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(54) Title: METHODS AND COMPOSITIONS FOR MODIFYING SHADE AVOIDANCE IN PLANTS

(57) Abstract: This invention relates to compositions and methods for modifying an *Elongated Hypocotyl5 (HY5)* transcription factor in plants to suppress the shade avoidance response. The invention further relates to plants and plant parts produced using the methods and compositions of the invention.



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## METHODS AND COMPOSITIONS FOR MODIFYING SHADE AVOIDANCE IN PLANTS

### STATEMENT REGARDING ELECTRONIC FILING OF A SEQUENCE LISTING

5 A Sequence Listing in XML format, entitled 1499-102\_ST26.xml, 314,170 bytes in size, generated on June 3, 2023 and filed herewith, is hereby incorporated by reference into the specification for its disclosures.

### STATEMENT OF PRIORITY

10 This application claims the benefit, under 35 U.S.C. § 119 (e), of U.S. Provisional Application No. 63/355,730 filed on June 27, 2022, the entire content of which is incorporated by reference herein.

### FIELD OF THE INVENTION

15 This invention relates to compositions and methods for modifying an *Elongated Hypocotyl5 (HY5)* transcription factor in plants to suppress the shade avoidance response. The invention further relates to plants and plant parts produced using the methods and compositions of the invention.

### BACKGROUND OF THE INVENTION

20 Shade avoidance response (SAR) is a response to a decrease in the quality or quantity of available light (Kebrom and Brutnell, *J Exp Bot* 58:3079-3089 (2007)) in which a plant attempts to outcompete neighboring plants by growing toward resources (primarily light). Overcrowding of plants can lead to shade avoidance syndrome (SAS) where plants lack vigor and decreased yield. Shade avoidance relates to the relative proportion of red light to far-red light that is present in a plant's environment (Ballare et al. *Science*, 247:329-332 (1990)). Plants absorb most of the red light available to them, but reflect far-red light, including reflecting this light on nearby plants. When a plant detects consistent far-red light in its environment, it will undergo a morphological and physiological response. These responses can include reduced branching, increased plant height, decreased leaf blade area, redistribution of auxin, enhanced ethylene production and acceleration of flowering. SAS is characterized by increased root/shoot ratio, increased plant height, and reduced individual plant yield. In a typical monoculture crop setting, interplant competition through shade avoidance is considered a wasteful survival mechanism.

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Novel strategies for modulating the shade avoidance response in plants are needed to improve crop performance.

### SUMMARY OF THE INVENTION

5 One aspect of the invention provides a plant or part thereof comprising at least one mutation in an endogenous gene encoding an *Elongated Hypocotyl5 (HY5)* transcription factor, optionally wherein the mutation disrupts the binding of the *HY5* transcription factor by a Constitutive Photomorphogenic 1 (COP1) polypeptide (i.e., disrupts binding of COP1 to *HY5* transcription) in the plant or part thereof, optionally wherein the mutation may be a non-natural  
10 mutation.

A second aspect of the invention provides a plant cell comprising an editing system, the editing system comprising: (a) a CRISPR-Cas associated effector protein; and (b) a guide nucleic acid (gRNA, gDNA, crRNA, crDNA) having a spacer sequence with complementarity to an endogenous target gene encoding an endogenous *HY5* transcription factor.

15 A third aspect of the invention provides a plant cell comprising a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene, wherein the mutation is a substitution, insertion and/or a deletion that is introduced into an endogenous *HY5* gene encoding the *HY5* polypeptide using an editing system that comprises a nucleic acid binding domain that binds to a target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene: (a) comprises a sequence  
20 having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID**  
25 **NO:106**, optionally wherein the mutation may be a non-natural mutation.

A fourth aspect of the invention provides a method of providing a plurality of plants having increased yield when each plant of the plurality of plants is planted in close proximity to one another, the method comprising planting two or more plants of the invention in close proximity to one another, thereby providing a plurality of plants having increased yield as  
30 compared to a plurality of control plants devoid of the at least one mutation that are planted in close proximity to one another.

In a fifth aspect, a method of producing/breeding a transgene-free genome-edited plant is provided, comprising: (a) crossing a plant of the invention with a transgene-free plant, thereby introducing the mutation into the plant that is transgene-free; and (b) selecting a progeny plant

that comprises the mutation but is transgene-free, thereby producing a transgene-free genome-edited plant.

In a sixth aspect, a method of creating a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene in a plant is provided, comprising: (a) targeting a gene editing system to a portion of the *HY5* gene that comprises a sequence having at least 80% sequence identity to any one of **SEQ ID NOs:75-105**; and (b) selecting a plant that comprises a modification located in a region of the *HY5* gene having at least 80% sequence identity to any one of **SEQ ID NOs:75-105**, optionally wherein the mutation may be a non-natural mutation.

In a seventh aspect, a method of generating variation in Elongated Hypocotyl5 (HY5) polypeptide is provided, comprising: introducing an editing system into a plant cell, wherein the editing system is targeted to a region of an endogenous *HY5* gene that encodes the HY5 polypeptide, and contacting the region of the endogenous *HY5* gene with the editing system, thereby introducing a mutation into the endogenous *HY5* gene and generating variation in the HY5 polypeptide of the plant cell.

An eighth aspect provides a method of detecting a mutant *Elongated Hypocotyl5 (HY5)* gene (a mutation in an endogenous *HY5* gene) in a plant comprising detecting in the genome of the plant a *HY5* gene having at least one mutation within a region having at least 80% sequence identity to a nucleotide sequence of any one of **SEQ ID NOs:75-105**.

A ninth aspect provides a method for editing a specific site in the genome of a plant cell, the method comprising cleaving, in a site-specific manner, a target site within an endogenous *Elongated Hypocotyl5 (HY5)* gene in the plant cell, the endogenous *HY5* gene: (a) comprising a sequence having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby generating an edit in the endogenous *HY5* gene of the plant cell.

In a tenth aspect, a method for making a plant is provided, the method comprising: (a) contacting a population of plant cells that comprise an endogenous gene encoding an Elongated Hypocotyl5 (HY5) polypeptide with a nuclease targeted to the endogenous gene, wherein the nuclease is linked to a nucleic acid binding domain that binds to a target site within the endogenous gene, the endogenous gene (i) comprising a sequence having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NO:72** or **SEQ ID NO:73**; (ii)

comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (iii) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (iv) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**; (b) selecting a plant cell from the population comprising a mutation in the endogenous gene encoding a HY5 transcription factor, wherein the mutation is a base insertion or base deletion in a region of the endogenous gene of (ii), optionally wherein the mutation reduces or eliminates the ability of the HY5 transcription factor to bind a Constitutive Photomorphogenic 1 (COP1) polypeptide; and (c) growing the selected plant cell into a plant comprising the mutation within the endogenous gene encoding the HY5 transcription factor.

In an eleventh aspect, a method for reducing/suppressing a Shade Avoidance Response in a plant is provided, the method comprising: (a) contacting a plant cell comprising an endogenous *Elongated Hypocotyl5 (HY5)* gene with a nuclease targeted to the endogenous gene, wherein the nuclease is linked to a nucleic acid binding domain that binds to a target site within the endogenous *HY5* gene, the endogenous *HY5* gene: (i) comprising a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (ii) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (iii) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (iv) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby producing a plant cell comprising a mutation in the endogenous *HY5* gene; and (b) growing the plant cell into a plant, thereby reducing/suppressing the Shade Avoidance Response in the plant.

In a twelfth aspect, a method for producing a plant or part thereof comprising at least one cell (e.g., one or more cells) having a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene, the method comprising contacting a target site within an endogenous *HY5* gene in the plant or plant part with a nuclease comprising a cleavage domain and a nucleic acid binding domain, wherein the nucleic acid binding domain of the nuclease binds to a target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene (a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**,

thereby producing the plant or part thereof comprising at least one cell having a mutation in the endogenous *HY5* gene.

In a thirteenth aspect, a method is provided for producing a plant or part thereof comprising a mutation in an *Elongated Hypocotyl5 (HY5)* gene that produces a mutated HY5 transcription factor having reduced binding by a Constitutive Photomorphogenic 1 (COP1) polypeptide comprising contacting a target site within an endogenous *HY5* gene in the plant or part thereof with a nuclease comprising a cleavage domain and a nucleic acid binding domain, wherein the nucleic acid binding domain of the nuclease binds to the target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene: (a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby producing the plant or part thereof comprising a mutation in the *HY5* gene that produces a mutated HY5 transcription factor having reduced binding by the COP1.

In a fourteenth aspect, a method is provided for modifying an endogenous *Elongated Hypocotyl5 (HY5)* gene in a corn plant or part thereof for reducing/suppressing a Shade Avoidance Response in the corn plant or part thereof, the method comprising modifying a target site within the endogenous *HY5* gene in the corn plant or a part thereof, wherein the endogenous *HY5* gene: (a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby modifying the endogenous *HY5* gene and reducing/suppressing a Shade Avoidance Response in the corn plant or part thereof.

In a fifteenth aspect, a guide nucleic acid is provided that binds to a target site within an endogenous gene encoding an *Elongated Hypocotyl5 (HY5)* polypeptide, the endogenous gene comprising a nucleotide sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid

sequence of **SEQ ID NO:74**; and/or encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

A further aspect provides a system comprising a guide nucleic acid of the invention and a CRISPR-Cas effector protein that associates with the guide nucleic acid.

5 In another aspect gene editing system is provided that comprises a CRISPR-Cas effector protein in association with a guide nucleic acid, wherein the guide nucleic acid comprises a spacer sequence that binds to an *Elongated Hypocotyl5 (HY5)* gene.

10 An additional aspect provides a complex comprising a CRISPR-Cas effector protein comprising a cleavage domain and a guide nucleic acid, wherein the guide nucleic acid binds to a target site within an *Elongated Hypocotyl5 (HY5)* gene, wherein the endogenous *HY5* gene: (a) comprising a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, wherein the cleavage domain cleaves a target strand in the *HY5* gene.

15 In a further aspect, an expression cassette is provided comprising (a) a polynucleotide encoding CRISPR-Cas effector protein comprising a cleavage domain and (b) a guide nucleic acid that binds to a target site within an endogenous *Elongated Hypocotyl5 (HY5)* gene, wherein the guide nucleic acid comprises a spacer sequence that is complementary to and binds to (i) a portion of a nucleic acid having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (ii) a portion of a nucleic acid having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (iii) a portion of a nucleic acid encoding an amino acid sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (iv) a portion of a nucleic acid sequence encoding an amino acid sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

20 An additional aspect provides a mutated endogenous *Elongated Hypocotyl5 (HY5)* gene encoding a *HY5* transcription factor having a mutated COP1 binding domain, optionally wherein the one or more mutated *HY5* genes comprise sequences having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139**, optionally wherein the sequence identity may be at least 95% or the sequence identity may be 100%, and/or encoding a mutated *HY5* polypeptide having at least 90%

sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**, optionally wherein the sequence identity may be at least 95% or the sequence identity may be 100%.

5 A further aspect, a corn plant or plant part thereof comprising at least one mutation within an endogenous *Elongated Hypocotyl5 (HY5)* gene having the gene identification number (gene ID) of Zm00001d015743 (**SEQ ID NO:72**), optionally wherein the mutation is a non-natural mutation.

10 A still further aspect of the invention provides a guide nucleic acid that binds to a target nucleic acid within an endogenous *Elongated Hypocotyl5 (HY5)* gene having the gene identification number (gene ID) (Maize Genetics and Genomics Database (Maize GDB)) of Zm00001d015743.

15 Further provided are plants, plant cells, and plant parts produced by the methods of the invention and comprising in their genomes one or more than one mutated *Elongated Hypocotyl5 (HY5)* gene(s), as well as polypeptides, polynucleotides, nucleic acid constructs, expression cassettes and vectors for making a plant, plant cell, and/or plant part of this invention.

These and other aspects of the invention are set forth in more detail in the description of the invention below.

### **BRIEF DESCRIPTION OF THE SEQUENCES**

20 **SEQ ID NOs:1-17** are exemplary Cas12a amino acid sequences useful with this invention.

**SEQ ID NOs:18-20** are exemplary Cas12a nucleotide sequences useful with this invention.

**SEQ ID NO:21-22** are exemplary regulatory sequences encoding a promoter and intron.

25 **SEQ ID NOs:23-29** are exemplary cytosine deaminase sequences useful with this invention.

**SEQ ID NOs:30-40** are exemplary adenine deaminase amino acid sequences useful with this invention.

30 **SEQ ID NO:41** is an exemplary uracil-DNA glycosylase inhibitor (UGI) sequences useful with this invention.

**SEQ ID NOs:42-44** provides an example of a protospacer adjacent motif position for a Type V CRISPR-Cas12a nuclease.

**SEQ ID NOs:45-47** provide example peptide tags and affinity polypeptides useful with this invention.



**SEQ ID NOs:48-58** provide example RNA recruiting motifs and corresponding affinity polypeptides useful with this invention.

**SEQ ID NOs:59-60** are example Cas9 polypeptide sequences useful with this invention.

**SEQ ID NOs:61-71** are example Cas9 polynucleotide sequences useful with this  
5 invention.

**SEQ ID NO:72** is an example *HY5* genomic sequence.

**SEQ ID NO:73** is example *HY5* coding (cds) sequence.

**SEQ ID NO:74** is an example *HY5* transcription factor polypeptide sequence.

**SEQ ID NOs:75-105** are example nucleic acid sequences (regions) from *HY5*  
10 polynucleotides.

**SEQ ID NO:106** is an example region from a *HY5* transcription factor polypeptide.

**SEQ ID NOs:107-113** are example spacer sequences for nucleic acid guides useful with  
this invention.

**SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, and 139**  
15 are example edited *HY5* nucleic acid sequences produced as described herein.

**SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, and 138** are  
example mutated *HY5* polypeptides produced by the edited nucleic acid sequences of **SEQ ID  
NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137 and 139**.

**SEQ ID NOs:141, 143, 144, 146, 147, and 149** are deleted portions of consecutive  
20 nucleotides from **SEQ ID NOs:123, 125, 129, 133, 135, and 137**, respectively.

**SEQ ID NOs:142, 145, 148, and 150** is a portion of consecutive amino acid residues  
deleted from the mutated *HY5* polypeptide of **SEQ ID NOs:124, 130, 136, and 138**,  
respectively.

## 25 **DETAILED DESCRIPTION**

The present invention now will be described hereinafter with reference to the  
accompanying examples, in which embodiments of the invention are shown. This description is  
not intended to be a detailed catalog of all the different ways in which the invention may be  
implemented, or all the features that may be added to the instant invention. For example,  
30 features illustrated with respect to one embodiment may be incorporated into other  
embodiments, and features illustrated with respect to a particular embodiment may be deleted  
from that embodiment. Thus, the invention contemplates that in some embodiments of the  
invention, any feature or combination of features set forth herein can be excluded or omitted. In  
addition, numerous variations and additions to the various embodiments suggested herein will be

apparent to those skilled in the art in light of the instant disclosure, which do not depart from the instant invention. Hence, the following descriptions are intended to illustrate some particular embodiments of the invention, and not to exhaustively specify all permutations, combinations and variations thereof.

5 Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. The terminology used in the description of the invention herein is for the purpose of describing particular embodiments only and is not intended to be limiting of the invention.

10 All publications, patent applications, patents and other references cited herein are incorporated by reference in their entireties for the teachings relevant to the sentence and/or paragraph in which the reference is presented.

Unless the context indicates otherwise, it is specifically intended that the various features of the invention described herein can be used in any combination. Moreover, the present invention also contemplates that in some embodiments of the invention, any feature or  
15 combination of features set forth herein can be excluded or omitted. To illustrate, if the specification states that a composition comprises components A, B and C, it is specifically intended that any of A, B or C, or a combination thereof, can be omitted and disclaimed singularly or in any combination.

20 As used in the description of the invention and the appended claims, the singular forms "a," "an" and "the" are intended to include the plural forms as well, unless the context clearly indicates otherwise.

Also as used herein, "and/or" refers to and encompasses any and all possible combinations of one or more of the associated listed items, as well as the lack of combinations when interpreted in the alternative ("or").

25 The term "about," as used herein when referring to a measurable value such as an amount or concentration and the like, is meant to encompass variations of  $\pm 10\%$ ,  $\pm 5\%$ ,  $\pm 1\%$ ,  $\pm 0.5\%$ , or even  $\pm 0.1\%$  of the specified value as well as the specified value. For example, "about X" where X is the measurable value, is meant to include X as well as variations of  $\pm 10\%$ ,  $\pm 5\%$ ,  $\pm 1\%$ ,  $\pm 0.5\%$ , or even  $\pm 0.1\%$  of X. A range provided herein for a measurable value may include  
30 any other range and/or individual value therein.

As used herein, phrases such as "between X and Y" and "between about X and Y" should be interpreted to include X and Y. As used herein, phrases such as "between about X and Y" mean "between about X and about Y" and phrases such as "from about X to Y" mean "from about X to about Y."

Recitation of ranges of values herein are merely intended to serve as a shorthand method of referring individually to each separate value falling within the range, unless otherwise indicated herein, and each separate value is incorporated into the specification as if it were individually recited herein. For example, if the range 10 to 15 is disclosed, then 11, 12, 13, and 14 are also disclosed.

The term "comprise," "comprises" and "comprising" as used herein, specify the presence of the stated features, integers, steps, operations, elements, and/or components, but do not preclude the presence or addition of one or more other features, integers, steps, operations, elements, components, and/or groups thereof.

As used herein, the transitional phrase "consisting essentially of" means that the scope of a claim is to be interpreted to encompass the specified materials or steps recited in the claim and those that do not materially affect the basic and novel characteristic(s) of the claimed invention. Thus, the term "consisting essentially of" when used in a claim of this invention is not intended to be interpreted to be equivalent to "comprising."

As used herein, the terms "increase," "increasing," "increased," "enhance," "enhanced," "enhancing," and "enhancement" (and grammatical variations thereof) describe an elevation of at least about 15%, 20%, 25%, 50%, 75%, 100%, 150%, 200%, 300%, 400%, 500% or more as compared to a control.

As used herein, the terms "reduce," "reduced," "reducing," "reduction," "diminish," and "decrease" (and grammatical variations thereof), describe, for example, a decrease of at least about 5%, 10%, 15%, 20%, 25%, 35%, 50%, 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99%, 99.5%, 99.6%, 99.7%, 99.8%, 99.9%, or 100% as compared to a control. In some embodiments, the reduction can result in no or essentially no (*i.e.*, an insignificant amount, *e.g.*, less than about 10% or even 5%) detectable activity or amount.

As used herein, the terms "express," "expresses," "expressed" or "expression," and the like, with respect to a nucleic acid molecule and/or a nucleotide sequence (*e.g.*, RNA or DNA) indicates that the nucleic acid molecule and/or a nucleotide sequence is transcribed and, optionally, translated. Thus, a nucleic acid molecule and/or a nucleotide sequence may express a polypeptide of interest or, for example, a functional untranslated RNA.

As used herein, the term "heterologous" refers to a nucleotide/polypeptide that originates from a foreign species, or, if from the same species, is substantially modified from its native form in composition and/or genomic locus by deliberate human intervention. A "heterologous" or a "recombinant" nucleotide sequence is a nucleotide sequence not naturally associated with a

host cell into which it is introduced, including non- naturally occurring multiple copies of a naturally occurring nucleotide sequence.

A "native" or "wild type" nucleic acid, nucleotide sequence, polypeptide or amino acid sequence refers to a naturally occurring or endogenous nucleic acid, nucleotide sequence, polypeptide, or amino acid sequence. Thus, for example, a "wild type mRNA" is an mRNA that is naturally occurring in or endogenous to the reference organism.

As used herein, the term "heterozygous" refers to a genetic status wherein different alleles reside at corresponding loci on homologous chromosomes.

As used herein, the term "homozygous" refers to a genetic status wherein identical alleles reside at corresponding loci on homologous chromosomes.

As used herein, the term "allele" refers to one of two or more different nucleotides or nucleotide sequences that occur at a specific locus.

A "null allele" is a nonfunctional allele caused by a genetic mutation that results in a complete lack of production of the corresponding protein or produces a protein that is non-functional.

A "knock-out mutation" is a mutation that results in a non-functional protein, but which may have a detectable transcript or protein.

A "recessive mutation" is a mutation in a gene that produces a phenotype when homozygous but the phenotype is not observable when the locus is heterozygous.

A "dominant mutation" is a mutation in a gene that produces a mutant phenotype in the presence of a non-mutated copy of the gene. A dominant mutation may be a loss or a gain of function mutation, a hypomorphic mutation, a hypermorphic mutation or a weak loss of function or a weak gain of function.

A "dominant negative mutation" is a mutation that produces an altered gene product (e.g., having an aberrant function relative to wild type), which gene product adversely affects the function of the wild-type allele or gene product. For example, a "dominant negative mutation" may block a function of the wild type gene product. A dominant negative mutation may also be referred to as an "antimorphic mutation."

A "semi-dominant mutation" refers to a mutation in which the penetrance of the phenotype in a heterozygous organism is less than that observed for a homozygous organism.

A "weak loss-of-function mutation" is a mutation that results in a gene product having partial function or reduced function (partially inactivated) as compared to the wild type gene product.

A "hypomorphic mutation" is a mutation that results in a partial loss of gene function, which may occur through reduced expression (e.g., reduced protein and/or reduced RNA) or reduced functional performance (e.g., reduced activity), but not a complete loss of function/activity. A "hypomorphic" allele is a semi-functional allele caused by a genetic mutation that results in production of the corresponding protein that functions at anywhere  
5 between 1% and 99% of normal efficiency.

A "hypermorphic mutation" is a mutation that results in increased expression of the gene product and/or increased activity of the gene product.

A "gain-of-function" allele or mutation is a mutation that confers a new function on the encoded gene product and/or confers a new gene expression pattern. In some embodiments, a  
10 gain-of-function mutation may be dominant or semi-dominant.

As used herein, a "non-natural mutation" refers to a mutation that is generated through human intervention and differs from mutations found in the same gene that have occurred in nature (e.g., occurred naturally and not as a result of a modification made by a human).

A "locus" is a position on a chromosome where a gene or marker or allele is located. In  
15 some embodiments, a locus may encompass one or more nucleotides.

As used herein, the terms "desired allele," "target allele" and/or "allele of interest" are used interchangeably to refer to an allele associated with a desired trait. In some embodiments, a desired allele may be associated with either an increase or a decrease (relative to a control) of or in a given trait, depending on the nature of the desired phenotype.—In some embodiments of  
20 this invention, the phrase "desired allele," "target allele" or "allele of interest" refers to an allele(s) that is associated with increased yield under non-water stress conditions in a plant relative to a control plant not having the target allele or alleles.

A marker is "associated with" a trait when said trait is linked to it and when the presence  
25 of the marker is an indicator of whether and/or to what extent the desired trait or trait form will occur in a plant/germplasm comprising the marker. Similarly, a marker is "associated with" an allele or chromosome interval when it is linked to it and when the presence of the marker is an indicator of whether the allele or chromosome interval is present in a plant/germplasm comprising the marker.

As used herein, the terms "backcross" and "backcrossing" refer to the process whereby a  
30 progeny plant is crossed back to one of its parents one or more times (e.g., 1, 2, 3, 4, 5, 6, 7, 8, etc.). In a backcrossing scheme, the "donor" parent refers to the parental plant with the desired gene or locus to be introgressed. The "recipient" parent (used one or more times) or "recurrent" parent (used two or more times) refers to the parental plant into which the gene or locus is being

introgressed. For example, see Ragot, M. et al. *Marker-assisted Backcrossing: A Practical Example*, in *TECHNIQUES ET UTILISATIONS DES MARQUEURS MOLECULAIRES LES COLLOQUES*, Vol. 72, pp. 45-56 (1995); and Openshaw et al., *Marker-assisted Selection in Backcross Breeding*, in *PROCEEDINGS OF THE SYMPOSIUM "ANALYSIS OF MOLECULAR MARKER DATA,"* pp. 5 41-43 (1994). The initial cross gives rise to the F1 generation. The term "BC1" refers to the second use of the recurrent parent, "BC2" refers to the third use of the recurrent parent, and so on.

As used herein, the terms "cross" or "crossed" refer to the fusion of gametes via pollination to produce progeny (e.g., cells, seeds, or plants). The term encompasses both sexual 10 crosses (the pollination of one plant by another) and selfing (self-pollination, e.g., when the pollen and ovule are from the same plant). The term "crossing" refers to the act of fusing gametes via pollination to produce progeny.

As used herein, the terms "introgression," "introgressing" and "introgressed" refer to both the natural and artificial transmission of a desired allele or combination of desired alleles of a 15 genetic locus or genetic loci from one genetic background to another. For example, a desired allele at a specified locus can be transmitted to at least one progeny via a sexual cross between two parents of the same species, where at least one of the parents has the desired allele in its genome. Alternatively, for example, transmission of an allele can occur by recombination between two donor genomes, e.g., in a fused protoplast, where at least one of the donor 20 protoplasts has the desired allele in its genome. The desired allele may be a selected allele of a marker, a QTL, a transgene, or the like. Offspring comprising the desired allele can be backcrossed one or more times (e.g., 1, 2, 3, 4, or more times) to a line having a desired genetic background, selecting for the desired allele, with the result being that the desired allele becomes fixed in the desired genetic background. For example, a marker associated with increased yield 25 under non-water stress conditions may be introgressed from a donor into a recurrent parent that does not comprise the marker and does not exhibit increased yield under non-water stress conditions. The resulting offspring could then be backcrossed one or more times and selected until the progeny possess the genetic marker(s) associated with increased yield under non-water stress conditions in the recurrent parent background.

30 A "genetic map" is a description of genetic linkage relationships among loci on one or more chromosomes within a given species, generally depicted in a diagrammatic or tabular form. For each genetic map, distances between loci are measured by the recombination frequencies between them. Recombination between loci can be detected using a variety of markers. A genetic map is a product of the mapping population, types of markers used, and the

polymorphic potential of each marker between different populations. The order and genetic distances between loci can differ from one genetic map to another.

As used herein, the term "genotype" refers to the genetic constitution of an individual (or group of individuals) at one or more genetic loci, as contrasted with the observable and/or detectable and/or manifested trait (the phenotype). Genotype is defined by the allele(s) of one or more known loci that the individual has inherited from its parents. The term genotype can be used to refer to an individual's genetic constitution at a single locus, at multiple loci, or more generally, the term genotype can be used to refer to an individual's genetic make-up for all the genes in its genome. Genotypes can be indirectly characterized, e.g., using markers and/or directly characterized by nucleic acid sequencing.

