina, Spinacea, Tanacetum, Taxus, Theobroma, Triticosecale, Triticum, Uniola, Veratrum, Vinca, Vitis, and Zea.

Suitable species include *Panicum* spp. or hybrids thereof, 65 Sorghum spp. or hybrids thereof, sudangrass, *Miscanthus* spp. or hybrids thereof, *Saccharum* spp. or hybrids thereof,

Erianthus spp., Populus spp., Andropogon gerardii (big bluestem), Pennisetum purpureum (elephant grass) or hybrids thereof (e.g., Pennisetum purpureum×Pennisetum typhoidum), Phalaris arundinacea (reed canarygrass), Cynodon dactylon (bermudagrass), Festuca arundinacea (tall fescue), Spartina pectinata (prairie cord-grass), Medicago sativa (alfalfa), Arundo donax (giant reed) or hybrids thereof, Secale cereale (rye), Salix spp. (willow), Eucalyptus spp. (eucalyptus), Triticosecale (Triticum—wheatxrye), Tripsicum dactyloides (Eastern gammagrass), Leymus cinereus (basin wildrye), Leymus condensatus (giant wildrye), and bamboo.

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In some embodiments, a suitable species can be a wild, weedy, or cultivated sorghum species such as, but not limited to, Sorghum almum, Sorghum amplum, Sorghum angustum, Sorghum arundinaceum, Sorghum bicolor (such as bicolor, guinea, caudatum, kafir, and durra), Sorghum brachypodum, Sorghum bulbosum, Sorghum burmahicum, Sorghum controversum, Sorghum drummondii, Sorghum ecarinatum, Sorghum exstans, Sorghum grande, Sorghum halepense, Sorghum interjectum, Sorghum intrans, Sorghum laxiflorum, Sorghum leiocladum, Sorghum macrospermum, Sorghum matarankense, Sorghum miliaceum, Sorghum nigrum, Sorghum nitidum, Sorghum plumosum, Sorghum propinquum, Sorghum purpureosericeum, Sorghum stipoideum, Sorghum sudanensese, Sorghum timorense, Sorghum trichocladum, Sorghum versicolor, Sorghum virgatum, Sorghum vulgare, or hybrids such as Sorghum×almum, Sorghum×sudangrass or Sorghum×drummondii.

Suitable species also include *Helianthus annuus* (sunflower), *Carthamus tinctorius* (safflower), *Jatropha curcas* (jatropha), *Ricinus communis* (castor), *Elaeis guineensis* (palm), *Linum usitatissimum* (flax), and *Brassica juncea*.

Suitable species also include *Beta vulgaris* (sugarbeet), and *Manihot esculenta* (cassava).

Suitable species also include Lycopersicon esculentum (tomato), Lactuca sativa (lettuce), Musa paradisiaca (banana), Solanum tuberosum (potato), Brassica oleracea (broccoli, cauliflower, brusselsprouts), Camellia sinensis (tea), Fragaria ananassa (strawberry), Theobroma cacao (cocoa), Coffea arabica (coffee), Vitis vinifera (grape), Ananas comosus (pineapple), Capsicum annum (hot & sweet pepper), Allium cepa (onion), Cucumis melo (melon), Cucumis sativus (cucumber), Cucurbita maxima (squash), Cucurbita moschata (squash), Spinacea oleracea (spinach), Citrullus lanatus (watermelon), Abelmoschus esculentus (okra), and Solanum melongena (eggplant).

Suitable species also include Papaver somniferum (opium poppy), Papaver orientale, Taxus baccata, Taxus brevifolia, Artemisia annua, Cannabis sativa, Camptotheca acuminate, Catharanthus roseus, Vinca rosea, Cinchona officinalis, Colchicum autumnale, Veratrum californica, Digitalis lanata, Digitalis purpurea, Dioscorea spp., Andrographis paniculata, Atropa belladonna, Datura stomonium, Berberis spp., Cephalotaxus spp., Ephedra sinica, Ephedra spp., Erythroxylum coca, Galanthus wornorii, Scopolia spp., Lycopodium serratum (=Huperzia serrata), Lycopodium spp., Rauwolfia serpentina, Rauwolfia spp., Sanguinaria canadensis, Hyoscyamus spp., Calendula officinalis, Chrysanthemum parthenium, Coleus forskohlii, and Tanacetum parthenium.

Suitable species also include *Parthenium argentatum* (guayule), *Hevea* spp. (rubber), *Mentha spicata* (mint), *Mentha piperita* (mint), *Bixa orellana*, and *Alstroemeria* spp.

Suitable species also include Rosa spp. (rose), Dianthus caryophyllus (carnation), Petunia spp. (petunia) and Poinsettia pulcherrima (poinsettia).

Suitable species also include *Nicotiana tabacum* (to-bacco), *Lupinus albus* (lupin), *Uniola paniculata* (oats), bentgrass (*Agrostis* spp.), *Populus tremuloides* (aspen), *Pinus* spp. (pine), *Abies* spp. (fir), Acer spp. (maple, *Hordeum vulgare* (barley), *Poa pratensis* (bluegrass), *Lolium* spp. (ryegrass) and *Phleum pratense* (timothy).

Thus, the methods and compositions can be used over a broad range of plant species, including species from the dicot genera Brassica, Carthamus, Glycine, Gossypium, Helianthus, Jatropha, Parthenium, Populus, and Ricinus; and the monocot genera Elaeis, Festuca, Hordeum, Lolium, 15 Oryza, Panicum, Pennisetum, Phleum, Poa, Saccharum, Secale, Sorghum, Triticosecale, Triticum, and Zea. In some embodiments, a plant is a member of the species *Panicum* virgatum (switchgrass), Sorghum bicolor (sorghum, sudangrass), Miscanthus giganteus (miscanthus), Saccharum sp. 20 (energycane), Populus balsamifera (poplar), Zea mays (corn), Glycine max (soybean), Brassica napus (canola), Triticum aestivum (wheat), Gossypium hirsutum (cotton), Oryza sativa rice), Heianthus annuus (sunflower), Medicago sativa (alfalfa), Beta vulgaris (sugarbeet), or Pennisetum 25 glaucum (pearl millet).

In certain embodiments, the polynucleotides and vectors described herein can be used to transform a number of monocotyledonous and dicotyledonous plants and plant cell systems, wherein such plants are hybrids of different species or varieties of a specific species (e.g., Saccharum sp.x Miscanthus sp., Panicum virgatum×Panicum amarum, Panicum virgatum×Panicum amarulum, and Pennisetum purpureum×Pennisetum typhoidum).

In another embodiment of the current invention, expression constructs can be used for gene expression in callus culture for the purpose of expressing marker genes encoding peptides or polypeptides that allow identification of transformed plants. Here, a promoter that is operatively linked to 40 a polynucleotide to be transcribed is transformed into plant cells and the transformed tissue is then placed on callusinducing media. If the transformation is conducted with leaf discs, for example, callus will initiate along the cut edges. Once callus growth has initiated, callus cells can be trans- 45 ferred to callus shoot-inducing or callus root-inducing media. Gene expression will occur in the callus cells developing on the appropriate media: callus root-inducing promoters will be activated on callus root-inducing media, etc. Examples of such peptides or polypeptides useful as trans- 50 formation markers include, but are not limited to barstar, glyphosate, chloramphenicol acetyltransferase (CAT), kanamycin, spectinomycin, streptomycin or other antibiotic resistance enzymes, green fluorescent protein (GFP), and β-glucuronidase (GUS), etc. Some of the promoters provided in SEQ ID NOs: 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26 will also be capable of sustaining expression in some tissues or organs after the initiation or completion of regeneration. Examples of these tissues or 60 organs are somatic embryos, cotyledon, hypocotyl, epicotyl, leaf, stems, roots, flowers and seed.

Integration into the host cell genome also can be accomplished by methods known in the art, for example, by the homologous sequences or T-DNA discussed above or using 65 the cre-lox system (A. C. Vergunst et al. (1998) *Plant Mol. Biol.* 38:393).

7. Uses of the Promoters of the Invention

7.1 Use of the Promoters to Study and Screen for Expression
The promoters of the present invention can be used to
further understand developmental mechanisms. For
example, promoters that are specifically induced during
callus formation, somatic embryo formation, shoot formation or root formation can be used to explore the effects of
overexpression, repression or ectopic expression of target
genes, or for isolation of trans-acting factors.

The vectors of the invention can be used not only for expression of coding regions but may also be used in exon-trap cloning, or promoter trap procedures to detect differential gene expression in various tissues (see Lindsey et al. (1993) *Transgenic Research* 2:3347. Auch and Reth (1990) *Nucleic Acids Research* 18: 6743).

Entrapment vectors, first described for use in bacteria (Casadaban and Cohen (1979) *Proc. Nat. Aca. Sci. U.S.A.* 76: 4530; Casadaban et al. (1980) *J. Bacteriol.* 143: 971) permit selection of insertional events that lie within coding sequences. Entrapment vectors can be introduced into pluripotent ES cells in culture and then passed into the germline via chimeras (Gossler et al. aaa91989) *Science* 244: 463; Skarnes (1990) *Biotechnology* 8: 827). Promoter or gene trap vectors often contain a reporter gene, e.g., lacZ, lacking its own promoter and/or splice acceptor sequence upstream. That is, promoter gene traps contain a reporter gene with a splice site but no promoter. If the vector lands in a gene and is spliced into the gene product, then the reporter gene is expressed.

Recently, the isolation of preferentially-induced genes has been made possible with the use of sophisticated promoter traps (e.g. IVET) that are based on conditional auxotrophy complementation or drug resistance. In one IVET approach, various bacterial genome fragments are placed in front of a necessary metabolic gene coupled to a reporter gene. The DNA constructs are inserted into a bacterial strain otherwise lacking the metabolic gene, and the resulting bacteria are used to infect the host organism. Only bacteria expressing the metabolic gene survive in the host organism; consequently, inactive constructs can be eliminated by harvesting only bacteria that survive for some minimum period in the host. At the same time, broadly active constructs can be eliminated by screening only bacteria that do not express the reporter gene under laboratory conditions. The bacteria selected by such a method contain constructs that are selectively induced only during infection of the host. The IVET approach can be modified for use in plants to identify genes induced in either the bacteria or the plant cells upon pathogen infection or root colonization. For information on IVET see the articles by Mahan et al. (1993) Science 259:686-688, Mahan et al. (1995) Proc. Natl. Acad. Sci. USA 92:669-673, Heithoff et al. (1997) Proc. Natl. Acad. Sci USA 94:934-939, and Wang et al. (1996) Proc. Natl. Acad. 55 Sci USA 93:10434.

7.2 Use of the Promoters to Transcribe Genes of Interest

In one embodiment of the invention, a nucleic acid molecule as shown in SEQ ID NOs: 1-26 or nucleic acid residues 601-1000 of SEQ ID NO: 26 is incorporated into a construct such that a promoter of the present invention is operably linked to a transcribable nucleic acid molecule that is a gene of agronomic interest. As used herein, the term "gene of agronomic interest" refers to a transcribable nucleic acid molecule that includes but is not limited to a gene that provides a desirable characteristic associated with plant morphology, physiology, growth and development, yield, nutritional enhancement, disease or pest resistance, or envi-

ronmental or chemical tolerance. The expression of a gene of agronomic interest is desirable in order to confer an agronomically important trait. A gene of agronomic interest that provides a beneficial agronomic trait to crop plants may be, for example, including, but not limited to genetic ele- 5 ments comprising herbicide resistance, increased yield, increased biomass, insect control, fungal disease resistance, virus resistance, nematode resistance, bacterial disease resistance, starch production, modified oils production, high oil production, modified fatty acid content, high protein pro- 10 duction, fruit ripening, enhanced animal and human nutrition, biopolymers, environmental stress resistance, pharmaceutical peptides, improved processing traits, improved digestibility, industrial enzyme production, improved flavor, nitrogen fixation, hybrid seed production, and biofuel pro- 15 duction. The genetic elements, methods, and transgenes described in the patents listed above are hereby incorporated by reference.

Alternatively, a transcribable nucleic acid molecule can effect the above mentioned phenotypes by encoding a RNA 20 molecule that causes the targeted inhibition of expression of an endogenous gene, for example via antisense, inhibitory RNA (RNAi), or cosuppression-mediated mechanisms. The RNA could also be a catalytic RNA molecule (i.e., a ribozyme) engineered to cleave a desired endogenous 25 mRNA product. Thus, any nucleic acid molecule that encodes a protein or mRNA that expresses a phenotype or morphology change of interest may be useful for the practice of the present invention.

7.3. Stress Induced Preferential Transcription

Promoters and control elements providing modulation of transcription under oxidative, drought, oxygen, wound, and methyl jasmonate stress are particularly useful for producing host cells or organisms that are more resistant to biotic and abiotic stresses. In a plant, for example, modulation of 35 genes, transcripts, and/or polypeptides in response to oxidative stress can protect cells against damage caused by oxidative agents, such as hydrogen peroxide and other free

Drought induction of genes, transcripts, and/or polypep- 40 tides are useful to increase the viability of a plant, for example, when water is a limiting factor. In contrast, genes, transcripts, and/or polypeptides induced during oxygen stress can help the flood tolerance of a plant.

tion can modulate stresses similar to those described in, for example, stress conditions are VuPLD1 (drought stress; Cowpea; see Pham-Thi et al. (1999) Plant Mol Biol 39:1257-65), pyruvate decarboxylase (oxygen stress; rice; see Rivosal et al. (1997) Plant Physiol 114(3): 1021-29), 50 chromoplast specific carotenoid gene (oxidative stress; Capsicum; see Bouvier et al. (1998) J Biol Chem 273: 30651-

Promoters and control elements providing preferential transcription during wounding or induced by methyl jas- 55 monate can produce a defense response in host cells or organisms. In a plant, for example, preferential modulation of genes, transcripts, and/or polypeptides under such conditions is useful to induce a defense response to mechanical wounding, pest or pathogen attack or treatment with certain 60

Promoters and control elements of the present invention also can trigger a response similar to those described for cf9 (viral pathogen; tomato; see O'Donnell et al. (1998) Plant J 14(1): 137-42), hepatocyte growth factor activator inhibitor 65 type 1 (HAI-1), which enhances tissue regeneration (tissue injury; human; Koono et al. (1999) J Histochem Cytochem

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47: 673-82), copper amine oxidase (CuAO), induced during ontogenesis and wound healing (wounding; chick-pea; Rea et al. (1998) FEBS Lett 437: 177-82), proteinase inhibitor II (wounding; potato; see Pena-Cortes et al. (1988) *Planta* 174: 84-89), protease inhibitor II (methyl jasmonate; tomato; see Farmer and Ryan (1990) Proc Natl Acad Sci USA 87: 7713-7716), two vegetative storage protein genes VspA and VspB (wounding, jasmonic acid, and water deficit; soybean; see Mason and Mullet (1990) Plant Cell 2: 569-579).

Up-regulation and transcription down-regulation are useful for these applications. For instance, genes, transcripts, and/or polypeptides that increase oxidative, flood, or drought tolerance may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription in wounding or under methyl jasmonate induction, produce transcript levels that are statistically significant as compared to cell types, organs or tissues under other conditions.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.4. Light Induced Preferential Transcription

Promoters and control elements providing preferential transcription when induced by light exposure can be utilized to modulate growth, metabolism, and development; to increase drought tolerance; and decrease damage from light stress for host cells or organisms. In a plant, for example, modulation of genes, transcripts, and/or polypeptides in response to light is useful

- (1) to increase the photosynthetic rate;
- (2) to increase storage of certain molecules in leaves or green parts only, e.g. silage with high protein or starch content;
- (3) to modulate production of exogenous compositions in green tissue, e.g. certain feed enzymes;
- (4) to induce growth or development, such as fruit development and maturity, during extended exposure to light;
- (5) to modulate guard cells to control the size of stomata in leaves to prevent water loss, or
- (6) to induce accumulation of beta-carotene to help plants cope with light induced stress.

The promoters and control elements of the present inven-The promoters and control elements of the present inven- 45 tion also can trigger responses similar to those described in: abscisic acid insensitive3 (ABI3) (dark-grown Arabidopsis seedlings, see Rohde et al. (2000) Plant Cell 12: 35-52), asparagine synthetase (pea root nodules, see Tsai and Coruzzi (1990) EMBO J 9: 323-32), mdm2 gene (human tumor, see Saucedo et al. (1998) Cell Growth Differ 9: 119-30).

Up-regulation and transcription down-regulation are useful for these applications. For instance, genes, transcripts, and/or polypeptides that increase drought or light tolerance may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription in cells, tissues or organs exposed to light, produce transcript levels that are statistically significant as compared to cells, tissues, or organs under decreased light exposure (intensity or length of time).

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.5. Dark Induced Preferential Transcription

Promoters and control elements providing preferential transcription when induced by dark or decreased light intensity or decreased light exposure time can be utilized to time growth, metabolism, and development, to modulate photo-

synthesis capabilities for host cells or organisms. In a plant, for example, modulation of genes, transcripts, and/or polypeptides in response to dark is useful, for example,

- to induce growth or development, such as fruit development and maturity, despite lack of light;
- (2) to modulate genes, transcripts, and/or polypeptide active at night or on cloudy days; or
- (3) to preserve the plastid ultra structure present at the onset of darkness.

The present promoters and control elements can also trigger response similar to those described in the section above

Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/ or polypeptides that increase or decrease growth and development may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription under exposure to dark or decrease light intensity or decrease exposure time, produce transcript 20 levels that are statistically significant.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.6. Leaf Preferential Transcription

Promoters and control elements providing preferential transcription in a leaf can modulate growth, metabolism, and development or modulate energy and nutrient utilization in host cells or organisms. In a plant, for example, preferential modulation of genes, transcripts, and/or polypeptide in a 30 leaf, is useful, for example,

- (1) to modulate leaf size, shape, and development;
- (2) to modulate the number of leaves; or
- (3) to modulate energy or nutrient usage in relation to other organs and tissues

Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/ or polypeptides that increase growth, for example, may require up-regulation of transcription.