As used herein, the term "germplasm" refers to genetic material of or from an individual (e.g., a plant), a group of individuals (e.g., a plant line, variety, or family), or a clone derived from a line, variety, species, or culture. The germplasm can be part of an organism or cell or can be separate from the organism or cell. In general, germplasm provides genetic material with a specific genetic makeup that provides a foundation for some or all of the hereditary qualities of an organism or cell culture. As used herein, germplasm includes cells, seed or tissues from which new plants may be grown, as well as plant parts that can be cultured into a whole plant (e.g., leaves, stems, buds, roots, pollen, cells, etc.).

As used herein, the terms "cultivar" and "variety" refer to a group of similar plants that by structural or genetic features and/or performance can be distinguished from other varieties within the same species.

As used herein, the terms "exotic," "exotic line" and "exotic germplasm" refer to any plant, line or germplasm that is not elite. In general, exotic plants/germplasms are not derived from any known elite plant or germplasm, but rather are selected to introduce one or more desired genetic elements into a breeding program (e.g., to introduce novel alleles into a breeding program).

As used herein, the term "hybrid" in the context of plant breeding refers to a plant that is the offspring of genetically dissimilar parents produced by crossing plants of different lines or breeds or species, including but not limited to the cross between two inbred lines.

As used herein, the term "inbred" refers to a substantially homozygous plant or variety. The term may refer to a plant or plant variety that is substantially homozygous throughout the entire genome or that is substantially homozygous with respect to a portion of the genome that is of particular interest.

A "haplotype" is the genotype of an individual at a plurality of genetic loci, i.e., a combination of alleles. Typically, the genetic loci that define a haplotype are physically and genetically linked, i.e., on the same chromosome segment. The term "haplotype" can refer to polymorphisms at a particular locus, such as a single marker locus, or polymorphisms at multiple loci along a chromosomal segment.

As used herein, "shade avoidance response" is defined as the growth of a plant in response to a low red:far-red (R:FR) light ratio. Suppression of a shade avoidance response refers to the suppression of the growth changes in response to a low R:FR light ratio. In one aspect, suppression of a shade avoidance response can be shown by measuring the height of a plant comprising the trait of the invention (e.g., a mutated HY5 transcription factor as described herein) and an isogenic plant without the trait in a controlled environment with a low R:FR light ratio. When grown under identical conditions in the presence of a R:FR ratio of 0.16, a plant comprising the trait of the invention will be at least 5% shorter (e.g., height measured at coleoptile, V1 sheath or V2 sheath) than an isogenic plant not comprising the trait (e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150%, or more shorter, or any range or value therein; e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25% shorter to about 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150%, or more shorter) (e.g., about 5% to about 10% shorter, about 5% to about 15% shorter, about 5% to about 20% shorter, about 5% to about 25% shorter, about 5% to about 30% shorter, about 5% to about 40% shorter, about 5% to about 50% shorter, about 10% to about 20% shorter, about 10% to about 30% shorter, about 10% to about 50% shorter, 10% to about 70% shorter, about 15% to about 20% shorter, about 15% to about 30% shorter, 15% to about 50% shorter, about 20% to about 30% shorter, about 20% to about 50% shorter, about 20% to about 70% shorter, about 40% to about 50% shorter, about 40% to about 60% shorter, about 40% to about 80% shorter, about 40% to about 100% shorter, about 50% to about 70% shorter, about 50% to about 100% shorter, about 50% to about 125% shorter, about 75% to about 100% shorter, about 75% to about 120% shorter, about 75% to about 140% shorter, and the like).



Plants exhibiting reduced SAR exhibit increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity.

A plant in which SAR is reduced as described herein may have increased yield as compared to a plant that does not comprise the reduction in SAR. As used herein, "increased yield" refers to any plant trait associated with growth, for example, biomass, yield, nitrogen use efficiency (NUE), inflorescence size/weight, fruit yield, fruit quality, fruit size, seed size, seed number, foliar tissue weight, nodulation number, nodulation mass, nodulation activity, number of seed heads, number of tillers, number of flowers, number of tubers, tuber mass, bulb mass, number of seeds, total seed mass, rate of leaf emergence, rate of tiller emergence, rate of seedling emergence, length of roots, number of roots, size and/or weight of root mass, or any combination thereof. Thus, in some aspects, "increased yield" may include, but is not limited to, increased inflorescence production, increased fruit production (e.g., increased number, weight and/or size of fruit; e.g., increase number, weight, and/or size of ears for, e.g., maize), increased fruit quality, increased number, size and/or weight of roots, increased meristem size, increased seed size, increased biomass, increased nitrogen use efficiency, as compared to a control plant or part thereof (e.g., a plant that does not comprise a mutated endogenous nucleic acid encoding a HY5 transcription factor as described herein grown in an environment with a low R:FR light ratio (e.g., a shaded environment; e.g., a R:FR ratio of about 0.16) including when grown in close proximity with other plants). In some aspects, increased yield can be expressed as quantity of grain produced per area of land (e.g., bushels per acre of land).

As used herein "decreased height" means repression of stem elongation in response to enriched far-red light. Thus, for example, a plant having a mutation in an *HY5* gene as described herein exhibits a decreased height when grown in shade conditions as compared to a control plant not comprising the mutation and also growing in shade conditions. A decrease in height may be a decrease of about 15% to about 80% (e.g., about 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, or 80% or any range or value therein) compared to a control.

As used herein, "decreased shoot:root ratio" means reduction of the proportion of above ground biomass relative to below ground biomass. Thus, for example, a plant having a mutation

in an *HY5* gene as described herein exhibits a decreased shoot:root ratio when grown in shade conditions as compared to a control plant not comprising the mutation and also growing in shade conditions. A decrease in shoot:root ratio may be a decrease of about 5% to about 50% (e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50% or any range or value therein) compared to a control.

As used herein, "increased upright growth" means that a plant having a mutation in an *HY5* gene as described herein continues to grow upright when experiencing shade, such as when planted at a high density, rather than "leaning" or growing in the direction of the light as compared to a control plant without the mutation that is also experiencing shade. A plant having increased upright growth grows more vertically than the control plant. In some embodiments, an increase in upright growth may be an increase of about 5% to about 50% (e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50% or any range or value therein) compared to a control.

As used herein, "decreased leaf length" means that the leaf of a plant having a mutation in an *HY5* gene as described herein has a reduction in the distance from leaf apex to leaf base as compared to a control plant not comprising the mutation. In some embodiments, a decrease in leaf length may be a decrease of about 5% to about 50% (e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50% or any range or value therein) compared to a control.

As used herein, "increased mechanical strength of stems" means that a plant having a mutation in an *HY5* gene as described herein has an improvement in one or more of bending strength, elastic modulus, tensile strength, critical buckling load, compressive strength, shear strength and/or shear modulus as compared to a control plant not comprising the mutation. In some embodiments, a decrease in mechanical strength of a stem may be an increase of about 5% to about 65% (e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, or 65% or any range or value therein) compared to a control.

As used herein, "lodging rate" means the ratio of the number of plants inclined at  $>45^\circ$  relative to the vertical axis to the total number of plants in the community being evaluated. Thus, for example, a plant having a mutation in an *HY5* gene as described herein exhibits a "decreased

lodging rate" as compared to a control plant not comprising the mutation. In some embodiments, a "decrease in lodging rate" may be a decrease of about 20% to about 100% (e.g., about 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99 or 100% or any range or value therein) compared to a control.

As used herein, "senescence" means a process of aging in plants. For example, a plant having a mutation in an *HY5* gene as described herein has a delay in one or both of stress-induced or age-related developmental aging (senescence) as compared to a control plant not comprising the mutation. In some embodiments, a delay in senescence may be a delay of about 10% to about 80% (e.g., about 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, or 80% or any range or value therein) compared to a control.

As used herein, "increased photosynthesis efficiency" means that a plant having a mutation in an *HY5* gene as described herein exhibits an improvement in the efficiency of the energy transfer from the harvested light into the electron transport chain as compared to a control plant not comprising the mutation. In some embodiments, an increase in photosynthesis efficiency may be an increase of about 10% to about 100% (e.g., about 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99 or 100% or any range or value therein) compared to a control.

As used herein, "grain filling" refers to the time between anthesis and physiological maturity. For example, a plant having a mutation in an *HY5* gene as described herein has an increase in grain filling (e.g., a longer grain filling period) resulting in larger grain weight, higher harvest index and/or higher grain yield per plant as compared to a control plant not comprising the mutation. In some embodiments, an increase in grain filling may be an increased length of the grain filling period by about 10% to about 100% (e.g., about 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99 or 100% or any range or value therein) compared to a control.

As used herein, "enhanced defense responses against pathogens and herbivores" means that a plant having a mutation in an *HY5* gene as described herein has an improvement in basal resistance (e.g., innate immunity) and/or systemic acquired resistance in response to pathogens (e.g., viral, bacterial, nematode, and/or fungal pathogen) and herbivores as compared to a control plant not comprising the mutation. In some embodiments, an enhanced defense response may be an enhancement/improvement of or increase in defense response by about 30% to about 100% (e.g., about 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99 or 100% or any range or value therein) compared to a control.

As used herein, "substantially no change in flowering time" means that a plant having a mutation in an *HY5* gene as described herein, when experiencing shade, such as when planted at a high density, the plant retains its normal flowering time substantially unchanged as compared to a control plant (not comprising the mutation) experiencing shade. In some embodiments, a plant having a mutation in *HY5* as described herein may have substantially no change in flowering time, wherein "substantially no change" refers to a change in flowering time of less than 15% (e.g., no change or a change of less than 1, 2, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15% or any range or value therein) as compared to a control. A plant with substantially no change in flowering time may also be described as having a flowering time that is at least 85% (e.g., 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100% or any range or value therein) that of a control plant.

Thus, a plant in which at least one (e.g., one or more, e.g., 1, 2, 3, or 4, or more) endogenous *HY5* gene is modified as described herein (e.g., comprises a modification as described herein) may have improved yield traits as compared to a plant that does not comprise (is devoid of) the modification in the at least one endogenous *HY5* gene. As used herein, "improved yield traits" refers to any plant trait associated with growth, for example, biomass, yield, nitrogen use efficiency (NUE), inflorescence size/weight, fruit yield, fruit quality, fruit size, seed size (e.g., seed area, seed size), seed number, foliar tissue weight, nodulation number, nodulation mass, nodulation activity, number of seed heads, number of tillers, number of branches, number of flowers, number of tubers, tuber mass, bulb mass, number of seeds, total seed mass, rate of leaf emergence, rate of tiller/branch emergence, rate of seedling emergence, length of roots, number of roots, size and/or weight of root mass, or any combination thereof. In some aspects, "improved yield traits" may include, but are not limited to, increased inflorescence production, increased fruit production (e.g., increased number, weight and/or size of fruit; e.g.,

increased number, weight, and/or length of ears for, e.g., maize), increased fruit quality, increased number, size and/or weight of roots, increased meristem size, increased seed size (e.g., seed area and/or seed weight), increased biomass, increased leaf size, increased nitrogen use efficiency, increased height, increased internode number and/or increased internode length as compared to a control plant or part thereof (e.g., a plant that does not comprise a mutated endogenous *HY5* nucleic acid as described herein). In some aspects, improved yield traits can be expressed as quantity of grain/seed produced per area of land (e.g., bushels per acre of land). In some embodiments, the one or more improved yield traits may be an increased kernel row number, optionally without a decrease in ear length.

As used herein a "control plant" means a plant that does not contain an edited *HY5* transcription factor gene as described herein. A control plant is used to identify and select a plant edited as described herein and that has an enhanced trait or altered phenotype as compared to the control plant. A suitable control plant can be a plant of the parental line used to generate a plant comprising a mutated *HY5* transcription factor gene(s), for example, a wild type plant devoid of an edit in an endogenous *HY5* transcription factor gene as described herein. A suitable control plant can also be a plant that contains recombinant nucleic acids that impart other traits, for example, a transgenic plant having enhanced herbicide tolerance. A suitable control plant can in some cases be a progeny of a heterozygous or hemizygous transgenic plant line that is devoid of the mutated *HY5* transcription factor gene as described herein, known as a negative segregant, or a negative isogenic line.

An enhanced trait (e.g., improved yield trait) may include, for example, decreased days from planting to maturity, increased stalk size, increased number of leaves, increased plant height growth rate in vegetative stage, increased ear size, increased ear dry weight per plant, increased number of kernels per ear, increased weight per kernel, increased number of kernels per plant, decreased ear void, extended grain fill period, reduced plant height, increased number of root branches, increased total root length, increased yield (e.g., increase in harvestable seed), increased nitrogen use efficiency, and/or increased water use efficiency as compared to a control plant. An altered phenotype may be, for example, plant height, biomass, canopy area, anthocyanin content, chlorophyll content, water applied, water content, and water use efficiency.

In some embodiments, a plant of this invention may comprise one or more improved yield traits. In some embodiments, the one or more improved yield traits includes higher yield (bu/acre), increased biomass, increased plant height, increased stem diameter, increased leaf area, increased number of flowers, increased kernel row number, optionally wherein ear length is not substantially reduced, increased kernel number, increased kernel size, increased ear

length, decreased tiller number, decreased tassel branch number, increased number of pods, including an increased number of pods per node and/or an increased number of pods per plant, increased number of seeds per pod, increased number of seeds, increased seed size, and/or increased seed weight (e.g., increase in 100-seed weight) as compared to a control plant devoid  
5 of the at least one non-natural mutation. In some embodiments, a plant of this invention may comprise one or more improved yield traits including, but not limited to, optionally an increase in yield (bu/acre), seed size (including kernel size), seed weight (including kernel weight), increased kernel row number (optionally wherein ear length is not substantially reduced), increased number of pods, increased number of seeds per pod and an increase in ear length as  
10 compared to a control plant or part thereof.

As used herein a "trait" is a physiological, morphological, biochemical, or physical characteristic of a plant or particular plant material or cell. In some instances, this characteristic is visible to the human eye and can be measured mechanically, such as seed or plant size, weight, shape, form, length, height, growth rate and development stage, or can be measured by  
15 biochemical techniques, such as detecting the protein, starch, certain metabolites, or oil content of seed or leaves, or by observation of a metabolic or physiological process, for example, by measuring tolerance to water deprivation or particular salt or sugar concentrations, or by the measurement of the expression level of a gene or genes, for example, by employing Northern analysis, RT-PCR, microarray gene expression assays, or reporter gene expression systems, or  
20 by agricultural observations such as hyperosmotic stress tolerance or yield. However, any technique can be used to measure the amount of, the comparative level of, or the difference in any selected chemical compound or macromolecule in the transgenic plants.

As used herein an "enhanced trait" means a characteristic of a plant resulting from mutations in a HY5 transcription factor gene as described herein. Such traits include, but are not  
25 limited to, an enhanced agronomic trait characterized by enhanced plant morphology, physiology, growth and development, yield, nutritional enhancement, disease, or pest resistance, or environmental or chemical tolerance. In some embodiments, an enhanced trait/altered phenotype may be, for example, decreased days from planting to maturity, increased stalk size, increased number of leaves, increased plant height growth rate in vegetative stage, increased ear  
30 size, increased ear dry weight per plant, increased number of kernels per ear, increased weight per kernel, increased number of kernels per plant, decreased ear void, extended grain fill period, reduced plant height, increased number of root branches, increased total root length, drought tolerance, increased water use efficiency, cold tolerance, increased nitrogen use efficiency, and/or increased yield. In some embodiments, a trait is increased yield under nonstress

conditions or increased yield under environmental stress conditions. Stress conditions can include both biotic and abiotic stress, for example, drought, shade, fungal disease, viral disease, bacterial disease, insect infestation, nematode infestation, cold temperature exposure, heat exposure, osmotic stress, reduced nitrogen nutrient availability, reduced phosphorus nutrient availability and high plant density. "Yield" can be affected by many properties including without limitation, plant height, plant biomass, pod number, pod position on the plant, number of internodes, incidence of pod shatter, grain size, ear size, ear tip filling, kernel abortion, efficiency of nodulation and nitrogen fixation, efficiency of nutrient assimilation, resistance to biotic and abiotic stress, carbon assimilation, plant architecture, resistance to lodging, percent seed germination, seedling vigor, and juvenile traits. Yield can also be affected by efficiency of germination (including germination in stressed conditions), growth rate (including growth rate in stressed conditions), flowering time and duration, ear number, ear size, ear weight, seed number per ear or pod, seed size, composition of seed (starch, oil, protein) and characteristics of seed fill.

Also used herein, the term "trait modification" encompasses altering the naturally occurring trait by producing a detectable difference in a characteristic in a plant comprising a mutation in an endogenous *HY5* transcription factor gene as described herein relative to a plant not comprising the mutation, such as a wild-type plant, or a negative segregant. In some cases, the trait modification can be evaluated quantitatively. For example, the trait modification can entail an increase or decrease in an observed trait characteristic or phenotype as compared to a control plant. It is known that there can be natural variations in a modified trait. Therefore, the trait modification observed can entail a change of the normal distribution and magnitude of the trait characteristics or phenotype in the plants as compared to a control plant.

The present disclosure relates to a plant with improved economically relevant characteristics, more specifically reduced shade avoidance. More specifically the present disclosure relates to a plant comprising a mutation(s) in a *HY5* transcription factor gene(s) as described herein, wherein the plant has reduced shade avoidance response as compared to a control plant devoid of said mutation(s) (e.g., a plant comprising a mutation as described herein when grown in shade conditions results in a shorter plant than a plant devoid of the mutation grown under the same conditions). In some embodiments, a plant of the present disclosure further exhibits an improved trait that is related to yield, including but not limited to increased nitrogen use efficiency, increased nitrogen stress tolerance, increased water use efficiency and/or increased drought tolerance, as defined and discussed *infra*.

Yield can be defined as the measurable produce of economic value from a crop. Yield can be defined in the scope of quantity and/or quality. Yield can be directly dependent on several factors, for example, the number and size of organs (e.g., number of flowers), plant architecture (such as the number of branches, plant biomass, e.g., increased root biomass, steeper root angle and/or longer roots, and the like), flowering time and duration, grain fill period. Root architecture and development, photosynthetic efficiency, nutrient uptake, stress tolerance, early vigor, delayed senescence, and functional stay green phenotypes may be factors in determining yield. Optimizing the above-mentioned factors can therefore contribute to increasing crop yield.

Reference herein to an increase/improvement in yield-related traits can also be taken to mean an increase in biomass (weight) of one or more parts of a plant, which can include above ground and/or below ground (harvestable) plant parts. In particular, such harvestable parts are seeds, and performance of the methods of the disclosure results in plants with increased yield and in particular increased seed yield relative to the seed yield of suitable control plants. The term "yield" of a plant can relate to vegetative biomass (root and/or shoot biomass), to reproductive organs, and/or to propagules (such as seeds) of that plant. In some embodiments, performance of the methods of the disclosure results in plants having a shade avoidance response that is reduced relative to suitable control plants.

Increased yield of a plant of the present disclosure can be measured in a number of ways, including test weight, seed number per plant, seed weight, seed number per unit area (for example, seeds, or weight of seeds, per acre), bushels per acre, tons per acre, or kilo per hectare. Increased yield can result from improved utilization of key biochemical compounds, such as nitrogen, phosphorous and carbohydrate, or from improved responses to environmental stresses, such as cold, heat, drought, salt, shade, high plant density, and attack by pests or pathogens.

"Increased yield" can manifest as one or more of the following: (i) increased plant biomass (weight) of one or more parts of a plant, particularly aboveground (harvestable) parts, of a plant, increased root biomass (increased number of roots, increased root thickness, increased root length) or increased biomass of any other harvestable part; or (ii) increased early vigor, defined herein as an improved seedling aboveground area approximately three weeks post-germination.

"Early vigor" refers to active healthy plant growth especially during early stages of plant growth, and can result from increased plant fitness due to, for example, the plants being better adapted to their environment (for example, optimizing the use of energy resources, uptake of nutrients and partitioning carbon allocation between shoot and root). Early vigor, for example,



can be a combination of the ability of seeds to germinate and emerge after planting and the ability of the young plants to grow and develop after emergence. Plants having early vigor also show increased seedling survival and better establishment of the crop, which often results in highly uniform fields with the majority of the plants reaching the various stages of development at substantially the same time, which often results in increased yield. Therefore, early vigor can be determined by measuring various factors, such as kernel weight, percentage germination, percentage emergence, seedling growth, seedling height, root length, root and shoot biomass, canopy size and color and others.

Further, increased yield can also manifest as increased total seed yield, which may result from one or more of an increase in seed biomass (seed weight) due to an increase in the seed weight on a per plant and/or on an individual seed basis an increased number of, for example, flowers/panicles per plant; an increased number of pods; an increased number of nodes; an increased number of flowers ("florets") per panicle/plant; increased seed fill rate; an increased number of filled seeds; increased seed size (length, width, area, perimeter, and/or weight), which can also influence the composition of seeds; and/or increased seed volume, which can also influence the composition of seeds. In one embodiment, increased yield can be increased seed yield, for example, increased seed weight; increased number of filled seeds; and/or increased harvest index.

Increased yield can also result in modified architecture, or can occur because of modified plant architecture.

Increased yield can also manifest as increased harvest index, which is expressed as a ratio of the yield of harvestable parts, such as seeds, over the total biomass

The disclosure also extends to harvestable parts of a plant such as, but not limited to, seeds, leaves, fruits, flowers, bolls, pods, siliques, nuts, stems, rhizomes, tubers, and bulbs. The disclosure furthermore relates to products derived from a harvestable part of such a plant, such as dry pellets, powders, oil, fat and fatty acids, starch, or proteins.

The present disclosure provides a method for increasing "yield" of a plant or "broad acre yield" of a plant or plant part defined as the harvestable plant parts per unit area, for example seeds, or weight of seeds, per acre, pounds per acre, bushels per acre, tones per acre, tons per acre, kilo per hectare.

As used herein "nitrogen use efficiency" refers to the processes which lead to an increase in the plant's yield, biomass, vigor, and growth rate per nitrogen unit applied. The processes can include the uptake, assimilation, accumulation, signaling, sensing, retranslocation (within the plant) and use of nitrogen by the plant.

As used herein "increased nitrogen use efficiency" refers to the ability of plants to grow, develop, or yield faster or better than normal when subjected to the same amount of available/applied nitrogen as under normal or standard conditions; ability of plants to grow, develop, or yield normally, or grow, develop, or yield faster or better when subjected to less than optimal amounts of available/applied nitrogen, or under nitrogen limiting conditions.

As used herein "nitrogen limiting conditions" refers to growth conditions or environments that provide less than optimal amounts of nitrogen needed for adequate or successful plant metabolism, growth, reproductive success and/or viability.

As used herein the "increased nitrogen stress tolerance" refers to the ability of plants to grow, develop, or yield normally, or grow, develop, or yield faster or better when subjected to less than optimal amounts of available/applied nitrogen, or under nitrogen limiting conditions.

Increased plant nitrogen use efficiency can be translated in the field into either harvesting similar quantities of yield, while supplying less nitrogen, or increased yield gained by supplying optimal/sufficient amounts of nitrogen. The increased nitrogen use efficiency can improve plant nitrogen stress tolerance and can also improve crop quality and biochemical constituents of the seed such as protein yield and oil yield. The terms "increased nitrogen use efficiency", "enhanced nitrogen use efficiency", and "nitrogen stress tolerance" are used inter-changeably in the present disclosure to refer to plants with improved productivity under nitrogen limiting conditions.

As used herein "water use efficiency" refers to the amount of carbon dioxide assimilated by leaves per unit of water vapor transpired. It constitutes one of the most important traits controlling plant productivity in dry environments. "Drought tolerance" refers to the degree to which a plant is adapted to arid or drought conditions. The physiological responses of plants to a deficit of water include leaf wilting, a reduction in leaf area, leaf abscission, and the stimulation of root growth by directing nutrients to the underground parts of the plants. Typically, plants are more susceptible to drought during flowering and seed development (the reproductive stages), as plant's resources are deviated to support root growth. In addition, abscisic acid (ABA), a plant stress hormone, induces the closure of leaf stomata (microscopic pores involved in gas exchange), thereby reducing water loss through transpiration, and decreasing the rate of photosynthesis. These responses improve the water-use efficiency of the plant on the short term. The terms "increased water use efficiency", "enhanced water use efficiency", and "increased drought tolerance" are used inter-changeably in the present disclosure to refer to plants with improved productivity under water-limiting conditions.

As used herein "increased water use efficiency" refers to the ability of plants to grow, develop, or yield faster or better than normal when subjected to the same amount of available/applied water as under normal or standard conditions; ability of plants to grow, develop, or yield normally, or grow, develop, or yield faster or better when subjected to reduced amounts of available/applied water (water input) or under conditions of water stress or water deficit stress.

As used herein "increased drought tolerance" refers to the ability of plants to grow, develop, or yield normally, or grow, develop, or yield faster or better than normal when subjected to reduced amounts of available/applied water and/or under conditions of acute or chronic drought; ability of plants to grow, develop, or yield normally when subjected to reduced amounts of available/applied water (water input) or under conditions of water deficit stress or under conditions of acute or chronic drought.

As used herein, "drought stress" refers to a period of dryness (acute or chronic/prolonged) that results in water deficit and subjects plants to stress and/or damage to plant tissues and/or negatively affects grain/crop yield; a period of dryness (acute or chronic/prolonged) that results in water deficit and/or higher temperatures and subjects plants to stress and/or damage to plant tissues and/or negatively affects grain/crop yield.

As used herein, "water deficit" refers to the conditions or environments that provide less than optimal amounts of water needed for adequate/successful growth and development of plants.

As used herein, "water stress" refers to the conditions or environments that provide improper (either less/insufficient or more/excessive) amounts of water than that needed for adequate/successful growth and development of plants/crops thereby subjecting the plants to stress and/or damage to plant tissues and/or negatively affecting grain/crop yield.

As used herein "water deficit stress" refers to the conditions or environments that provide less/insufficient amounts of water than that needed for adequate/successful growth and development of plants/crops thereby subjecting the plants to stress and/or damage to plant tissues and/or negatively affecting grain yield.

As used herein, the terms "nucleic acid," "nucleic acid molecule," "nucleotide sequence" and "polynucleotide" refer to RNA or DNA that is linear or branched, single or double stranded, or a hybrid thereof. The term also encompasses RNA/DNA hybrids. When dsRNA is produced synthetically, less common bases, such as inosine, 5-methylcytosine, 6-methyladenine, hypoxanthine and others can also be used for antisense, dsRNA, and ribozyme pairing. For example, polynucleotides that contain C-5 propyne analogues of uridine and cytidine have been

shown to bind RNA with high affinity and to be potent antisense inhibitors of gene expression. Other modifications, such as modification to the phosphodiester backbone, or the 2'-hydroxy in the ribose sugar group of the RNA can also be made.

As used herein, the term "nucleotide sequence" refers to a heteropolymer of nucleotides or the sequence of these nucleotides from the 5' to 3' end of a nucleic acid molecule and includes DNA or RNA molecules, including cDNA, a DNA fragment or portion, genomic DNA, synthetic (*e.g.*, chemically synthesized) DNA, plasmid DNA, mRNA, and anti-sense RNA, any of which can be single stranded or double stranded. The terms "nucleotide sequence" "nucleic acid," "nucleic acid molecule," "nucleic acid construct," "oligonucleotide" and "polynucleotide" are also used interchangeably herein to refer to a heteropolymer of nucleotides. Nucleic acid molecules and/or nucleotide sequences provided herein are presented herein in the 5' to 3' direction, from left to right and are represented using the standard code for representing the nucleotide characters as set forth in the World Intellectual Property Organization (WIPO) Standard ST.26. A "5' region" as used herein can mean the region of a polynucleotide that is nearest the 5' end of the polynucleotide. Thus, for example, an element in the 5' region of a polynucleotide can be located anywhere from the first nucleotide located at the 5' end of the polynucleotide to the nucleotide located halfway through the polynucleotide. A "3' region" as used herein can mean the region of a polynucleotide that is nearest the 3' end of the polynucleotide. Thus, for example, an element in the 3' region of a polynucleotide can be located anywhere from the first nucleotide located at the 3' end of the polynucleotide to the nucleotide located halfway through the polynucleotide.

As used herein with respect to nucleic acids, the term "fragment" or "portion" refers to a nucleic acid that is reduced in length relative (*e.g.*, reduced by 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 210, 220, 230, 240, 250, 260, 270, 280, 290, 300, 310, 320, 330, 340, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950 or 1000 or more nucleotides or any range or value therein) to a reference nucleic acid and that comprises, consists essentially of and/or consists of a nucleotide sequence of contiguous nucleotides identical or almost identical (*e.g.*, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% identical) to a corresponding portion of the reference nucleic acid. Such a nucleic acid fragment may be, where appropriate, included in a larger polynucleotide of which it is a constituent. As an example, a repeat sequence of guide nucleic acid of this invention may comprise a portion of a wild type CRISPR-Cas repeat sequence (*e.g.*, a wild Type CRISPR-Cas repeat; *e.g.*, a repeat from

the CRISPR Cas system of, for example, a Cas9, Cas12a (Cpf1), Cas12b, Cas12c (C2c3), Cas12d (CasY), Cas12e (CasX), Cas12g, Cas12h, Cas12i, C2c4, C2c5, C2c8, C2c9, C2c10, Cas14a, Cas14b, and/or a Cas14c, and the like). In some embodiments, a nucleic acid fragment may comprise, consist essentially of or consist of about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 125, 150, 175, 200, 225, 250, 300, 350, 400, 450, 500, 550, 600, 660, 700, 750, 800, 850, 900, 950, 1000, 1050, 1100, 1150, 1200, 1250, 1300, 1350, 1400, 1450, 1500, 1550, 1600, 1650, 1700, 1750, 1800, 1850, 1900, 1950, 2000 or more consecutive nucleotides of a nucleotide sequence encoding a *HY5* gene for which a reduction in an activity, e.g., a reduction in COP1 binding to *HY5*, can result in a reduced shade avoidance response in a plant.

In some embodiments, a nucleic acid fragment or portion may comprise, consist essentially of or consist of about 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 102, 103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, 115, 116, 117, 118, 119, 120, 130, 140, 141, 142, 143, 145, 146, 147, 148, 149, 150, 175, 200, 210, 220, 225, 23, 231, 232, 233, 234, 235, 236, 237, 238, 239, 240, 241, 242, 243, 245, 245, 246, 247, 248, 249, 250, 260, 270, 280, 290, 300, 350, 360, 361, 362, 363, 364, 365, 366, 367, 368, 369, 370, 380, 390, 400, 410, 411, 412, 413, 414, 415, 416, 417, 418, 419, 420, 430, 440, 450, 460, 470, 480, 490, 491, 492, 493, 494, 495, 496, 497, 498, 499, 500, 510, 520, 530, 531, 532, 533, 534, 535, 536, 537, 538, 539, 540, 550, 560, 570, 580, 590, 600, 605, 610, 611, 612, 613, 614, 615, 616, 618, 619, or 620, or more consecutive nucleotides or any range or value therein, of a *HY5* polynucleotide (e.g., genomic DNA or coding DNA), optionally a fragment of a *HY5* polynucleotide may be about 20 nucleotides to about 120 nucleotides, about 20 nucleotides to about 250 nucleotides, about 20 nucleotides to about 620 nucleotides, about 100 nucleotides to about 250 nucleotides, about 100 nucleotides to about 400 nucleotides, about 150 nucleotides to about 400 nucleotides, about 200 nucleotides to about 620 nucleotides e.g., about 60, 80, 100, 120, 140, 160, 180 or 200 nucleotides to about 210, 220, 240, 260, 280, 300, 350, 400, 450, 500, 550, 600, 615 or 620 or more consecutive nucleotides (e.g., consecutive nucleotides of **SEQ ID NO:72**, e.g., **SEQ ID NO:75-105** optionally at least 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**).

As used herein with respect to polypeptides, the term "fragment" or "portion" may refer to a polypeptide that is reduced in length relative to a reference polypeptide and that comprises,

consists essentially of and/or consists of an amino acid sequence of contiguous amino acids identical or almost identical (e.g., 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% identical) to a corresponding portion of the reference polypeptide. Such a polypeptide fragment may be, where appropriate, included in a larger polypeptide of which it is a constituent. In some  
5 embodiments, the polypeptide fragment comprises, consists essentially of or consists of at least about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 35, 40, 45, 46, 47, 48, 49, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 125, 150, 175, 200, 225, 250, 300, 350, 400 or more consecutive amino acids of a reference polypeptide. In some embodiments, a HY5 transcription factor fragment comprises, consists essentially of, or consists of at least about 15,  
10 20, 25, 30, 35, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 125, 150, or 160 or more consecutive amino acids (e.g., a fragment or portion of **SEQ ID NO:74**, e.g., **SEQ ID NO:106**).

In some embodiments, a "portion" may be related to the number of amino acids that are deleted from a polypeptide. Thus, for example, a deleted "portion" of a HY5 transcription factor  
15 polypeptide may comprise at least one amino acid residue (e.g., at least 1, or at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50 or more consecutive amino acid residues) deleted from the amino acid sequence of **SEQ ID NO:74** (or from a sequence having at least 80% sequence identity (e.g., at least 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91,  
20 92, 93, 94, 95, 96, 97, 98, 99 or 100% identity; optionally wherein the sequence identity may be at least 85%, it may be at least 90% or it may be at least 95% or the sequence identity may be 100%) to the amino acid sequence of **SEQ ID NO:74**) (e.g., a deletion of about 1, 2, 3, 4, 5, 6, 7, 8, 9, or 10 to about 30, 31, 32, 33, 34, 35, 36, 37, 38, 37, or 40 residues, e.g., **SEQ ID NO:106** or a portion thereof), optionally a deletion in the region adjacent to or in the first and/or  
25 second exons of the *HY5* gene may result in one or more than one amino acid residue being deleted from the encoded HY5 polypeptide. In some embodiments, a deleted portion of a *HY5* transcription factor gene may be, for example, an in-frame deletion or an out-of-frame deletion in which at least one amino acid (e.g., one or more than one amino acid) is deleted from the N-terminal portion of the encoded HY5 transcription factor, optionally, a deletion of at least a  
30 portion of consecutive amino acid residues of **SEQ ID NO:74** (e.g., at least 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or more consecutive amino acid residues). In some embodiments, when the mutation is an out-of-frame deletion an early stop codon may be generated that results in a truncated HY5 protein.

In some embodiments, a deletion of one or more amino acid residues from a HY5 transcription factor polypeptide as described herein may result in a dominant mutation and/or or a dominant negative mutation, which when comprised in a plant can result in the plant exhibiting a reduced shade avoidance response (SAR) as compared to a plant not comprising the deletion. In some embodiments, deletion of one or more amino acid residues from a HY5 transcription factor polypeptide as described herein may result in a plant exhibiting increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity. In some embodiments, such a mutation results in a HY5 transcription factor that has reduced binding by a Constitutive Photomorphogenic 1 (COP1) polypeptide. In some embodiments, the mutation is a dominant negative mutation.

A "region" of a polynucleotide or a polypeptide refers to a portion of consecutive nucleotides or consecutive amino acid residues of that polynucleotide or a polypeptide, respectively. For example, a region of a HY5 polynucleotide sequence may include, but is not limited to, to any one of the nucleic acid sequences of **SEQ ID NOs:75-105** optionally 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%. In some embodiments, a region of a HY5 polypeptide sequence may include, but is not limited to, to the amino acid sequence of **SEQ ID NO:106**.

In some embodiments, a "sequence-specific nucleic acid binding domain" (e.g., sequence-specific DNA binding domain) may bind to a *HY5* transcription factor gene (e.g., **SEQ ID NO:72** or **SEQ ID NO:73**) and/or to one or more fragments, portions, or regions of a *HY5* transcription factor nucleic acid (e.g., **SEQ ID NOs:75-105**).

As used herein with respect to nucleic acids, the term "functional fragment" refers to nucleic acid that encodes a functional fragment of a polypeptide. A "functional fragment" with respect to a polypeptide is a fragment of a polypeptide that retains one or more of the activities of the native reference polypeptide.

The term "gene," as used herein, refers to a nucleic acid molecule capable of being used to produce mRNA, antisense RNA, miRNA, anti-microRNA antisense oligodeoxyribonucleotide (AMO) and the like. Genes may or may not be capable of being used to produce a functional

protein or gene product. Genes can include both coding and non-coding regions (e.g., introns, regulatory elements, promoters, enhancers, termination sequences and/or 5' and 3' untranslated regions). A gene may be "isolated" by which is meant a nucleic acid that is substantially or essentially free from components normally found in association with the nucleic acid in its natural state. Such components include other cellular material, culture medium from recombinant production, and/or various chemicals used in chemically synthesizing the nucleic acid.

The term "mutation" refers to point mutations (e.g., missense, or nonsense, or insertions or deletions of single base pairs that result in frame shifts), insertions, deletions, and/or truncations. When the mutation is a substitution of a residue within an amino acid sequence with another residue, or a deletion or insertion of one or more residues within a sequence, the mutations are typically described by identifying the original residue followed by the position of the residue within the sequence and by the identity of the newly substituted residue. A truncation can include a truncation at the C-terminal end of a polypeptide or at the N-terminal end of a polypeptide. A truncation of a polypeptide can be the result of a deletion of the corresponding 5' end or 3' end of the gene encoding the polypeptide. A frameshift mutation can occur when deletions or insertions of one or more base pairs are introduced into a gene, optionally resulting in an out-of-frame mutation or an in-frame mutation. Frameshift mutations in a gene can result in the production of a polypeptide that is longer, shorter or the same length as the wild type polypeptide depending on when the first stop codon occurs following the mutated region of the gene. As an example, an out-of-frame mutation that produces a premature stop codon can produce a polypeptide that is shorter than the wild type polypeptide, or, in some embodiments, the polypeptide may be absent/undetectable. A DNA inversion is the result of a rotation of a genetic fragment within a region of a chromosome.

The terms "complementary" or "complementarity," as used herein, refer to the natural binding of polynucleotides under permissive salt and temperature conditions by base-pairing. For example, the sequence "A-G-T" (5' to 3') binds to the complementary sequence "T-C-A" (3' to 5'). Complementarity between two single-stranded molecules may be "partial," in which only some of the nucleotides bind, or it may be complete when total complementarity exists between the single stranded molecules. The degree of complementarity between nucleic acid strands has significant effects on the efficiency and strength of hybridization between nucleic acid strands.

"Complement," as used herein, can mean 100% complementarity with the comparator nucleotide sequence or it can mean less than 100% complementarity (e.g., about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%,



88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, and the like, complementarity; e.g., substantial complementarity) to the comparator nucleotide sequence.

Different nucleic acids or proteins having homology are referred to herein as "homologues." The term homologue includes homologous sequences from the same and from other species and orthologous sequences from the same and other species. "Homology" refers to the level of similarity between two or more nucleic acid and/or amino acid sequences in terms of percent of positional identity (*i.e.*, sequence similarity or identity). Homology also refers to the concept of similar functional properties among different nucleic acids or proteins. Thus, the compositions and methods of the invention further comprise homologues to the nucleotide sequences and polypeptide sequences of this invention. "Orthologous," as used herein, refers to homologous nucleotide sequences and/ or amino acid sequences in different species that arose from a common ancestral gene during speciation. A homologue of a nucleotide sequence of this invention has a substantial sequence identity (e.g., at least about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.5% or 100%) to said nucleotide sequence of the invention.

As used herein "sequence identity" refers to the extent to which two optimally aligned polynucleotide or polypeptide sequences are invariant throughout a window of alignment of components, *e.g.*, nucleotides or amino acids. "Identity" can be readily calculated by known methods including, but not limited to, those described in: *Computational Molecular Biology* (Lesk, A. M., ed.) Oxford University Press, New York (1988); *Biocomputing: Informatics and Genome Projects* (Smith, D. W., ed.) Academic Press, New York (1993); *Computer Analysis of Sequence Data, Part I* (Griffin, A. M., and Griffin, H. G., eds.) Humana Press, New Jersey (1994); *Sequence Analysis in Molecular Biology* (von Heinje, G., ed.) Academic Press (1987); and *Sequence Analysis Primer* (Gribskov, M. and Devereux, J., eds.) Stockton Press, New York (1991).

As used herein, the term "percent sequence identity" or "percent identity" refers to the percentage of identical nucleotides in a linear polynucleotide sequence of a reference ("query") polynucleotide molecule (or its complementary strand) as compared to a test ("subject") polynucleotide molecule (or its complementary strand) when the two sequences are optimally aligned. In some embodiments, "percent sequence identity" can refer to the percentage of identical amino acids in an amino acid sequence as compared to a reference polypeptide. In regard to a *HY5* gene, a sequence may have at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**. In some embodiments, a *HY5* gene may have at

least 85% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**. In some embodiments, a *HY5* gene may have at least 90% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**. In some embodiments, a *HY5* gene may have at least 95% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**,  
5 optionally wherein the *HY5* gene may have at least 100% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**. A *HY5* polypeptide as described herein may have at least 80% sequence identity to the polypeptide sequence of **SEQ ID NO:74**. In some embodiments, a *HY5* polypeptide may have at least 85% sequence identity to the polypeptide sequence of **SEQ ID NO:74**. In some embodiments, a *HY5* polypeptide may have at least 90%  
10 sequence identity to the polypeptide sequence of **SEQ ID NO:74**. In some embodiments, a *HY5* polypeptide may have at least 95% sequence identity to the polypeptide sequence of **SEQ ID NO:74**, optionally wherein the *HY5* polypeptide may have at least 100% sequence identity to the polypeptide sequence of **SEQ ID NO:74**. With regard to regions or portions of a *HY5* gene, the region or portion may have at least 80% sequence identity to the nucleotide sequence  
15 of any one of **SEQ ID NOs:75-105**, optionally at least 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**. In some embodiments, a region or portion of a *HY5* gene may have at least 85% sequence identity to the nucleotide sequence of any one of **SEQ ID NOs:75-105**, optionally at least 85% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**. In some  
20 embodiments, a region or portion of a *HY5* gene may have at least 90% sequence identity to the nucleotide sequence of any one of **SEQ ID NOs:75-105**, optionally at least 90% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**. In some embodiments, a region or portion of a *HY5* gene may have at least 95% sequence identity to the nucleotide sequence of any one of **SEQ ID NOs:75-105**, optionally at least 95% sequence  
25 identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**. In some embodiments, a region or portion of a *HY5* gene may have at least 100% sequence identity to the nucleotide sequence any one of **SEQ ID NOs:75-105**, optionally at least 100% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**.  
With regard to regions or portions of a *HY5* polypeptide, the region or portion may have at least  
30 at least 80% sequence identity to the polypeptide sequence of **SEQ ID NO:106**. In some embodiments, a region or portion of a *HY5* polypeptide may have at least 85% sequence identity to the polypeptide sequence of **SEQ ID NO:106**. In some embodiments, a region or portion of a *HY5* polypeptide may have at least 90% sequence identity to the polypeptide sequence of **SEQ ID NO:106**. In some embodiments, a region or portion of a *HY5* polypeptide may have at least

95% sequence identity to the polypeptide sequence of **SEQ ID NO:106**. In some embodiments, a region or portion of a HY5 polypeptide may have at least 100% sequence identity to the polypeptide sequence of **SEQ ID NO:106**. With regard to an edited HY5 nucleic acid sequences, an edited HY5 nucleic acid sequence may have at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139**. In some embodiments, an edited HY5 nucleic acid sequence may have at least 95% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139**. In some embodiments, an edited HY5 nucleic acid sequence may have 100% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139**. With regard to an mutated HY5 amino acid sequences, a mutated HY5 polypeptide may have at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, and 138**. In some embodiments, a mutated HY5 polypeptide may have at least 95% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, and 138**. In some embodiments, a mutated HY5 polypeptide may have 100% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, and 138**.

As used herein, the phrase "substantially identical," or "substantial identity" in the context of two nucleic acid molecules, nucleotide sequences or polypeptide sequences, refers to two or more sequences or subsequences that have at least about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.5% or 100% nucleotide or amino acid residue identity, when compared and aligned for maximum correspondence, as measured using one of the following sequence comparison algorithms or by visual inspection. In some embodiments of the invention, the substantial identity exists over a region of consecutive nucleotides of a nucleotide sequence of the invention that is about 10 nucleotides to about 20 nucleotides, about 10 nucleotides to about 25 nucleotides, about 10 nucleotides to about 30 nucleotides, about 15 nucleotides to about 25 nucleotides, about 30 nucleotides to about 40 nucleotides, about 50 nucleotides to about 60 nucleotides, about 70 nucleotides to about 80 nucleotides, about 90 nucleotides to about 100 nucleotides, about 100 nucleotides to about 200 nucleotides, about 100 nucleotides to about 300 nucleotides, about 100 nucleotides to about 400 nucleotides, about 100 nucleotides to about 500 nucleotides, about 100 nucleotides to about 600 nucleotides, about 100 nucleotides to about 800 nucleotides, about 100 nucleotides to about 900 nucleotides, or more nucleotides in length, and any range therein, up to the full length of the sequence. In some embodiments, nucleotide sequences can be substantially identical over at

least about 20 consecutive nucleotides (e.g., about 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 50, 60, 70, 80, 90, 100, 200, 300, 400, 500, 600, 700, 800, 900, 1000, 1100, 1200, 1300, 1400, 1500, 1600, 1700, 1800, 1900, 2000, 2100, 2200, 2300 or more nucleotides). In some embodiments, two or more *HY5* transcription factor genes may be  
5 substantially identical to one another over at least about 30 or more consecutive nucleotides (e.g., 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 54, 56, 57, 58, 59, 60, 65, 70, 75, 80, 85, 90, 95, 100, 110, 120, 130, 140, 150, 160, 170, 180, 200, 220, 240, 260, 280, 300, 320, 340, 360, 380, 400, 420, 440, 460, 480, 500, 520, 540, 560, 580, 600, or more consecutive nucleotides) of **SEQ ID NO:72** (see, e.g., **SEQ ID NOs:75-105**).

10 In some embodiments of the invention, the substantial identity exists over a region of consecutive amino acid residues of a polypeptide of the invention that is about 3 amino acid residues to about 20 amino acid residues, about 5 amino acid residues to about 10 amino acid residues, about 5 amino acid residues to about 55 amino acid residues, about 5 amino acid residues to about 25 amino acid residues, about 7 amino acid residues to about 30 amino acid  
15 residues, about 10 amino acid residues to about 25 amino acid residues, about 15 amino acid residues to about 30 amino acid residues, about 20 amino acid residues to about 40 amino acid residues, about 25 amino acid residues to about 40 amino acid residues, about 25 amino acid residues to about 50 amino acid residues, about 30 amino acid residues to about 50 amino acid residues, about 40 amino acid residues to about 50 amino acid residues, about 40 amino acid  
20 residues to about 70 amino acid residues, about 50 amino acid residues to about 70 amino acid residues, about 60 amino acid residues to about 80 amino acid residues, about 70 amino acid residues to about 80 amino acid residues, about 90 amino acid residues to about 100 amino acid residues, or more amino acid residues in length, and any range therein, up to the full length of the sequence. In some embodiments, polypeptide sequences can be substantially identical to one  
25 another over at least about 8, 9, 10, 11, 12, 13, 14, or more consecutive amino acid residues (e.g., about 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 102, 103, 104, 105,  
30 106, 107, 108, 109, 110, 111, 112, 113, 114, 115, 116, 117, 118, 119, 120, 130, 140, 150, 175, 200, 225, 250, 275, 300, 325, 350, 400, 450, 500, or more amino acids in length or more consecutive amino acid residues). In some embodiments, two or more *HY5* transcription factor polypeptides may be substantially identical to one another over at least about 10 to about 150 (e.g., about 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31,

32, 33, 34, 35, 36, 37, 38, 39, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 105, 110, 115, 120, 125, 130, 135, 140, 145 or 150 residues or more) consecutive amino acid residues of **SEQ ID NO:74**, e.g., **SEQ ID NO:106**, or any range or value therein). In some embodiments, a substantially identical nucleotide or protein sequence may perform substantially the same  
5 function as the nucleotide (or encoded protein sequence) to which it is substantially identical.

For sequence comparison, typically one sequence acts as a reference sequence to which test sequences are compared. When using a sequence comparison algorithm, test and reference sequences are entered into a computer, subsequence coordinates are designated if necessary, and sequence algorithm program parameters are designated. The sequence comparison algorithm  
10 then calculates the percent sequence identity for the test sequence(s) relative to the reference sequence, based on the designated program parameters.

Optimal alignment of sequences for aligning a comparison window are well known to those skilled in the art and may be conducted by tools such as the local homology algorithm of Smith and Waterman, the homology alignment algorithm of Needleman and Wunsch, the search  
15 for similarity method of Pearson and Lipman, and optionally by computerized implementations of these algorithms such as GAP, BESTFIT, FASTA, and TFASTA available as part of the GCG® Wisconsin Package® (Accelrys Inc., San Diego, CA). An "identity fraction" for aligned segments of a test sequence and a reference sequence is the number of identical components which are shared by the two aligned sequences divided by the total number of components in the  
20 reference sequence segment, e.g., the entire reference sequence or a smaller defined part of the reference sequence. Percent sequence identity is represented as the identity fraction multiplied by 100. The comparison of one or more polynucleotide sequences may be to a full-length polynucleotide sequence or a portion thereof, or to a longer polynucleotide sequence. For purposes of this invention "percent identity" may also be determined using BLASTX version 2.0  
25 for translated nucleotide sequences and BLASTN version 2.0 for polynucleotide sequences.

Two nucleotide sequences may also be considered substantially complementary when the two sequences hybridize to each other under stringent conditions. In some embodiments, two nucleotide sequences considered to be substantially complementary hybridize to each other under highly stringent conditions.

30 "Stringent hybridization conditions" and "stringent hybridization wash conditions" in the context of nucleic acid hybridization experiments such as Southern and Northern hybridizations are sequence dependent and are different under different environmental parameters. An extensive guide to the hybridization of nucleic acids is found in Tijssen *Laboratory Techniques in Biochemistry and Molecular Biology-Hybridization with Nucleic Acid Probes* part I chapter 2

"Overview of principles of hybridization and the strategy of nucleic acid probe assays" Elsevier, New York (1993). Generally, highly stringent hybridization and wash conditions are selected to be about 5°C lower than the thermal melting point ( $T_m$ ) for the specific sequence at a defined ionic strength and pH.

5           The  $T_m$  is the temperature (under defined ionic strength and pH) at which 50% of the target sequence hybridizes to a perfectly matched probe. Very stringent conditions are selected to be equal to the  $T_m$  for a particular probe. An example of stringent hybridization conditions for hybridization of complementary nucleotide sequences which have more than 100 complementary residues on a filter in a Southern or northern blot is 50% formamide with 1 mg of heparin at 42°C, with the hybridization being carried out overnight. An example of highly 10 stringent wash conditions is 0.1 5M NaCl at 72°C for about 15 minutes. An example of stringent wash conditions is a 0.2x SSC wash at 65°C for 15 minutes (*see*, Sambrook, *infra*, for a description of SSC buffer). Often, a high stringency wash is preceded by a low stringency wash to remove background probe signal. An example of a medium stringency wash for a duplex of, 15 e.g., more than 100 nucleotides, is 1x SSC at 45°C for 15 minutes. An example of a low stringency wash for a duplex of, e.g., more than 100 nucleotides, is 4-6x SSC at 40°C for 15 minutes. For short probes (e.g., about 10 to 50 nucleotides), stringent conditions typically involve salt concentrations of less than about 1.0 M Na ion, typically about 0.01 to 1.0 M Na ion concentration (or other salts) at pH 7.0 to 8.3, and the temperature is typically at least about 20 30°C. Stringent conditions can also be achieved with the addition of destabilizing agents such as formamide. In general, a signal to noise ratio of 2x (or higher) than that observed for an unrelated probe in the particular hybridization assay indicates detection of a specific hybridization. Nucleotide sequences that do not hybridize to each other under stringent conditions are still substantially identical if the proteins that they encode are substantially 25 identical. This can occur, for example, when a copy of a nucleotide sequence is created using the maximum codon degeneracy permitted by the genetic code.