Typically, promoter or control elements, which provide 40 preferential transcription in the cells, tissues, or organs of a leaf, produce transcript levels that are statistically significant as compared to other cells, organs or tissues.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above 45 background of the assay.

7.7. Root Preferential Transcription

Promoters and control elements providing preferential transcription in a root can modulate growth, metabolism, development, nutrient uptake, nitrogen fixation, or modulate 50 energy and nutrient utilization in host cells or organisms. In a plant, for example, preferential modulation of genes, transcripts, and/or polypeptide in a root, is useful,

- (1) to modulate root size, shape, and development;
- (2) to modulate the number of roots, or root hairs;
- (3) to modulate mineral, fertilizer, or water uptake;
- (4) to modulate transport of nutrients; or
- (4) to modulate energy or nutrient usage in relation to other cells, organs and tissues.

Up-regulation and transcription down-regulation is useful 60 for these applications. For instance, genes, transcripts, and/ or polypeptides that increase or decrease growth, for example, may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription in cells, tissues, or organs of a root, produce transcript levels that are statistically significant as compared to other cells, organs or tissues.

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For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.8. Stem/Shoot Preferential Transcription

Promoters and control elements providing preferential transcription in a stem or shoot can modulate growth, metabolism, and development or modulate energy and nutrient utilization in host cells or organisms. In a plant, for example, preferential modulation of genes, transcripts, and/or polypeptide in a stem or shoot, is useful, for example,

- (1) to modulate stem/shoot size, shape, and development; or
- (2) to modulate energy or nutrient usage in relation to other organs and tissues

Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/ or polypeptides that increase growth, for example, may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription in the cells, tissues, or organs of a stem or shoot, produce transcript levels that are statistically significant as compared to other cells, organs or tissues.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.9. Fruit and Seed Preferential Transcription

Promoters and control elements providing preferential transcription in a silique or fruit can time growth, development, or maturity; or modulate fertility; or modulate energy and nutrient utilization in host cells or organisms. In a plant, for example, preferential modulation of genes, transcripts, and/or polypeptides in a fruit, is useful

- (1) to modulate fruit size, shape, development, and maturity:
- (2) to modulate the number of fruit or seeds;
- (3) to modulate seed shattering;
- (4) to modulate components of seeds, such as, storage molecules, starch, protein, oil, vitamins, anti-nutritional components, such as phytic acid;
- (5) to modulate seed and/or seedling vigor or viability;
- (6) to incorporate exogenous compositions into a seed, such as lysine rich proteins;
- (7) to permit similar fruit maturity timing for early and late blooming flowers; or
- (8) to modulate energy or nutrient usage in relation to other organs and tissues.

Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/ or polypeptides that increase or decrease growth, for example, may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription in the cells, tissues, or organs of siliques or fruits, produce transcript levels that are statistically significant as compared to other cells, organs or tissues.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.10. Callus Preferential Transcription

Promoters and control elements providing preferential transcription in a callus can be useful to modulating transcription in dedifferentiated host cells. In a plant transformation, for example, preferential modulation of genes, transcripts, in callus is useful to modulate transcription of a marker gene, which can facilitate selection of cells that are transformed with exogenous polynucleotides.

Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/

or polypeptides that increase marker gene detectability, for example, may require up-regulation of transcription.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.11. Flower Specific Transcription

Promoters and control elements providing preferential transcription in flowers can modulate pigmentation; or modulate fertility in host cells or organisms. In a plant, for example, preferential modulation of genes, transcripts, and/ 10 or polypeptides in a flower, is useful,

- (1) to modulate petal color; or
- (2) to modulate the fertility of pistil and/or stamen.
- Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/ 15 or polypeptides that increase or decrease pigmentation, for example, may require up-regulation of transcription

Typically, promoter or control elements, which provide preferential transcription in flowers, produce transcript levels that are statistically significant as compared to other 20 for these applications. For instance, genes, transcripts, and/ cells, organs or tissues.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.12. Immature Bud and Inflorescence Preferential Tran- 25 scription

Promoters and control elements providing preferential transcription in a immature bud or inflorescence can time growth, development, or maturity; or modulate fertility or viability in host cells or organisms. In a plant, for example, 30 preferential modulation of genes, transcripts, and/or polypeptide in a immature bud and/or inflorescence, is useful,

- (1) to modulate embryo development, size, and maturity;
- (2) to modulate endosperm development, size, and composition;
- (3) to modulate the number of seeds and fruits; or
- (4) to modulate seed development and viability.

Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/ or polypeptides that increase or decrease growth, for 40 example, may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription in immature buds and inflorescences, produce transcript levels that are statistically significant as compared to other cell types, organs or tissues. 45

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.13. Senescence Preferential Transcription

Promoters and control elements providing preferential 50 transcription during senescence can be used to modulate cell degeneration, nutrient mobilization, and scavenging of free radicals in host cells or organisms. Other types of responses that can be modulated include, for example, senescence associated genes (SAG) that encode enzymes thought to be 55 involved in cell degeneration and nutrient mobilization (Arabidopsis; see Hensel et al. (1993) Plant Cell 5: 553-64), and the CP-2/cathepsin L gene (rat; Kim and Wright (1997) Biol Reprod 57: 1467-77), both induced during senescence.

In a plant, for example, preferential modulation of genes, 60 transcripts, and/or polypeptides during senescence is useful to modulate fruit ripening.

Up-regulation and transcription down-regulation is useful for these applications. For instance, genes, transcripts, and/ or polypeptides that increase or decrease scavenging of free 65 radicals, for example, may require up-regulation of transcription.

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Typically, promoter or control elements, which provide preferential transcription in cells, tissues, or organs during senescence, produce transcript levels that are statistically significant as compared to other conditions.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

7.14. Germination Preferential Transcription

Promoters and control elements providing preferential transcription in a germinating seed can time growth, development, or maturity; or modulate viability in host cells or organisms. In a plant, for example, preferential modulation of genes, transcripts, and/or polypeptide in a germinating seed, is useful,

- (1) to modulate the emergence of the hypocotyls, cotyledons and radical; or
- (2) to modulate shoot and primary root growth and development;

Up-regulation and transcription down-regulation is useful or polypeptides that increase or decrease growth, for example, may require up-regulation of transcription.

Typically, promoter or control elements, which provide preferential transcription in a germinating seed, produce transcript levels that are statistically significant as compared to other cell types, organs or tissues.

For preferential up-regulation of transcription, promoter and control elements produce transcript levels that are above background of the assay.

8. GFP Experimental Procedures and Results

Procedures

The polynucleotide sequences of the present invention 35 were tested for promoter activity using Green Fluorescent Protein (GFP) assays in the following manner.

Approximately 1-3 kb of genomic sequence occurring immediately upstream of the ATG translational start site of the gene of interest was isolated using appropriate primers tailed with BstXI restriction sites. Standard PCR reactions using these primers and genomic DNA were conducted. The resulting product was isolated, cleaved with BstXI and cloned into the BstXI site of an appropriate vector, such as pNewBin4-HAP1-GFP (see FIG. 1).

Agrobacterium-Mediated Transformation of Arabidopsis

Host Plants and Transgenes: Wild-type Arabidopsis thaliana Wassilewskia (WS) plants are transformed with Ti plasmids containing nucleic acid sequences to be expressed, as noted in the respective examples, in the sense orientation relative to the 35S promoter in a Ti plasmid. A Ti plasmid vector useful for these constructs, CRS 338, contains the Ceres-constructed, plant selectable marker gene phosphinothricin acetyltransferase (PAT), which confers herbicide resistance to transformed plants.

Ten independently transformed events are typically selected and evaluated for their qualitative phenotype in the

Preparation of Soil Mixture: 24 L Sunshine Mix #5 soil (Sun Gro Horticulture, Ltd., Bellevue, WA) is mixed with 16 L Therm-O-Rock vermiculite (Therm-O-Rock West, Inc., Chandler, AZ) in a cement mixer to make a 60:40 soil mixture. To the soil mixture is added 2 Tbsp Marathon 1% granules (Hummert, Earth City, MO), 3 Tbsp OSMO-COTE® 14-14-14 (Hummert, Earth City, MO) and 1 Tbsp Peters fertilizer 20-20-20 (J. R. Peters, Inc., Allentown, PA), which are first added to 3 gallons of water and then added to the soil and mixed thoroughly. Generally, 4-inch diameter pots are filled with soil mixture. Pots are then covered with 8-inch squares of nylon netting.

Planting: Using a 60 mL syringe, 35 mL of the seed mixture is aspirated. 25 drops are added to each pot. Clear propagation domes are placed on top of the pots that are then placed under 55% shade cloth and subirrigated by adding 1 inch of water.

Plant Maintenance: 3 to 4 days after planting, lids and shade cloth are removed. Plants are watered as needed. After 7-10 days, pots are thinned to 20 plants per pot using forceps. After 2 weeks, all plants are subirrigated with Peters fertilizer at a rate of 1 Tsp per gallon of water. When bolts are about 5-10 cm long, they are clipped between the first node and the base of stem to induce secondary bolts. Dipping infiltration is performed 6 to 7 days after clipping.

Preparation of *Agrobacterium*: To 150 mL fresh YEB is added 0.1 mL each of carbenicillin, spectinomycin and rifampicin (each at 100 mg/ml stock concentration). *Agrobacterium* starter blocks are obtained (96-well block with 20 *Agrobacterium* cultures grown to an OD₆₀₀ of approximately 1.0) and inoculated one culture vessel per construct by transferring 1 mL from appropriate well in the starter block. Cultures are then incubated with shaking at 27° C. Cultures are spun down after attaining an OD₆₀₀ of approximately 1.0 (about 24 hours). 200 mL infiltration media is added to resuspend *Agrobacterium* pellets. Infiltration media is prepared by adding 2.2 g MS salts, 50 g sucrose, and 5 μL 2 mg/ml benzylaminopurine to 900 ml water.

Dipping Infiltration: The pots are inverted and submerged for 5 minutes so that the aerial portion of the plant is in the *Agrobacterium* suspension. Plants are allowed to grow normally and seed is collected.

High-throughput Screening of T_1 Transgenic Plants: Seed is evenly dispersed into water-saturated soil in pots and placed into a dark 4° C. cooler for two nights to promote uniform germination. Pots are then removed from the cooler and covered with 55% shade cloth for 4-5 days. Cotyledons are fully expanded at this stage. FINALE® (Sanofi Aventis, Paris, France) is sprayed on plants (3 ml FINALE® diluted into 48 oz. water) and repeated every 3-4 days until only transformants remain.

GFP Assay

Tissues are dissected by eye or under magnification using INOX 5 grade forceps and placed on a slide with water and coversliped. An attempt is made to record images of observed expression patterns at earliest and latest stages of development of tissues listed below. Specific tissues will be preceded with High (H), Medium (M), Low (L) designations.

Flower	Pedicel, receptacle, nectary, sepal, petal, filament,
	anther, pollen, carpel, style, papillae, vascular, epidermis, stomata, trichome
Silique	Stigma, style, carpel, septum, placentae,
	transmitting tissue, vascular, epidermis, stomata,
	abscission zone, ovule
Ovule	Pre-fertilization: inner integument, outer
	integument, embryo sac, funiculus, chalaza,
	micropyle, gametophyte
	Post-fertilization: zygote, inner integument, outer
	integument, seed coat, primordia, chalaza, micropyle,
	early endosperm, mature endosperm, embryo
Embryo	Suspensor, preglobular, globular, heart, torpedo,
	late mature, provascular, hypophysis, radicle,
	cotyledons, hypocotyl
Stem	Epidermis, cortex, vascular, xylem, phloem, pith,
	stomata, trichome

-continued

Leaf	Petiole, mesophyll, vascular, epidermis, trichorne,
	primordia, stomata, stipule, margin

T1 Mature: These are the T1 plants resulting from independent transformation events. These are screened between stage 6.50-6.90 (i.e. the plant is flowering and 50-90% of the flowers that the plant will make have developed), which is 4-6 weeks of age. At this stage the mature plant possesses flowers, siliques at all stages of development, and fully expanded leaves. The plants are initially imaged under UV with a Leica Confocal microscope to allow examination of the plants on a global level. If expression is present, they are re-imaged using scanning laser confocal microscopy.

T2 Seedling: Progeny are collected from the T1 plants giving the same expression pattern and the progeny (T2) are sterilized and plated on agar-solidified medium containing M&S salts. In the event that there is no expression in the T1 plants, T2 seeds are planted from all lines. The seedlings are grown in Percival incubators under continuous light at 22° C. for 10-12 days. Cotyledons, roots, hypocotyls, petioles, leaves, and the shoot meristem region of individual seedlings were screened until two seedlings were observed to have the same pattern. In general, the same expression pattern was found in the first two seedlings. However, up to 6 seedlings were screened before "no expression pattern" was recorded. All constructs are screened as T2 seedlings even if they did not have an expression pattern in the T1 generation.

T2 Mature: The T2 mature plants were screened in a similar manner to the T1 plants. The T2 seeds were planted in the greenhouse, exposed to selection and at least one plant screened to confirm the T1 expression pattern. In instances where there were any subtle changes in expression, multiple plants were examined and the changes noted in the tables.

T3 Seedling: This was done similar to the T2 seedlings except that only the plants for which we are trying to confirm the pattern are planted.

Image Data:

Images are collected by scanning laser confocal microscopy. Scanned images are taken as 2-D optical sections or 3-D images generated by stacking the 2-D optical sections collected in series. All scanned images are saved as TIFF files by imaging software, edited in Adobe Photoshop, and labeled in Powerpoint specifying organ and specific expressing tissues.

Results

The Promoter Expression Reports of the Tables present 55 the results of the GFP assays as reported by their corresponding construct number and line number.

	Promoter Exp	pression Report For PT0960 (SEQ ID NO: 1)
60	Promoter Tested In:	Arabidopsis thaliana,
		Wassilewskija (WS) ecotype
		Spatial expression summary:
	TT 4.1	- TI - 1
	Hypocotyl	H vascular
	Cotyledon	H vascular
65	Rosette Leaf	H vascular
	Primary Root	L epidermis L cortex H endodermis H vascular

40

-continued							-continued				
			ion pattern	:			TABLE 1-1. T1 M		Organs/Tissues screened		
T1 Mature expression: None observed. T2 Seedling expression: High GFP expression throughout vasculature of seedlings. Source Promoter Arabidopsis thaliana,							Events Screened: n = 3 Events Expressing: n = 0 No GFP Expression Detected				
Organism: Vector:	Columb	opsis inali oia (Col) o .n4-HAP1	ecotype				TABLE 2-1. T2 Seedl	ling Expression	Tissues Screened		
Marker Type: Generation Screened: Inductions completed.	GFP-EF	٤	2 Seedling			Events Screened: n = 3 Events Expression Expression Detected Hypocotyl H vascular		•	g: n = 3		
Treatment:	Age:	Gen:	Time points:	Events Screened/ Response	Response:		Cotyledon Rosette Leaf Primary Root Construct: Promoter candidate	H vascular	rtex H endodermis H vascular		
1. 14.3 mM KNO ₃ to 28.6 Mannitol	4 wks	T2	72 hrs post transfer	2/0	No	15	I.D: cDNA I.D: Events expressing:	23518786 01-03			

Promoter 1	Expression Report Y deletion of I	ZP2585 (SEQ ID NO: 2) PT0743
Promoter Tested In:	Arabidopsis the Spatial expression	aliana, Wassilewskija (WS) ecotype n summary:
Primary Root Mature Root	-	cortex H root hairs vascular H pericycle H stele ion pattern:
T1 Mature expression: No ex T2 Seedling expression: Expr T2 Mature expression: No ex Source Promoter Organism: Vector: Marker Type: Generation Screened:	pression in root epide pression detected Arabidopsis the pNewbin4-HAI GFP-ER	aliana, Columbia (Col) ecotype
TABLE 1-2. T1 Mature Pla	ant Expression	Organs/Tissues screened
No GFP Expression Detected		
TABLE 2-2. T2 Seedling	g Expression	Tissues Screened
Events Screened: n = 6 Expression Detected Primary Root	•	ing: n = 5 (02-06) cortex H root hairs
TABLE 3-2. T2 Mature Pla		Organs/Tissues screened
Events Screened: n = 6 Expression Detected	Events Express	ing: n = 3 (03, 04, 06
Mature Root	H epidermis H	vascular H pericycle H stele
TABLE 4-2.		Promoter utility
Trait Area: Sub-trait Area: Utility: Among other uses this water and nutrients from the	Drought Tolera s promoter sequence	ciency, Nutrient Use Efficiency nce e is useful to improve the uptake of
Construct: Promoter candidate I.D:	YP2585 40983033	

One or more fragments of the above described promoter are identified in the miscellaneous feature section of the relevant SEQ ID in the Sequence Listing. Those fragments were tested for promoter activity by the same procedures as described above, and the results are summarized below.

No expression is observed for Ceres Promoter YP2581.