A polynucleotide and/or recombinant nucleic acid construct of this invention (e.g., expression cassettes and/or vectors) may be codon optimized for expression. In some 30 embodiments, the polynucleotides, nucleic acid constructs, expression cassettes, and/or vectors of the editing systems of the invention (e.g., comprising/encoding a sequence-specific nucleic acid binding domain (e.g., a sequence-specific nucleic acid binding domain from a polynucleotide-guided endonuclease, a zinc finger nuclease, a transcription activator-like effector nuclease (TALEN), an Argonaute protein, and/or a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein) (e.g., a Type I CRISPR-Cas effector protein, a Type II CRISPR-

Cas effector protein, a Type III CRISPR-Cas effector protein, a Type IV CRISPR-Cas effector protein, a Type V CRISPR-Cas effector protein or a Type VI CRISPR-Cas effector protein)), a nuclease (e.g., an endonuclease (e.g., FokI), a polynucleotide-guided endonuclease, a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein), a zinc finger nuclease, and/or a  
5 transcription activator-like effector nuclease (TALEN)), deaminase proteins/domains (e.g., adenine deaminase, cytosine deaminase), a polynucleotide encoding a reverse transcriptase protein or domain, a polynucleotide encoding a 5'-3' exonuclease polypeptide, and/or affinity polypeptides, peptide tags, etc.) may be codon optimized for expression in a plant. In some  
10 embodiments, the codon optimized nucleic acids, polynucleotides, expression cassettes, and/or vectors of the invention have about 70% to about 99.9% (e.g., 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.5%, 99.9% or 100%) identity or more to the reference nucleic acids, polynucleotides, expression cassettes, and/or vectors that have not been codon optimized.

15 A polynucleotide or nucleic acid construct of the invention may be operatively associated with a variety of promoters and/or other regulatory elements for expression in a plant and/or a cell of a plant. Thus, in some embodiments, a polynucleotide or nucleic acid construct of this invention may further comprise one or more promoters, introns, enhancers, and/or  
20 terminators operably linked to one or more nucleotide sequences. In some embodiments, a promoter may be operably associated with an intron (e.g., Ubi1 promoter and intron). In some embodiments, a promoter associated with an intron maybe referred to as a "promoter region" (e.g., Ubi1 promoter and intron) (see, e.g., **SEQ ID NO:21** and **SEQ ID NO:22**).

By "operably linked" or "operably associated" as used herein in reference to polynucleotides, it is meant that the indicated elements are functionally related to each other and  
25 are also generally physically related. Thus, the term "operably linked" or "operably associated" as used herein, refers to nucleotide sequences on a single nucleic acid molecule that are functionally associated. Thus, a first nucleotide sequence that is operably linked to a second nucleotide sequence means a situation when the first nucleotide sequence is placed in a functional relationship with the second nucleotide sequence. For instance, a promoter is  
30 operably associated with a nucleotide sequence if the promoter effects the transcription or expression of said nucleotide sequence. Those skilled in the art will appreciate that the control sequences (e.g., promoter) need not be contiguous with the nucleotide sequence to which it is operably associated, as long as the control sequences function to direct the expression thereof. Thus, for example, intervening untranslated, yet transcribed, nucleic acid sequences can be

present between a promoter and the nucleotide sequence, and the promoter can still be considered "operably linked" to the nucleotide sequence.

As used herein, the term "linked," in reference to polypeptides, refers to the attachment of one polypeptide to another. A polypeptide may be linked to another polypeptide (at the N-terminus or the C-terminus) directly (e.g., via a peptide bond) or through a linker.

The term "linker" is art-recognized and refers to a chemical group, or a molecule linking two molecules or moieties, e.g., two domains of a fusion protein, such as, for example, a DNA binding polypeptide or domain and peptide tag and/or a reverse transcriptase and an affinity polypeptide that binds to the peptide tag; or a DNA endonuclease polypeptide or domain and peptide tag and/or a reverse transcriptase and an affinity polypeptide that binds to the peptide tag. A linker may be comprised of a single linking molecule or may comprise more than one linking molecule. In some embodiments, the linker can be an organic molecule, group, polymer, or chemical moiety such as a bivalent organic moiety. In some embodiments, the linker may be an amino acid or it may be a peptide. In some embodiments, the linker is a peptide.

In some embodiments, a peptide linker useful with this invention may be about 2 to about 100 or more amino acids in length, for example, about 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100 or more amino acids in length (e.g., about 2 to about 40, about 2 to about 50, about 2 to about 60, about 4 to about 40, about 4 to about 50, about 4 to about 60, about 5 to about 40, about 5 to about 50, about 5 to about 60, about 9 to about 40, about 9 to about 50, about 9 to about 60, about 10 to about 40, about 10 to about 50, about 10 to about 60, or about 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 amino acids to about 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100 or more amino acids in length (e.g., about 105, 110, 115, 120, 130, 140 150 or more amino acids in length). In some embodiments, a peptide linker may be a GS linker.

In some embodiments, two or more polynucleotide molecules may be linked by a linker that can be an organic molecule, group, polymer, or chemical moiety such as a bivalent organic moiety. A polynucleotide may be linked or fused to another polynucleotide (at the 5' end or the 3' end) via a covalent or non-covalent linkage or binding, including e.g., Watson-Crick base-



pairing, or through one or more linking nucleotides. In some embodiments, a polynucleotide motif of a certain structure may be inserted within another polynucleotide sequence (e.g., extension of the hairpin structure in the guide RNA). In some embodiments, the linking nucleotides may be naturally occurring nucleotides. In some embodiments, the linking

5 nucleotides may be non-naturally occurring nucleotides.

A "promoter" is a nucleotide sequence that controls or regulates the transcription of a nucleotide sequence (e.g., a coding sequence) that is operably associated with the promoter. The coding sequence controlled or regulated by a promoter may encode a polypeptide and/or a functional RNA. Typically, a "promoter" refers to a nucleotide sequence that contains a binding

10 site for RNA polymerase II and directs the initiation of transcription. In general, promoters are found 5', or upstream, relative to the start of the coding region of the corresponding coding sequence. A promoter may comprise other elements that act as regulators of gene expression; e.g., a promoter region. These include a TATA box consensus sequence, and often a CAAT box consensus sequence (Breathnach and Chambon, (1981) *Annu. Rev. Biochem.* 50:349). In plants,

15 the CAAT box may be substituted by the AGGA box (Messing et al., (1983) in *Genetic Engineering of Plants*, T. Kosuge, C. Meredith and A. Hollaender (eds.), Plenum Press, pp. 211-227).

Promoters useful with this invention can include, for example, constitutive, inducible, temporally regulated, developmentally regulated, chemically regulated, tissue-preferred and/or

20 tissue-specific promoters for use in the preparation of recombinant nucleic acid molecules, e.g., "synthetic nucleic acid constructs" or "protein-RNA complex." These various types of promoters are known in the art.

The choice of promoter may vary depending on the temporal and spatial requirements for expression, and also may vary based on the host cell to be transformed. Promoters for many

25 different organisms are well known in the art. Based on the extensive knowledge present in the art, the appropriate promoter can be selected for the particular host organism of interest. Thus, for example, much is known about promoters upstream of highly constitutively expressed genes in model organisms and such knowledge can be readily accessed and implemented in other systems as appropriate.

In some embodiments, a promoter functional in a plant may be used with the constructs of this invention. Non-limiting examples of a promoter useful for driving expression in a plant include the promoter of the RubisCo small subunit gene 1 (PrbcS1), the promoter of the actin gene (Pactin), the promoter of the nitrate reductase gene (Pnr) and the promoter of duplicated carbonic anhydrase gene 1 (Pdca1) (See, Walker et al. (2005) *Plant Cell Rep.* 23:727-735; Li et

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al. (2007) *Gene* 403:132-142; Li et al. (2010) *Mol Biol. Rep.* 37:1143-1154). PrbcS1 and Pactin are constitutive promoters and Pnr and Pdca1 are inducible promoters. Pnr is induced by nitrate and repressed by ammonium (Li et al. (2007) *Gene* 403:132-142) and Pdca1 is induced by salt (Li et al. (2010) *Mol Biol. Rep.* 37:1143-1154). In some embodiments, a promoter useful with  
5 this invention is RNA polymerase II (Pol II) promoter. In some embodiments, a U6 promoter or a 7SL promoter from *Zea mays* may be useful with constructs of this invention. In some embodiments, the U6c promoter and/or 7SL promoter from *Zea mays* may be useful for driving expression of a guide nucleic acid. In some embodiments, a U6c promoter, U6i promoter and/or 7SL promoter from *Glycine max* may be useful with constructs of this invention. In some  
10 embodiments, the U6c promoter, U6i promoter and/or 7SL promoter from *Glycine max* may be useful for driving expression of a guide nucleic acid.

Examples of constitutive promoters useful for plants include, but are not limited to, cestrum virus promoter (cmp) (US Patent No. 7,166,770), the rice actin 1 promoter (Wang et al. (1992) *Mol. Cell. Biol.* 12:3399-3406; as well as US Patent No. 5,641,876), CaMV 35S  
15 promoter (Odell et al. (1985) *Nature* 313:810-812), CaMV 19S promoter (Lawton et al. (1987) *Plant Mol. Biol.* 9:315-324), nos promoter (Ebert et al. (1987) *Proc. Natl. Acad. Sci USA* 84:5745-5749), Adh promoter (Walker et al. (1987) *Proc. Natl. Acad. Sci. USA* 84:6624-6629), sucrose synthase promoter (Yang & Russell (1990) *Proc. Natl. Acad. Sci. USA* 87:4144-4148), and the ubiquitin promoter. The constitutive promoter derived from ubiquitin accumulates in  
20 many cell types. Ubiquitin promoters have been cloned from several plant species for use in transgenic plants, for example, sunflower (Binet et al. (1991) *Plant Science* 79: 87-94), maize (Christensen et al. (1989) *Plant Molec. Biol.* 12: 619-632), and *Arabidopsis* (Norris et al. (1993) *Plant Molec. Biol.* 21:895-906). The maize ubiquitin promoter (UbiP) has been developed in transgenic monocot systems and its sequence and vectors constructed for monocot  
25 transformation are disclosed in the patent publication EP 0 342 926. The ubiquitin promoter is suitable for the expression of the nucleotide sequences of the invention in transgenic plants, especially monocotyledons. Further, the promoter expression cassettes described by McElroy et al. ((1991) *Mol. Gen. Genet.* 231: 150-160) can be easily modified for the expression of the nucleotide sequences of the invention and are particularly suitable for use in monocotyledonous  
30 hosts.

In some embodiments, tissue specific/tissue preferred promoters can be used for expression of a heterologous polynucleotide in a plant cell. Tissue specific or preferred expression patterns include, but are not limited to, green tissue specific or preferred, root specific or preferred, stem specific or preferred, flower specific or preferred or pollen specific or

preferred. Promoters suitable for expression in green tissue include many that regulate genes involved in photosynthesis and many of these have been cloned from both monocotyledons and dicotyledons. In one embodiment, a promoter useful with the invention is the maize PEPC promoter from the phosphoenol carboxylase gene (Hudspeth & Grula (1989) *Plant Molec. Biol.* 12:579-589). Non-limiting examples of tissue-specific promoters include those associated with genes encoding the seed storage proteins (such as  $\beta$ -conglycinin, cruciferin, napin and phaseolin), zein or oil body proteins (such as oleosin), or proteins involved in fatty acid biosynthesis (including acyl carrier protein, stearyl-ACP desaturase and fatty acid desaturases (fad 2-1)), and other nucleic acids expressed during embryo development (such as Bce4, see, e.g., Kridl et al. (1991) *Seed Sci. Res.* 1:209-219; as well as EP Patent No. 255378). Tissue-specific or tissue-preferential promoters useful for the expression of the nucleotide sequences of the invention in plants, particularly maize, include but are not limited to those that direct expression in root, pith, leaf, or pollen. Such promoters are disclosed, for example, in WO 93/07278, herein incorporated by reference in its entirety. Other non-limiting examples of tissue specific or tissue preferred promoters useful with the invention the cotton rubisco promoter disclosed in US Patent No. 6,040,504; the rice sucrose synthase promoter disclosed in US Patent No. 5,604,121; the root specific promoter described by de Framond ((1991) *FEBS* 290:103-106; EP 0 452 269 to Ciba-Geigy); the stem specific promoter described in US Patent No. 5,625,136 (to Ciba-Geigy) and which drives expression of the maize trpA gene; the cestrum yellow leaf curling virus promoter disclosed in WO 01/73087; and pollen specific or preferred promoters including, but not limited to, ProOsLPS10 and ProOsLPS11 from rice (Nguyen et al. (2015) *Plant Biotechnol. Reports* 9(5):297-306), ZmSTK2\_USP from maize (Wang et al. (2017) *Genome* 60(6):485-495), LAT52 and LAT59 from tomato (Twell et al. (1990) *Development* 109(3):705-713), Zm13 (US Patent No. 10,421,972), PLA<sub>2</sub>- $\delta$  promoter from *Arabidopsis* (US Patent No. 7,141,424), and/or the ZmC5 promoter from maize (International PCT Publication No. WO1999/042587).

Additional examples of plant tissue-specific/tissue preferred promoters include, but are not limited to, the root hair-specific cis-elements (RHEs) (Kim et al. (2006) *The Plant Cell* 18:2958-2970), the root-specific promoters RCc3 (Jeong et al. (2010) *Plant Physiol.* 153:185-197) and RB7 (US Patent No. 5459252), the lectin promoter (Lindstrom et al. (1990) *Dev. Genet.* 11:160-167; and Vodkin (1983) *Prog. Clin. Biol. Res.* 138:87-98), corn alcohol dehydrogenase 1 promoter (Dennis et al. (1984) *Nucleic Acids Res.* 12:3983-4000), S-adenosyl-L-methionine synthetase (SAMS) (Vander Mijnsbrugge et al. (1996) *Plant and Cell Physiology* 37(8):1108-1115), corn light harvesting complex promoter (Bansal et al. (1992) *Proc. Natl.*

*Acad. Sci. USA* 89:3654-3658), corn heat shock protein promoter (O'Dell et al. (1985) *EMBO J.* 5:451-458; and Rochester et al. (1986) *EMBO J.* 5:451-458), pea small subunit RuBP carboxylase promoter (Cashmore, "Nuclear genes encoding the small subunit of ribulose-1,5-bisphosphate carboxylase" pp. 29-39 In: *Genetic Engineering of Plants* (Hollaender ed., Plenum Press 1983; and Poulsen et al. (1986) *Mol. Gen. Genet.* 205:193-200), Ti plasmid mannopine synthase promoter (Langridge et al. (1989) *Proc. Natl. Acad. Sci. USA* 86:3219-3223), Ti plasmid nopaline synthase promoter (Langridge et al. (1989) *supra*), petunia chalcone isomerase promoter (van Tunen et al. (1988) *EMBO J.* 7:1257-1263), bean glycine rich protein 1 promoter (Keller et al. (1989) *Genes Dev.* 3:1639-1646), truncated CaMV 35S promoter (O'Dell et al. (1985) *Nature* 313:810-812), potato patatin promoter (Wenzler et al. (1989) *Plant Mol. Biol.* 13:347-354), root cell promoter (Yamamoto et al. (1990) *Nucleic Acids Res.* 18:7449), maize zein promoter (Kriz et al. (1987) *Mol. Gen. Genet.* 207:90-98; Langridge et al. (1983) *Cell* 34:1015-1022; Reina et al. (1990) *Nucleic Acids Res.* 18:6425; Reina et al. (1990) *Nucleic Acids Res.* 18:7449; and Wandelt et al. (1989) *Nucleic Acids Res.* 17:2354), globulin-1 promoter (Belanger et al. (1991) *Genetics* 129:863-872),  $\alpha$ -tubulin cab promoter (Sullivan et al. (1989) *Mol. Gen. Genet.* 215:431-440), PEPCase promoter (Hudspeth & Grula (1989) *Plant Mol. Biol.* 12:579-589), R gene complex-associated promoters (Chandler et al. (1989) *Plant Cell* 1:1175-1183), and chalcone synthase promoters (Franken et al. (1991) *EMBO J.* 10:2605-2612).

Useful for seed-specific expression is the pea vicilin promoter (Czako et al. (1992) *Mol. Gen. Genet.* 235:33-40; as well as the seed-specific promoters disclosed in US Patent No. 5,625,136. Useful promoters for expression in mature leaves are those that are switched at the onset of senescence, such as the SAG promoter from *Arabidopsis* (Gan et al. (1995) *Science* 270:1986-1988).

In addition, promoters functional in chloroplasts can be used. Non-limiting examples of such promoters include the bacteriophage T3 gene 9 5' UTR and other promoters disclosed in US Patent No. 7,579,516. Other promoters useful with the invention include but are not limited to the S-E9 small subunit RuBP carboxylase promoter and the Kunitz trypsin inhibitor gene promoter (Kti3).

Additional regulatory elements useful with this invention include, but are not limited to, introns, enhancers, termination sequences and/or 5' and 3' untranslated regions.

An intron useful with this invention can be an intron identified in and isolated from a plant and then inserted into an expression cassette to be used in transformation of a plant. As would be understood by those of skill in the art, introns can comprise the sequences required for self-excision and are incorporated into nucleic acid constructs/expression cassettes in frame. An

intron can be used either as a spacer to separate multiple protein-coding sequences in one nucleic acid construct, or an intron can be used inside one protein-coding sequence to, for example, stabilize the mRNA. If they are used within a protein-coding sequence, they are inserted "in-frame" with the excision sites included. Introns may also be associated with promoters to improve or modify expression. As an example, a promoter/intron combination useful with this invention includes but is not limited to that of the maize Ubi1 promoter and intron (see, e.g., SEQ ID NO:21 and SEQ ID NO:22).

Non-limiting examples of introns useful with the present invention include introns from the ADHI gene (e.g., Adh1-S introns 1, 2 and 6), the ubiquitin gene (Ubi1), the RuBisCO small subunit (*rbcS*) gene, the RuBisCO large subunit (*rbcL*) gene, the actin gene (e.g., actin-1 intron), the pyruvate dehydrogenase kinase gene (*pdh*), the nitrate reductase gene (*nr*), the duplicated carbonic anhydrase gene 1 (*Tdca1*), the *psbA* gene, the *atpA* gene, or any combination thereof.

In some embodiments, a polynucleotide and/or a nucleic acid construct of the invention can be an "expression cassette" or can be comprised within an expression cassette. As used herein, "expression cassette" means a recombinant nucleic acid molecule comprising, for example, a one or more polynucleotides of the invention (e.g., a polynucleotide encoding a sequence-specific nucleic acid binding domain, a polynucleotide encoding a deaminase protein or domain, a polynucleotide encoding a reverse transcriptase protein or domain, a polynucleotide encoding a 5'-3' exonuclease polypeptide or domain, a guide nucleic acid and/or reverse transcriptase (RT) template), wherein polynucleotide(s) is/are operably associated with one or more control sequences (e.g., a promoter, terminator and the like). Thus, in some embodiments, one or more expression cassettes may be provided, which are designed to express, for example, a nucleic acid construct of the invention (e.g., a polynucleotide encoding a sequence-specific nucleic acid binding domain (e.g., sequence-specific DNA binding domain), a polynucleotide encoding a nuclease polypeptide/domain, a polynucleotide encoding a deaminase protein/domain, a polynucleotide encoding a reverse transcriptase protein/domain, a polynucleotide encoding a 5'-3' exonuclease polypeptide/domain, a polynucleotide encoding a peptide tag, and/or a polynucleotide encoding an affinity polypeptide, and the like, or comprising a guide nucleic acid, an extended guide nucleic acid, and/or RT template, and the like). When an expression cassette of the present invention comprises more than one polynucleotide, the polynucleotides may be operably linked to a single promoter that drives expression of all of the polynucleotides or the polynucleotides may be operably linked to one or more separate promoters (e.g., three polynucleotides may be driven by one, two or three promoters in any combination). When two or more separate promoters are used, the promoters

may be the same promoter, or they may be different promoters. Thus, a polynucleotide encoding a sequence specific nucleic acid binding domain, a polynucleotide encoding a nuclease protein/domain, a polynucleotide encoding a CRISPR-Cas effector protein/domain, a polynucleotide encoding an deaminase protein/domain, a polynucleotide encoding a reverse transcriptase polypeptide/domain (e.g., RNA-dependent DNA polymerase), and/or a  
5 polynucleotide encoding a 5'-3' exonuclease polypeptide/domain, a guide nucleic acid, an extended guide nucleic acid and/or RT template when comprised in a single expression cassette may each be operably linked to a single promoter, or separate promoters in any combination.

An expression cassette comprising a nucleic acid construct of the invention may be  
10 chimeric, meaning that at least one of its components is heterologous with respect to at least one of its other components (e.g., a promoter from the host organism operably linked to a polynucleotide of interest to be expressed in the host organism, wherein the polynucleotide of interest is from a different organism than the host or is not normally found in association with that promoter). An expression cassette may also be one that is naturally occurring but has been  
15 obtained in a recombinant form useful for heterologous expression.

An expression cassette can optionally include a transcriptional and/or translational termination region (i.e., termination region) and/or an enhancer region that is functional in the selected host cell. A variety of transcriptional terminators and enhancers are known in the art and are available for use in expression cassettes. Transcriptional terminators are responsible for  
20 the termination of transcription and correct mRNA polyadenylation. A termination region and/or the enhancer region may be native to the transcriptional initiation region, may be native to, for example, a gene encoding a sequence-specific DNA binding protein, a gene encoding a nuclease, a gene encoding a reverse transcriptase, a gene encoding a deaminase, and the like, or may be native to a host cell, or may be native to another source (e.g., foreign or heterologous to,  
25 for example, to a promoter, to a gene encoding a sequence-specific DNA binding protein, a gene encoding a nuclease, a gene encoding a reverse transcriptase, a gene encoding a deaminase, and the like, or to the host cell, or any combination thereof).

An expression cassette of the invention also can include a polynucleotide encoding a selectable marker, which can be used to select a transformed host cell. As used herein,  
30 "selectable marker" means a polynucleotide sequence that when expressed imparts a distinct phenotype to the host cell expressing the marker and thus allows such transformed cells to be distinguished from those that do not have the marker. Such a polynucleotide sequence may encode either a selectable or screenable marker, depending on whether the marker confers a trait that can be selected for by chemical means, such as by using a selective agent (e.g., an antibiotic

and the like), or on whether the marker is simply a trait that one can identify through observation or testing, such as by screening (e.g., fluorescence). Many examples of suitable selectable markers are known in the art and can be used in the expression cassettes described herein.

In addition to expression cassettes, the nucleic acid molecules/constructs and polynucleotide sequences described herein can be used in connection with vectors. The term "vector" refers to a composition for transferring, delivering, or introducing a nucleic acid (or nucleic acids) into a cell. A vector comprises a nucleic acid construct (e.g. expression cassette(s)) comprising the nucleotide sequence(s) to be transferred, delivered or introduced. Vectors for use in transformation of host organisms are well known in the art. Non-limiting examples of general classes of vectors include viral vectors, plasmid vectors, phage vectors, phagemid vectors, cosmid vectors, fosmid vectors, bacteriophages, artificial chromosomes, minicircles, or Agrobacterium binary vectors in double or single stranded linear or circular form which may or may not be self-transmissible or mobilizable. In some embodiments, a viral vector can include, but is not limited, to a retroviral, lentiviral, adenoviral, adeno-associated, or herpes simplex viral vector. A vector as defined herein can transform a prokaryotic or eukaryotic host either by integration into the cellular genome or exist extrachromosomally (e.g. autonomous replicating plasmid with an origin of replication). Additionally included are shuttle vectors by which is meant a DNA vehicle capable, naturally or by design, of replication in two different host organisms, which may be selected from actinomycetes and related species, bacteria and eukaryotic (e.g. higher plant, mammalian, yeast, or fungal cells). In some embodiments, the nucleic acid in the vector is under the control of, and operably linked to, an appropriate promoter or other regulatory elements for transcription in a host cell. The vector may be a bi-functional expression vector which functions in multiple hosts. In the case of genomic DNA, this may contain its own promoter and/or other regulatory elements and in the case of cDNA this may be under the control of an appropriate promoter and/or other regulatory elements for expression in the host cell. Accordingly, a nucleic acid or polynucleotide of this invention and/or expression cassettes comprising the same may be comprised in vectors as described herein and as known in the art.

As used herein, "contact," "contacting," "contacted," and grammatical variations thereof, refer to placing the components of a desired reaction together under conditions suitable for carrying out the desired reaction (e.g., transformation, transcriptional control, genome editing, nicking, and/or cleavage). As an example, a target nucleic acid may be contacted with a sequence-specific DNA binding protein (e.g., polynucleotide-guided endonuclease, a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein), a zinc finger nuclease, a transcription

activator-like effector nuclease (TALEN) and/or an Argonaute protein)) and a deaminase or a nucleic acid construct encoding the same, under conditions whereby the sequence-specific DNA binding protein, the reverse transcriptase and the deaminase are expressed and the sequence-specific DNA binding protein binds to the target nucleic acid, and the reverse transcriptase and/or deaminase may be fused to either the sequence-specific DNA binding protein or recruited to the sequence-specific DNA binding protein (via, for example, a peptide tag fused to the sequence-specific DNA binding protein and an affinity tag fused to the reverse transcriptase and/or deaminase) and thus, the deaminase and/or reverse transcriptase is positioned in the vicinity of the target nucleic acid, thereby modifying the target nucleic acid. Other methods for recruiting reverse transcriptase and/or deaminase may be used that take advantage of other protein-protein interactions, and also RNA-protein interactions and chemical interactions may be used for protein-protein and protein-nucleic acid recruitment.

As used herein, "modifying" or "modification" in reference to a target nucleic acid includes editing (e.g., mutating), covalent modification, exchanging/substituting nucleic acids/nucleotide bases, deleting, cleaving, nicking, and/or altering transcriptional control of a target nucleic acid. In some embodiments, a modification may include one or more single base changes (SNPs) of any type.

The term "regulating" as used in the context of a transcription factor "regulating" a phenotype, for example, a response to illumination (e.g., a light response, e.g., a shade avoidance response), means the ability of the transcription factor to affect the expression of a gene or genes such that a phenotype, for instance, a response to illumination, is modified.

"Introducing," "introduce," "introduced" (and grammatical variations thereof) in the context of a polynucleotide of interest means presenting a nucleotide sequence of interest (e.g., polynucleotide, RT template, a nucleic acid construct, and/or a guide nucleic acid) to a plant, plant part thereof, or cell thereof, in such a manner that the nucleotide sequence gains access to the interior of a cell.

The terms "transformation" or "transfection" may be used interchangeably and as used herein refer to the introduction of a heterologous nucleic acid into a cell. Transformation of a cell may be stable or transient. Thus, in some embodiments, a host cell or host organism (e.g., a plant) may be stably transformed with a polynucleotide/nucleic acid molecule of the invention. In some embodiments, a host cell or host organism may be transiently transformed with a polynucleotide/nucleic acid molecule of the invention.

"Transient transformation" in the context of a polynucleotide means that a polynucleotide is introduced into the cell and does not integrate into the genome of the cell.



By "stably introducing" or "stably introduced" in the context of a polynucleotide introduced into a cell is intended that the introduced polynucleotide is stably incorporated into the genome of the cell, and thus the cell is stably transformed with the polynucleotide.

"Stable transformation" or "stably transformed" as used herein means that a nucleic acid molecule is introduced into a cell and integrates into the genome of the cell. As such, the integrated nucleic acid molecule is capable of being inherited by the progeny thereof, more particularly, by the progeny of multiple successive generations. "Genome" as used herein includes the nuclear and the plastid genome, and therefore includes integration of the nucleic acid into, for example, the chloroplast or mitochondrial genome. Stable transformation as used herein can also refer to a transgene that is maintained extrachromasomally, for example, as a minichromosome or a plasmid.

Transient transformation may be detected by, for example, an enzyme-linked immunosorbent assay (ELISA) or Western blot, which can detect the presence of a peptide or polypeptide encoded by one or more transgene introduced into an organism. Stable transformation of a cell can be detected by, for example, a Southern blot hybridization assay of genomic DNA of the cell with nucleic acid sequences which specifically hybridize with a nucleotide sequence of a transgene introduced into an organism (e.g., a plant). Stable transformation of a cell can be detected by, for example, a Northern blot hybridization assay of RNA of the cell with nucleic acid sequences which specifically hybridize with a nucleotide sequence of a transgene introduced into a host organism. Stable transformation of a cell can also be detected by, e.g., a polymerase chain reaction (PCR) or other amplification reactions as are well known in the art, employing specific primer sequences that hybridize with target sequence(s) of a transgene, resulting in amplification of the transgene sequence, which can be detected according to standard methods. Transformation can also be detected by direct sequencing and/or hybridization protocols well known in the art.

Accordingly, in some embodiments, nucleotide sequences, polynucleotides, nucleic acid constructs, and/or expression cassettes of the invention may be expressed transiently and/or they can be stably incorporated into the genome of the host organism. Thus, in some embodiments, a nucleic acid construct of the invention (e.g., one or more expression cassettes comprising polynucleotides for editing as described herein) may be transiently introduced into a cell with a guide nucleic acid and as such, no DNA is maintained in the cell.

A nucleic acid construct of the invention may be introduced into a plant cell by any method known to those of skill in the art. Non-limiting examples of transformation methods include transformation via bacterial-mediated nucleic acid delivery (e.g., via *Agrobacteria*),

viral-mediated nucleic acid delivery, silicon carbide or nucleic acid whisker-mediated nucleic acid delivery, liposome mediated nucleic acid delivery, microinjection, microparticle bombardment, calcium-phosphate-mediated transformation, cyclodextrin-mediated transformation, electroporation, nanoparticle-mediated transformation, sonication, infiltration, PEG-mediated nucleic acid uptake, as well as any other electrical, chemical, physical (mechanical) and/or biological mechanism that results in the introduction of nucleic acid into the plant cell, including any combination thereof. Procedures for transforming both eukaryotic and prokaryotic organisms are well known and routine in the art and are described throughout the literature (See, for example, Jiang et al. (2013) *Nat. Biotechnol.* 31:233-239; Ran et al. (2013) *Nature Protocols* 8:2281-2308). General guides to various plant transformation methods known in the art include Miki et al. ("Procedures for Introducing Foreign DNA into Plants" in *Methods in Plant Molecular Biology and Biotechnology*, Glick, B. R. and Thompson, J. E., Eds. (CRC Press, Inc., Boca Raton, 1993), pages 67-88) and Rakowoczy-Trojanowska ((2002) *Cell. Mol. Biol. Lett.* 7:849-858).

In some embodiments of the invention, transformation of a cell may comprise nuclear transformation. In other embodiments, transformation of a cell may comprise plastid transformation (e.g., chloroplast transformation). In still further embodiments, nucleic acids of the invention may be introduced into a cell via conventional breeding techniques. In some embodiments, one or more of the polynucleotides, expression cassettes and/or vectors may be introduced into a plant cell via *Agrobacterium* transformation.

A polynucleotide therefore can be introduced into a plant, plant part, plant cell in any number of ways that are well known in the art. The methods of the invention do not depend on a particular method for introducing one or more nucleotide sequences into a plant, only that they gain access to the interior the cell. Where more than polynucleotide is to be introduced, they can be assembled as part of a single nucleic acid construct, or as separate nucleic acid constructs, and can be located on the same or different nucleic acid constructs. Accordingly, the polynucleotide can be introduced into the cell of interest in a single transformation event, or in separate transformation events, or, alternatively, a polynucleotide can be incorporated into a plant as part of a breeding protocol.

Plants respond to proximal neighboring plants to better compete with neighboring plants for resources, particularly for access to light. While this is an adaptive advantage in a natural or wild environment, in monocrop agriculture, plants are competing with same species crop plants that collectively contribute to yield, so the net gain from an individual plant is lost when averaged over a farm. This response to proximal plants is called Shade Avoidance Response

(SAR), which can result in poor plant vigor and decreased plant yield (Shade Avoidance Syndrome; SAS). As an example, yield (bushels/acre) in corn has increased steadily through high intensity breeding. However, incremental increases in yield have recently started to plateau and require large investments in field evaluation and breeding to demonstrate genetic gain. New approaches to genetic modification are required to deliver significant improvements to yield that are not possible through traditional methods. Crop yields may be improved in two fundamentally distinct ways: 1) yield improvements *per se*, where an engineered plant has gained an advantage such as improved photosynthesis or optimized carbohydrate partitioning or 2) the removal of vestigial survival mechanisms that are not consistent with high production agriculture. Shade avoidance response (SAR) or shade avoidance syndrome (SAS) is such a survival mechanism. SAS/SAR is characterized by increased root/shoot ratio, increased plant height, and reduced individual plant yield and in a typical monoculture crop setting, this response to competition is a wasteful survival mechanism.

The environmental signal that plants use to detect neighboring plants is a shift in the R:FR light ratio, where light rebounding off leaf tissue is shifted toward FR wavelengths relative to light that comes directly to the plant. The phytochrome system is used by plants to detect light wavelength and to signal changes in the plant.

The present invention addresses problems associated with increased planting density and reduction in yield loss (on an acre basis) due to planting variability by using gene editing to modify key regulatory factors that trigger shade avoidance in crops. Plants with such edited genomes will have reduced shade avoidance capability. HY5 encodes a basic leucine zipper (bZIP) transcription factor and has a role in the promotion of photomorphogenesis. HY5 genes are regulators of shade avoidance response through their interaction with Constitutive Photomorphogenic 1 (COP1) polypeptides. The activity of HY5 is negatively regulated by COP1. COP1 contains three conserved domains that are important for protein–protein interaction: an N-terminal Ring-finger domain, a coiled-coil for dimerization and a WD-40 repeat domain for substrate recognition. In darkness, COP1 protein is localized primarily in the nucleus and becomes abundant in the cytosol when exposed to light. COP1 acts as an E3 ligase and targets transcription factors (such as HY5) for degradation via the 26S proteasome in darkness, and thus desensitizes light signaling. COP1 is degraded in the absence of shade. The abundance of HY5 protein inversely correlates with light intensity and is regulated via darkness-dependent degradation, mediated by the 26S proteasome through the interaction with COP1.

The methods of the invention are directed to editing of *HY5* genes so that the effect of the shade response in a plant or part thereof comprising the edited *HY5* gene is reduced or

eliminated, e.g., so that the activity of the encoded HY5 polypeptide is no longer regulated by COP1, e.g., so that regulation of HY5 transcription factor by COP1 is reduced. In some embodiments, a HY5 gene may be edited so as to modify a COP1 binding site in the encoded HY5 transcription factor, thereby reducing or eliminating binding of COP1 to the modified HY5 transcription factor. When plants are in light (e.g., not experiencing far-red light), COP1 is in the cytosol, where it is not interacting with other proteins. When the plant is experiencing far-red light, COP1 is in the nucleus, where it can interact with the HY5 transcription factor, leading to the degradation of the HY5 transcription factor. Without wishing to be limited by any particular theory, a HY5 transcription factor modified as described herein will not be degraded when the plants experience far-red light and as such, the plants will not display the morphological changes associated with shade avoidance. A single copy of a *HY5* gene modified as described herein may be sufficient to reduce or eliminate SAR/SAS, because production of a HY5 transcription factor polypeptide that is modified so as to not be regulated by COP1 should accumulate when plants experience far-red light.

Types of mutations that may be useful for producing plants with a mutation in an endogenous *HY5* gene, resulting in the plants having a reduced/suppressed Shade Avoidance Response, for example, substitutions, deletions, and/or insertions. In some aspects, a mutation in an endogenous *HY5* gene may result in a mutated *HY5* polypeptide having increased activity. In some aspects, a mutation generated by the editing technology can be a point mutation. In some embodiments, a mutation may be a non-natural mutation.

Accordingly, the present invention provides a plant or part thereof comprising at least one mutation (e.g., one or more mutations) in an endogenous *Elongated Hypocotyl5 (HY5)* gene encoding a HY5 transcription factor, wherein the HY5 transcription factor encoded by the endogenous gene is capable of regulating response to illumination in the plant (e.g., shade avoidance response (SAR)). A HY5 transcription factor useful with this invention comprises a Constitutive Photomorphogenic 1 (COP1) binding site. COP1 is a ubiquitin ligase (e.g., an E3 ubiquitin ligase, a RING finger ubiquitin ligase). In some embodiments, the at least one mutation may be in the COP1 binding site of the HY5 transcription factor that is encoded by the *HY5* gene. In some embodiments, the at least one mutation may disrupt the binding of the HY5 transcription factor by a COP1 polypeptide in the plant or part thereof, optionally wherein the at least one mutation results in a HY5 transcription factor having no or reduced COP1 binding. In some embodiments, the binding of COP1 to a HY5 transcription factor that is mutated as described herein may be reduced by about 30% to about 100% (e.g., about 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61,

62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100%, or any range or value therein).

In some embodiments, an endogenous gene encoding a HY5 transcription factor useful with this invention: (a) comprises a sequence having at least 80% sequence identity (e.g., at least  
5 about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO: 73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at  
10 least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**. In some embodiments, the at least one mutation may be in a region of the endogenous *HY5* transcription factor having at least 80% sequence identity to **SEQ ID NO:106** (SSSERSSSSGHHVDMEVKEGMESDDEIRRVPGLLEL), optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%,  
15 optionally the sequence identity may be at least 100%.

In some embodiments, a plant or plant part of the invention comprises at least one mutation (e.g., one or more mutations) in an endogenous *Elongated Hypocotyl5 (HY5)* gene encoding a HY5 transcription factor, optionally wherein the mutation disrupts the binding of the  
20 HY5 transcription factor by COP1 in the plant or part thereof, wherein the endogenous HY5 transcription factor gene (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO: 73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80%  
25 sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%. In some  
30 embodiments, the HY5 transcription factor comprises a COP1 binding domain in a region of the HY5 transcription factor polypeptide, the region having at least about 80% identity to **SEQ ID NO:106**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%.

A mutation in an endogenous *Elongated Hypocotyl5 (HY5)* transcription factor gene in a plant may be any type of mutation including, but not limited to, a base substitution, a base

deletion and/or a base insertion. In some embodiments, mutation in an endogenous *HY5* gene results in a *HY5* transcription factor having disrupted in the binding of a Constitutive Photomorphogenic 1 (COP1) ubiquitin ligase to the *HY5* transcription factor. For example, the mutation may be a substitution, a deletion and/or an insertion of one or more bases of the transcription factor gene or one or more amino acid residues of the *HY5* transcription factor. In some embodiments, the at least one mutation is an out-of-frame deletion or an in-frame deletion. In some embodiments, the at least one mutation is an in-frame deletion. In some embodiments, at least one mutation may comprise a base substitution to an A, a T, a G, or a C, which results in an amino acid substitution, thereby disrupting the binding of the *HY5* transcription factor by a COP1 polypeptide. In some embodiments, a mutation useful with the invention is a non-natural mutation.

In some embodiments, the at least one mutation (e.g., one or more mutations) in an endogenous *HY5* gene may comprise a deletion. In some embodiments, an endogenous *HY5* gene encodes a *HY5* transcription factor polypeptide comprising a COP1 binding domain and the deletion in the *HY5* gene is an in-frame deletion comprising a deletion of at least three consecutive base pairs (e.g., at least 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 95, 100, 105, 110, 115, 120, 125, 130, 135, 140, 145, 150, 160, 170, 180, 190, 200, 210, 220, 230, 240, 250, or 260 or more consecutive base pairs or any range or value therein of a *HY5* transcription factor gene, (e.g., **SEQ ID NO:72**), optionally about 3, 6, 9, 12, 15, 18, 21, 24, 27, 30, 33, 36, 39, 42, 45, 48, 51, 54, 57, 60, 63, 66, 69, 72, 75, 78, 81, 84, 87, 90, 93, 96, 99, 102, 105, 108, 111, 114, 117, 120 or more consecutive base pairs. In some embodiments, a deletion in the endogenous *HY5* transcription factor gene may be a deletion of a portion or of the entire COP1 binding region, optionally wherein the deletion is an in-frame deletion.

In some embodiments, at least one mutation (e.g., one or more mutations) may produce a dominant negative mutation or a dominant mutation. In some embodiments, a mutation in an endogenous *HY5* gene useful with this invention is a dominant negative mutation. In some embodiments, the dominant negative mutation results in a loss of the COP1 binding to the *HY5* transcription factor, thereby reducing the function of the encoded *HY5* transcription factor.

In some embodiments, a plant comprising a mutation in an *HY5* gene as described herein may be planted at an increased density without a decrease in plant yield on a per plant basis as compared to a control, optionally wherein the planting density is increased by about 5% to about 75% (e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53,

54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100%, or any range or value therein) over a standard planting density, without a decrease in plant yield on a per plant basis. For comparative purposes, a standard planting density, for example, for corn, can be about 25,000 plants per acre to about 35,000 plants per acre, while a standard planting density, for example, for soybean, can be in a range of about 100,000 plants per acre to about 125,000 plants per acre. As understood in the art, a standard planting density will vary by at least the crop species. In some embodiments, a plant comprising a mutation in an *HY5* gene may exhibit increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity.

"Close proximity" refers to a high planting density for any particular plant species that can result in SAR. For example, in some embodiments, "close proximity" includes a density of plants resulting from planting seeds of the plant about 6.1 inches or less apart (e.g., about 6.1, 6, 5.9, 5.8, 5.7, 5.6, 5.5, 5.4, 5.3, 5.2, 5.2, 5.1, 5, 4.9, 4.8, 4.7, 4.6, 4.5, 4.4, 4.3, 4.2, 4.1, 4.3.9, 3.8, 3.7, 3.6, 3.5, 3.4, 3.3, 3.2, 3.1, 3, 2.9, 2.8, 2.7, 2.6, 2.5, 2.4, 2.3, 2.2, 2.1, 2, 1.9, 1.8, 1.7, 1.6, 1.5, 1.4, 1.3, 1.2, 1.1, 1, 0.9, 0.8, 0.7, 0.6, 0.5, inches apart, and the like or any range or value therein). As would be understood by one of skill in the art, the number of seeds planted per acre to achieve a high density planting will vary by plant species. As an example, a high-density planting for corn includes more than 35,000 seeds per acre at 30 inches or more row spacing.

In some embodiments, a mutation may result in a mutated *HY5* gene having at least 90% sequence identity to a nucleotide sequence of any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encoding a mutated *HY5* polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

In some embodiments, a plant cell comprising an editing system is provided, the editing system comprising: (a) a CRISPR-Cas associated effector protein; and (c) a guide nucleic acid (gRNA, gDNA, crRNA, crDNA) having a spacer sequence with complementarity to an endogenous target gene encoding an Elongated Hypocotyl5 (*HY5*) transcription factor. The endogenous *HY5* transcription factor may be any *HY5* transcription factor involved in the shade avoidance response. In some embodiments, an endogenous *HY5* gene encodes a basic leucine

zipper (bZIP) transcription factor (e.g., a HY5 transcription factor). In some embodiments, the *HY5* transcription factor gene to which a spacer sequence of the guide nucleic acid shares complementarity (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%. In some embodiments, a spacer sequence useful with this invention can include, but is not limited to, a nucleotide sequence of any one of **SEQ ID NOs:107-113**, or reverse complement thereof, or a combination thereof.

In some embodiments, a plant cell is provided that comprises at least one mutation in an endogenous *Elongated Hypocotyl5* (*HY5*) gene, wherein the mutation is a substitution, an insertion and/or a deletion that is introduced into the endogenous *HY5* gene using an editing system that comprises a nucleic acid binding domain that binds to a target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene: (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%. In some embodiments, the nucleic acid binding domain of the editing system may be from a polynucleotide-guided endonuclease, a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein), a zinc finger nuclease, a transcription activator-like effector nuclease (TALEN) and/or an Argonaute protein.

In some embodiments, a deletion or insertion generated in an endogenous *HY5* gene may be a deletion or insertion of 1 base pair to about 100 base pairs (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 30, 31, 32, 33, 34, 35,



36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100 base pairs, or any value or range therein) or more. In some embodiments, the deletion may be an in-frame deletion or an out-of-frame  
5 deletion. In some embodiments, a mutation may be a deletion in the region of the HY5 gene that encodes the COP1 binding site, optionally wherein the region may encode a sequence having at least 80% sequence identity (optionally the sequence identity may be at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%) to the amino acid sequence of **SEQ ID NO:106**. In some embodiments, the mutation may be a deletion of all or a  
10 portion of a COP1 binding domain of the HY5 transcription factor. In some embodiments, the mutation can be a base substitution to an A, a T, a G, or a C, optionally wherein the base substitution results in an amino acid substitution. In some embodiments, the mutation is a non-natural mutation.

A plant or plant part useful with this invention may be a dicot or a monocot. Non-  
15 limiting examples of a plant or part thereof useful with this invention include, but are not limited to, corn, soy, canola, wheat, rice, cotton, sugarcane, sugar beet, barley, oats, alfalfa, sunflower, safflower, oil palm, sesame, coconut, tobacco, potato, sweet potato, cassava, coffee, apple, plum, apricot, peach, cherry, pear, fig, banana, citrus, cocoa, avocado, olive, almond, walnut, strawberry, watermelon, pepper, grape, tomato, cucumber, blackberry, raspberry, black  
20 raspberry or a *Brassica* spp. In some embodiments, the plant part may be a cell from a plant that includes, but is not limited to, corn, soy, canola, wheat, rice, cotton, sugarcane, sugar beet, barley, oats, alfalfa, sunflower, safflower, oil palm, sesame, coconut, tobacco, potato, sweet potato, cassava, coffee, apple, plum, apricot, peach, cherry, pear, fig, banana, citrus, cocoa, avocado, olive, almond, walnut, strawberry, watermelon, pepper, grape, tomato, cucumber,  
25 blackberry, raspberry, black raspberry or a *Brassica* spp. In some embodiments, a plant or part thereof may be corn. In some embodiments, a plant may be regenerated from a plant cell or plant part of this invention. In some aspects, a plant cell can be non-propagating plant cell that does not regenerate into a plant. Plants of this invention comprising at least one mutation in a *HY5* transcription factor gene may comprise an attenuated shade avoidance response (SAR) as  
30 compared to a plant that is devoid of the at least one mutation. In some embodiments, a plant comprising at least one mutation in mutation in a *HY5* gene may exhibit increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or

enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity.

Also provided herein is a method of providing a plurality of plants having an increase in yield (e.g., an increase in floret fertility, an increase in seed number, and/or an increase in seed weight) when each plant of the plurality of plants is planted in close proximity to one another, the method comprising planting two or more plants of the invention (e.g., 2, 3, 4, 5, 6, 7, 8, 9, 10, 20, 30, 40, 50, 100, 200, 300, 400, 500, 1000, 2000, 3000, 400, 5000, or 10,000 or more plants of the invention) (e.g., comprising a mutation in a *HY5* transcription factor gene and having an attenuated Shade Avoidance Response, optionally exhibiting increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity) in a growing area, thereby providing a plurality of plants having increased yield as compared to a plurality of control plants not comprising the mutation (e.g., as compared to an isogenic wild type plant not comprising the mutation). A growing area can be any area in which a plurality of plants can be planted together, including, but not limited to, a field (e.g., a cultivated field, an agricultural field), a growth chamber, a greenhouse, a recreational area, a lawn, and/or a roadside, and the like.

In some embodiments, a method of producing/breeding a transgene-free edited plant is provided, the method comprising: crossing a plant of the present invention (e.g., a plant comprising a mutation in a *HY5* transcription factor gene as described herein (and having an attenuated Shade Avoidance Response and optionally exhibiting increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity) with a transgene-free plant, thereby introducing the at least one mutation (e.g., one or more mutations) into the plant that is transgene-free (e.g., into progeny plants); and selecting a progeny plant that comprises the at least one mutation and is transgene-free, thereby producing a transgene-free edited (e.g., base edited) plant.

In some embodiments, the present invention provides a method of creating a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene in a plant, comprising: (a) targeting a gene

editing system to a portion of the *HY5* gene that comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally the sequence identity may be at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%) to **SEQ ID NOs:75-105**; and (b) selecting a plant that comprises a modification located in a region of the *HY5* gene having at least 80% sequence identity to any one of **SEQ ID NOs:75-105**. In some embodiments, a plant having a mutation in an endogenous *HY5* gene exhibits increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity.

In some embodiments, a method of generating variation in an *Elongated Hypocotyl5* (*HY5*) gene is provided, the method comprising: introducing an editing system into a plant cell, wherein the editing system is targeted to a region of a *HY5* gene that encodes a *HY5* transcription factor and contacting the region of the *HY5* gene with the editing system, thereby introducing a mutation into the *HY5* gene and generating variation in the *HY5* gene of the plant cell. In some embodiments, a *HY5* gene into which variation is generated comprises a nucleotide sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to any one of **SEQ ID NO:72** or **SEQ ID NO:73**, and/or encodes an amino acid sequence having at least 80% sequence identity to **SEQ ID NO:74**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%. In some embodiments, a region of the *HY5* gene that may be targeted comprises at least 80% sequence identity to a nucleotide sequence of any one of **SEQ ID NOs:75-105**, optionally where the region of the *HY5* gene that may be targeted encodes an amino acid sequence having at least 80% sequence identity to **SEQ ID NO:106**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%. In some embodiments, contacting a region of an endogenous *HY5* gene in a plant cell with the editing system produces a plant cell comprising in its genome an edited endogenous *HY5* gene. In some embodiments, the method may further comprise (a) regenerating a plant from the plant cell; (b) selfing the plant to produce progeny plants (E1); (c) assaying the progeny plants of (b) for reduced shade avoidance response (SAR)/shade avoidance syndrome (SAS); and (d) selecting the progeny plants exhibiting reduced shade avoidance response

(SAR)/shade avoidance syndrome (SAS) as compared to a control plant devoid of the mutation. In some embodiments, the method may further comprise (e) selfing the selected progeny plants of (d) to produce progeny plants (E2); (f) assaying the progeny plants of (e) for reduced shade avoidance response (SAR)/shade avoidance syndrome (SAS); and (g) selecting the progeny plants exhibiting reduced shade avoidance response (SAR)/shade avoidance syndrome (SAS) as compared to a control plant, optionally repeating (e) through (g) one or more additional times.

In some embodiments, a method of detecting a mutant *HY5* transcription factor gene (a mutation in an endogenous *HY5* transcription factor gene) is provided, the method comprising detecting in the genome of a plant a mutation in any one of the nucleotide sequences of, for example, **SEQ ID NOs:72** or **SEQ ID NO:73**, optionally wherein the mutation is a deletion of at least one nucleotide (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more), optionally an in-frame deletion. In some embodiments, a method of detecting a mutant *HY5* gene (a mutation in an endogenous *HY5* gene) in a plant or plant part (e.g., plant cell) is provided, the method comprising detecting in the genome of the plant a *HY5* gene having at least one mutation within a region having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally the sequence identity may be at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%) to a nucleotide sequence of any one of **SEQ ID NOs:75-105**, optionally wherein the mutation is a deletion or an insertion of at least one nucleotide (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, or more).

In some embodiments, a method of detecting a mutation in an endogenous *HY5* gene is provided, comprising detecting in the genome of a plant a mutated *HY5* gene. In some embodiments, the mutated *HY5* gene comprises a sequence having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated *HY5* polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

In some embodiments, a method for editing a specific site in the genome of a plant cell is provided, the method comprising cleaving, in a site-specific manner, a target site within an endogenous *Elongated Hypocotyl5 (HY5)* gene in the plant cell, the endogenous *HY5* gene: (a) comprising a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encoding a region having at least 80%

sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, thereby generating an edit in the endogenous *HY5* gene of the plant cell. In some embodiments, the *HY5* gene encodes a *HY5* transcription factor that comprises a COP1 binding region and the edit results in a mutation in the COP1 binding region of the *HY5* transcription factor. In some embodiments, the edit may result in a *HY5* transcription factor having no or reduced COP1 binding, optionally wherein the binding of the COP1 polypeptide to the *HY5* transcription factor may be reduced by about 30% to about 100% (e.g., about 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, or 100%). In some embodiments, the edit may be located in a region of the *HY5* transcription factor having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally at least 90% or at least 95%, optionally or at least 100%) to **SEQ ID NO:106**.

In some embodiments, an edit in an endogenous *HY5* gene may result in an endogenous *HY5* gene having at least 90% (e.g., 90, 91, 92, 93, 94, 95, 96, 97, 98, 99 or 100%, optionally the sequence identity may be at least 95%, optionally the sequence identity may be at least 100%) sequence identity to any one of **SEQ ID NOS:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated *HY5* polypeptide having at least 90% sequence identity to any one of **SEQ ID NOS:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

In some embodiments, a plant may be regenerated from the plant cell comprising the edit in the endogenous *HY5* transcription factor gene to produce a plant comprising the edit in its endogenous *HY5* transcription factor gene. In some embodiments, a plant is not regenerated from a plant cell. In some embodiments, a plant comprising an edit in its endogenous *HY5* transcription factor gene exhibits an attenuated Shade Avoidance Response compared to a control plant that does not comprise the edit. In some embodiments, a plant comprising an edit in an endogenous *HY5* gene exhibits increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants as compared to a control plant planted in close proximity.