	41				42				
Promoter Expr	ession Report For I	PT0998 (SEQ ID N	NO: 3)	•		-contin	ued		
Promoter Tested In:	Arabidopsis that	liana, Wassilewski		. 5	cDNA I.D: Events expressing:	23506262 + 01, 02, 03, 0			
Flower Silique Mature root Primary Root	H anther H tape L vascular L ep H mature root H cortex H end H phloem H pe Observed expression	etum H silique idermis odermis H vascula ricycle	ır L xylem	-	One or more fragments of the miscellaneous feature s Listing. Those fragments w procedures as described ab No expression is observed	vere tested for promove, and the results	nnt SEQ ID in noter activity b are summariz	the Sequence by the same	
T1 Mature expression: developing anthers and		on in roots, tapetui	m cells of		Promoter Ex	pression Report	YP2219 (SE	Q ID NO: 4	.)
T2 Seedling expression and surrounding vascul T2 Mature expression:	n: High GFP express lar bundle.			15	Promoter Tested In: Are	abidopsis thaliar Spatial expression			ecotype
mature plants. Source Promoter Organism: Vector: Marker Type: Generation Screened:	Arabidopsis tha Columbia (Col) pNewbin4-HAF GFP-ER	liana, ecotype		20	Hypocotyl Cotyledon Rosette Leaf Primary Root Lateral root	L epidermis L hydathode L epidermis L epidermis H lateral roo Observed expres	L petiole L H petiole H cortex H t cap	epidermis vascular	
TABLE 1-3. T1 M Expression		Organs/Tissues	screened		T1 Mature expression: T2 Seedling expression and lateral root				nary,
Events Screened: n = 4 Expression Detected Flower H Silique Root	H anther H tape L vascular L ep H Yes	etum H silique		• 25 • 30	T2 Mature expression: Source Promoter Organ (Col) ecotype Vector: pNewbin4-HAH Marker Type: GFP-ER Generation Screened: X Inductions completed.	P1-GFP		Columbia	
TABLE 2-3. T2 Seedl Events Screened: n = 4		Tissues Scre	eened	•			Time	Events Screened/	Re-
GFP Expression Detect X Primary Root	ted	odermis H vascula	ır L xylem	35	1. Drought	Age: Gen: 4 wks T2	points: 1.0% moisture	Response	yes Yes
TABLE 3-3. T2 M				•	I	nducible express		y:	
Expression Events Screened: n = 4		Organs/Tissues	screened	- 40	Treatment:	Time point induced:	Organs induced:	Tissues in	duced:
Expression Detected Mature Root	H mature root				1. Drought	1.0% moisture	Stem, leaf leaf	,	
	TABLE 4-3. R	Γ-PCR			TABLE 1-4. T1 M	lature Plant Expr	ession Orga	ns/Tissues so	reened
Results: Plants were gr collection. Roots and a	erial tissues (Aerial	s) were harvested:	separately	45	Events Screened: n = 6 No GFP Expression De		essing: n = 0)	
in liquid nitrogen. For for qRT-PCR analysis.		•			TABLE 2-4.	T2 Seedling Exp	oression Tiss	ues Screene	d
and aerials between two	PT0998-2 Ratio (Roots/ Aerials)	PT0998-3 Ratio (Roots/ Aerials)	PT0998-4 Ratio (Roots/ Aerials)	50	Events Screened: n = 6 Expression Detected Hypocotyl Cotyledon Rosette Leaf Primary Root Lateral root	L epidermis L epidermis L epidermis L epidermis L epidermis H epidermis	L cortex L v l L petiole L H petiole H cortex H	ascular hydathode	
HAP GFP	6.33 253.98	23.16 575.31	33.01 555.41	55		TABLE 4-4. Pro	moter utility	,	
TABLE 5	-3.	Promoter u	tility	•	Trait Area: Sub-trait Area:	Water Use E Drought Tole	erance		
Trait Area: Sub-trait Area: Utility: Among other u Modulate nutrients, wa plants. Provide drought Construct: Promoter candidate I.D	UV-B tolerance ses this promoter se ter uptake from soi t or UV protection. PT0998	tion, drought tolera	o improve:	60	Utility: Among other usengineer drought tolera. Construct: Promoter candidate I.D. cDNA I.D: Events expressing: One or more fragments in the miscellaneous fethe Sequence Listing. Tactivity by the same pr	The in plants. YP2219 37172464 23494283 01, 02, 04, 0 s of the above deature section of	6 scribed pror the relevant were tested	noter are ide SEQ ID in for promote:	

-continued									-C(ontinu	ed		
Promoter Expression Report YP2219 (SEQ ID NO: 4)							Promoter Expression Report For YP0286 (SEQ ID NO: 5)						
results are summarized below. No induction is observed for Ceres Promoter YP2229.						5 -	Modulation growth and development. Modulation of nutrient uptake and loading. Expression of nitrate transports and water pumps. Modulation of drought responses, including modulation of water uptake and transport under drought conditions. Notes: Candidate to drive genes involved in osmotic stresses such as						er
Promo	oter Expression	n Repo	ort For YP0286 (S	SEQ ID NO	D: 5)	10	NCED. Endogenor		_	uced un			icii as
Promoter Teste		-	aliana, Wassilew ression summary		ecotype	-	Promoter candidate cDNA I.D: Lines expressing:	e I.D:	1266	,	OCKHAM , -06; 7/3		
Flower Stem Hypocotyl	L pedicel : L epiderm H epiderm	is	rmis			15							
Cotyledon Rosette Leaf	H mesoph H epiderm	yll H v is H pe	ascular H epidem tiole		Promote	er Expi	ession R	eport YI	P1692 (SE	EQ ID NO: (5)		
Primary Root Lateral root	H epiderm H lateral r	oot cap	xpression pattern				Promoter Tested In				Wassilev summary		ecotype
T1 mature: Low epidermal expression in stem and pedicles near inflorescence apical meristem. T2 seedling: High epidermal expression in cotyledons, petioles of emerging rosette leaves, hypocotyl, and root. Expression observed in vascular and mesophyll cells of cotyledons. Source Promoter Organism: <i>Arabidopsis thaliana</i> , WS ecotype Vector: pNewbin4.HAP1-GFP Marker Type: GFP-ER Generation Screened: XT1 Mature XT2 Seedling						- 20 25	Flower Hypocotyl Cotyledon Primary Root Lateral root Root T1 mature express T2 seedling expres]]]] O sion: G	on pattern: vasculature of petals in flowers.				
			ns completed:			- 30	Low GFP expressi epidermis of hypo	ion in l	ypocoty.	l and co	yledons.		
Treatment: 1. Drought	Age: 7 days 4 weeks	Gen: T2 T2	Time points: 3 Hrs Air dry	Events Screened/ Response 2/0 2/2		35	T1 mature express Source Promoter A Organism: (Col) et Vector: pNewbin4- Marker Type: GFF Generation Screen	Arabida cotype -HAP1 P-ER ed: X	psis thai -GFP	liana, Co	olumbia		e
2. Drought	4 weeks	12	10-12 day No H20	212	ies	_	Inductions comple	iea.				Events	
		ible ex	pression summar	y:		-	Treatment:	1	Age:	Gen:	Time points:	Screened/ Response	
Treatment:	Time point induced:	Orgai	ns induced:	Tissues in	nduced:	40	1. Drought	4	1 wks	Т2	1.0% moistur	6/2 re	Yes
2. Drought	10-12 day			Pedicel, I		•		Inc	lucible ex	kpression	ı summar	y:	
	No H20	Siliqu Leaf Stem	ies	Epidermis Epidermis Epidermis	s, Vascular	45	Treatment:		Γime poi nduced:		ns induce	d:	Tissues induced
TABLE 1-	-5. T1 Matur	e Plant	Expression Orga	ns/Tissues s	creened	•	TABLE 1-6.	Γ1 Mat	ure Plant	Express	sion Orga	ns/Tissues s	creened
Events Screene GFP Expression Flower	n Detected L pedicel		Events Express	ing: n = 2		50	Events Screened: 1 Expression Detector Flower	ed	L petal L		•	sing: $n = 3$	
Stem	L epiderm		PTi	6	- 4	-	TABLE	2-6. T	2 Seedlii	ıg Expre	ssion Tis	sues Screene	ed
Events Screene Seedlings expr Event-04: 6/6 Event-06: 4/6 Expression De	ed: n = 2 ressing/Seedli	ngs scr	g Expression Tiss Events Express eened		ed	- 55	Events Screened: a Expression Detects Hypocotyl Cotyledon Primary Root Lateral root				dermis L cular scular	sing: n = 1 vascular	
Hypocotyl Cotyledon		yll H v	ascular H epidem	nis H petiol	le		TABLE 3-6. T	Γ2 Mat	ure Plant	Express	sion Orga	ns/Tissues s	creened
Rosette Leaf Primary Root Lateral root	H epiderm H epiderm H lateral r	.is				60	Events Screened: 1 Expression Detector					sing: $n = 2$	
	TAE	BLE 3-5	. Promoter utility	ī		-	X Root Construct: Promoter candidat	e I.D:		H ma YP16 1537			
Trait Area: Sub-trait Area:			cy noter sequence is			65	cDNA I.D: Events expressing:			3653 ₄ 01-06	1367		

Promoter F	xpression	Report `	YP1894 (S	SEQ ID NO: 1			Promoter Expression	on Report For YP1976 (SEQ ID NO: 8)
Promoter Tested In: A			-			•		Opsis thaliana, Wassilewskija (WS) ecotype
			n summai			. 5		tial expression summary:
Cotyledon Primary Root Root	L epide H epide H matu	rmis H	cortex			3	Hypocotyl Primary Root Lateral root	L epidennis H cortex H cortex
Observed expression T1 Mature expression T2 Seedling expression cortex cells of root. I T2 Mature expression Inductions: Expression	:: No expre on: High G ow GFP e :: High GF n enhanced	ession de FP expr xpression P expression I by hig	etected. ression in on in cotyl ssion in ro	edons. oots.	I	10	T2 seedling expression: Hig root transition zone. GFP is root and lateral root.	GFP expression observed in aerial organs. gh GFP expressed in epidermal cells at s expressed in cortex cells of lower main
conditions (see Induct Source Promoter Organism: Vector: Marker Type:	Arabido	<i>psis tha</i> oia (Col) n4-H A F	ecotype			15	No GFP expression observe	n GFP expression in roots of mature plant. ed in aerial tissues. : Arabidopsis thaliana, Columbia (Col) ecotype pNewbin4-HAP1-GFP GFP-ER X T1 Mature X T2 Seedling X T2 Mature
Generation Screened: Inductions Table. Upr low-nitrogen conditio	egulation of	of gene	expression	was expected	d in			e Plant Expression Organs/Tissues screened
GFP was detected in (control test) (see Tab	response to	the hig			inced	20	Events Screened: n = 6 No GFP Expression Detector	Events Expressing: n = 0 ed
			Time	Events Screened/	Re-		TABLE 2-8. T2	Seedling Expression Tissues Screened
Treatment:	Age:	Gen:	points:	Response	sponse:	. 25	Events Screened: n = 6 Expression Detected	Events Expressing: n = 6
1. Nitrogen (High to Low)-14.3 mM KNO ₃ to 28.6	4 wks	T2	90 hrs	6/0	No		X Hypocotyl X Primary Root X Lateral rant	L epidemnis H cortex H cortex
mM Mannitol						30	TABLE 3-8. T2 Mature	e Plant Expression Organs/Tissues screened
	Inducible Time	express	ion summ	ary:			Events Screened: n = 6 Expression Detected X Root	Events Expressing: n = 6 H mature root
Treatment:	point induced	Organ : induc		Tissues indu	ıced:			BLE 4-8 Promoter utility
High nitrogen	90 hrs	Leave stems				35	Trait Area: Sub-trait Area:	Nutrient and water economy. Nitrogen use efficiency Water use efficiency
TABLE 1-7. T1				-	creened		Nitrogen use efficiency in le	his promoter sequence is useful to improve: ower/non-limiting nitrogen environments, trought and non-limiting water environments,
Events Screened: n = No GFP Expression I		Event	s Express	ing: n = 0		40	and bacterial pathogens.	borne nematodes, root worms, fungal
TABLE 2-7	7. T2 Seed	ling Exp	ression T	issues Screene	ed		Construct: Promoter candidate I.D: cDNA I.D:	YP1976 15371806 23523729
Events Screened: n = Expression Detected Cotyledon Primary Root	L epide H epide	rmis	-	ing: n = 6 (01	-06)	45	Events expressing:	01-06
TABLE 3-7. T2				gans/Tissues s	creened	•	Promoter Express	sion Report YP2016 (SEQ ID NO: 9)
Events Screened: n = Expression Detected	6	Event	s Express	ing: n = 6 (01	-06)	50	Promoter Tested In: Arabid	lopsis thaliana, Wassilewskija (WS) ecotype tial expression summary:
Root H mature root	TABLE :	5-7. Pro	moter util	ity			Flower	H pedicel H receptacle H nectary H sepal H petal H filament H tapetum H carpel
Trait Area: Sub-trait Area: Utility: Among other	Drough uses this p	t, Nitrog romoter		ficiency, is useful to i		55	Silique	H style H papillae H vascular H epidermis H stomata H silique H stigma H style H carpel H septum H vascular H epidermis H abscission zone
Expression in the root uptake into the root n	nass withou	ut undes	irable affe	ects on the abo	ove		Ovule	H ovule Post-fertilization: H inner integument H outer integument H seed coat H embryo
ground tissues. The h toxicity associated wi the feedback loop tha	th very hig t limits nit	gh levels rogen u	s of nitrog ptake whe	en and/or byp n nitrogen lev	assing els are not	60	Embryo Stem	H torpedo H late H mature H vascular H xylem H phloem H pith H epidermis H cortex
limiting, thus accelera use or for enhancing Construct:		gen upta	ke for eith	ier storage foi YP1894	later		Leaf	H petiole H mesophyll H epidermis H stomata
Promoter candidate I. cDNA I.D:	D:			25518825 23499704		65	Hypocotyl Cotyledon	H epidermis H cortex H vascular H xylem H phloem H mesophyll H vascular H epidermis
				23.33701			Cotyledon	H petiole

-continued

	-continued	_	Prome	oter Expressio	n Renor	t YP2097 (S	SEO ID NO:	10)
Promoter Expre	ession Report YP2016 (SEQ ID NO: 9)	_	Promoter Tested			,	•	· · · · · · · · · · · · · · · · · · ·
Rosette Leaf	H mesophyll H vascular H epidermis	•	Fromoter restet	-		sion summa) ecotype
Primary Root	H cortex H vascular	5					,-	
Lateral root	H root cap		Flower		H pol			
Observed expression patte	rn: P broadly expressed throughout mature plant		Primary Root			dermis		
	inflorescence meristems. High GFP expression		Root			ture root		
	ers. Not expressed in anther walls of mature	10	Observed expre			magion and	oifia ta mallar	
stamen. High GFP express	sion in silique, developing ovules, seed and	10	T1 mature expr T2 seedling exp	_	-	-	-	1.
	in epidermis, vasculature and mesophyll in		root transition z		-		illiai celis at	
•	epidermis, cortex, pith, and vascular bundles		T2 mature expr		-	-	ollen. GFP	
GFP expression in mature	near apex decreasing toward rosette leaves.		expression in ep	_	_	_		
•	FP expression throughout epidermis, cortex,	15	Source Promote	er Organism:	Arabi	dopsis thali	ana,	
	ells in aerial tissues of seedling.	13				nbia (Col) e		
In root, GFP expressed in			Vector:			bin4-HAP1	-GFP	
	P broadly expressed throughout mature plant		Marker Type:	anadi V T1 N	GFP-		V TO Mate	140
with highest expression at Source Promoter	inflorescence meristems and root. Arabidopsis thaliana,		Generation Screen Inductions com		viature A	12 Seedin	ig A 12 Mau	ire
Organism:	Columbia (Col) ecotype	20	- Inductions com	preteu.				
Vector:	pNewbin4-HAP1-GFP						Events	
Marker Type:	GFP-ER					Time	Screened/	
Generation Screened: X T	1 Mature X T2 Seedling X T2 Mature		Treatment:	Age:	Gen:	points:	Response	Response:
TABLE 1-9. T1 Matu	ure Plant Expression Organs/Tissues screened	- 25	1. Drought	4 wks	T2	1.0%	6/2	Yes
Events Screened: n = 6 Expression Detected	Events Expressing: n = 6	- 23		T 1 11	1	moisture		
Flower	H pedicel H receptacle H nectary			Inducit	ole expre	ession summ	ıary:	
	H sepal H petal H filament			Time poi	nt.	Organs		Tissues
	H tapetum H carpel H style	30	Treatment:	induced:		induced:		induced:
	H papillae H vascular H epidermis H stomata H silique	50						
Silique	H stigma H style H carpel		1. Drought	1.0%		Flower		Pollen,
	H septum H vascular			moisture		Leaf		Epidermis
	H epidermis H abscission zone H ovule					Stem		Guard cells
Ovule	Post- fertilization:H inner integument	35				Root		Epidermis
Embryo	H outer integument H seed coat H embryo H torpedo H late H mature							Guard cells Vascular
Stem	H epidermis H cortex H vascular							vasculai
	H xylem H phloem H pith		TABLE 1-1	0. T1 Mature	Plant E	xpression O	rgans/Tissues	screened
Leaf	H petiole H mesophyll H epidermis					•		
	H stomata	40	Events Screene			Events Ex	pressing: n =	= 4
TABLE 2-9. T2	Seedling Expression Tissues Screened	•	Expression Dete Flower	ected H pollen				
Events Screened: n = 4 Expression Detected	Events Expressing: n = 4	•	TABL	E 2-10. T2 S	eedling 1	Expression '	Tissues Scree	ened
Hypocotyl	H epidermis H cortex H vascular	45				_		
	H xylem H phloem	45	Events Screene			Events Ex	pressing: n =	= 2
Cotyledon	H mesophyll H vascular H epidermis		Expression Determinary Root	ectea L epidern	nie			
Rosette Leaf	H petiole H mesophyll H vascular H epidermis		Tilliary Root	L epidem	115			
Primary Root	H cortex H vascular		TABLE 3-1	0. T2 Mature	Plant E:	xpression O	rgans/Tissues	screened
Lateral root	H root cap	50				1	8	
		30	Events Screene	d: n = 6		Events Ex	pressing: n =	= 3
TABLE 3-9. T2 Matu	ure Plant Expression Organs/Tissues screened	•	Expression Det					
TABLE 3-9. T2 Matu Events Screened: n = 4		-		ected				
Events Screened: n = 4	ure Plant Expression Organs/Tissues screened	- - - 55	Expression DeteRoot L mature	ected root	E 5-10.	Promoter ut	ility	
Events Screened: n = 4 TA	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility	- - - 55	Expression Determine Root L mature Trait Area:	ected root	PG&l	D, water use	e efficiency	
Events Screened: n = 4 TA Trait Area:	Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development	• • 55	Expression Determine Root L mature Trait Area: Sub-trait Area:	root TABL	PG&l	D, water use	e efficiency e,	
Events Screened: n = 4 TA Trait Area: Sub-trait Area:	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development Size and source capacity	- - 55	Expression Det Root L mature Trait Area: Sub-trait Area: Utility: Among	TABL	PG&l Droug	D, water use ght tolerance ter sequence	e efficiency e, e is useful to	-
Events Screened: n = 4 TA Trait Area: Sub-trait Area: Utility: Among other uses	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development Size and source capacity this promoter sequence is useful to improve:	-	Expression Det Root L mature Trait Area: Sub-trait Area: Utility: Among Desiccation told	TABLE other uses the erance, recover	PG&l Droug s promo	D, water use ght tolerance ter sequence drought, dr	e efficiency e, e is useful to ought toleran	ce,
Events Screened: n = 4 TA Trait Area: Sub-trait Area: Utility: Among other uses Plant size and architecture	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development Size and source capacity this promoter sequence is useful to improve: c, growth rate, seedling establishment,	55	Expression Det Root L mature Trait Area: Sub-trait Area: Utility: Among Desiccation told improve water	TABL other uses this erance, recoveruse efficiency,	PG&l Droug s promo	D, water use ght tolerance ter sequence drought, dr	e efficiency e, e is useful to ought toleran	ce,
Events Screened: n = 4 TA Trait Area: Sub-trait Area: Utility: Among other uses Plant size and architecture responses to shade and lov	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development Size and source capacity this promoter sequence is useful to improve:	-	Expression Determine Root L mature Trait Area: Sub-trait Area: Utility: Among Desiccation tole improve water and nitrogen us	TABL other uses this erance, recoveruse efficiency,	PG&l Droug is promo ery from seed siz	D, water use ght tolerance ter sequence drought, dro ze and nutrie	e efficiency e, e is useful to ought toleran	ce,
Events Screened: n = 4 TA Trait Area: Sub-trait Area: Utility: Among other uses Plant size and architecture responses to shade and lov capacity and sucrose loadi	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development Size and source capacity this promoter sequence is useful to improve: c, growth rate, seedling establishment, w light, responses to drought and cold, source	-	Expression Determine Trait Area: Sub-trait Area: Utility: Among Desiccation tole improve water and nitrogen us Construct:	TABL other uses thi erance, recove use efficiency, e efficiency.	PG&l Droug is promo ry from seed siz	D, water use ght tolerance ster sequence drought, drought, drought, drought, drought, drought	e efficiency e, e is useful to ought toleran	ce,
Events Screened: n = 4 TA Trait Area: Sub-trait Area: Utility: Among other uses Plant size and architecture responses to shade and lov capacity and sucrose loadi Construct:	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development Size and source capacity this promoter sequence is useful to improve: c, growth rate, seedling establishment, w light, responses to drought and cold, source ing, seed filling, seed size and plant yield.	-	Expression Determine Root L mature Trait Area: Sub-trait Area: Utility: Among Desiccation tole improve water and nitrogen us	TABL other uses thi erance, recove use efficiency, e efficiency.	PG&l Droug is promo ery from seed siz	D, water use ght tolerance ster sequence drought, dro ze and nutrie	e efficiency e, e is useful to ought toleran	ce,
Events Screened: n = 4 TA Trait Area: Sub-trait Area: Utility: Among other uses Plant size and architecture responses to shade and lov	re Plant Expression Organs/Tissues screened Events Expressing: n = 4 BLE 4-9. Promoter utility Plant growth and development Size and source capacity this promoter sequence is useful to improve: c, growth rate, seedling establishment, w light, responses to drought and cold, source ing, seed filling, seed size and plant yield. YP2016	-	Expression Determine Expression Determine Expression Determine Expression Exp	TABL other uses the erance, recove use efficiency, e efficiency.	PG&l Droug is promo ery from seed siz YP20 29223	D, water use ght tolerance ster sequence drought, droze and nutrie	e efficiency e, e is useful to ought toleran	ce,

49 					50 -continued								
Promoter E	xpression Re	ort YP	2538 (SEQ	ID NO: 1	1)					COIIII	iucu		
Promoter Tested In: <i>Arabidopsis thaliana</i> , Wassilewskija (WS) ecotype Spatial expression summary:			-	Promote	r Expr	ession l	Report	YP2538 (SEQ ID NO	: 11)			
Flower Silique		H anthe	er H silique	e		- 5	Aerial organs Root		H infloi H matu		e H flowe	rs H silique	H stem
Ovule Embryo	Post-fertil H mature	ization:	H embryo					TA	ABLE 5	5-11. Pı	omoter ut	ility	
Hadure Expressed in stem and pedicels near apex. High GFP expression in				10	Sub-trait Area: Drought Tolerance, Low Ni Phosphorous Tolerance, Seed Size and Yiel Utility: Among other uses this promoter sec response to drought conditions and low soi in the embryo could be valuable for engine				v Nitroger Yield er sequenc v soil nutr gineering	Nitrogen Tolerance, Low ield sequence is useful to improve: oil nutrient levels. Expression neering of seed size and yield.			
anthers of developing in mature siliques. TZ Seedling expressic hypocotyl and cotylet mesophyll and vascul emerging rosette leav TZ Mature expression flowers, stem and roo	on: High GFF dons. High G ature of coty es. a: High GFP	express FP expr ledons.	sion in vas ession in e High GFP	culature of pidermis, expression	root,	20	Promoter candidat cDNA I.D: Events expressing			23	519856 -05		
Source Promoter	A rabidops						Promote	r Expr	ession l	Report	YP2552 (SEQ ID NO	: 12)
Organism: Vector: Marker Type: Generation Screened: Inductions completed.				25	Primary Root H epidermis H vascular H root hairs Mature Root H epidermis H vasculature H xylem H phloem H root hairs lateral root L stele Observed expression pattern:					S) ecotype			
Treatment: 1. Drought				30									
	Inducible ex	pression	summary:	:		- - 35	expression in epid Source Promoter (. Columbia	(Col) ecotyn
Treatment:	Time point induced:	Organ		Tissues induced		_ 33	Vector: pNewbin4 Marker Type: GFI Generation Screen Inductions comple	-HAP1 P-ER ed: X	-GFP				
1. Drought	1.0% moisture	Inflore Flower Silique Stem	que	Pedicel Silique, Stem S Stamen	ilique,	- 40	Treatment:		Age:	Gen:	Time points:	Events Screened/ Response	Response:
		Root		Epidern	nis,		1. Cold		10	T2	4 hr	6/0	None
				Vascula Vascula Epidern	ır	45	2. Nitrogen - high to low N [14.3 ml KNO ₃ to 28.6 mM	N	day 4 weeks	T2	4 hr	6/0	None
TABLE 1-11. T1	Mature Plant	Expres	sion Organ	ıs/Tissues s	creened	_	Mannitol]						
Events Screened: n =	5		Events E (02, 05)	xpressing:	n = 2	_	3. Far Red Far Re 525 μW/cm	d ₇₃₀ =	10 day	Т2	4 hr	6/0	None
Expression Detected Flower	H pedicel	H anthe	er H silique			50		Inc	ducible	express	sion sumn	ıary:	
Silique Ovule Embryo	H carpel I Post-fertil H mature	I epider	mis				Treatment:		Time indu	•		gans uced:	Tissues induced:
		г	·			-	Table 1-12. T	1 Mat	ure Pla	nt Expi	ession Or	gans/Tissues	screened
TABLE 2-11. T2 Seedling Expression Tissues Screened Events Screened: n = 3 Events Expressing:			- 55	Events Screened:				Expressin sion Detec					
Expression Detected			n = 2 (03)	3, 05)			Table 2	-12. T	2 Seedl	ing Ex	oression T	issues Scree	ned
Hypocotyl Cotyledon Rosette Leaf	H epidern H mesoph H mesoph	yll H va	ascular H e	pidermis		60	Events Screened:			Events		g: n = 4 (01	
Primary Root TABLE 3-11. T2	H vascula		-	ns/Tigennee e	screened	_	X Primary Root					re H root ha	iirs
		pres				-	Table 3-12. T	2 Mat	ure Pla	nt Expi	ession Or	gans/Tissues	screened
Events Screened: n =	4		Events E n = 2 (03)	expressing: 3, 05)		65	Events Screened:	1 = 6		Events	Expressin	g: n = 5 (01	, 02, 03,
Expression Detected			. (,,					Ü		05, 06)		, , (°1	, , - ~ ,

-continued

Promoter Ex	pression Report YP2552 (SEQ ID NO: 12)	-		
	Expression Detected	- - 5		
Mature Root	H epidermis H vasculature H xylem H phloem H root hairs lateral root L stele	,		
	Table 4-12. Promoter utility	_		
Trait Area: Sub-trait Area: Utility: Among other umprove the uptake of	Water Use Efficiency, Nutrient Use Efficiency Drought Tolerance, Nitrogen and Phosphorous Use Efficiency uses this promoter sequence is useful to water and nutrients.	10		
Construct: Promoter candidate I.I cDNA I.D:	YP2552 D: 25659462 23504306	15		
Events expressing:	01, 02, 03, 05, 06	_		
Promoter Ex	pression Report YP2563 (SEQ ID NO: 13)	- ²⁰		
Promoter Tested In: A Spatial expression sun	mbidopsis thaliana, Wassilewskija (WS) ecotype nmary:	_		
Flower H carpel H silique Silique H carpel Ovule Post-fertilization: H funiculus H seed coat Hypocotyl H epidermis L vascular Cotyledon L epidermis H mesophyll H epidermis Primary Root L epidermis L cortex H vascular H root cap H root hairs				
	I anidamaia			
Lateral root Observed expression p	L epidermis oattern:	_		
Observed expression properties of the coats and funicul T2 Seedling expression throughout root and in elongation zone. GFP T2 Mature expressions	eattern: High GFP expression in carpels and in	35		
Observed expression procession procession of the coats and funicular 2 Seedling expression throughout root and in elongation zone. GFP T2 Mature expression lateral inflorescences. Source Promoter Organism: Vector: Marker Type:	hattern: High GFP expression in carpels and in us of mature siliques. High GFP expression in vasculature expidermis and cortex cells near root tip at expressed in root hair and root cap. High GFP expression in roots and Arabidopsis thaliana, Columbia (Col) ecotype pNewbin4-HAP1-GFP GFP-ER	35		
Observed expression proceed coats and funicular 2 Seedling expression throughout root and in elongation zone. GFP T2 Mature expression: lateral inflorescences. Source Promoter Organism:	attern: High GFP expression in carpels and in us of mature siliques. High GFP expression in vasculature expidermis and cortex cells near root tip at expressed in root hair and root cap. High GFP expression in roots and Arabidopsis thaliana, Columbia (Col) ecotype pNewbin4-HAP1-GFP			

inductions completed.					
Treatment:	Age:	Gen:		Events Screened/ Response	Response:
1. Nitrogen-high N to low N [14.3 mM KNO ₃ to 28.6 mM Mannitol]	4 wks	T2	72 Hr	12/0	None
Inducible expression summary:					
Treatment:		point uced:		Organs nduced:	Tissues induced:

Table 1-13. T1	Mature Plant	Expression	Organs/Tissues	screened

Events Sc	reened: n = 12	Events Expressing: Expression Detected	n = 12 (01-12)	
Flower H carpel H silique Silique H carpel Ovule Post-fertilization: H funiculus H seed coat				
Table 2-13. T2 Seedling Expression Tissues Screened				
Events Sci	reened: n = 11	Events Expressing: n = 9 (02-04, 06, 09-11, 13, 14)		

	52					
	-continued					
	Promoter Expression Report YP2563 (SEQ ID NO: 13)					
	Expression Detected					
0	Hypocotyl H epidermis L vascular Cotyledon L epidermis Rosette Leaf H mesophyll H epidermis Primary Root L epidermis L cortex H vascular H root cap H root hairs Lateral root L epidermis					
	Table 3-13. T2 Mature Plant Expression Organs/Tissues screened					
	Events Screened: n = 5 Events Expressing: n = 4 (11-15) Expression Detected					
5	Aerial organs H inflorescence L stem Root H mature root					
	Table 4-13. Promoter utility					
20	Trait Area: Nutrient Use Efficiency, Water Use Efficiency, Yield, PG&D, Confinement Sub-trait Area: Nitrogen and Phosphorous Utilization, Drought Tolerance, Seed Size, Plant Establishment, Seed Confinement Utility: Among other uses this promoter sequence could be useful to improve enhance the uptake of nutrients and water from the soil					
5	(root), and seed size (seed coat). Expression in the seed could also be used to engineer seed ablation and seed confinement. Construct: YP2563 Promoter candidate I.D: 25518834 cDNA I.D: 36516796					
0	Events expressing: 01-12					
35	Promoter Expression Report YP2571 (SEQ ID NO: 14) Promoter Tested In: <i>Arabidopsis thaliana</i> , Wassilewskija (WS) ecotype Spatial expression summary:					
	Flower H sepal H petal H anther H vascular H stomata H silique					
Ю	Silique H style H carpel H funiculus H vascular Leaf L vascular Aerial organs H inflorescence Root H mature root Hypocotyl H vascular Cotyledon H mesophyll H vascular H epidermis					
-5	Primary Root H vascular Observed expression pattern: T1 Mature expression: High GFP expression detected in inflorescences. T2 Seedling expression: High GFP expression in vasculature of hypocotyls, cotyledons and root.					
0	GFP expression in epidermis and mesophyll cells of cotyledons. T2 Mature expression: High GFP expression in vasculature and guard cells of sepals and in anthers, petals and silique of flowers. GFP expressed in style, carpels and vasculature of silique. Not expressed in ovules or seed. Low GFP expression in leaf vasculature. High GFP expression in root. Source Promoter Organism: Arabidopsis thaliana,					
	Vector: Columbia (Col) ecotype Vertor: pNewbin4-HAP1-GFP Marker Type: GFP-ER					

60	Treatment:	Age:	Gen:	Time points:	Events Screened/ Response	Re- sponse:
	1. ABA—[uM]	14 days	T2	4 hrs	6/0	No
	2. Heat—28 C.	15 days	T2	24 hrs	3/3	Low
65	3. Heat—36 C.	9 days	T2	>24 hrs	6/3	High

-continued						-continued		
Promot	ter Expressio	n Report	YP2571 (S	SEQ ID NO:	14)	-	Silique	H ovule
4. Heat—41 C.	14	T2	4 hrs	3/3	Medium	-	Ovule	Pre-fertilization: H funiculus Post-fertilization: H funiculus
5. Heat—41 C.	days 15	Т2	24 hrs	3/0	No	5	Aerial organs Root	H flowers H mature root
	days						Rice	
6. Heat—46 C.	14 days	T2	4 hrs	3/0	No		Root Meristem	H not-specific H not-specific
7. Heat—40 C.	4 wks	T2	24 hrs	6/6	Low	10		Observed expression pattern:
8. Drought	4 wks		1.0% moisture	6/6	High	10	Arabidopsis:	
	Inducib	le expres	sion summ	arv.		-	T1 Mature expression: end of the funiculus in	High GFP expression at the distal
		те екртев	SIOH SHITH	ary.		-	and seed. GFP highly	localized to adaxial side at the
	Time point	Organs		Tissues		15	base of the pedicel, in stipules of leaves.	structures resembling
Γreatment:	induced:	induce		induced:		_	T2 Seedling expression	n: No GFP expression detected.
2. Heat—28 C.	24 hrs	Cotyled					T2 Mature expression: Rice:	GFP detected at the base of pedicles and roots.
3. Heat—36 C.	>24hrs	Rosette Cotylec		Ep, Me, V	·s		T0 Seedling expression	n: GFP expression was
	· 2-mis	Rosette		Ep, Me, V		20	detected strongly throu meristematic tissues co	aghout the root and in orresponding to the stem nodes.
4. Heat—41 C.	4 hrs	Root Cotyled	dons	Vs				nism: Arabidopsis thaliana,
		Rosette	e leaf				Columbia (Col) ecotyp	
7. Heat—40 C.	24 hrs	Flower Stem		Se, Pe, Vs Ph, Vs			Vector: Marker Type:	pNewbin4-HAP1-GFP GFP-ER
		Leaf		Vs		25	Generation Screened:	X T1 Mature X T2 Seedling X T2 Mature
3. Drought	1.0% moisture	Flower Stem Leaf		Se, Pe, Ca Xy, Ph, Vs Ep, Me		23	Table 1-14. T1 M	fature Plant Expression Organs/Tissues screened
		Root		Ep			Events Screened: n = 6	Events Expressing: $n = 3$
TABLE 1-14	. T1 Mature	Plant Exp	oression O	rgans/Tissues	screened	•	Expression Detected Flower	H pedicel
						- 30	Silique	H ovule
Events Screened: Expression Detec		Events	Expressin	$g: n = 1 \ (04)$			Ovule	Pre-fertilization: H funiculus
nflorescence	H flowers							Post-fertilization: H funiculus
TABLE	E 2-14. T2 Se	edling E	xpression [Tissues Scree	ened	-	Table 2-14.	T2 Seedling Expression Tissues Screened
Events Screened: Expression Detec		Events	Expressin	g: n = 2 (04,	05)	- 35	Events Screened: n = 6 No GFP Expression D	
Hypocotyl Cotyledon Primary Root	H vascula H mesoph H vascula	ıyll H vas	scular H ep	oidermis			Table 3-14. T2 M	fature Plant Expression Organs/Tissues screened
TABLE 3-14			pression O	rgans/Tissues	screened	- 40	Events Screened: n = 6 Expression Detected	
Events Screened:	: n = 6	Events	Expressin	g: n = 6 (01-	06)	-	Aerial organs Root	H flowers H mature root
Expression Detec	cted				/			
Flower	H sepai H		anther H v e	ascular			Table 4-14,	To Rice Seedlings Organs/Tissues screened
Silique	H style H	carpel H		H vascular		45	Events Screened: n = 1	13 Events Expressing: n = 4 (01, 02, 06, 09)
Leaf Aerial organs	L vascular H inflores						Expression Detected	IIt:G-
Root	H mature	root					Root Meristem	H not-specific L not-specific
	TABLI	∃ 5-14. Pı	romoter ut	ility		- - 50		Table 5-14. Promoter utility
Γrait Area: Water							Trait Area:	Yield, PG&D, Confinement,
Sub-trait Area: D Utility: Among o								Water Use Efficiency
mprove plant tol							Sub-trait Area:	Seed Number, Seed Growth,
Construct:			YP2571			55	Hility: Among other :	Plant Size, Growth Rate uses this promoter sequence is
Promoter candidate I.D: 29223786 cDNA I.D: 23618816 Events expressing: 01-06					33	useful to improve plan		
							increase the number of	f secondary floral branches.
	o·		-1 00			-	Expression in the root	
							improved uptake and t Arabidopsis Construct:	ransport of water and nutrients. YP2590-4rabidonsis
						60	•	-
						- "	cDNA I.D:	23523207
Events expressing	ton Ever'-	n Dominat	VD2500 (DEO ID NO	15)			
Events expressing	ter Expressio	n Report	YP2590 (S	SEQ ID NO:	15)	_	Events expressing:	02, 03, 04
Events expressing						-	Rice Construct:	02, 03, 04 PD3146
Events expressing Promot	ed In: Arabid	opsis that		silewskija (V		- -	Rice Construct: Promoter candidate I.I.	02, 03, 04 PD3146 D: 55210297
Events expressing Promot	ed In: Arabid	opsis that	liana, Was	silewskija (V		- - 65	Rice Construct:	02, 03, 04 PD3146