A plant comprising an endogenous *HY5* transcription factor gene that is edited as described herein to provide a *HY5* transcription factor with reduced COP1 binding may exhibit an attenuated Shade Avoidance Response (optionally also exhibiting increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores) when compared to a control plant that does not comprise the edited endogenous *HY5* transcription factor gene. A plant comprising an edited endogenous *HY5* transcription factor gene as described herein may be compared to a plant that is not so edited when grown under the same environmental conditions, e.g., an environment with a low R:FR light ratio, e.g., shaded conditions (e.g., an R:FR ratio of about 0.16; or a range of an R:FR ratio of about 0.09 to about 0.7 (e.g., about 0.09, 0.10, 0.11, 0.12, 0.13, 0.14, 0.15, 0.16, 0.17, 0.18, 0.19, 0.2, 0.21, 0.23, 0.24, 0.25, to about 0.26, 0.27, 0.28, 0.29, 0.3, 0.35, 0.4, 0.45, 0.5, 0.55, 0.6, 0.65, 0.7, or any range or value therein))

In some embodiments, a method for making a plant is provided, the method comprising:

- (a) contacting a population of plant cells that comprise an endogenous gene encoding an Elongated Hypocotyl5 (*HY5*) transcription factor with a nuclease targeted to the endogenous gene, wherein the nuclease is linked to a nucleic acid binding domain that binds to a target site within the endogenous gene, the endogenous gene (i) comprising a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (ii) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105** optionally 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**; (iii) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (iv) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (i), (ii), (iii) and/or (iv) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%;
- (b) selecting a plant cell from the population of plant cells in which the endogenous gene encoding a *HY5* transcription factor has been mutated, thereby producing a plant cell comprising a mutation in the endogenous gene; and
- (c) growing the selected plant cell into a plant comprising the mutation in the endogenous gene encoding a *HY5* transcription factor, optionally wherein the mutation reduces or eliminates the

ability of the HY5 transcription factor to bind a Constitutive Photomorphogenic 1 (COP1) polypeptide.

In some embodiments, a method for reducing/suppressing a Shade Avoidance Response in a plant is provided, the method comprising (a) contacting a plant cell comprising an  
5 endogenous *Elongated Hypocotyl5* (*HY5*) gene with a nuclease targeted to the endogenous gene, wherein the nuclease is linked to a nucleic acid binding domain that binds to a target site within the endogenous gene, the endogenous gene: (i) comprising a sequence having at least 80%  
10 sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (ii) comprising a region having at least 80% sequence identity to any one of the  
15 nucleotide sequences of **SEQ ID NOs:75-105**, optionally 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**; (iii) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid  
20 sequence of **SEQ ID NO:74**; and/or (iv) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, thereby producing a plant cell comprising a mutation in the endogenous *HY5* gene; and (b) growing the plant cell into a plant, thereby  
25 reducing/suppressing the Shade Avoidance Response in the plant, optionally increasing yield, increasing upright growth, decreasing height, decreasing shoot:root ratio, decreasing leaf length, increasing mechanical strength of stems, reducing lodging rate, delaying senescence, substantially no change in flowering time, increasing photosynthesis efficiency and grain filling, and/or enhancing defense responses against pathogens and herbivores when planted in close  
30 proximity with one or more plants as compared to a control plant planted in close proximity.

In some embodiments, a method for producing a plant or part thereof comprising at least one cell (e.g., one or more cells) having a mutation in an endogenous *Elongated Hypocotyl5* (*HY5*) gene, the method comprising contacting a target site within the endogenous *HY5* gene in the plant or plant part with a nuclease comprising a cleavage domain and a nucleic acid binding domain, wherein the nucleic acid binding domain of the nuclease binds to a target site within the  
35 endogenous *HY5* gene, wherein the endogenous *HY5* gene: (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a

sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, thereby producing a plant or part thereof comprising at least one cell having a mutation in the endogenous *HY5* gene. In some embodiments, the at least one cell in the plant or part thereof having a mutated endogenous *HY5* transcription factor gene produces a HY5 transcription factor having reduced binding of COP1.

In some embodiments, a method is provided for producing a plant or part thereof comprising a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene that produces in a mutated HY5 transcription factor having reduced binding of a Constitutive Photomorphogenic 1 polypeptide, the method comprising contacting a target site within an endogenous *HY5* gene in the plant or part thereof with a nuclease comprising a cleavage domain and a nucleic acid binding domain, wherein the nucleic acid binding domain of the nuclease binds to the target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene: (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**, optionally 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, thereby producing a plant or part thereof having a mutation in the endogenous *HY5* gene that produces a mutated HY5 transcription factor having reduced binding by the COP1.

In some embodiments, a target site may be a region or within a region of the *HY5* gene having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally the sequence identity may be at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%) to a nucleotide sequence of any one of **SEQ ID NOs:75-105**, optionally at least 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**.



In some embodiments, the nuclease may be a zinc finger nuclease, transcription activator-like effector nucleases (TALEN), endonuclease (e.g., Fok1) or a CRISPR-Cas effector protein, wherein the nuclease cleaves the endogenous *HY5* gene and a mutation is introduced into the *HY5* gene.

5 In some embodiments, a mutation may be a base substitution, a base deletion and/or a base insertion, optionally a non-natural mutation. In some embodiments, a mutation is a base substitution to an A, a T, a G, or a C. In some embodiments, the mutation in the *HY5* gene may be a deletion or an insertion. In some embodiments, a mutation may be located in the region of the *HY5* gene that encodes the COP1 binding site, optionally wherein the region may encode a  
10 sequence having at least 80% sequence identity (optionally the sequence identity may be at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%) to the amino acid sequence of **SEQ ID NO:106**. In some embodiments, the mutation results in a mutated *HY5* transcription factor having modified activity. In some embodiments, the mutation may reduce or eliminate the ability of the *HY5* transcription factor to bind a Constitutive  
15 Photomorphogenic 1 (COP1) polypeptide. In some embodiments, a mutation in an endogenous *HY5* gene may be dominant mutation and/or a dominant negative mutation.

In some embodiments, the plant or part thereof that is produced by the methods of this invention comprises a mutated endogenous *HY5* transcription factor gene as described herein and exhibits an attenuated/reduced shade avoidance response as compared to a control plant that  
20 does not comprise the mutation in the endogenous *HY5* transcription factor gene, e.g., the plant or plant part has not had a target site within its endogenous *HY5* transcription factor gene contacted with an editing system or a nuclease comprising a cleavage domain and a nucleic acid binding domain (e.g., a DNA binding domain). In some embodiments, the comparison with a control plant may be made between the edited plant and control plant when grown under the  
25 same environmental conditions; e.g., a shaded environment, e.g., a low R:FR ratio environment.

A plant that comprises a mutated endogenous *HY5* transcription factor as described herein and exhibits an attenuated/reduced shade avoidance response further exhibits one or more phenotypes that can include, but are not limited to, increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length; increased mechanical  
30 strength of stems; reduced lodging rate; delayed senescence; increased photosynthesis efficiency and grain filling; substantially no change in flowering time, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more other plants as compared to a plant that does not comprise the mutated endogenous *HY5* transcription factor as described herein that is planted in close proximity with one or more other plants. In some

embodiments, a plant having a reduced SAR is at least about 5% shorter than the control plant grown under the same environmental conditions (e.g., a shaded environment, e.g., a low R:FR ratio environment) (e.g., about 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 110, 120, 130, 140, 150%, or more shorter, or any range or value therein).

In some embodiments, the methods of the invention produce plants or parts thereof having a mutated *HY5* gene having at least 90% sequence identity (e.g., at least about 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally the sequence identity may be at least 95%, optionally the sequence identity may be at least 100%) to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encoding a mutated *HY5* polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

In some embodiments, a nuclease contacting a plant cell, a population of plant cells and/or a target site cleaves an endogenous *HY5* transcription factor gene, thereby introducing a mutation into the DNA binding site of the endogenous *HY5* transcription factor gene. A nuclease useful with the invention may be any nuclease that can be utilized to edit/modify a target nucleic acid. Such nucleases include, but are not limited to, a zinc finger nuclease, transcription activator-like effector nucleases (TALEN), endonuclease (e.g., FokI) and/or a CRISPR-Cas effector protein. Likewise, any nucleic acid binding domain (e.g., DNA binding domain) useful with the nuclease of the invention may be any nucleic acid binding domain that can be utilized to edit/modify a target nucleic acid. Such a nucleic acid binding domain includes, but is not limited to, a zinc finger, transcription activator-like DNA binding domain (TAL), an argonaute and/or a CRISPR-Cas effector DNA binding domain.

In some embodiments a method is provided for modifying an endogenous *Elongated Hypocotyl5 (HY5)* gene in a corn plant or part thereof for attenuating or reducing a shade avoidance response in the corn plant or part thereof, the method comprising modifying a target site within the endogenous *HY5* gene in the corn plant or a part thereof, wherein the endogenous *HY5* gene, (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence

identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, thereby modifying the endogenous *HY5* gene and attenuating or reducing a shade avoidance response in the corn plant or part thereof. In some embodiments, the target site is a region of the *HY5* gene having at least 80% sequence identity to a nucleotide sequence of any one of **SEQ ID NOs:75-105**.

In some embodiments, a method of editing an endogenous *HY5* transcription factor gene in a plant or plant part is provided, the method comprising contacting a target site within the *HY5* transcription factor gene in the plant or plant part with a cytosine base editing system comprising a cytosine deaminase and a nucleic acid binding domain that binds to a target site within the *HY5* transcription factor gene, wherein the *HY5* transcription factor gene (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, thereby producing the plant or part thereof comprising an endogenous *HY5* transcription factor gene having a mutation. In some embodiments, the mutation reduces the COP1 binding to the *HY5* transcription factor. In some embodiments, a plant comprising the endogenous *HY5* transcription factor gene having a mutation exhibits a reduced shade avoidance response (SAR) (e.g., increased yield when planted in close proximity to other plants).

In some embodiments, a method of editing an endogenous *HY5* transcription factor gene in a plant or plant part is provided, the method comprising contacting a target site within the *HY5* transcription factor gene in the plant or plant part with an adenosine base editing system comprising an adenosine deaminase and a nucleic acid binding domain that binds to a target site within the *HY5* transcription factor gene, wherein the *HY5* transcription factor gene (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at

least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the  
5 sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, thereby producing the plant or part thereof comprising an endogenous *HY5* transcription factor gene having a mutation. In some embodiments, the mutation reduces the COP1 binding to the *HY5* transcription factor. In some embodiments, a plant comprising the endogenous *HY5* transcription factor gene having a  
10 mutation exhibits a reduced shade avoidance response (SAR) (e.g., increased yield when planted in close proximity to other plants).

In some embodiments, the present invention provides a method of producing a plant comprising a mutation in an endogenous *HY5* transcription factor gene and at least one polynucleotide of interest, the method comprising crossing a plant of the invention comprising at  
15 least one mutation in an endogenous *HY5* transcription factor gene (a first plant) with a second plant that comprises the at least one polynucleotide of interest to produce progeny plants; and selecting progeny plants comprising at least one mutation in the *HY5* transcription factor gene and the at least one polynucleotide of interest, thereby producing the plant comprising a mutation in an endogenous *HY5* transcription factor gene and at least one polynucleotide of  
20 interest.

Further provided is a method of producing a plant comprising a mutation in an endogenous *HY5* transcription factor gene and at least one polynucleotide of interest, the method comprising introducing at least one polynucleotide of interest into a plant of the present invention comprising at least one mutation in a *HY5* transcription factor gene, thereby producing  
25 a plant comprising at least one mutation in a *HY5* transcription factor gene and at least one polynucleotide of interest.

Additionally provided is a method of producing a plant comprising a mutation in an endogenous *HY5* transcription factor gene and exhibiting a phenotype of improved yield traits, improved plant architecture and/or improved defense traits, comprising crossing a first plant,  
30 which is the plant of the present invention (e.g., comprising at least one mutation in an endogenous *HY5* transcription factor gene), with a second plant that exhibits a phenotype of improved yield traits, improved plant architecture and/or improved defense traits; and selecting progeny plants comprising the mutation in the *HY5* transcription factor gene and a phenotype of improved yield traits, improved plant architecture and/or improved defense traits, thereby

producing the plant comprising a mutation in an endogenous *HY5* transcription factor gene and exhibiting a phenotype of improved yield traits, improved plant architecture and/or improved defense traits as compared to a control plant.

5 In some embodiments, the invention provides a method of producing a plant comprising a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene and at least one polynucleotide of interest, the method comprising crossing a first plant, which is the plant of the present invention (e.g., comprising at least one mutation in an endogenous *HY5* gene), with a second plant that comprises the at least one polynucleotide of interest to produce progeny plants; and selecting progeny plants comprising the mutation in the *HY5* transcription factor gene and the at  
10 least one polynucleotide of interest, thereby producing the plant comprising a mutation in an endogenous *HY5* gene and at least one polynucleotide of interest.

Further provided is a method of controlling weeds in a container (e.g., pot, or seed tray and the like), a growth chamber, a greenhouse, a field, a recreational area, a lawn, or on a roadside, the method comprising applying an herbicide to one or more (a plurality) plants of the  
15 present invention (e.g., comprising at least one mutation in an endogenous *HY5* transcription factor gene) growing in a container, a growth chamber, a greenhouse, a field, a recreational area, a lawn, or on a roadside, thereby controlling the weeds in the container, the growth chamber, the greenhouse, the field, the recreational area, the lawn, or on the roadside in which the one or more plants are growing.

20 In some embodiments, a method of reducing insect predation on a plant is provided, the method comprising applying an insecticide to one or more plants of the invention (e.g., comprising at least one mutation in an endogenous *HY5* transcription factor gene), thereby reducing insect predation on the one or more plants.

In some embodiments, a method of reducing fungal disease on a plant is provided, the  
25 method comprising applying a fungicide to one or more plants of the invention (e.g., comprising at least one mutation in an endogenous *HY5* transcription factor gene), thereby reducing fungal disease on the one or more plants, optionally wherein the one or more plants are growing in a container, a growth chamber, a greenhouse, a field, a recreational area, a lawn, or on a roadside.

In some embodiments, a method of reducing bacterial disease on a plant is provided, the  
30 method comprising applying a bactericide to one or more plants of the invention (e.g., comprising at least one mutation in an endogenous *HY5* transcription factor gene), thereby reducing bacterial disease on the one or more plants, optionally wherein the one or more plants are growing in a container, a growth chamber, a greenhouse, a field, a recreational area, a lawn, or on a roadside.

A polynucleotide of interest may be any polynucleotide that can confer a desirable phenotype or otherwise modify the phenotype or genotype of a plant. In some embodiments, a polynucleotide of interest may include, but is not limited to, a polynucleotide that confers herbicide tolerance, insect resistance, nematode resistance, disease resistance, increased yield, increased nutrient use efficiency and/or abiotic stress resistance.

Thus, plants or plant cultivars which are to be treated with preference in accordance with the invention include all plants which, through genetic modification, received genetic material which imparts particular advantageous useful properties ("traits") to these plants. Examples of such properties are better plant growth, vigor, stress tolerance, standability, lodging resistance, nutrient uptake, plant nutrition, and/or yield, in particular improved growth, increased tolerance to high or low temperatures, increased tolerance to drought or to levels of water or soil salinity, enhanced flowering performance, easier harvesting, accelerated ripening, higher yields, higher quality and/or a higher nutritional value of the harvested products, better storage life and/or processability of the harvested products.

Further and particularly emphasized examples of such properties are an increased resistance against animal and microbial pests, such as against insects, arachnids, nematodes, mites, slugs, and snails owing, for example, to toxins formed in the plants. Among DNA sequences encoding proteins which confer properties of tolerance to such animal and microbial pests, in particular insects, mention will particularly be made of the genetic material from *Bacillus thuringiensis* encoding the Bt proteins widely described in the literature and well known to those skilled in the art. Mention will also be made of proteins extracted from bacteria such as *Photorhabdus* (WO97/17432 and WO98/08932). In particular, mention will be made of the Bt Cry or VIP proteins which include the CryIA, CryIAb, CryIAc, CryIIA, CryIIIA, CryIIIB2, Cry9c Cry2Ab, Cry3Bb and CryIF proteins or toxic fragments thereof and also hybrids or combinations thereof, especially the CryIF protein or hybrids derived from a CryIF protein (e.g. hybrid CryIA-CryIF proteins or toxic fragments thereof), the CryIA-type proteins or toxic fragments thereof, preferably the CryIAc protein or hybrids derived from the CryIAc protein (e.g. hybrid CryIAb-CryIAc proteins) or the CryIAb or Bt2 protein or toxic fragments thereof, the Cry2Ae, Cry2Af or Cry2Ag proteins or toxic fragments thereof, the CryIA.105 protein or a toxic fragment thereof, the VIP3Aa19 protein, the VIP3Aa20 protein, the VIP3A proteins produced in the COT202 or COT203 cotton events, the VIP3Aa protein or a toxic fragment thereof as described in Estruch et al. (1996), Proc Natl Acad Sci US A. 28;93(11):5389-94, the Cry proteins as described in WO2001/47952, the insecticidal proteins from *Xenorhabdus* (as described in WO98/50427),

*Serratia* (particularly from *S. entomophila*) or *Photorhabdus* species strains, such as Tc-proteins from *Photorhabdus* as described in WO98/08932. Also any variants or mutants of any one of these proteins differing in some amino acids (1-10, preferably 1-5) from any of the above named sequences, particularly the sequence of their toxic fragment, or which are fused to a transit peptide, such as a plastid transit peptide, or another protein or peptide, is included herein.

Another and particularly emphasized example of such properties is conferred tolerance to one or more herbicides, for example imidazolinones, sulphonylureas, glyphosate or phosphinothricin. Among DNA sequences encoding proteins (i.e., polynucleotides of interest) which confer properties of tolerance to certain herbicides on the transformed plant cells and plants, mention will be particularly be made to the bar or PAT gene or the *Streptomyces coelicolor* gene described in WO2009/152359 which confers tolerance to glufosinate herbicides, a gene encoding a suitable EPSPS (5-Enolpyruvylshikimat-3-phosphat-Synthase) which confers tolerance to herbicides having EPSPS as a target, especially herbicides such as glyphosate and its salts, a gene encoding glyphosate-n-acetyltransferase, or a gene encoding glyphosate oxidoreductase. Further suitable herbicide tolerance traits include at least one ALS (acetolactate synthase) inhibitor (e.g. WO2007/024782), a mutated Arabidopsis ALS/AHAS gene (e.g. U.S. Patent 6,855,533), genes encoding 2,4-D-monooxygenases conferring tolerance to 2,4-D (2,4-dichlorophenoxyacetic acid) and genes encoding Dicamba monooxygenases conferring tolerance to dicamba (3,6-dichloro-2- methoxybenzoic acid).

Further examples of such properties are increased resistance against phytopathogenic fungi, bacteria and/or viruses owing, for example, to systemic acquired resistance (SAR), systemin, phytoalexins, elicitors and also resistance genes and correspondingly expressed proteins and toxins.

Particularly useful transgenic events in transgenic plants or plant cultivars which can be treated with preference in accordance with the invention include Event 531/ PV-GHBK04 (cotton, insect control, described in WO2002/040677), Event 1143-14A (cotton, insect control, not deposited, described in WO2006/128569); Event 1143-51B (cotton, insect control, not deposited, described in WO2006/128570); Event 1445 (cotton, herbicide tolerance, not deposited, described in US-A 2002-120964 or WO2002/034946); Event 17053 (rice, herbicide tolerance, deposited as PTA-9843, described in WO2010/117737); Event 17314 (rice, herbicide tolerance, deposited as PTA-9844, described in WO2010/117735); Event 281-24-236 (cotton, insect control - herbicide tolerance, deposited as PTA-6233, described in WO2005/103266 or US-A 2005-216969); Event 3006-210-23

(cotton, insect control - herbicide tolerance, deposited as PTA-6233, described in US-A 2007-143876 or WO2005/103266); Event 3272 (corn, quality trait, deposited as PTA-9972, described in WO2006/098952 or US-A 2006-230473); Event 33391 (wheat, herbicide tolerance, deposited as PTA-2347, described in WO2002/027004), Event 40416 (corn, insect control - herbicide tolerance, deposited as ATCC PTA-11508, described in WO 11/075593); Event 43A47 (corn, insect control - herbicide tolerance, deposited as ATCC PTA-11509, described in WO2011/075595); Event 5307 (corn, insect control, deposited as ATCC PTA-9561, described in WO2010/077816); Event ASR-368 (bent grass, herbicide tolerance, deposited as ATCC PTA-4816, described in US-A 2006-162007 or WO2004/053062); Event B16 (corn, herbicide tolerance, not deposited, described in US-A 2003-126634); Event BPS-CV127- 9 (soybean, herbicide tolerance, deposited as NCIMB No. 41603, described in WO2010/080829); Event BLRI (oilseed rape, restoration of male sterility, deposited as NCIMB 41193, described in WO2005/074671), Event CE43-67B (cotton, insect control, deposited as DSM ACC2724, described in US-A 2009-217423 or WO2006/128573); Event CE44-69D (cotton, insect control, not deposited, described in US-A 2010- 0024077); Event CE44-69D (cotton, insect control, not deposited, described in WO2006/128571); Event CE46-02A (cotton, insect control, not deposited, described in WO2006/128572); Event COT102 (cotton, insect control, not deposited, described in US-A 2006-130175 or WO2004/039986); Event COT202 (cotton, insect control, not deposited, described in US-A 2007-067868 or WO2005/054479); Event COT203 (cotton, insect control, not deposited, described in WO2005/054480); ); Event DAS21606-3 / 1606 (soybean, herbicide tolerance, deposited as PTA-11028, described in WO2012/033794), Event DAS40278 (corn, herbicide tolerance, deposited as ATCC PTA-10244, described in WO2011/022469); Event DAS-44406-6 / pDAB8264.44.06.1 (soybean, herbicide tolerance, deposited as PTA-11336, described in WO2012/075426), Event DAS-14536-7 /pDAB8291.45.36.2 (soybean, herbicide tolerance, deposited as PTA-11335, described in WO2012/075429), Event DAS-59122-7 (corn, insect control - herbicide tolerance, deposited as ATCC PTA 11384, described in US-A 2006-070139); Event DAS-59132 (corn, insect control - herbicide tolerance, not deposited, described in WO2009/100188); Event DAS68416 (soybean, herbicide tolerance, deposited as ATCC PTA-10442, described in WO2011/066384 or WO2011/066360); Event DP-098140-6 (corn, herbicide tolerance, deposited as ATCC PTA-8296, described in US-A 2009- 137395 or WO 08/112019); Event DP-305423-1 (soybean, quality trait, not deposited, described in US-A 2008-312082 or WO2008/054747); Event DP-32138-1 (corn, hybridization system, deposited as ATCC



PTA-9158, described in US-A 2009-0210970 or WO2009/103049); Event DP-356043-5 (soybean, herbicide tolerance, deposited as ATCC PTA-8287, described in US-A 2010-0184079 or WO2008/002872); Event EE-I (brinjal, insect control, not deposited, described in WO 07/091277); Event Fil 17 (corn, herbicide tolerance, deposited as ATCC 209031, described in US-A 2006-059581 or WO 98/044140); Event FG72 (soybean, herbicide tolerance, deposited as PTA-11041, described in WO2011/063413), Event GA21 (corn, herbicide tolerance, deposited as ATCC 209033, described in US-A 2005-086719 or WO 98/044140); Event GG25 (corn, herbicide tolerance, deposited as ATCC 209032, described in US-A 2005-188434 or WO98/044140); Event GHB119 (cotton, insect control - herbicide tolerance, deposited as ATCC PTA-8398, described in WO2008/151780); Event GHB614 (cotton, herbicide tolerance, deposited as ATCC PTA-6878, described in US-A 2010-050282 or WO2007/017186); Event GJ11 (corn, herbicide tolerance, deposited as ATCC 209030, described in US-A 2005-188434 or WO98/044140); Event GM RZ13 (sugar beet, virus resistance, deposited as NCIMB-41601, described in WO2010/076212); Event H7-1 (sugar beet, herbicide tolerance, deposited as NCIMB 41158 or NCIMB 41159, described in US-A 2004-172669 or WO 2004/074492); Event JOPLINI (wheat, disease tolerance, not deposited, described in US-A 2008-064032); Event LL27 (soybean, herbicide tolerance, deposited as NCIMB41658, described in WO2006/108674 or US-A 2008-320616); Event LL55 (soybean, herbicide tolerance, deposited as NCIMB 41660, described in WO 2006/108675 or US-A 2008-196127); Event LLcotton25 (cotton, herbicide tolerance, deposited as ATCC PTA-3343, described in WO2003/013224 or US- A 2003-097687); Event LLRICE06 (rice, herbicide tolerance, deposited as ATCC 203353, described in US 6,468,747 or WO2000/026345); Event LLRice62 ( rice, herbicide tolerance, deposited as ATCC 203352, described in WO2000/026345), Event LLRICE601 (rice, herbicide tolerance, deposited as ATCC PTA-2600, described in US-A 2008-2289060 or WO2000/026356); Event LY038 (corn, quality trait, deposited as ATCC PTA-5623, described in US-A 2007-028322 or WO2005/061720); Event MIR162 (corn, insect control, deposited as PTA-8166, described in US-A 2009-300784 or WO2007/142840); Event MIR604 (corn, insect control, not deposited, described in US-A 2008-167456 or WO2005/103301); Event MON15985 (cotton, insect control, deposited as ATCC PTA-2516, described in US-A 2004-250317 or WO2002/100163); Event MON810 (corn, insect control, not deposited, described in US-A 2002-102582); Event MON863 (corn, insect control, deposited as ATCC PTA-2605, described in WO2004/011601 or US-A 2006-095986); Event MON87427 (corn, pollination control, deposited as ATCC PTA-7899,

described in WO2011/062904); Event MON87460 (corn, stress tolerance, deposited as ATCC PTA-8910, described in WO2009/111263 or US-A 2011-0138504); Event MON87701 (soybean, insect control, deposited as ATCC PTA- 8194, described in US-A 2009-130071 or WO2009/064652); Event MON87705 (soybean, quality trait - herbicide tolerance, deposited as ATCC PTA-9241, described in US-A 2010-0080887 or 5 WO2010/037016); Event MON87708 (soybean, herbicide tolerance, deposited as ATCC PTA-9670, described in WO2011/034704); Event MON87712 (soybean, yield, deposited as PTA-10296, described in WO2012/051199), Event MON87754 (soybean, quality trait, deposited as ATCC PTA-9385, described in WO2010/024976); Event MON87769 10 (soybean, quality trait, deposited as ATCC PTA- 8911, described in US-A 2011-0067141 or WO2009/102873); Event MON88017 (corn, insect control - herbicide tolerance, deposited as ATCC PTA-5582, described in US-A 2008-028482 or WO2005/059103); Event MON88913 (cotton, herbicide tolerance, deposited as ATCC PTA-4854, described in WO2004/072235 or US-A 2006-059590); Event MON88302 (oilseed rape, herbicide tolerance, deposited as PTA-10955, described in WO2011/153186), Event MON88701 15 (cotton, herbicide tolerance, deposited as PTA-11754, described in WO2012/134808), Event MON89034 (corn, insect control, deposited as ATCC PTA-7455, described in WO 07/140256 or US-A 2008-260932); Event MON89788 (soybean, herbicide tolerance, deposited as ATCC PTA-6708, described in US-A 2006-282915 or WO2006/130436); 20 Event MSI 1 (oilseed rape, pollination control - herbicide tolerance, deposited as ATCC PTA-850 or PTA-2485, described in WO2001/031042); Event MS8 (oilseed rape, pollination control - herbicide tolerance, deposited as ATCC PTA-730, described in WO2001/041558 or US-A 2003-188347); Event NK603 (corn, herbicide tolerance, deposited as ATCC PTA-2478, described in US-A 2007-292854); Event PE-7 (rice, insect control, not deposited, described in WO2008/114282); Event RF3 (oilseed rape, pollination control - herbicide tolerance, deposited as ATCC PTA-730, described in WO2001/041558 or US-A 2003-188347); Event RT73 (oilseed rape, herbicide tolerance, not deposited, described in WO2002/036831 or US-A 2008-070260); Event SYHT0H2 / SYN-000H2-5 (soybean, herbicide tolerance, deposited as PTA-11226, described in WO2012/082548), 25 Event T227-1 (sugar beet, herbicide tolerance, not deposited, described in WO2002/44407 or US-A 2009-265817); Event T25 (corn, herbicide tolerance, not deposited, described in US-A 2001-029014 or WO2001/051654); Event T304-40 (cotton, insect control - herbicide tolerance, deposited as ATCC PTA-8171, described in US-A 2010-077501 or WO2008/122406); Event T342-142 (cotton, insect control, not deposited, described in 30