	55		.,731 D 2	56			
Promoter Expression Report YP2606 (SEQ ID NO: 16)			-continued				
Promoter Tested In: Arabide	opsis thaliana, Wassilewskija (WS) ecotype		Generation Screened: X T1 Matu X T2 Mature T3 Seedling	re X T2 Seedling			
•	expression summary:	5	Table 1-16. T1 Mature Plant	t Expression Organs/Tissues screened			
Primary Root Mature Root	H epidermis H cortex H epidermis L cortex H stele H vasculature //ed expression pattern:		Events Screened: n = 4 No GFP Expression Detected	Events Expressing: n = 0			
	<u> </u>		Table 2-16. T2 Seedlin	ng Expression Tissues Screened			
T1 Mature expression: No GF T2 Seedling expression: Root GFP expressed in root epider T2 Mature expression: Root s GFP expressed in root epider	specific GFP expression. nis and cortex cells. pecific GFP expression.	10	Events Screened: n = 5 Expression Detected	Events Expressing: n = 5 (01-05)			
stele, non-ground cell vascula	r region of root.		Primary Root	H epidermis H cortex			
Source Promoter Organism: A Columbia (Col) ecotype	rabidopsis thaliana,	15	Table 3-16. T2 Mature Plant	t Expression Organs/Tissues screened			
Vector: Marker Type: Generation Screened: X T1 M.	pNewbin4-HAP1-GFP GFP-ER fature X T2 Seedling X T2 Mature		Events Screened: n = 4 Expression Detected	Events Expressing: n = 3 (02, 03, 04)			
Table 1-15. T1 Mature P	lant Expression Organs/Tissues screened	20	Flower	H stamen H filament H anther			
Events Screened: n = 6	Events Expressing: n = 0	20	Aerial organs Mature Root	H flowers H epidermis L cortex H stele			
No Expression Detected	dling Evenossian Tiemes Company		Table 4-1	6. Promoter utility			
	Expression Tissues Screened	25	Trait Area:	Water Use Efficiency, Nutrient			
Events Screened: n = 6 Expression Detected Primary Root	Events Expressing: n = 3 (04-06) H epidermisH cortex	25	Use Efficiency, Sterility Sub-trait Area: Drought Tolerance, Nitrogen and Phosphorous Utilization, Male Sterility				
Table 3-15. T2 Mature P	lant Expression Organs/Tissues screened		Utility: Among other uses this pruseful to improve the uptake of v				
Events Screened: n = 6 Expression Detected	Events Expressing: n = 2 (04, 06)	30	the soil. It could also be used to Construct: Promoter candidate I.D:	engineer male sterility. YP2608 25656951			
Mature Root	H epidermis H cortex H stele H vasculature		cDNA I.D: Events expressing:	5680676 02, 03,04			
Table	4-15. Promoter utility			scribed promoter are identified in the miscellaneous			
Trait Area: Sub-trait Area:	Water Use Efficiency, Nutrient Use Efficiency Drought Tolerance, Nitrogen and Phosphorous Utilization	35	feature section of the relevant SEQ ID) in the Sequence Listing. Those fragments were procedures as described above, and the results are			
Utility: Among other uses this to improve the uptake of water	er and nutrients.	40	Promoter Expression Re	eport YP2683 (SEQ ID NO: 18)			
Construct: Promoter candidate I.D:	YP2606 25086987						
cDNA I.D: Events expressing:	36511228 04-06			is thaliana, Wassilewskija (WS) ecotype pression summary:			
feature section of the relevant SEC	described promoter are identified in the miscellaneous Q ID in the Sequence Listing. Those fragments were ame procedures as described above, and the results are weakly in roots in rice.	45 50	Stem L H H Leaf H Cotyledon L Primary Root L Aerial Organs L	petal L anther H pollen cortex L vascular H xylem I procambium I vascular L epidermis L stomata epidermis L hydathode cortex L epidermis H vascular flower epidemis H cortex H vascular			
Promoter Expression	n Report YP2608 (SEQ ID NO: 17)	30	Н	I quiescent center expression pattern:			
	opsis thaliana, Wassilewskija (WS) ecotype expression summary:		T1 Mature expression: Expression	n is primarily in pollen and in			
Primary Root Aerial organs Flower Mature Root Observ	H epidermis H cortex H flowers H stamen H filament H anther H epidermis L cortex H stele red expression pattern:	55	vascular tissues in leaves and ster Expression appears to be stronges vascular cells (procambium), whi to the xylem and phloem. T2 Seedling expression: Expressi in the root vasculature. Weak exp	st in the meristematic ich gives rise on is observed primarily pression			
T1 Mature expression: No GF T2 Seedling expression: GFP T2 Mature expression: High s expression corresponding to the Arabidopsis flower. GFP expr Source Promoter Organism: A Columbia (Col) ecotype Vector: Marker Type:	expressed in root epidermis. tamen specific GFP hird organ whorl in essed in sub-epidermal cells in root.	60	Vector: pl	s observed in the floral tissue. In the vascular tissues as oidopsis thaliana, Columbia (Col) ecotype Newbin4-HAP1-GFP FF-ER			

continued

Table 1-17. T1 Mature	Plant Expression Organs/Tissues screened
Events Screened: n = 8	Events Expressing: n = 3 (02, 03, 06)
Expression Detected Flower Stem	L petal L anther H pollen L cortex L vascular H xylem
Leaf	H procambium H vascular L epidermis L stomata
Table 2-17. T2 S	eedling Expression Tissues Screened
Events Screened: n = 6	Events Expressing: n = 3 (02, 05, 06)
Expression Detected Cotyledon	L epidermis L hydathode
Primary Root	L epidermis L cortex H vascular
	Plant Expression Organs/Tissues screened
Events Screened: n = 6	Events Expressing: $n = 4$ (02, 03, 05, 06)
Expression Detected Aerial organs	L flower
Mature Root	L epidermis H cortex H vascular H quiescent center
Tabl	e 4-17. Promoter utility
Trait Area:	Sterility, Water Use Efficiency,
Sub-trait Area:	Nutrient Use Efficiency Male-Sterility, Drought Tolerance, Nitrogen and Phosphorous Utilization
Utility: Among other uses the useful to improve the uptake	nis promoter sequence is
nutrients from the soil. The	pollen expression could be used r gene-confinement.
Construct:	YP2683
	YP2683 41958148 36558613

Flower	H vascular H ovule
Silique	H septum
Ovule	Pre-fertilization:
	H outer integument
	Post-fertilization:
	H seed coat H embryo
Embryo	H radicle H late mature
Stem	H epidermis H vascular
	H xylem H phloem
Leaf	L vascular H trichome
Hypocotyl	L epidermis
Cotyledon	L epidermis
Rosette Leaf	H epidermis H trichome
	H primordia
Primary Root	H epidermis H cortex H vascular
	L quiescent center L columella
Mature Root	H epidermis H cortex H vascular
	H endodermis
	H parenchyma H stele
	Observed expression pattern: All cells in root.

T1 Mature expression: Promoter expression is observed in the pre-fertilized ovule integuments as well as the seed coat and the developing radicle of the embryo. Expression is also observed weakly in the vasculature of the stem and leaf trichomes. T2 Seedling expression: Seedling expression is primarily in all cells of the root.

-continued

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5	T2 Mature: Expression is strong Source Promoter Organism: <i>Arab</i> Vector: Marker Type: Generation Screened: X T1 Matu	nidopsis thaliana, Columbia (Col) ecotyp pNewbin4-HAP1-GFP GFP-ER					
	Table 1-18. T1 Mature Plan	t Expression Organs/Tissues screened					
10	Events Screened: n = 8 Expression Detected Flower Silique Ovule	Events Expressing: n = 5 (02, 03, 05, 07, 08) H vascular H ovule H septum Pre-fertilization: H outer integument Post-fertilization:					
15	Embryo Stem	H seed coat H embryo H late H radicle H epidermis H vascular H xylem H phloem L vascular H trichome					
20	Table 2-18. T2 Seedling Expression Tissues Screened						
	Events Screened: n = 6 Expression Detected	Events Expressing: n = 3 (02, 03, 05)					
25	Hypocotyl Cotyledon Rosette Leaf	L epidemis L epidemis H epidemis H trichome H primordia Root H epidemis					
		H cortex H vascular L quiescent center L columella					
30	Table 3-18. T2 Mature Plant Expression Organs/Tissues screen						
	Events Screened: $n = 6$	Events Expressing: n = 5 (01-05)					
35	Expression Detected Leaf Aerial organs Mature Root	H trichome L stem H epidermis H cortex H vascular H endodermis H parenchyma H stele					
	Table 4-18. Promoter utility						
40	Trait Area: Water Use Efficiency, Sub-trait Area: Drought Tolerance Phosphorous Use Efficiency Utility: Among other uses this pr	e, Nitrogen and					
45	useful to improve the uptake and water and nutrients from the soil water throughout the vasculature. Construct: Promoter candidate I.D:	utilization of and the transport of					

One or more fragments of the above described promoter are identified in the miscellaneous feature section of the relevant SEQ ID in the Sequence Listing. Those fragments were tested for promoter activity by the same procedures as described above, and the results are summarized below.

Ceres Promoter PD3238 expresses weakly in roots in rice. No expression is observed for Ceres Promoter PD3229 and Ceres Promoter PD3243.

36540030

01-05, 07, 08

cDNA I.D:

55

Events expressing:

Promoter Expression Report YP2832 (SEQ ID NO: 20)

Promoter Tested In: Arabidopsis thaliana, Wassilewskija (WS) ecotype Spatial expression summary:

60	Flower	H pedicel H receptacle H style
	Silique	H style H septum
	-	H abscission zone
	Stem	H epidermis H cortex
	Leaf	L stomata
	Primary Root	L epidermis H cortex
65		H endodermis H phloem
	Lateral Root	H primordia

-continued		-continued				
Aerial Organs Mature Root	H stem L epidermis H cortex H endodermis H phloem H lateral roots	5	Stem Leaf Inflorescence	H epidermis H pith L epidermis H mesophyll H floral meristem H floral primordia		
Observed expression pattern: T1 Mature expression: Expression is primarily observed in			Hypocotyl Cotyledon Rosette Leaf	H epidermis H cortex H vascular H epidermis H mesophyll H epidermis H mesophyll		
the epidermis and cortical layers of the stem tissue. T2 Seedling expression: The promoter is expressed in the most mature parts of the primary root, and			Primary Root	H stipule H epidermis H cortex H vascular H quiescent center		
is not detected in the root tips. The root expression is also observed in the buds giving rise to the secondary root branches. No expression is observed			Lateral root	H root meristem H root cap H primordia H flanking cells H lateral root cap		
in the hypocotyl or cotyledons. T2 Mature expression: Expression observed in non-leaf tissues. Source Promoter Organism: <i>Arabidopsis thaliana</i> , Columbia (Col) ecotype			Aerial organs	H inflorescence H floral meristem H floral primordia H flowers H silique		
Vector: pNewbin4-HAP1-GFP Marker Type: GFP-ER Generation Screened: X T1 Mature X T2 Seedling XT2 Mature			Root Obser	H stem H leaf H mature root ved expression pattern:		
Table 1-19. T1 Mature	Plant Expression Organs/Tissues screened			broad GFP expression throughout		
Events Screened: n = 6 Expression Detected Flower Silique	Events Expressing: n = 5 (01-05) H pedicel H receptacle H style H style H septum	20	aerial organs. High GFP expr in flowers, stems, and leaves. meristems and primordia. Hig in carpels, ovules and develo	. GFP expressed in floral gh GFP expression ping seed in silique. GFP		
Stem Leaf	H abscission zone H epidermis H cortex L stomata	25	expressed in outer integumen seed coat and embryos in ove pith and epidermis in stem ar mesophyll of leaf.	ules. GFP expressed in		
Table 2-19. T2 S	Seedling Expression Tissues Screened		T2 Seedling expression: High organs and root. High GFP expression organs are root.			
Events Screened: n = 6	Events Expressing: n = 4 (01, 02, 04, 05)	20	epidermis and mesophyll cells of cotyledon and rosette leaf. High GFP expression in epidermis,			
Expression Detected Primary Root L epidermis H cortex H endodermis H phloem		30	expressed in root meristem cells and root cap. T2 Mature Expression: High boad GFP expression			
Lateral root H primordia Table 3-19. T2 Mature Plant Expression Organs/Tissues screened			throughout aerial organs and Source Promoter Organism:			
Events Screened: n = 6 Events Expression Organis rissues screened Events Screened: n = 6 (01-06) Expression Detected			Columbia (Col) ecotype Vector: CRS338-GFP Marker Type: GFP-ER			
Aerial organs H stem Mature Root H lateral root L epidermis H cortex H endodermis H phloem			Generation Screened: X T1 Mature X T2 Seedling X T2 Mature Table 1-20. T1 Mature Plant Expression Organs/Tissues screened			
Tab	le 4-19. Promoter utility	40	Events Screened: n = 6	Events Expressing: n = 6 (01-06)		
Trait Area:	Source, Water Use Efficiency, Nutrient Use Efficiency		Expression Detected X Flower	H pedicel L petal H carpel H style H epidermis H silique		
Sub-trait Area: Carbon/Nitr Tolerance, Nitrogen and Ph Utility: Among other uses t		45	X Silique X Ovule	H style H carpel H septum H epidermis H ovule Pre-fertilization:		
to engineer the storage of c	earbon into mprove the uptake of water and n the soil, and		A Ovuic	H outer integument Post-fertilization: H outer integument		
Construct: Promoter candidate I.D:	YP2832 32258957	50	X Embryo	H seed coat H mature endosperm H mature		
cDNA I.D: Events expressing:	36551046 01-06		X Stem X Leaf X Inflorescence	H epidermis H pith H mesophyll L epidermis H floral meristem H floral primordia		
		55	Table 2-20. T2 Se	edling Expression Tissues Screened		
Promoter Expression Rep	oort PD2995 (aka PD2263) (SEQ ID NO: 21)	-	Events Screened: n = 6	Events Expressing: n = 6 (01-06)		
Promoter Tested In: <i>Arabidopsis thaliana</i> , Wassilewskija (WS) ecotype Spatial expression summary:			GFP Expression Detected X Hypocotyl H epidermis H cortex H vascular X Cotyledon H mesophyll H epidermis			
Flower	H epidermis L petal H carpel H style H silique H pedicel	60	X Rosette Leaf	H mesophyll H epidermis H stipule		
Silique	H epidermis H style H carpel H septum H ovule Pre-fertilization:		X Primary Root	H epidermis H cortex H vascular H quiescent center H root meristem H root cap		
Ovule Pre-fertilization: H outer integument Post-fertilization: H seed coat H outer integument Embryo H mature endosperm H mature		65	X Lateral root	H primordia H flanking cells □ H lateral root cap		

-continued

Events Screened: n = 6 GFP Expression Detected	Events Expressing: $n = 6 (01-06)$
X Aerial organs	H inflorescence H floral meristem H floral primordia H flowers H silique H stem H leaf
X Root	H mature root

Trait Area:	Water Use Efficiency, PG&D,			
	Seed, Nutrient, and Yield			
Sub-trait Area: Water use efficiency, growth rate,				
seed yield, and nutrient utilizar	tion			
Utility: Among other uses this	promoter sequence is			
useful to modulate plant growt	h and architecture			
and the utilization and water and nutrients.				
Construct: PD2263				
Promoter candidate I.D: 38960222				
cDNA I.D: 36549595				
Events expressing:	01-06			

One or more fragments of the above described promoter are identified in the miscellaneous feature section of the relevant SEQ ID in the Sequence Listing. Those fragments were tested for promoter activity by the same procedures as described above, and the results are summarized below.

summarzed below.

For Ceres Promoter PD2926, expression is weak compared to the full-length promoter PD2995; the promoter remains active in all tissues except the embryo.

For Ceres Promoter PD3048, expression is weak compared to the full-length promoter PD2995; the promoter remains active in all tissues.

For Ceres Promoter PD3182, no expression is observed.

For Ceres Promoter PD3345, expression is very weak.

For Ceres Promoter PD3503, no expression is observed.

For Ceres Promoter PD3676, expression is weak compared to the full-length promoter PD2995; the promoter expresses at higher levels in vegetative tissues than in reproductive

Promoter Expression Report PD2999 (aka PD2258) (SEQ ID NO: 22) Promoter Tested In: Arabidopsis thaliana, Wassilewskija (WS) ecotype

Flower	H pedicel H sepal H carpel
	H style H stigma H silique
Silique	H stigma H style H carpel
*	H placentae H funiculus
	H epidermis H ovule
Ovule	Pre-fertilization: H primordia
	H inner integument
	H outer integument
	H funiculus
	Post-fertilization: H outer
	integument H seed coat
	H early endosperm
	H mature endosperm
	H embryo

H suspensor H heart H mature Embryo H radicle H cotyledons Stem H epidermis H cortex H interfascicular region H vascular H xylem H phloem H pith

Leaf H mesophyll H epidermis Inflorescence H floral meristem H floral primordia

Hypocotyl H epidermis H cortex H vascular Cotyledon H epidermis H vascular Rosette Leaf H epidermis Primary Root H vascular H root cap

Observed expression pattern:

T1 Mature expression: Broad high GFP expression throughout aerial organs. High GFP expression in inflorescence, floral meristem, flowers, siliques. High GFP expression throughout tissues of silique. GFP expressed in carpels, placenta, ovule primordia, developing ovules, embryo, and endosperm. High GFP expression in outer integuments and seed coats of developing ovules and seed.

-continued

High GFP expression in vascular and ground tissues of stem.
High GFP expression in epidermis and mesophyll cells of leaf.
T2 Seedling expression: Broad expression throughout

aerial organs and vasculature of root.

Source Promoter Organism: Arabidopsis thaliana,

Columbia (Col) ecotype

Vector: CRS338-GFP GFP-ER Marker Type: 10 Generation Screened: X T1 Mature X 12 Seedling

Table 1-21. T1 Mature Plant Expression Organs/Tissues screened

	Events Screened: n = 6	Events Expressing: n = 4 (01, 02, 04, 06)
15	GFP Expression Detected	(01, 02, 04, 00)
	X Flower	H pedicel H sepal H carpel
		H style H stigma H silique
	X Silique	H stigma H style H carpel
	-	H placentae H funiculus
20		H epidermis H ovule
	X Ovule	Pre-fertilization: H primordia
		H inner integument
		H outer integument H
		funiculus
25		Post-fertilization: H outer
25		integument H seed coat
		H early endosperm H
		mature endosperm H embryo
	X Embryo	H suspensor H heart H mature
		H radicle H cotyledons
30	X Stem	H epidermis H cortex
		H interfascicular region
		H vascular H xylem H
		phloem H pith
	X Leaf	H mesophyll H epidermis
35	X Inflorescence	H floral meristem
33		H floral primordia

Table 2-21. T2 Seedling Expression Tissues Screened

	Events Screened: n = 6	Events Expressing: $n = 5$	
40		(01-04, 06)	
	☐ No GFP Expression Detected		
	X Hypocotyl	H epidermis H cortex H vascular	
	X Cotyledon	H epidermis H vascular	
	X Rosette Leaf	H epidermis	
	X Primary	Root H vascular H root cap	
45			
	Table 3-21. Promoter utility		

	Trait Area:	Water use efficiency, PG&D,	
50	Sub-trait Area:	Seed, Nutrient, Yield Water use efficiency, growth rate, seed size and yield, and nutrient use	
	Utility: Among other uses this promoter sequence is useful to improve: Water use efficiency, PG&D, Seed, Nutrient, Yield		
55	Construct:	PD2258	

Promoter candidate I.D: 38960200 23478038 cDNA I.D: Events expressing: 01, 02, 04, 06

For Ceres Promoter PD2929, expression is very weak compared to the full-length promoter PD2999; the promoter remains active in all tissues.

For Ceres Promoter PD3183, expression is only detected in anthers and stigma.

For Ceres Promoter PD3240, expression is very weak compared to the full-length promoter

PD2999; the promoter remains active in all tissues. For Ceres Promoter PD3266, no expression is detected in rice.

One or more fragments of the above described promoter are identified in the miscellaneous feature section of the relevant SEQ ID in the Sequence Listing. Those fragments were tested for promoter activity by the same procedures as described above, and the results are summarized below.

64 -continued cDNA I.D: NA Events expressing: 01, 03-04, 06-11,14 Promoter Expression Report YP2680 (SEQ ID NO: 24) Promoter Tested In: Arabidopsis thaliana 10 Spatial expression summary: Leaf: epidermis, vasculature Flower: vascular Silique: abscission zone, vascular Primary root: cortex, epidermis, root hairs, vascular Lateral root: cortex, epidermis, vascular Mature root: not-specific Observed expression pattern: Primarily root expression Source Promoter Organism: Arabidopsis thaliana Vector: Binary TC(815) Marker Type: erGFP Generation Screened: T1 Mature, T2 Seedling, T2 Mature Table 1-23. T1 Mature Plant Expression Organs/Tissues screened Events Screened: n = 8Events Expressing: n = 7(01, 03-08)Organs 25 Leaf: epidermis, vasculature Flower: vascular Silique: abscission zone, vascular Table 2-23. T1 Seedling Expression Tissues Screened 30 Events Screened: n = 5Events Expressing: n = 5 (01-05)Primary root: cortex, epidermis, root hairs, vascular Lateral root: cortex, epidermis, vascular Table 3-23. T2 Mature Plant Expression Organs/Tissues screened 35 Events Screened: n = 6 Events Expressing: n = 5(01-03, 06, 08)Organs Mature root: not-specific Primary root: cortex, epidermis, vascular Lateral root: cortex, epidermis, vascular Promoter utility: 40 Trait Area: Drought Tolerance, Nutrient Utilization Sub-trait Area: Water Utilization, Phosphate and Nitrate Utilization Utility: Among other uses, this promoter sequence is useful to improve: the uptake and transport of water and nutrients from the soil to support vegetative growth. Notes: 1000 nt upstream of atg YP2664 Construct:

SR/OS Line: SR04406 Promoter candidate I.D: 41958160 cDNA I.D: 36511557 Events expressing: 01-08

One or more fragments of the above described promoter are identified in the miscellaneous feature section of the relevant SEQ ID in the Sequence Listing. Those fragments were tested for promoter activity by the same procedures as described above, and the results are susumarized below.

Ceres Promoter PD3584 is a strong, broadly expressing promoter; comparable to the full-length promoter PD3141.

Promoter Expression Report PD3147 (aka YP2663) (SEQ ID NO: 25)

Promoter Tested In: Arabidopsis thaliana, Wassilewskija (WS) ecotype Spatial expression summary:

H pedicel H sepal

H petal H filament H anther

H epidermis H cortex H vascular

Promoter Expression Report PD3141 (SEQ TD NO: 23)

Promoter Tested In: Orvza sativa Spatial expression summary:

Tiller: not-specific

Main culm: bundle sheath, endodermis, epidermis, internode, ligule, node, pericycle, phloem,

sclerenchyma layer, vasculature, xylem

Root: cortex, epidermis, vascular

Leaf: epidermis, leaf blade, leaf sheath, mesophyll,

petiole, stipule, stomata, vasculature

Meristem: floral meristem, shoot apical meristem, vegetative meristem Panicle: flag leaf, ovary, peduncle, primary branch, rachilla, rachis, spiklet Spikelet: floral meristem, shoot apical meristem, vegetative meristem

Observed expression pattern: Broad Source Promoter Organism: Oryza sativa

Vector: Binary DF EGFP Marker Type: EGFP

Generation Screened: To Seedling and To Mature

Induction Data

Table 1-22. To Seedling Expression Organs/Tissues screened

Events Screened: n = 12Events Expressing:

Organs

Tiller: not-specific Main culm: not-specific Root: not-specific Leaf: not-specific Meristem: not-specific

Table 2-22. T0 Mature Plant Expression Tissues Screened

Events Screened: n = 6

Events Expressing: n = 6(01, 04, 07-08, 11, 14)

n = 9 (01, 03-04, 06-11)

Organs

Main culm: bundle sheath, endodermis, epidermis, internode, ligule, node, pericycle, phloem,

sclerenchyma layer, vasculature, xylem

Root: cortex, vascular

Panicle: flag leaf, ovary, peduncle, primary branch,

rachilla, rachis, spiklet

Spiklet: flag leaf, floret(palea), lemma, ovule,

pedicle, pollen, seed, stigma

Leaf: epidermis, leaf blade, leaf sheath, mesophyll

Meristem: floral meristem, shoot apical meristem, vegetative meristem

Promoter utility

Trait Area: Yield, Composition, Disease, Stress Tolerance, Nutrient Use Efficiency, Nutrient Utilization Sub-trait Area: Biomass, Lignin composition, Disease resistance, Salt tolerance, Drought tolerance,

Phosphate and Nitrate Use Efficiency, Phosphate and Nitrate Utilization Utility: Among other uses, this promoter sequence is useful to improve: the biomass of the plants under normal and stressful conditions

through the

overexpression of trait-specific transgenes.

Construct: PD3141 OS00486 SR/OS Line: 54507599 Promoter candidate I.D:

Flower

L carpel L silique L carpel L funiculus

Silique Stem

H xylem H phloem L pith

-continued

Leaf	H epidermis H mesophyll H vascular	
Hypocotyl	H epidermis H cortex	
Cotyledon	H epidermis H mesophyll	
Rosette Leaf	H epidermis H mesophyll	
Aerial organs	H inflorescence H flowers	
	L silique H stem H leaf	
	Observed expression pattern:	

- T1 Mature expression: High GFP expressed in aerial tissues. High GFP expression in leaf, stem and inflorescence. High GFP expressed in pedicels, sepals, petals and stamens. Low GFP expression in epidermis of silique. No GFP expression observed in embryo. High GFP expression in epidermis, cortex, phloem and xylem cells of vasculature. Low GFP expression in pith sells of stem. High GFP expression in epidermis, mesophyll and vasculature of leaves.
- T2 Seedling expression: High GFP expressed in aerial tissues. High GFP expression in epidermis and mesophyll cells of cotyledons and rosette leaves and in epidermis and cortex of hypocotyls. No GFP expression observed in roots.
- T2 Mature expression: GFP expressed in aerial tissues. High GFP expression in leaf, stem and inflorescence. No GFP expression observed in roots.

Source Promoter Organism: Arabidopsis thaliana, Columbia (Col) ecotype Vector: pNewbin4-HAP1-GFP

Marker Type: pNewbin4
GFP-ER

Generation Screened: X T1 Mature X T2 Seedling X T2 Mature

Inductions completed.					
Treatment:	Age:	Gen:	Time points:	Events Screened/ Response	Response:
1. Nitrogen-high N to low N [14.3 mM KNO ₃ to 28.6 mM Mannitol]	4 wks	Т2	72 hrs	6/0	No
Inducible expression summary: Treatment: Time point induced:	Organs	induced	: Tissues induced	1:	
Table 1-24. T1 M	ature Plant	Expres	sion Organs/Tiss	ues screened	
Events Screened: n = 6 Events Expressing: n = 4 (02, 04-06) GFP Expression Detected					
X Flower	H pedicel H sepal H petal H filament H anther L carpel L silique				
X Silique X Stem	L carpel L fimiculus				
A Stelli	H epidermis H cortex H vascular H xylem H phloem L pith				
X Leaf	· · · · · · · · · · · · · · · · · · ·				
Table 2-24.	T2 Seedlir	ıg Expr	ession Tissues Sc	reened	
Events Screened: n = 6 Events Expressing: n = 4 (02, 04-06) GFP Expression Detected					
X Hypocotyl	H epide	rmis H	cortex		
X Cotyledon	H mesophyll H epidermis				
X Rosette Leaf	H mesophyll H epidermis				
Table 3-24. T2 M	ature Plant	Expres	sion Organs/Tiss	ues screened	
Events Screened: n = 5 GFP Expression Detected	1 2 , , ,				
X Aerial organs	H inflorescence H flowers L silique H stem H leaf				
	Table 5-2	4. Prom	oter utility		
Trait Area: PG&D, Confinement, Hybrid Production Sub-trait Area: Plant Size, Growth Rate, Carbon Fixation Utility: Among other uses this promoter sequence is useful to improve carbon fixation in above ground tissues to increase biomass and seed yield. Expression is useful for engineering of carbon and nitrogen ratios. Expression in flowers is deployed for sterility and confinement. Construct: Y132663 Promoter candidate LD: 25087104 cDNA I.D: 36567145 Events expressing: 02, 04-06					

One or more fragments of the above described promoter are identified in the miscellaneous feature section of the relevant SEQ ID in the Sequence Listing. Those fragments were tested for promoter activity by the same procedures as described above, and the results are summarized below. Ceres Promoter PD3721 is a strong, vegetatively expressing promoter; comparable to the full-length promoter PD3147.

Promoter 1	Expression Report For PT0822 (SEQ ID NO: 26)
Promoter Tested In: Arabia	lopsis thaliana, Wassilewskija (WS) ecotype Observed expression pattern:
T2 Seedling expression: Ex Source Promoter Organism Vector: Marker Type:	pression observed in mature root only. (spression specific to epidermis and cortex cells of root. (arabidopsis thatiana, Columbia (Col) ecotype pNewbin4-HAP1-GFP
	Mature T2 Seedling T2 Mature
Root	1 Mature Plant Expression Organs/Tissues screened
	H epidermis
18616 2	-25. T2 Seedling Expression Tissues Screened
	Expression Detected
Primary Root	H epidermis H cortex
Promoter Expression R	Leport for PD3389 (nucleotides 601-1000 of SEQ ID NO: 26)
Promoter Tested In: Oryza	sativa Spatial expression summary:
T0 Seedling	X Root expression in:
CORTEX EPIDERMIS ROOT CAP T0 Mature	
X Root expression: non-sp	ecific. Observed expression pattern:
Deletion of root-specific pr the 5'end of the 1000 nucle	expression specific to epidermis and cortex cells of root. comoter PT0822. 600 nucleotides were deleted from eotide PT0822 promoter (SEQ ID NO: 26). : Arabidopsis thaliana, Columbia (Col) ecotype
Table 1-26.	. To Seedling Expression Organs/Tissues screened
Events Screened: n = 16	Events Expressing: n = 3 (01, 05,13) Organs
X Root expression in: CORTEX EPIDERMIS ROOT CAP	
Table 2-26. T	O Mature Plant Expression Organs/Tissues screened
Events Screened: n = 4 Organs	Events Expressing: $n = 2 (05,13)$
X Root expression: nonspe	cific Promoter utility
Sub-trait Area: Salt toleran Use Efficiency, Phosphate : Utility: Among other uses,	Drought Tolerance, Nutrient Use Efficiency, Nutrient Utilization ce, Drought tolerance, Phosphate and Nitrate

One or more fragments of the above described promoter are identified in the miscellaneous feature section of the relevant SEQ ID in the Sequence Listing. Those fragments were tested for promoter activity by the same procedures as described above, and the results are summarized below. For Ceres Promoter PD3389, expression is observed in the roots of rice seedlings.

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.