WO2006/128568); Event TC1507 (corn, insect control - herbicide tolerance, not deposited, described in US-A 2005-039226 or WO2004/099447); Event VIP1034 (corn, insect control - herbicide tolerance, deposited as ATCC PTA-3925, described in WO2003/052073), Event 32316 (corn, insect control-herbicide tolerance, deposited as PTA-11507, described in

5 WO2011/084632), Event 4114 (corn, insect control-herbicide tolerance, deposited as PTA-11506, described in W02011/084621), event EE-GM3 / FG72 (soybean, herbicide tolerance, ATCC Accession N° PTA-11041) optionally stacked with event EE-GM1/LL27 or event EE-GM2/LL55 (WO2011/063413A2), event DAS-68416-4 (soybean, herbicide tolerance, ATCC Accession N° PTA-10442, WO2011/066360A1), event DAS-68416-4

10 (soybean, herbicide tolerance, ATCC Accession N° PTA-10442, WO2011/066384A1), event DP-040416-8 (corn, insect control, ATCC Accession N° PTA-11508, WO2011/075593A1), event DP-043A47-3 (corn, insect control, ATCC Accession N° PTA-11509, WO2011/075595A1), event DP- 004114-3 (corn, insect control, ATCC Accession N° PTA-11506, WO2011/084621A1), event DP-032316-8 (corn, insect control, ATCC

15 Accession N° PTA-11507, WO2011/084632A1), event MON-88302-9 (oilseed rape, herbicide tolerance, ATCC Accession N° PTA-10955, WO2011/153186A1), event DAS-21606-3 (soybean, herbicide tolerance, ATCC Accession No. PTA-11028, WO2012/033794A2), event MON-87712-4 (soybean, quality trait, ATCC Accession N° PTA-10296, WO2012/051199A2), event DAS-44406-6 (soybean, stacked herbicide

20 tolerance, ATCC Accession N°. PTA-11336, WO2012/075426A1), event DAS-14536-7 (soybean, stacked herbicide tolerance, ATCC Accession N°. PTA-11335, WO2012/075429A1), event SYN-000H2-5 (soybean, herbicide tolerance, ATCC Accession N°. PTA-11226, WO2012/082548A2), event DP-061061-7 (oilseed rape, herbicide tolerance, no deposit N° available, WO2012071039A1), event DP-073496-4 (oilseed rape,

25 herbicide tolerance, no deposit N° available, US2012131692), event 8264.44.06.1 (soybean, stacked herbicide tolerance, Accession N° PTA-11336, WO2012075426A2), event 8291.45.36.2 (soybean, stacked herbicide tolerance, Accession N°. PTA-11335, WO2012075429A2), event SYHT0H2 (soybean, ATCC Accession N°. PTA-11226, WO2012/082548A2), event MON88701 (cotton, ATCC Accession N° PTA-11754,

30 WO2012/134808A1), event KK179-2 (alfalfa, ATCC Accession N° PTA-11833, WO2013/003558A1), event pDAB8264.42.32.1 (soybean, stacked herbicide tolerance, ATCC Accession N° PTA-11993, WO2013/010094A1), event MZDT09Y (corn, ATCC Accession N° PTA-13025, WO2013/012775A1).

The genes/events (e.g., polynucleotides of interest), which impart the desired traits in question, may also be present in combinations with one another in the transgenic plants. Examples of transgenic plants which may be mentioned are the important crop plants, such as cereals (wheat, rice, triticale, barley, rye, oats), maize, soya beans, potatoes, sugar beet, sugar cane, tomatoes, peas and other types of vegetable, cotton, tobacco, oilseed rape and also fruit plants (with the fruits apples, pears, citrus fruits and grapes), with particular emphasis being given to maize, soya beans, wheat, rice, potatoes, cotton, sugar cane, tobacco and oilseed rape. Traits which are particularly emphasized are the increased resistance of the plants to insects, arachnids, nematodes and slugs and snails, as well as the increased resistance of the plants to one or more herbicides.

Commercially available examples of such plants, plant parts or plant seeds that may be treated with preference in accordance with the invention include commercial products, such as plant seeds, sold or distributed under the GENUITY®, DROUGHTGARD®, SMARTSTAX®, RIB COMPLETE®, ROUNDUP READY®, VT DOUBLE PRO®, VT TRIPLE PRO®, BOLLGARD II®, ROUNDUP READY 2 YIELD®, YIELDGARD®, ROUNDUP READY® 2 XTEN<sup>DTM</sup>, INTACTA RR2 PRO®, VISTIVE GOLD®, and/or XTENDFLEX<sup>TM</sup> trade names.

A *HY5* transcription factor gene useful with this invention includes any *HY5* transcription factor gene that is capable of regulating response to illumination in the plant (e.g., shade avoidance response (SAR)) and in which a mutation as described herein can confer reduced SAR/SAS in a plant or part thereof comprising the mutation. In some embodiments, a *HY5* gene encodes a basic leucine zipper (bZIP) transcription factor and has a role in the promotion of photomorphogenesis, e.g., a *HY5* transcription factor. In some embodiments, a *HY5* transcription factor gene (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity of (a), (b), (c) and/or (d) may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%.

In some embodiments, the mutation in an endogenous *HY5* gene may be a non-natural mutation. In some embodiments, the mutation may be any mutation in an endogenous *HY5* gene that results in a *HY5* transcription factor that can confer a reduced SAR response in a plant

comprising the mutated *HY5* gene and/or an increased yield when planted in close proximity with other plants. In some embodiments, the at least one mutation (e.g., one or more mutations) in an endogenous *HY5* transcription factor gene is a point mutation, optionally a base substitution, a base insertion and/or a base deletion. In some embodiments, the at least one mutation in an endogenous *HY5* transcription factor gene is a dominant negative mutation. In some embodiments, the at least one mutation in an endogenous *HY5* transcription factor gene in a plant may be a base substitution, a base deletion and/or a base insertion that results in a plant having a reduced SAR response and/or an increased yield when planted in close proximity with other plants. In some embodiments, the at least one mutation in an endogenous *HY5* transcription factor gene in a plant may be a substitution, a deletion and/or an insertion that results in a dominant negative mutation or a dominant mutation and a plant having a reduced SAR response and/or an increased yield when planted in close proximity with other plants. For example, the mutation may be a substitution, a deletion and/or an insertion of 1 nucleotide or of 2, 3, 4, or 5 consecutive nucleotides to about 100 consecutive nucleotides, optionally 3, 6, 9, 12, 18, 21, 24, 27, 30 or more consecutive nucleotides (e.g., an in-frame insertion or in-frame deletion). In some embodiments, the at least one mutation may be a base substitution to an A, a T, a G, or a C. In some embodiments, the at least one mutation may be a deletion of a portion or the entire encoded COP1 binding domain of the *HY5* transcription factor. In some embodiments, the at least one mutation may be an in-frame deletion of 3 or more consecutive nucleotides.

In some embodiments, a guide nucleic acid (e.g., gRNA, gDNA, crRNA, crDNA) is provided that binds to a target site within an endogenous *Elongated Hypocotyl5 (HY5)* gene, the endogenous *HY5* gene comprising a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**, optionally at least 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**; encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or encoding a region having at least at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%.

In some embodiments, the guide nucleic acid binds to a target nucleic acid within an endogenous *Elongated Hypocotyl5 (HY5)* gene having a gene identification number (gene ID) of Zm00001d015743.

In some embodiments, the target site to which a guide nucleic acid of the invention may  
5 bind may comprise a nucleotide sequence, or portion thereof, having at least 80% sequence  
identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97,  
99 or 100% sequence identity, optionally the sequence identity may be at least 90% or it may be  
at least 95%, optionally the sequence identity may be at least 100%) to any one of the nucleotide  
10 sequences of **SEQ ID NO:75-105**, optionally at least 80% sequence identity to any one of **SEQ  
ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101**, or **103-105**, and/or may encode a sequence  
(or portion thereof) having at least 80% sequence identity to the amino acid sequence of **SEQ  
ID NO:106**.

Example spacer sequences useful with a guide of this invention may comprise  
complementarity to a fragment or portion of a nucleotide sequence having at least 80% sequence  
15 identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97,  
99 or 100% sequence identity, optionally the sequence identity may be at least 90% or it may be  
at least 95%, optionally the sequence identity may be at least 100%) to any one of the nucleotide  
sequences of **SEQ ID NO:72** or **SEQ ID NO: 73**; optionally at least about 80, 81, 82, 83, 84,  
85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity (optionally at  
20 least 90% sequence identity or at least 95% sequence identity, or at least 100% sequence  
identity) to any one of the nucleotide sequences of **SEQ ID NOs:75-105**, optionally 80%  
sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101**, or  
**103-105**; and/or a fragment or portion of a nucleotide sequence encoding a polypeptide  
comprising a sequence having at least 80% (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88,  
25 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally the sequence identity  
may be at least 90% or it may be at least 95%, optionally the sequence identity may be at least  
100%) sequence identity to the amino acid sequence **SEQ ID NO:74**; optionally at least about  
80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence  
identity (optionally at least 90% sequence identity or at least 95% sequence identity, or at least  
30 100% sequence identity) to the amino acid sequence of **SEQ ID NO:106**.

In some embodiments, a target nucleic acid is an endogenous *HY5* transcription factor  
gene that is capable of regulating response to illumination in a plant. In some embodiments, a  
target site within a target nucleic acid may comprise a sequence having at least 80% sequence  
identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97,

99 or 100% sequence identity, optionally the sequence identity may be at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%) to a region, portion or fragment of **SEQ ID NO:72** or **SEQ ID NO:73** (e.g., **SEQ ID NOs:75-105**, optionally at least 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, 5** or **103-105**), or may encode a region of an amino acid sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74** (e.g., **SEQ ID NO:106**).

In some embodiments, a guide nucleic acid may comprise a spacer having the nucleotide sequence of any one of **SEQ ID NOs:107-113**, or a reverse complement thereof, or any combination thereof.

10 In some embodiments, a system is provided that comprises a guide nucleic acid of the present invention and a CRISPR-Cas effector protein that associates with the guide nucleic acid. In some embodiments, the system may further comprise a tracr nucleic acid that associates with the guide nucleic acid and a CRISPR-Cas effector protein, optionally wherein the tracr nucleic acid and the guide nucleic acid are covalently linked.

15 As used herein, "a CRISPR-Cas effector protein in association with a guide nucleic acid" refers to the complex that is formed between a CRISPR-Cas effector protein and a guide nucleic acid in order to direct the CRISPR-Cas effector protein to a target site within a gene.

In some embodiments, a gene editing system is provided, the gene editing system comprising a CRISPR-Cas effector protein in association with a guide nucleic acid, wherein the 20 guide nucleic acid comprises a spacer sequence that binds to a *HY5* gene. In some embodiments, a *HY5* transcription factor gene useful with the gene editing system: (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% 25 sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may 30 be at least 100%.

In some embodiments, the guide nucleic acid of a gene editing system can comprise a spacer sequence that has complementarity to a region, portion or fragment of a nucleotide sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally the sequence

identity may be least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%) to any one of the nucleotide sequences of **SEQ ID NO:72** or **SEQ ID NO:73** (e.g., **SEQ ID NOs:75-105**, optionally at least 80% sequence identity to any one of **SEQ ID NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**), or may encode a region, portion or  
5 fragment of a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74** (e.g., **SEQ ID NO:106**). In some embodiments, a gene editing system may further comprise a tracr nucleic acid that associates with the guide nucleic acid and a CRISPR-Cas effector protein, optionally wherein the tracr nucleic acid and the guide nucleic acid are covalently linked. In some embodiments, a guide nucleic acid is provided that binds to a  
10 target nucleic acid in an endogenous *HY5* gene having a gene identification number (gene ID) (Maize Genetics and Genomics Database (Maize GDB)) of Zm00001d015743 (**SEQ ID NO:72**), optionally wherein (a) a target region within Zm00001d015743 (**SEQ ID NO: 72**) may comprise a portion of consecutive nucleotides of any one or more of the nucleotide sequences of **SEQ ID NOs:75-105**, optionally at least 80% sequence identity to any one of **SEQ ID NOs:79-15 81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**.

The present invention further provides a complex comprising a CRISPR-Cas effector protein comprising a cleavage domain and a guide nucleic acid, wherein the guide nucleic acid binds to a target site within an *Elongated Hypocotyl5 (HY5)* transcription factor gene, wherein the *HY5* gene: (a) comprises a nucleotide sequence having at least 80% sequence identity (e.g.,  
20 at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes an amino acid sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes an amino acid  
25 sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%, wherein the cleavage domain cleaves a target strand in the *HY5* transcription factor gene.

Also provided herein are expression cassettes comprising a (a) polynucleotide encoding  
30 CRISPR-Cas effector protein comprising a cleavage domain and (b) a guide nucleic acid that binds to a target site within an *Elongated Hypocotyl5 (HY5)* gene, wherein the guide nucleic acid comprises a spacer sequence that is complementary to and binds to the target site within the *HY5* gene, wherein the *HY5* gene (a) comprises a nucleotide sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95,



96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; (c) encodes an amino acid sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or (d) encodes an amino acid sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the sequence identity may be at least 100%.

In some embodiments, a nucleic acid is provided that encodes an Elongated Hypocotyl5 (HY5) transcription factor having a mutated COP1 binding site, wherein the mutated COP1 binding site comprises a mutation that disrupts binding of the HY5 transcription factor by a COP1 polypeptide. In some embodiments, the mutation may eliminate the binding of the HY5 transcription factor to COP1 or may reduce the ability of the HY5 transcription factor to bind to a COP1 polypeptide by at least 75% (e.g., at least about 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% or more). In some embodiments, a nucleic acid is provided that comprises a mutated *HY5* gene having at least 90% sequence identity (e.g., at least about 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity, optionally the sequence identity may be at least 95%, optionally the sequence identity may be at least 100%) to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encoding a mutated HY5 polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

Further provided are plants or parts thereof comprising a mutated *HY5* nucleic acid as described herein. In some embodiments, the plant may be a corn (maize) plant. In some embodiments, the plant is a wheat plant. In some embodiments, a plant, a corn plant and/or a wheat plant comprising a mutated *HY5* as described herein and having a reduced SAR may exhibit increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate; delayed senescence; increased photosynthesis efficiency and grain filling, substantially no change in flowering time, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants comprising a mutated *HY5* as described herein and having a reduced SAR as compared to one or more plants that are planted in close proximity with one another but are devoid of the mutation and do not exhibit a reduced shade avoidance response. In some embodiments, when planted in close proximity to one another, plants of this invention comprising reduced SAR may be at least about 5% shorter than control

plants grown under the same conditions (e.g., a shaded environment, e.g., a low R:FR ratio environment). In some embodiments, a corn plant or part thereof is provided that comprises a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene having the gene identification number (gene ID) of Zm00001d015743 (**SEQ ID NO:72**), optionally wherein the mutation is a non-natural mutation.

In some embodiments, a method of the present invention may further comprise regenerating a plant from a plant cell or plant part comprising at least one mutation (e.g., one or more mutations) in an endogenous *Elongated Hypocotyl5 (HY5)* gene, optionally, wherein the mutation disrupts the binding of the encoded HY5 polypeptide by a COP1 polypeptide. In some embodiments, a plant comprising at least one mutation in an endogenous *HY5* gene may exhibit increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate; delayed senescence; increased photosynthesis efficiency and grain filling, substantially no change in flowering time, and/or enhanced defense responses against pathogens and herbivores when they are planted in close proximity with one or more other plants compared to a control plant that does not comprise the at least one mutation in an endogenous *HY5* gene, and therefore, does not comprise a reduced shade avoidance response when planted in close proximity with one or more other plants. In some embodiments, the mutation is a non-naturally occurring mutation. In some embodiments, the mutation is a deletion, optionally an in-frame deletion. In some embodiments, the deletion results in a dominant negative mutation or a dominant mutation.

An editing system useful with this invention can be any site-specific (sequence-specific) genome editing system now known or later developed, which system can introduce mutations in target specific manner. For example, an editing system (e.g., site- or sequence-specific editing system) can include, but is not limited to, a CRISPR-Cas editing system, a meganuclease editing system, a zinc finger nuclease (ZFN) editing system, a transcription activator-like effector nuclease (TALEN) editing system, a base editing system and/or a prime editing system, each of which can comprise one or more polypeptides and/or one or more polynucleotides that when expressed as a system in a cell can modify (mutate) a target nucleic acid in a sequence specific manner. In some embodiments, an editing system (e.g., site- or sequence-specific editing system) can comprise one or more polynucleotides and/or one or more polypeptides, including but not limited to a nucleic acid binding domain (DNA binding domain), a nuclease, and/or other polypeptide, and/or a polynucleotide.

In some embodiments, an editing system can comprise one or more sequence-specific nucleic acid binding domains (DNA binding domains) that can be from, for example, a

polynucleotide-guided endonuclease, a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein), a zinc finger nuclease, a transcription activator-like effector nuclease (TALEN) and/or an Argonaute protein. In some embodiments, an editing system can comprise one or more cleavage domains (e.g., nucleases) including, but not limited to, an endonuclease (e.g., Fok1), a polynucleotide-guided endonuclease, a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein), a zinc finger nuclease, and/or a transcription activator-like effector nuclease (TALEN). In some embodiments, an editing system can comprise one or more polypeptides that include, but are not limited to, a deaminase (e.g., a cytosine deaminase, an adenine deaminase), a reverse transcriptase, a Dna2 polypeptide, and/or a 5' flap endonuclease (FEN). In some embodiments, an editing system can comprise one or more polynucleotides, including, but is not limited to, a CRISPR array (CRISPR guide) nucleic acid, extended guide nucleic acid, and/or a reverse transcriptase template.

In some embodiments, a method of modifying or editing a *HY5* gene may comprise contacting a target nucleic acid (e.g., a target region of the *HY5* gene) with a base-editing fusion protein (e.g., a sequence specific DNA binding protein (e.g., a CRISPR-Cas effector protein or domain) fused to a deaminase domain (e.g., an adenine deaminase and/or a cytosine deaminase) and a guide nucleic acid, wherein the guide nucleic acid is capable of guiding/targeting the base editing fusion protein to the target nucleic acid, thereby editing a locus within the target nucleic acid. In some embodiments, a base editing fusion protein and guide nucleic acid may be comprised in one or more expression cassettes. In some embodiments, the target nucleic acid may be contacted with a base editing fusion protein and an expression cassette comprising a guide nucleic acid. In some embodiments, the sequence-specific DNA binding fusion proteins and guides may be provided as ribonucleoproteins (RNPs). In some embodiments, a cell may be contacted with more than one base-editing fusion protein and/or one or more guide nucleic acids that may target one or more target nucleic acids in the cell.

In some embodiments, a method of modifying or editing a *HY5* gene may comprise contacting a target nucleic acid (e.g., a target region of the *HY5* gene) with a sequence-specific DNA binding fusion protein (e.g., a sequence-specific DNA binding protein (e.g., a CRISPR-Cas effector protein or domain) fused to a peptide tag, a deaminase fusion protein comprising a deaminase domain (e.g., an adenine deaminase and/or a cytosine deaminase) fused to an affinity polypeptide that is capable of binding to the peptide tag, and a guide nucleic acid, wherein the guide nucleic acid is capable of guiding/targeting the sequence-specific DNA binding fusion protein to the target nucleic acid and the sequence-specific DNA binding fusion protein is capable of recruiting the deaminase fusion protein to the target nucleic acid via the peptide tag-

affinity polypeptide interaction, thereby editing a locus within the target nucleic acid. In some embodiments, the sequence-specific DNA binding fusion protein may be fused to the affinity polypeptide that binds the peptide tag and the deaminase may be fused to the peptide tag, thereby recruiting the deaminase to the sequence-specific DNA binding fusion protein and to the target nucleic acid. In some embodiments, the sequence-specific binding fusion protein, deaminase fusion protein, and guide nucleic acid may be comprised in one or more expression cassettes. In some embodiments, the target nucleic acid may be contacted with a sequence-specific binding fusion protein, deaminase fusion protein, and an expression cassette comprising a guide nucleic acid. In some embodiments, the sequence-specific DNA binding fusion proteins, deaminase fusion proteins and guides may be provided as ribonucleoproteins (RNPs).

In some embodiments, methods such as prime editing may be used to generate a mutation in an endogenous *HY5* transcription factor gene. In prime editing, RNA-dependent DNA polymerase (reverse transcriptase, RT) and reverse transcriptase templates (RT template) are used in combination with sequence specific nucleic acid binding domains that confer the ability to recognize and bind the target in a sequence-specific manner, and which can also cause a nick of the PAM-containing strand within the target. The nucleic acid binding domain may be a CRISPR-Cas effector protein and in this case, the CRISPR array or guide RNA may be an extended guide that comprises an extended portion comprising a primer binding site (PSB) and the edit to be incorporated into the genome (the template). Similar to base editing, prime editing can take advantage of the various methods of recruiting proteins for use in the editing to the target site, such methods including both non-covalent and covalent interactions between the proteins and nucleic acids used in the selected process of genome editing.

In some embodiments, the mutation or modification of an endogenous *HY5* transcription factor gene may be an insertion, a deletion and/or a point mutation in that produces a mutated *HY5* transcription factor having reduced binding by a Constitutive Photomorphogenic 1 (COP1) polypeptide (e.g., a mutated *HY5* transcription factor). In some embodiments, a plant part may be a cell. In some embodiments, the plant or plant part thereof may be any plant or part thereof as described herein. In some embodiments, a plant useful with this invention may be corn, soy, canola, wheat, rice, cotton, sugarcane, sugar beet, barley, oats, alfalfa, sunflower, safflower, oil palm, sesame, coconut, tobacco, potato, sweet potato, cassava, coffee, apple, plum, apricot, peach, cherry, pear, fig, banana, citrus, cocoa, avocado, olive, almond, walnut, strawberry, watermelon, pepper, grape, tomato, cucumber, blackberry, raspberry, black raspberry, or a *Brassica* spp. In some embodiments, a plant comprising a mutated endogenous *HY5* transcription factor having reduced COP1 binding may comprise increased yield, increased

upright growth, decreased height, decreased shoot:root ratio, decreased leaf length; increased mechanical strength of stems, reduced lodging rate, delayed senescence; increased photosynthesis efficiency and grain filling, substantially no change in flowering time, and/or enhanced defense responses against pathogens and herbivores when they are planted in close proximity with one or more other plants compared to a control plant that does not comprise the at least one non-natural mutation in an endogenous *HY5* gene, and therefore, does not comprise a reduced shade avoidance response, which plant is planted in close proximity with one or more other plants. In some embodiments, the plant may be a corn plant, the corn plant comprising a mutated endogenous *HY5* transcription factor having reduced COP1 binding and, optionally, exhibiting increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length; increased mechanical strength of stems, reduced lodging rate; delayed senescence; increased photosynthesis efficiency and grain filling, substantially no change in flowering time, and/or enhanced defense responses against pathogens and herbivores.

In some embodiments, a mutation that is introduced into an endogenous *HY5* gene that optionally results in reduced COP1 binding to the encoded *HY5* polypeptide may be a non-natural mutation. In some embodiments, a mutation that is introduced into an endogenous *HY5* gene may optionally result in reduced COP1 binding to the encoded *HY5* polypeptide. In some embodiments, a mutation that is introduced into an endogenous *HY5* gene may be a substitution, an insertion and/or a deletion of one nucleotide or at least two consecutive nucleotides (e.g., 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, or 30 or more), wherein the mutation optionally results in reduced COP1 binding to the encoded *HY5* polypeptide. In some embodiments, a mutation that is introduced into an endogenous *HY5* gene may be a deletion, optionally an in-frame deletion. In some embodiments, the deletion may result in a deletion of all or a portion of a COP1 binding region in the *HY5* transcription factor polypeptide, e.g., a deletion of one, two, or three or more nucleotides (e.g., consecutive nucleotides) from a region of the *HY5* gene that encodes the amino acid sequence of **SEQ ID NO:106**.