SEQUENCE LISTING

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                       note = Motif name:GLMHVCHORD
misc_feature
                       909..916
                       note = Consensus TATABOX
5'UTR
                       956..1000
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source
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
misc_feature
                       1..1000
                       note = Ceres Promoter PT0960
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                                                                    420
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                                                                    840
tttacgtatg cttcagccac ttggattagt tattagtgta aggtaacact actttcgatt
                                                                    900
tattcatcta taaaaaccaa aattagagtg catttcattg atctatacca tcatacttgt
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source
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                       mol_type = genomic DNA
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misc_feature
                       1..400
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ataaatcata ggatcgaata tttacacggt atcaaaacct actcgccgct actatataaa
aattgaagtc aaatatcaac cgcaattatt aaaccagcca gaccaataat tcataaactt
aatataaaca taaataaatt aatgttacac aacgatatat ggtagggtta ttactatctt
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                       note = Ceres Promoter PT0998
misc_feature
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misc_feature
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misc_feature
                       199..209
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                       277..283
misc_feature
                       note = Motif name:ANAERO1CONSENSUS
misc feature
                       note = Motif name: ANAERO1CONSENSUS
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misc_feature
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source
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                       organism = Arabidopsis thaliana
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aaagcttact tgatccaaca tacatactaa ctctcaacag atacaaacac aagtttatta
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                       note = Ceres Promoter YP2219
misc_feature
                       505..509
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misc_feature
                       627..635
                       note = Motif name: TATABOX4
5'UTR
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misc feature
                       542..730
                       note = Ceres Promoter YP2229
source
                       1..730
                       mol type = genomic DNA
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SEQ ID NO: 5
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misc feature
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                       148..157
misc_feature
                       note = Motif name:02F3BE2S1
misc_feature
                       149..155
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misc feature
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                       note = Motif name:ABRELATERD1
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                       774..778
misc feature
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source
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taaaggctac aacaccacaa aggatcatca gtcatcacaa ccacattaac tcttcaccac
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                                                                    1000
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Location/Qualifiers
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misc feature
                       523..528
                       note = Motif name: ACGTCBOX
                       553..559
misc_feature
                       note = Motif name:ABRERATCAL
misc_feature
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misc_feature
                       810..817
                       note = Motif name: INRNTPSADB
misc_feature
                       867..872
                       note = Motif name: TATABOX5
misc_feature
                       901..905
                       note = Putative TSS
5'UTR
                       905..1000
unsure
                       813
                       note = n is a, c, t, g, unknown, or other
source
                       1..1000
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
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gatcgagatg ccaccaaatc ttttcattaa aatgaattgt ggaggacata ccacttttaa
cgaggtcatt tccactgggt gacatgtgga ctctactttg ggtggcatgt tcatatcttt
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ccacatcacc atgtaaacgt gaaaacaccc accacactca cttacatctc aaacacatgt
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cgcaatgtat aatatacaac ttgtaaaaat aaaatatttg aataagcatt ataaataaac
ccaaagaggt gttagattta tatacttaat tgtagctact aaatagagaa tcagagagaa
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SEQ ID NO: 7
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                       Location/Qualifiers
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misc feature
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                       note = Motif name:SURE2STPAT21
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                       1245..1268
                       note = Motif name: PRECONSCRHSP70A
misc_feature
                       1389..1396
                       note = Motif name:P1BS
misc feature
                       1452..1456
                       note = Putative TSS
misc_feature
                       1425..1433
                       note = Motif name: CONSENSUS TATA BOX
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source
                       1..1500
                       mol_type = genomic DNA
organism = Arabidopsis thaliana
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                                                                    1320
ttgcttgctt agtttttgtt ctcgaagctt gacttttgaa acagaatgct gtttaacctc
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                       90..99
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                       202..210
                       note = Motif name: HDZIP2ATATHB2
misc feature
                       454..463
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misc feature
                       479..486
                       note = Motif name:P1BS
misc_feature
                       491..500
                       note = Motif name: CARGCW8GAT
misc_feature
                       742..751
                       note = Motif name: CARGCW8GAT
misc feature
                       945..951
                       note = Motif name:TATABOX4
5'UTR
                       972..999
source
                       1..999
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
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aatatgtcaa gtataacaac tcacatcacc cttttggctt ttggggaaat atcggattta
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taatcaaatt ttatgataat gtctttgtaa acgctataaa ctaaaaattt ataatagtta
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cgtcgtacgt cgactttgta tactacttta tactccgtca aactaaaata acttaaaata
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taataaaatt gtattcatta tattcacaga aaaataaaaa tgttattctt gtaacaagtc
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misc_feature
                       207..214
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                       260..268
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5'UTR
                       978..1000
misc_feature
                       294..300
                       note = Motif name: TATABOX2
                       327..348
5'UTR
source
                       1..1000
                       mol type = genomic DNA
                       organism = Arabidopsis thaliana
SEOUENCE: 9
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gactetteaa ggeecaaaga aaaagagaat atteggeggg ataggggtae gtttgtaatt
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ctcagttgat agagcaccac atttttgtg gtagaaatcg gtttgaatcc gatagcggct
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misc_feature
                       36..41
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misc_feature
                       161..167
                       note = Motif name:ABRERATCAL
misc feature
                       237..246
                       note = Motif name:ABREATRD22
misc feature
                       240..246
                       note = Motif name: ABRERATCAL
misc_feature
                       283..289
                       note = Motif name: TATABOX3
5/UTR
                       317..397
source
                       1..397
                       mol type = genomic DNA
                       organism = Arabidopsis thaliana
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caaagttatc taaaaacaaa aaacaattaa ttagcattcg acgtgtacat atcactcgcc
acqtgtacaa qaqccttqqc ctttttqctt cttcttcttq tctattaata tcatctcctq
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misc_feature
                       530..534
                       note = Motif name:ABRELATERD1
misc feature
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misc_feature
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misc feature
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source
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aattggttgt attcgccctc ataaccaatt ggaaagtggc ctaaataaga gaatgatgcg
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SEO ID NO: 12
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FEATURE
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misc feature
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misc feature
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misc feature
                       216..223
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misc_feature
                       647..653
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                       710..718
misc_feature
                       note = Motif name: SEF1MOTIF
misc_feature
                       835..872
                       note = Motif name:TATABOX
source
                       1..1000
                       mol_type = genomic DNA
organism = Arabidopsis thaliana
SEOUENCE: 12
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cttcagtttt tttttacacg agaatcagta ctcattagca cagcctcatt ttatgctttc
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agatcatcct tttaaataaa tagtcttcac tatagtaaat tcgatttcat tatatctgtc
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tatatatata tatatata tatatatata tactetteta agtatatgat geaatggagt
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SEQ ID NO: 13
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misc feature
                       689..695
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misc feature
                       112..118
                       note = Motif name:DPBFCOREDCDC3
misc feature
                       653..659
                       note = Motif name:ABRERATCAL
misc feature
                       689..695
                       note = Motif name:DPBFCOREDCDC3
misc_feature
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source
                       1..1500
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
SEQUENCE: 13
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attcagtact tgaggttctc tattggacag aagatgaaca tatggactac aacacttgga
                                                                    120
caatccgtaa tggacaacca cacattttgg ttgcagcgta tgtgtcgtta caaaattgaa
                                                                    240
aatcaatgtc cacgaccata catgactgca cagcttggtg tttgataatt caaggaaaaa
gccacataaa ggttctattt cttaatctta cattttggaa gaaatcaaat cattgattta
agaaataata tgtaagcttg gatgctagag ctttaggtga tcctgtggcc agattaggat
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attgatttaa cttttcttac tgctactcaa cattgattgt ctggtattaa gttgtaaaag
agtagtttga cttatccatt tgtgttgata taattggtta ttataatgtg attagtcaat
agaaaacctt agagtacata atatcaaact tttaactttg ttaatttttt taggcgagtg
                                                                   540
ggtgaataca tcagttagac ttgcttttgc acatcatgtg atcatcgttt aattgttata
                                                                   600
tgatcggacg gtcattattt ctcattaact tctatttaaa ctgcaaatct taccacgttc
                                                                   660
atttgatccg aattaaccca ttattctaca cgtgtttggt acacacaatt acaaaccaaa
                                                                   720
accaatgacc ggttattctt ttttcgtgat tcagtttatt tgggggtataa tggtctattt
                                                                   780
tcactccaaa aacaaagaaa aatatctcaa ccttggaaaa tacctgtttt gtctaaactt
                                                                   840
tttcccaaag tgccttatta cggaacattc cctacaaaaa aaccatcgtc atgtactgga
                                                                   900
caaaatattc ttcaagtttg ttttcatgtt gtgttgggcc gtttaattcg catgtatccc
                                                                   960
atatoggaco ttataaactt ataagttata aacattaaaa tataatocat ttgtcaatoo
                                                                   1020
ttttttccac tttttttaat taagataaat cttaagtatt accaaatcat tattaaattt
                                                                   1080
ttatatttat tattctagct ttaccattta cacatacttt acccccattg tattttcatt
gccgaaatgt ttcaaaaaag gatgatttaa ggggcaataa taatatttaa aaaaactgtg
tttagtgatg aaacaaaacc cgccgttgga aattaaacca gcccatagag agaaaaacct
ctcttaacga agaaaaacaa caacatggcc cctggtttct cttctctttc gaatctatct
tettetttta teeaateett teetattttt atteteacet teteetegtt ttteeegaaa
ctgttctttt gccctcttct ctcaatttta atccaccaaa caaatcgaac agtgttcgat
aacttttaga ttgcaagtcc tgtttttgat ttggtcggga gaaagaaaac tagggttttg 1500
SEQ ID NO: 14
                      moltype = DNA length = 2001
FEATURE
                      Location/Qualifiers
misc feature
                       1..2001
                       note = Ceres Promoter YP2571
misc feature
                       397..404
                       note = Motif name: ABREMOTIFAOSOSEM
                       1231..1238
misc_feature
                       note = Motif name:ABREMOTIFAOSOSEM
                      1558..1565
misc_feature
                       note = Motif name: ABREATCONSENSUS
misc_feature
                       1794..1800
                       note = Motif name: TATABOX4
5'UTR
                       1829..2000
                       1..2001
source
                      mol_type = genomic DNA
                      organism = Arabidopsis thaliana
SEOUENCE: 14
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acagttgaga cttgagagac acattaaagc ttgtcatcgt tcactggaga gagtagagtg
                                                                   120
tgtgagagac caagtgacct aacttcactg caacagtagt cggtaaatat ccaaacataa
gagctacgat ggtaacacgt gtacggtgga ggaaaagaga gccctcagat tccgttataa
                                                                   240
aggaggtgag acccactcgc cggaatggaa acaaaggaga aacgtagagg taaatggttg
                                                                   300
ggccactgct tcaccaaaaa gccttttcta tcttcttttt atttttgttt tgtggctttt
                                                                   360
aacggttaac aatataactc ctcattccaa gacaattacg tgtcctctgt acggcttttt
                                                                   420
catttttcac tttctacttc atagtgactt tagggaggcc tcaattttta attactcgag
                                                                   480
gattatttag gtaatcatga atgtaactac agctttacag gtaattaaac agatgagatt
                                                                   540
tagttgcgtg aaatctagct gatctggtgc ttatatcaaa tccaattgat ggagacagat
                                                                   600
ttggacttaa attcatgatt gtatttgtaa tcttatatac gttatagtta aatttctttc
                                                                   660
tctccgtacg tactctaact ataattatta ttaaatgctg aacgcaatgc ttttgagtgc
                                                                   720
tgagaatett tttggtetga tteaaaaatg atgtattagt actetgaatt eaatettaae
                                                                   780
ttctgcaacg aatcaatgta ttaatttata ggagatccgg ataaaattat ggatatatgc
                                                                   840
acgctacttc tttcattttt aattaggtaa atggttataa ctttatttta tatatcaatt
aaatgatttt ggtatgagat tactagtaca ctttctttgc aaatgtttta aacacgacaa
                                                                   960
gacaaaaata ttacaagcat attttggtaa aaaatatcat aagctttcat atcaaaatca
ttagttatga tgttagattt ttttttttt tttttaacac tacaaaaagc tctggtctta
                                                                   1080
atatgttaga aattttagtc caaaccagcc tacagaggat ttagctaaac aattcccaag
caccttttaa gtgttaaccg aaataacgta atatgatgtt aaaggttaca taaaaacaaa
actaaagaat tttcatatga aaagttaacg tacgtgtctt agtgtaacct aattttagtt
cacagtatat aaattettta atgagatgat egeaaaateg etgtatacaa tttegtaett
aattcgttag tcttgaaaag ttgacctaat ttagatcaaa ttaaggttaa ctacaataaa
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tatetttaag ggagacacag agacaatttg gacaaaaaag gtetteetga gaaagaagtg
gaccacaatc gtggcgcgaa aggaacttcc tcctccctc tgttgccttg tcattgggcc
                                                                   1560
acgtatatct ccacctgatc gtgatgctta cgtggtccat ttctagatac tatagtgacc
agatcaacqq tcaaqattqa ttctaattta qacqaaaqac caaccqtca cqtcqctaqa
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gtaaaagatt ttttgaaggc ggagggagaa aaatcaaaag ttaaaagtaa tttgaaaacg
                                                                   1740
aggaagagaa aaaggaattt taaaatgttt aatgaagcgg taggccgcat gggtatataa
                                                                   1800
atgggcccgc tttgtaacgt gtaacgatga tatttattca actgcgtggt ataaccaaaa
                                                                   1860
aaaaaaagaa acactattcg actcttttt tgtttttgta ataaacataa aataaatttt
ggagagagat atttcgtcgg aaattaattg acgttcaaaa gattcgttac cggaatattt
                                                                   1980
agtagaggcg gcgatcgagt t
                                                                   2001
SEQ ID NO: 15
                      moltype = DNA length = 514
                      Location/Qualifiers
FEATURE
misc feature
                       1..514
                       note = Ceres Promoter YP2590
misc_feature
                       77..86
                       note = Motif name: CARGCW8GAT
                       255..262
misc feature
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note = Motif name: MYBPLANT
misc_feature
                       279..287
                       note = Motif name:ATHB6COREAT
misc_feature
                       283..288
                       note = Motif name: TATA BOX
5'UTR
                       314..514
source
                       1..514
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
SEQUENCE: 15
agaagggctt gctgtccgtt gctcatctca tatagtaagg cccaaaaggc ccaaaggctt
ctttttccaa ttaaaactaa tttttgtata gcgaaattat gatgaagaaa atcgtttcat
ggcacgtcaa aaaactttcg cacaacaatt ttttttaca aaagggtcac ttttatcaga
acctaatcta tcatagagtt cccatcaaca aaaaacctat catagagtct agtcctgtgt
taacgtaggt gatgggttgg tttcatggtt tacatggaca attattatgg tgtaagaaaa
agatgatggg tacacatatt ccatttaacc aatgtgaaag attcacatca cctttttcga
tttttaatag attcggtttt accttctcgt aacgtaattc taagaaaaga aaacgtcaat
catatttact aggaaagata attataattt attgcttttt ctctcactct ctgacctgaa
atctcctact gagatttttg aagtgtattc aaca
SEQ ID NO: 16
                       moltype = DNA length = 1002
                       Location/Qualifiers
FEATURE
misc feature
                       1..1002
                       note = Ceres Promoter YP2606
misc feature
                       44..51
                       note = Motif name:XYLAT
                       339..348
misc_feature
                       note = Motif name: CARGCW8GAT
                       593..600
misc_feature
                       note = Motif name: UP2ATMSD
misc_feature
                       636..645
                       note = Motif name: CARGCW8GAT
                       655..664
misc_feature
                       note = Motif name: CARGCW8GAT
misc_feature
                       807..812
                       note = Motif name: WBBOXPCWRKY1
misc_feature
                       829 834
                       note = Motif name: WBBOXPCWRKY1
misc_feature
                       840..849
                       note = Motif name: CARGCW8GAT
misc_feature
                       862..868
                       note = Motif name: TATA BOX
5'UTR
                       916..1002
misc_feature
                       666..1000
                       note = Ceres Promoter PD3464
source
                       1..1002
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
SEQUENCE: 16
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ctgtaatgct tgtatatata tatatatatt gatcttttct ttattattca catgataaaa
acatctaaac agaaataaaa ataaaggaaa gtgaatgaat gttgaaaaat gtaagattga
ctacattgat ttctatatgc accttctcaa catgttaaaa ataatagcaa ttaacttaat
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tcatagaaat aattattata tttccatgtg gtcatgtgta atacatagat acaaataatt
attaacgtta tcccatggtt ttctatccat tttttatact ttaaaaagtg aattcaaagt
tcaaataaat aattatgaga ttaacattat cccatgcgtt gattctctat ccataaactc
ttttttttt tttgtcaaac gatctttcca tatacatact ttaaaagttg tattcaaagg
ccaacaaaaa attattccct taaacttgtc ccaggaacta aaaattagaa aatatatatc
cctttcccca aaaatttaaa aaaacctttt tcgtcgcaat atttttttt tatagggttt
atacgggatt tgtatcccaa ccaggattat ataatcaatt ttttgcccca aaaacaaaat
aatgaaaaaa aatgaagggt ttttttgccc aaaagggaaa aagttttttt ttttttttt
ttttaccttt actttaaata aaatatatat aagcatacaa actaataata tatgatgctt
attgttttt atttaaaatg tagaactttg actatatcaa ttattttggt caaagtcttc
ataattttgt atatatgttt cctataaaga tttattatag atcatgtacc atttttcaca
gtctataaaa aggttatttc tctctaccat tttcaatatc aaaaagtttc ttcaaacaaa
caagtattag attaacatta tcctcttcat tttcagcaat cc
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SEQ ID NO: 17
                       moltype = DNA length = 1000
                       Location/Qualifiers
FEATURE
misc_feature
                       1..1000
                       note = Ceres Promoter YP2608
                       490..901
intron
misc_feature
                       28..35
                       note = Motif name:P1BS
misc feature
                       67..74
                       note = Motif name: INRNTPSADB
misc_feature
                       155..162
                       note = Motif name: PYRIMIDINEBOXHVEPB1
misc feature
                       156..161
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note = Motif name:GT1GMSCAM4
misc_feature
                       209..214
                       note = Motif name:GT1GMSCAM4
misc_feature
                      216..228
                       note = Motif name: TATABOX
5'UTR
                       279..489
misc_feature
                       401..1000
                      note = Ceres Promoter PD3739
source
                       1..1000
                       mol_type = genomic DNA
                      organism = Arabidopsis thaliana
SEQUENCE: 17
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gtttgaaaat tgagttccag ttgttatgat atttggggac atccattaaa aagtatagta
tgaaatttta acaaggtcta attaatttt tgaaggaaaa aatatataac taagcataaa
taaaaaatctt atagaaacat aaatataaga aaaagattaa tatatgatgc ttataatgat
gagetgtegt eteatattet egtttgatte aaaaegatag aaategacaa agaaggtgtg
aagaagttgg gaaaacaaaa ttaacgcttg aagaatctaa aggtttagct tttttctcaa
tccgatcatt caccgagaaa tattctattt tatatattta tgtgaagctc ttcaattgaa
gaaagaagaa gaaaagacaa tcgtcaaatg cttgactttg gtttcagttc ttcttcacga
cageteaagg tgatgateat ttttatttta etettetace taateatate aetgegaate
ttttccaaag tttcgctctt tattctctca atgtgctttt tttctgagtt gttgttgaac
tagggttctt ctgggtacta gtaactgttt tgctgatttt ctgagctgct tttggctata
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gaatttcagt tttcaaacca aaccttgtcg atttcaaatc aattctagtc ggcagtagat
                                                                   720
ctgatctgtc tctattgggc taggttttac agtgtatcaa ttcaccacta gttcaaagtt
catcagtgct tacacaagtt tactttttt tgtatgtgtt catgatttag ctaatgatca
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agatacttga gatataataa cacaccttat ttaatgcaaa tgggcctaca aattctacta
                                                                  900
ggaagatcgg ctctttgatg ggatgggaca ccaagaaatc accatgggtt tatgatgcta
                                                                  960
tcaattacta gtagtacgtg ttatcttctg aatatatac
                                                                   1000
SEQ ID NO: 18
                      moltype = DNA length = 995
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misc_feature
                      1..995
                      note = Ceres Promoter YP2683
misc_feature
                      162..168
                      note = Motif name:AMYBOX2
                      271 278
misc_feature
                      note = Motif name:P1BS
misc_feature
                      395..399
                       note = Motif name:ABRELATED1
misc_feature
                       601..608
                       note = Motif name: CACGCAATGMGH3
misc_feature
                       797..801
                      note = Motif name:ABRELATED1
misc feature
                      891..896
                      note = Motif name:GT1Consensus
misc_feature
                      917..923
                       note = Motif name: TATABOX4
5'UTR
                      956..995
                      1..995
source
                      mol_type = genomic DNA
organism = Arabidopsis thaliana
SEQUENCE: 18
caatcggtcg cgaaagactt gatctaggta gagatatact taattaaaat cgatgaatga
ccaagtcaat aaatcaagat tcaatgttct ttcgacgaat tctgttcatt ttgcggttgg
gatcatgcat tcatttaaaa taaaccatta ttgaatacgt tatggataat gatagtgatt
aattatccaa tgaggaagag atttggttgg attgttttgg agagtaataa taaaaaataa
aggcatgcaa tagaaggggc agaccaagtg gaatattctg tctaccacaa cgaagtttca
atcaaagata tcagaggtta atgtcttcca ataagcgacc aaaaatcttg attaataatt
aattatatcg cccaaagtca ctatgaactt gtgaacgtgt tattttggag acttttctca
cctaaataca tcgagatcgc cttgttagtt ggtacttggt aaacattatg ctctaatggc
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cacgcaatga aaaccaaacc gactaaattt tgtgtttttt tcagggtgat ttttaagaac
atteettaat caatgggate taaaaaagte acttgeetaa gaaagteace gaaactteac
attcaaagta actttatcct cttctcagct tttcacatta tgaacctccc gtgggaacaa
agacatttat accettacgt gagettecaa gaagaaacca acaattttta agatttttee
                                                                  840
ccattacatg tagattagtt ggcttcaagt ttctatggat gttccttctc ggaaatccat
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caatttetet caaactgttt taetttetet ttgta
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SEQ ID NO: 19
                      moltype = DNA length = 959
FEATURE
                       Location/Qualifiers
misc_feature
                      1..959
                      note = Ceres Promoter YP2816
misc_feature
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                      note = Motif name:TL1ATSAR
                       538..544
misc feature
                      note = Motif name: ANAERO1CONSENSUS
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misc_feature
                       672..679
                       note = Motif name:XYLAT
misc_feature
                       761..767
                       note = Motif name: TATABOXOSPAL
5'UTR
                       798..959
misc_feature
                       649..959
                       note = Ceres Promoter PD3229
misc feature
                       130..797
                       note = Ceres Promoter PD3243
misc_feature
                       448..959
                       note = Ceres Promoter PD3238
                       1..959
source
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
SEQUENCE: 19
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ttttatttgt aatttttcct gtaaggggga aatttatttt ttatacagct aataaatgat
aaaaactcga aaattgtgac tacagtggaa gagtttggtg gcagacattt acttgttgtc
tgaagaagaa attcaatgca atgactaatc tgaaattaca ctagaatcaa ataaagcttt
acctaatgta attaaatttc tacagagttg gtggcttatg aaagataatc taaggagatt
ttgatgctct gtattttcaa attttagatg ttggccataa agtaaaattt atttatactt
ccacactcaa tccccatttt cagtattact tcaagtcttc gacatacttt attctgtaag
ttttttttt qqcatcaaaa aqtqtqaaaa taaaaqtatc attttcttat tqaatqatqa
atacttatga aaaagaaaag caaactttat gacgaataaa gcctgaaact gtgttgtaaa
                                                                    540
caaagtctaa aacgaatgat ttttattttc tcgtcaagta atcattttat ttttattgat
gattittgat acticacata atactittga tattaggcca gittaggaata aattitcaaa
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tactttgatc tttctttgtt gaacttgagc ggttcgtacg aacatgcttt gatctttttg
catttactta tgaataaagc aaaaacttct aaactgtttc tatttaatag attggaactc
                                                                    780
gagtcggtgt acattgaaaa aataaaccaa aatttaataa cagagattat ttgcctttta
                                                                    840
atacatatac acacacgcac atatctacct gcatcttctt cataatcatt cctaagaaat
                                                                    900
tctctctctc acacactcaa gaacaagaca agagtcaaga aaaatcagcc atgcactga
                                                                    959
SEQ ID NO: 20
                       moltype = DNA length = 993
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                       Location/Qualifiers
misc_feature
                       1..993
                       note = Ceres Promoter YP2832
misc_feature
                       175..181
                       note = Motif name: MYBGAHV
misc_feature
                       438..446
                       note = Motif name:SURE2STPAT21
misc_feature
                       480..489
                       note = Motif name: CARGCW8GAT
misc_feature
                       525..533
                       note = Motif name:SURE2STPAT21
misc feature
                       529..538
                       note = Motif name: CARGCW8GAT
misc_feature
                       763..770
                       note = Motif name:XYLAT
misc_feature
                       816..822
                       note = Motif name: BOXIIPCCHS
misc_feature
                       816..823
                       note = Motif name:LRENPCABE
misc_feature
                       894..900
                       note = TATABOX2
5'UTR
                       927..993
misc feature
                       595..993
                       note = Ceres Promoter PD3300
                       1..993
source
                       mol_type = genomic DNA
organism = Arabidopsis thaliana
SEOUENCE: 20
tcaaaaaata aaaatttgag tcgttgctcg tcagatttta tgtcactaga cagcaacgaa
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catggaggca atttgcaaac aatgattagg attgtgtcat actgtagaaa aaattaacaa
aacaattttt ttattattca ttgactcgaa qqttgaacaa gaaggtcgaa aaaataaaca
totttacatg ggggttcata ttatttatgt acaaattgct accaaggaca aagatcacat
gctcatctac aacgataaga tcaatgaatt tgacagtata ttaataaatc atgcatctat
                                                                    360
acataaataa aaactagtag gaactagtct agtcaaaaag tacagtatta tggacaagat
                                                                    420
atgaaactag ctaggataat actaattgct gtagcttaag ccacttgaat gttgtccctc
attattatgt ccaaaaaatt caatgtagac aagaaaaaag aactaatact aatatatgtc
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                                                                    600
tgtaattctt atattccagt acgtatggag agactcggga gagtcgaacc aagaacatgc
                                                                    660
tcaggccttc tgagttgtga catagatagt acaaccaggc gaccgagaca aacaaaacca
                                                                    720
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ataatgaaac aaatgagcat gcaaagtatg tgacgacgtg gcaaaaaatg agaaggttca
                                                                    840
acgaagcaac aaaaagaagt atttcgtcgt ctttactccc atcaatatgt gactataaat
                                                                    900
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993

aaaaaaacac atcacacaaa agatctctac act

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moltype = DNA length = 600
SEQ ID NO: 21
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                       Location/Qualifiers
misc_feature
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                       note = Ceres Promoter PD2995
                       1..600
misc feature
                       note = Ceres Promoter PD2263
intron
                       365..574
misc_feature
                       1..10
                       note = Motif name:CIACADIANLELHC
misc_feature
                       3..11
                       note = Motif name: EVENINGAT
misc_feature
                       21..30
                       note = Motif name: CARGCW8GAT
misc feature
                       103..109
                       note = Motif name: ABRERATCAL
misc_feature
                       125..131
                       note = Motif name: RBCSCONSENSUS
misc_feature
                       185..194
                       note = Motif name:CIACADIANLELHC
misc feature
                       249..256
                       note = Motif name: MYBPLANT
misc feature
                       298..301
                       note = Transcription Start Site
misc_feature
                       272..281
                       note = Motif name: TATA Box
5'UTR
                       304..364
5'UTR
                       575..600
misc_feature
                       1..365
                       note = Ceres Promoter PD3048
                       1..342
misc_feature
                       note = Ceres Promoter PD3676
misc_feature
                       1..306
                       note = Ceres Promoter PD3182
                       40..600
misc_feature
                       note = Ceres Promoter PD2926
misc_feature
                       101..600
                       note = Ceres Promoter PD3345
                       176..600
misc_feature
                       note = Ceres Promoter PD3503
source
                       1..600
                       mol_type = genomic DNA
                       organism = Arabidopsis thaliana
SEOUENCE: 21
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gattegeaat ettetteatt agatgetgte aagttgtaet egeaegeggt ggteeagtga
agcaaatcca acggtttaaa accttcttac atttctagat ctaatctgaa ccgtcagata
                                                                    180
tctagatctc attgtctgaa cacagttaga tgaaactggg aatgaatctg gacgaaatta
                                                                    240
cgatcttaca ccaacccct cgacgagete gtatatataa agettatacg etecteette
                                                                    300
accttcgtac tactactacc accacatttc tttagctcaa ccttcattac taatctcctt
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ttaaggtatg ttcacttttc ttcgattcat actttctcaa gattcctgca tttctgtaga
atttgaacca agtgtcgatt tttgtttgag agaagtgttg atttatagat ctggttattg
                                                                    480
aatctagatt ccaattttta attgattcga gtttgttatg tgtgtttata ctacttctca
ttgatcttgt ttgatttctc tgctctgtat taggtttctt tcgtgaatca gatcggaaaa
SEQ ID NO: 22
                       moltype = DNA length = 900
FEATURE
                       Location/Qualifiers
misc_feature
                       1..900
                       note = Ceres Promoter PD2999
                       1..900
misc_feature
                       note = Ceres Promoter PD2258
                       393..898
misc feature
                       106..113
                       note = Motif name: MYBPLANT
misc feature
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catttttgta attctttgtt gggtaaattc acaaccaaaa aaatagaaag gcccaaaacg
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cgtaagggca aattagtaaa agtagaacca caaagagaaa gcgaaaaccc tagacacctc
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                                                                  1300
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                      466..472
misc feature
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misc_feature
                       709..715
                       note = Motif name: -300CORE
misc feature
                       842..849
                       note = Motif name: AUXRETGA1GMGH3
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source
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                                                                   780
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                       347..355
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                       576..583
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misc_feature
                       613..622
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misc feature
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                       823..828
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misc_feature
                       698..704
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cccattttct tattatgtgg ttaaccaata aaagactcaa ttttgcagtc ttactaatca
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                                                                   660
gctacatata tatatata tgattatttt cagtgtatat ataatatgta tagatattaa
                                                                   720
aaataaataa aagataagto gtgtgtagto cacatatott gaacatcaaa atgacaaaac
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tattaaaatc cgcggcaaaa tctcttatcc agtgacatgt tacttatcaa ccaatcacaa

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caegocaect cateacecca agaccaette tettatette etetecetee aetaateece
tcaagaatct aaagctcctc ccataagtct tctctccaca cttcacccaa aactccagaa
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misc feature
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misc feature
                       817..821
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misc feature
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misc feature
                       827..836
                       note = Motif name: CARGATCONSENSUS
misc feature
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                       1..1000
source
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organism = Arabidopsis thaliana
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                                                                    180
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                                                                    240
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                                                                     840
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                                                                    900
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                       note = ABREATRD22 motif
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000
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                       moltype =
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SEQUENCE: 31
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000
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SEQ ID NO: 37 FEATURE unsure	moltype = DNA length = 18 Location/Qualifiers 12	
unsure	note = n is a, c, t, g, unknown, or other 4	
unsure	note = n is a, c, t, g, unknown, or other 18	
source	note = n is a, c, t, g, unknown, or other 118	
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SEQ ID NO: 39 SEQUENCE: 39 000	moltype = length =	
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SEQ ID NO: 46 FEATURE source	moltype = DNA length = 10 Location/Qualifiers 110 mol_type = other DNA note = CARGATCONSENSUS motif organism = synthetic construct	
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SEQ ID NO: 47	moltype = DNA length = 10	

FEATURE	Location/Qualifiers
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	note = CARGCW8GAT motif
	note = Ceres Seed Line no. CW8
SEQUENCE: 47	organism = synthetic construct
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SEQ ID NO: 48	moltype = length =
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SEQUENCE: 49	
SEQ ID NO: 50 SEQUENCE: 50	moltype = length =
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CEO ID NO 50	waltuwa lawath
SEQ ID NO: 52 SEQUENCE: 52	moltype = length =
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CEO ID NO. E2	moltyme - length -
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SEQUENCE: 54	moreype - rengen -
000	
SEQ ID NO: 55	moltype = length =
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000	
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SEQUENCE: 56	
000	
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SEQ ID NO: 59	moltype = length =
SEQUENCE: 59	
SEQ ID NO: 60	moltype = length =
SEQUENCE: 60 000	
SEQ ID NO: 61 SEQUENCE: 61	moltype = length =
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source	110
	mol_type = other DNA
	note = MARTBOX motif
SEQUENCE: 62	organism = synthetic construct
ttwtwttwtt	10
CEO ID NO CO	weltyme - length -
SEQ ID NO: 63 SEQUENCE: 63	moltype = length =
000	
GEO TO WO	malthous longitude
SEQ ID NO: 64	moltype = length =

	Concinaca	
SEQUENCE: 64		
SEQ ID NO: 65 SEQUENCE: 65 000	moltype = length =	
SEQ ID NO: 66 FEATURE source	moltype = DNA length = 10 Location/Qualifiers 110 mol_type = other DNA note = 02F3BE2S1 motif organism = synthetic construct	
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SEQ ID NO: 74 SEQUENCE: 74 000	moltype = length =	
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SEQ ID NO: 82 SEQUENCE: 82	moltype = length =	

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SEQ ID NO: 83 SEQUENCE: 83 000	moltype = length =	
SEQ ID NO: 84 FEATURE source	moltype = DNA length = 11 Location/Qualifiers 111 mol_type = other DNA note = TLIATSAR motif organism = synthetic construct	
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SEQ ID NO: 85 SEQUENCE: 85 000	moltype = length =	
SEQ ID NO: 86 SEQUENCE: 86 000	moltype = length =	
SEQ ID NO: 87 SEQUENCE: 87 000	moltype = length =	

What is claimed is:

- 1. A vector construct comprising:
- a) a first nucleic acid molecule comprising the nucleotide sequence of SEQ ID NO:16 or a nucleotide sequence having at least 95% identity to the nucleotide sequence of SEQ ID NO:16, wherein said first nucleic acid molecule functions as a promoter; and
- b) a second nucleic acid molecule to be expressed, wherein said first nucleic acid molecule and said second nucleic acid molecule are heterologous to each other and are 35 operably linked.
- 2. The vector construct according to claim 1, wherein said first nucleic acid molecule comprises the nucleic acid sequence of SEQ ID NO:16.
- 3. The vector construct according to claim 1, wherein said 40 second nucleic acid molecule encodes a polypeptide.
- **4**. The vector construct according to claim **3**, wherein said second nucleic acid molecule is operably linked to said first nucleic acid molecule in the sense orientation.
- **5**. The vector construct according to claim **4**, wherein said 45 second nucleic acid molecule is transcribed into an RNA molecule that expresses the polypeptide encoded by said second nucleic acid molecule.
- **6**. The vector construct according to claim **1**, wherein said second nucleic acid molecule is operably linked to said first 50 nucleic acid molecule in the antisense orientation.
- 7. The vector construct according to claim 6, wherein transcription of said second nucleic acid molecule produces an antisense molecule.
- **8**. The vector construct according to claim **1**, wherein 55 transcription of said second nucleic acid molecule produces an RNAi molecule against an endogenous gene.
- 9. The vector construct according to claim 3, wherein said second nucleic acid molecule encodes a polypeptide of agronomic interest.
- 10. A plant or plant cell comprising the vector construct according to claim 1.
- 11. A plant or plant cell stably transformed with the vector construct according to claim 1.
 - 12. A transgenic seed of the plant according to claim 10. 65
- 13. A method of directing transcription comprising combining, in an environment suitable for transcription:

- a) a first nucleic acid molecule comprising the nucleotide sequence of SEQ ID NO: 16
- or a nucleotide sequence having at least 95% identity to the nucleotide sequence of SEQ ID NO: 16, wherein said first nucleic acid molecule functions as a promoter; and
- b) a second nucleic acid molecule;
- wherein said first nucleic acid molecule and said second nucleic acid molecule are heterologous to each other and operably linked, and transcribing said second nucleic acid molecule.
- 14. A method of expressing an exogenous coding region in a plant comprising:
 - (a) transforming a plant cell with the vector of claim 1;
 - (b) regenerating a stably transformed plant from the transformed plant cell of step (a); and
 - (c) selecting at least one plant containing the transformed plant cell,
 - wherein expression of the second nucleic acid molecule results in production of a polypeptide encoded by said second transcribable nucleic acid molecule.
- **15**. A method of altering the expression of an endogenous gene in a plant comprising:
 - a) transforming a plant cell with the vector construct according to claim 1, and
 - b) regenerating at least one stably transformed plant from said transformed plant cell, wherein transcription of said second nucleic acid molecule produces an RNAi molecule that suppresses expression of the endogenous gene.
- 16. A plant prepared according to the method of claim 14.17. A transgenic seed from the plant according to claim 16.
- 18. A method of producing a transgenic plant, said method comprising:
 - (a) transforming a plant cell comprising the vector according to claim 1; and
 - (b) growing a plant from said plant cell.

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19. The method of claim 18, wherein said second nucleic acid molecule comprises a nucleic acid sequence encoding a polypeptide.

- 20. The method of claim 18, wherein said second nucleic acid molecule is operably linked to said first nucleic acid molecule in an antisense orientation.
- 21. The method of claim 18, wherein transcription of said second nucleic acid molecule produces an RNAi molecule. 5
- 22. A plant or plant cell comprising the vector construct according to claim 2.
- 23. A plant or plant cell stably transformed with the vector construct according to claim 2.
- **24.** A transgenic seed of the plant according to claim **22**, 10 wherein the transgenic seed comprises the vector construct.
- **25**. A method of producing a transgenic plant, said method comprising:
 - (a) introducing into a plant cell the vector according to claim 2; and
 - (b) growing a plant from said plant cell.

* * * * *