In some embodiments, a sequence-specific nucleic acid binding domain (sequence-specific DNA binding domains) of an editing system useful with this invention can be from, for example, a polynucleotide-guided endonuclease, a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein), a zinc finger nuclease, a transcription activator-like effector nuclease (TALEN) and/or an Argonaute protein.

In some embodiments, a sequence-specific nucleic acid binding domain may be a CRISPR-Cas effector protein, optionally wherein the CRISPR-Cas effector protein may be from

a Type I CRISPR-Cas system, a Type II CRISPR-Cas system, a Type III CRISPR-Cas system, a Type IV CRISPR-Cas system, Type V CRISPR-Cas system, or a Type VI CRISPR-Cas system. In some embodiments, a CRISPR-Cas effector protein of the invention may be from a Type II CRISPR-Cas system or a Type V CRISPR-Cas system. In some embodiments, a CRISPR-Cas effector protein may be Type II CRISPR-Cas effector protein, for example, a Cas9 effector protein. In some embodiments, a CRISPR-Cas effector protein may be Type V CRISPR-Cas effector protein, for example, a Cas12 effector protein.

As used herein, a "CRISPR-Cas effector protein" is a protein or polypeptide or domain thereof that cleaves or cuts a nucleic acid, binds a nucleic acid (e.g., a target nucleic acid and/or a guide nucleic acid), and/or that identifies, recognizes, or binds a guide nucleic acid as defined herein. In some embodiments, a CRISPR-Cas effector protein may be an enzyme (e.g., a nuclease, endonuclease, nickase, etc.) or portion thereof and/or may function as an enzyme. In some embodiments, a CRISPR-Cas effector protein refers to a CRISPR-Cas nuclease polypeptide or domain thereof that comprises nuclease activity or in which the nuclease activity has been reduced or eliminated, and/or comprises nickase activity or in which the nickase has been reduced or eliminated, and/or comprises single stranded DNA cleavage activity (ss DNase activity) or in which the ss DNase activity has been reduced or eliminated, and/or comprises self-processing RNase activity or in which the self-processing RNase activity has been reduced or eliminated. A CRISPR-Cas effector protein may bind to a target nucleic acid.

In some embodiments, a CRISPR-Cas effector protein may include, but is not limited to, a Cas9, C2c1, C2c3, Cas12a (also referred to as Cpf1), Cas12b, Cas12c, Cas12d, Cas12e, Cas13a, Cas13b, Cas13c, Cas13d, Cas1, Cas1B, Cas2, Cas3, Cas3', Cas3", Cas4, Cas5, Cas6, Cas7, Cas8, Cas9 (also known as Csn1 and Csx12), Cas10, Csy1, Csy2, Csy3, Csel, Cse2, Csel, Csc2, Csa5, Csn2, Csm2, Csm3, Csm4, Csm5, Csm6, Cmr1, Cmr3, Cmr4, Cmr5, Cmr6, Csb1, Csb2, Csb3, Csx17, Csx14, Csx10, Csx16, CsaX, Csx3, Csx1, Csx15, Csf1, Csf2, Csf3, Csf4 (dinG), and/or Csf5 nuclease, optionally wherein the CRISPR-Cas effector protein may be a Cas9, Cas12a (Cpf1), Cas12b, Cas12c (C2c3), Cas12d (CasY), Cas12e (CasX), Cas12g, Cas12h, Cas12i, C2c4, C2c5, C2c8, C2c9, C2c10, Cas14a, Cas14b, and/or Cas14c effector protein.

In some embodiments, a CRISPR-Cas effector protein useful with the invention may comprise a mutation in its nuclease active site (e.g., RuvC, HNH, e.g., RuvC site of a Cas12a nuclease domain; e.g., RuvC site and/or HNH site of a Cas9 nuclease domain). A CRISPR-Cas effector protein having a mutation in its nuclease active site, and therefore, no longer comprising nuclease activity, is commonly referred to as "dead," e.g., dCas. In some embodiments, a CRISPR-Cas effector protein domain or polypeptide having a mutation in its nuclease active site

may have impaired activity or reduced activity as compared to the same CRISPR-Cas effector protein without the mutation, e.g., a nickase, e.g., Cas9 nickase, Cas12a nickase.

A CRISPR Cas9 effector protein or CRISPR Cas9 effector domain useful with this invention may be any known or later identified Cas9 nuclease. In some embodiments, a  
5 CRISPR Cas9 polypeptide can be a Cas9 polypeptide from, for example, *Streptococcus* spp. (e.g., *S. pyogenes*, *S. thermophilus*), *Lactobacillus* spp., *Bifidobacterium* spp., *Kandleria* spp., *Leuconostoc* spp., *Oenococcus* spp., *Pediococcus* spp., *Weissella* spp., and/or *Olsenella* spp. Example Cas9 sequences include, but are not limited to, the amino acid sequences of **SEQ ID NOs:59-60** or the polynucleotide sequences of **SEQ ID NOs:61-71**.

10 In some embodiments, the CRISPR-Cas effector protein may be a Cas9 polypeptide derived from *Streptococcus pyogenes* and recognizes the PAM sequence motif NGG, NAG, NGA (Mali et al, Science 2013; 339(6121): 823-826). In some embodiments, the CRISPR-Cas effector protein may be a Cas9 polypeptide derived from *Streptococcus thermophiles* and recognizes the PAM sequence motif NGGNG and/or NNAGAAW (W = A or T) (See, e.g.,  
15 Horvath et al, Science, 2010; 327(5962): 167-170, and Deveau et al, J Bacteriol 2008; 190(4): 1390-1400). In some embodiments, the CRISPR-Cas effector protein may be a Cas9 polypeptide derived from *Streptococcus mutans* and recognizes the PAM sequence motif NGG and/or NAAR (R = A or G) (See, e.g., Deveau et al, J BACTERIOL 2008; 190(4): 1390-1400). In some  
20 embodiments, the CRISPR-Cas effector protein may be a Cas9 polypeptide derived from *Streptococcus aureus* and recognizes the PAM sequence motif NNGRR (R = A or G). In some embodiments, the CRISPR-Cas effector protein may be a Cas9 protein derived from *S. aureus*, which recognizes the PAM sequence motif N GRRT (R = A or G). In some embodiments, the CRISPR-Cas effector protein may be a Cas9 polypeptide derived from *S. aureus*, which recognizes the PAM sequence motif N GRRV (R = A or G). In some embodiments, the  
25 CRISPR-Cas effector protein may be a Cas9 polypeptide that is derived from *Neisseria meningitidis* and recognizes the PAM sequence motif N GATT or N GCTT (R = A or G, V = A, G or C) (See, e.g., Hou et al (2013) *Proc. Natl. Acad. Sci. USA* 110 (39):15644-15649). In the aforementioned embodiments, N can be any nucleotide residue, e.g., any of A, G, C or T. In some embodiments, the CRISPR-Cas effector protein may be a Cas13a protein derived from  
30 *Leptotrichia shahii*, which recognizes a protospacer flanking sequence (PFS) (or RNA PAM (rPAM)) sequence motif of a single 3' A, U, or C, which may be located within the target nucleic acid.

In some embodiments, the CRISPR-Cas effector protein may be derived from Cas12a, which is a Type V Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)-Cas

nuclease (see, e.g., amino acid sequences of **SEQ ID NOs:1-17**, nucleic acid sequences of **SEQ ID NOs:18-20**). Cas12a differs in several respects from the more well-known Type II CRISPR Cas9 nuclease. For example, Cas9 recognizes a G-rich protospacer-adjacent motif (PAM) that is 3' to its guide RNA (gRNA, sgRNA, crRNA, crDNA, CRISPR array) binding site (protospacer, target nucleic acid, target DNA) (3'-NGG), while Cas12a recognizes a T-rich PAM that is located 5' to the target nucleic acid (5'-TTN, 5'-TTTN. In fact, the orientations in which Cas9 and Cas12a bind their guide RNAs are very nearly reversed in relation to their N and C termini. Furthermore, Cas12a enzymes use a single guide RNA (gRNA, CRISPR array, crRNA) rather than the dual guide RNA (sgRNA (e.g., crRNA and tracrRNA)) found in natural Cas9 systems, and Cas12a processes its own gRNAs. Additionally, Cas12a nuclease activity produces staggered DNA double stranded breaks instead of blunt ends produced by Cas9 nuclease activity, and Cas12a relies on a single RuvC domain to cleave both DNA strands, whereas Cas9 utilizes an HNH domain and a RuvC domain for cleavage.

A CRISPR Cas12a effector protein/domain useful with this invention may be any known or later identified Cas12a polypeptide (previously known as Cpf1) (see, e.g., U.S. Patent No. 9,790,490, which is incorporated by reference for its disclosures of Cpf1 (Cas12a) sequences). The term "Cas12a", "Cas12a polypeptide" or "Cas12a domain" refers to an RNA-guided nuclease comprising a Cas12a polypeptide, or a fragment thereof, which comprises the guide nucleic acid binding domain of Cas12a and/or an active, inactive, or partially active DNA cleavage domain of Cas12a. In some embodiments, a Cas12a useful with the invention may comprise a mutation in the nuclease active site (e.g., RuvC site of the Cas12a domain). A Cas12a domain or Cas12a polypeptide having a mutation in its nuclease active site, and therefore, no longer comprising nuclease activity, is commonly referred to as deadCas12a (e.g., dCas12a). In some embodiments, a Cas12a domain or Cas12a polypeptide having a mutation in its nuclease active site may have impaired activity, e.g., may have nickase activity.

Any deaminase domain/polypeptide useful for base editing may be used with this invention. In some embodiments, the deaminase domain may be a cytosine deaminase domain or an adenine deaminase domain. A cytosine deaminase (or cytidine deaminase) useful with this invention may be any known or later identified cytosine deaminase from any organism (see, e.g., U.S. Patent No. 10,167,457 and Thuronyi et al. *Nat. Biotechnol.* 37:1070–1079 (2019), each of which is incorporated by reference herein for its disclosure of cytosine deaminases). Cytosine deaminases can catalyze the hydrolytic deamination of cytidine or deoxycytidine to uridine or deoxyuridine, respectively. Thus, in some embodiments, a deaminase or deaminase domain useful with this invention may be a cytidine deaminase domain, catalyzing the hydrolytic



deamination of cytosine to uracil. In some embodiments, a cytosine deaminase may be a variant of a naturally-occurring cytosine deaminase, including but not limited to a primate (e.g., a human, monkey, chimpanzee, gorilla), a dog, a cow, a rat or a mouse. Thus, in some embodiments, an cytosine deaminase useful with the invention may be about 70% to about 5 100% identical to a wild type cytosine deaminase (e.g., about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% identical, and any range or value therein, to a naturally occurring cytosine deaminase).

In some embodiments, a cytosine deaminase useful with the invention may be an 10 apolipoprotein B mRNA-editing complex (APOBEC) family deaminase. In some embodiments, the cytosine deaminase may be an APOBEC1 deaminase, an APOBEC2 deaminase, an APOBEC3A deaminase, an APOBEC3B deaminase, an APOBEC3C deaminase, an APOBEC3D deaminase, an APOBEC3F deaminase, an APOBEC3G deaminase, an APOBEC3H deaminase, an APOBEC4 deaminase, a human activation induced deaminase 15 (hAID), an rAPOBEC1, FERNY, and/or a CDA1, optionally a pmCDA1, an atCDA1 (e.g., At2g19570), and evolved versions of the same (e.g., **SEQ ID NO:27**, **SEQ ID NO:28** or **SEQ ID NO:29**). In some embodiments, the cytosine deaminase may be an APOBEC1 deaminase having the amino acid sequence of **SEQ ID NO:23**. In some embodiments, the cytosine deaminase may be an APOBEC3A deaminase having the amino acid sequence of **SEQ ID** 20 **NO:24**. In some embodiments, the cytosine deaminase may be an CDA1 deaminase, optionally a CDA1 having the amino acid sequence of **SEQ ID NO:25**. In some embodiments, the cytosine deaminase may be a FERNY deaminase, optionally a FERNY having the amino acid sequence of **SEQ ID NO:26**. In some embodiments, a cytosine deaminase useful with the invention may be about 70% to about 100% identical (e.g., 70%, 71%, 72%, 73%, 74%, 75%, 25 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.5% or 100% identical) to the amino acid sequence of a naturally occurring cytosine deaminase (e.g., an evolved deaminase). In some embodiments, a cytosine deaminase useful with the invention may be about 70% to about 99.5% identical (e.g., about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 30 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 99.5% identical) to the amino acid sequence of **SEQ ID NO:23**, **SEQ ID NO:24**, **SEQ ID NO:25** or **SEQ ID NO:26** (e.g., at least 80%, at least 85%, at least 90%, at least 92%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or at least 99.5% identical to the amino acid sequence of **SEQ ID NO:23**, **SEQ ID NO:24**, **SEQ ID NO:25**, **SEQ ID NO:26**,

**SEQ ID NO:27**, **SEQ ID NO:28** or **SEQ ID NO:29**). In some embodiments, a polynucleotide encoding a cytosine deaminase may be codon optimized for expression in a plant and the codon optimized polypeptide may be about 70% to 99.5% identical to the reference polynucleotide.

In some embodiments, a nucleic acid construct of this invention may further encode an uracil glycosylase inhibitor (UGI) (e.g., uracil-DNA glycosylase inhibitor) polypeptide/domain. Thus, in some embodiments, a nucleic acid construct encoding a CRISPR-Cas effector protein and a cytosine deaminase domain (e.g., encoding a fusion protein comprising a CRISPR-Cas effector protein domain fused to a cytosine deaminase domain, and/or a CRISPR-Cas effector protein domain fused to a peptide tag or to an affinity polypeptide capable of binding a peptide tag and/or a deaminase protein domain fused to a peptide tag or to an affinity polypeptide capable of binding a peptide tag) may further encode a uracil-DNA glycosylase inhibitor (UGI), optionally wherein the UGI may be codon optimized for expression in a plant. In some embodiments, the invention provides fusion proteins comprising a CRISPR-Cas effector polypeptide, a deaminase domain, and a UGI and/or one or more polynucleotides encoding the same, optionally wherein the one or more polynucleotides may be codon optimized for expression in a plant. In some embodiments, the invention provides fusion proteins, wherein a CRISPR-Cas effector polypeptide, a deaminase domain, and a UGI may be fused to any combination of peptide tags and affinity polypeptides as described herein, thereby recruiting the deaminase domain and UGI to the CRISPR-Cas effector polypeptide and a target nucleic acid. In some embodiments, a guide nucleic acid may be linked to a recruiting RNA motif and one or more of the deaminase domain and/or UGI may be fused to an affinity polypeptide that is capable of interacting with the recruiting RNA motif, thereby recruiting the deaminase domain and UGI to a target nucleic acid.

An "uracil glycosylase inhibitor" useful with the invention may be any protein that is capable of inhibiting a uracil-DNA glycosylase base-excision repair enzyme. In some embodiments, a UGI domain comprises a wild type UGI or a fragment thereof. In some embodiments, a UGI domain useful with the invention may be about 70% to about 100% identical (e.g., 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.5% or 100% identical and any range or value therein) to the amino acid sequence of a naturally occurring UGI domain. In some embodiments, a UGI domain may comprise the amino acid sequence of **SEQ ID NO:41** or a polypeptide having about 70% to about 99.5% sequence identity to the amino acid sequence of **SEQ ID NO:41** (e.g., at least 80%, at least 85%, at least 90%, at least 92%, at least 95%, at least 96%, at least 97%, at least 98%, at least

99%, or at least 99.5% identical to the amino acid sequence of **SEQ ID NO:41**). For example, in some embodiments, a UGI domain may comprise a fragment of the amino acid sequence of **SEQ ID NO:41** that is 100% identical to a portion of consecutive nucleotides (e.g., 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80 consecutive nucleotides; e.g., about 10, 15, 20, 25, 30, 35, 40, 45, to about 50, 55, 60, 65, 70, 75, 80 consecutive nucleotides) of the amino acid sequence of **SEQ ID NO:41**. In some embodiments, a UGI domain may be a variant of a known UGI (e.g., **SEQ ID NO:41**) having about 70% to about 99.5% sequence identity (e.g., 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.5% sequence identity, and any range or value therein) to the known UGI. In some embodiments, a polynucleotide encoding a UGI may be codon optimized for expression in a plant (e.g., a plant) and the codon optimized polypeptide may be about 70% to about 99.5% identical to the reference polynucleotide.

An adenine deaminase (or adenosine deaminase) useful with this invention may be any known or later identified adenine deaminase from any organism (see, e.g., U.S. Patent No. 10,113,163, which is incorporated by reference herein for its disclosure of adenine deaminases). An adenine deaminase can catalyze the hydrolytic deamination of adenine or adenosine. In some embodiments, the adenine deaminase may catalyze the hydrolytic deamination of adenosine or deoxyadenosine to inosine or deoxyinosine, respectively. In some embodiments, the adenosine deaminase may catalyze the hydrolytic deamination of adenine or adenosine in DNA. In some embodiments, an adenine deaminase encoded by a nucleic acid construct of the invention may generate an A→G conversion in the sense (e.g., "+", template) strand of the target nucleic acid or a T→C conversion in the antisense (e.g., "-", complementary) strand of the target nucleic acid.

In some embodiments, an adenosine deaminase may be a variant of a naturally occurring adenine deaminase. Thus, in some embodiments, an adenosine deaminase may be about 70% to 100% identical to a wild type adenine deaminase (e.g., about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100% identical, and any range or value therein, to a naturally occurring adenine deaminase). In some embodiments, the deaminase or deaminase does not occur in nature and may be referred to as an engineered, mutated, or evolved adenosine deaminase. Thus, for example, an engineered, mutated or evolved adenine deaminase polypeptide or an adenine deaminase domain may be about 70% to 99.9% identical to a naturally occurring adenine deaminase polypeptide/domain (e.g., about 70%, 71%, 72%, 73%,

74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.1%, 99.2%, 99.3%, 99.4%, 99.5%, 99.6%, 99.7%, 99.8% or 99.9% identical, and any range or value therein, to a naturally occurring adenine deaminase polypeptide or adenine deaminase domain). In some  
5 embodiments, the adenosine deaminase may be from a bacterium, (e.g., *Escherichia coli*, *Staphylococcus aureus*, *Haemophilus influenzae*, *Caulobacter crescentus*, and the like). In some embodiments, a polynucleotide encoding an adenine deaminase polypeptide/domain may be codon optimized for expression in a plant.

In some embodiments, an adenine deaminase domain may be a wild type tRNA-specific  
10 adenosine deaminase domain, e.g., a tRNA-specific adenosine deaminase (TadA) and/or a mutated/evolved adenosine deaminase domain, e.g., mutated/evolved tRNA-specific adenosine deaminase domain (TadA\*). In some embodiments, a TadA domain may be from *E. coli*. In some embodiments, the TadA may be modified, e.g., truncated, missing one or more N-terminal and/or C-terminal amino acids relative to a full-length TadA (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11,  
15 12, 13, 14, 15, 6, 17, 18, 19, or 20 N-terminal and/or C terminal amino acid residues may be missing relative to a full length TadA. In some embodiments, a TadA polypeptide or TadA domain does not comprise an N-terminal methionine. In some embodiments, a wild type *E. coli* TadA comprises the amino acid sequence of **SEQ ID NO:30**. In some embodiments, a mutated/evolved *E. coli* TadA\* comprises the amino acid sequence of **SEQ ID NOs:31-40** (e.g.,  
20 **SEQ ID NOs: 31, 32, 33, 34, 35, 36, 37, 38, 39** or **40**). In some embodiments, a polynucleotide encoding a TadA/TadA\* may be codon optimized for expression in a plant.

A cytosine deaminase catalyzes cytosine deamination and results in a thymidine (through a uracil intermediate), causing a C to T conversion, or a G to A conversion in the complementary strand in the genome. Thus, in some embodiments, the cytosine deaminase  
25 encoded by the polynucleotide of the invention generates a C→T conversion in the sense (e.g., "+"; template) strand of the target nucleic acid or a G→A conversion in antisense (e.g., "-", complementary) strand of the target nucleic acid.

In some embodiments, the adenine deaminase encoded by the nucleic acid construct of the invention generates an A→G conversion in the sense (e.g., "+"; template) strand of the target  
30 nucleic acid or a T→C conversion in the antisense (e.g., "-", complementary) strand of the target nucleic acid.

The nucleic acid constructs of the invention encoding a base editor comprising a sequence-specific DNA binding protein and a cytosine deaminase polypeptide, and nucleic acid constructs/expression cassettes/vectors encoding the same, may be used in combination with

guide nucleic acids for modifying target nucleic acid including, but not limited to, generation of C→T or G →A mutations in a target nucleic acid including, but not limited to, a plasmid sequence; generation of C→T or G →A mutations in a coding sequence to alter an amino acid identity; generation of C→T or G →A mutations in a coding sequence to generate a stop codon; 5 generation of C→T or G →A mutations in a coding sequence to disrupt a start codon; generation of point mutations in genomic DNA to disrupt transcription factor binding; and/or generation of point mutations in genomic DNA to disrupt splice junctions.

The nucleic acid constructs of the invention encoding a base editor comprising a sequence-specific DNA binding protein and an adenine deaminase polypeptide, and expression 10 cassettes and/or vectors encoding the same may be used in combination with guide nucleic acids for modifying a target nucleic acid including, but not limited to, generation of A→G or T→C mutations in a target nucleic acid including, but not limited to, a plasmid sequence; generation of A→G or T→C mutations in a coding sequence to alter an amino acid identity; generation of A→G or T→C mutations in a coding sequence to generate a stop codon; generation of A→G or 15 T→C mutations in a coding sequence to disrupt a start codon; generation of point mutations in genomic DNA to disrupt function; and/or generation of point mutations in genomic DNA to disrupt splice junctions.

The nucleic acid constructs of the invention comprising a CRISPR-Cas effector protein or a fusion protein thereof may be used in combination with a guide RNA (gRNA, CRISPR 20 array, CRISPR RNA, crRNA), designed to function with the encoded CRISPR-Cas effector protein or domain, to modify a target nucleic acid. A guide nucleic acid useful with this invention comprises at least one spacer sequence and at least one repeat sequence. The guide nucleic acid is capable of forming a complex with the CRISPR-Cas nuclease domain encoded and expressed by a nucleic acid construct of the invention and the spacer sequence is capable of 25 hybridizing to a target nucleic acid, thereby guiding the complex (e.g., a CRISPR-Cas effector fusion protein (e.g., CRISPR-Cas effector domain fused to a deaminase domain and/or a CRISPR-Cas effector domain fused to a peptide tag or an affinity polypeptide to recruit a deaminase domain and optionally, a UGI) to the target nucleic acid, wherein the target nucleic acid may be modified (e.g., cleaved or edited) or modulated (e.g., modulating transcription) by 30 the deaminase domain.

As an example, a nucleic acid construct encoding a Cas9 domain linked to a cytosine deaminase domain (e.g., fusion protein) may be used in combination with a Cas9 guide nucleic acid to modify a target nucleic acid, wherein the cytosine deaminase domain of the fusion protein deaminates a cytosine base in the target nucleic acid, thereby editing the target nucleic

acid. In a further example, a nucleic acid construct encoding a Cas9 domain linked to an adenine deaminase domain (e.g., fusion protein) may be used in combination with a Cas9 guide nucleic acid to modify a target nucleic acid, wherein the adenine deaminase domain of the fusion protein deaminates an adenosine base in the target nucleic acid, thereby editing the target nucleic acid.

Likewise, a nucleic acid construct encoding a Cas12a domain (or other selected CRISPR-Cas nuclease, e.g., C2c1, C2c3, Cas12b, Cas12c, Cas12d, Cas12e, Cas13a, Cas13b, Cas13c, Cas13d, Cas1, Cas1B, Cas2, Cas3, Cas3', Cas3", Cas4, Cas5, Cas6, Cas7, Cas8, Cas9 (also known as Csn1 and Csx12), Cas10, Csy1, Csy2, Csy3, Cse1, Cse2, Cse1, Csc2, Csa5, Csn2, Csm2, Csm3, Csm4, Csm5, Csm6, Cmr1, Cmr3, Cmr4, Cmr5, Cmr6, Csb1, Csb2, Csb3, Csx17, Csx14, Csx10, Csx16, CsaX, Csx3, Csx1, Csx15, Csf1, Csf2, Csf3, Csf4 (dinG), and/or Csf5) linked to a cytosine deaminase domain or adenine deaminase domain (e.g., fusion protein) may be used in combination with a Cas12a guide nucleic acid (or the guide nucleic acid for the other selected CRISPR-Cas nuclease) to modify a target nucleic acid, wherein the cytosine deaminase domain or adenine deaminase domain of the fusion protein deaminates a cytosine base in the target nucleic acid, thereby editing the target nucleic acid.

A "guide nucleic acid," "guide RNA," "gRNA," "CRISPR RNA/DNA" "crRNA" or "crDNA" as used herein means a nucleic acid that comprises at least one spacer sequence, which is complementary to (and hybridizes to) a target DNA (e.g., protospacer), and at least one repeat sequence (e.g., a repeat of a Type V Cas12a CRISPR-Cas system, or a fragment or portion thereof; a repeat of a Type II Cas9 CRISPR-Cas system, or fragment thereof; a repeat of a Type V C2c1 CRISPR Cas system, or a fragment thereof; a repeat of a CRISPR-Cas system of, for example, C2c3, Cas12a (also referred to as Cpf1), Cas12b, Cas12c, Cas12d, Cas12e, Cas13a, Cas13b, Cas13c, Cas13d, Cas1, Cas1B, Cas2, Cas3, Cas3', Cas3", Cas4, Cas5, Cas6, Cas7, Cas8, Cas9 (also known as Csn1 and Csx12), Cas10, Csy1, Csy2, Csy3, Cse1, Cse2, Cse1, Csc2, Csa5, Csn2, Csm2, Csm3, Csm4, Csm5, Csm6, Cmr1, Cmr3, Cmr4, Cmr5, Cmr6, Csb1, Csb2, Csb3, Csx17, Csx14, Csx10, Csx16, CsaX, Csx3, Csx1, Csx15, Csf1, Csf2, Csf3, Csf4 (dinG), and/or Csf5, or a fragment thereof), wherein the repeat sequence may be linked to the 5' end and/or the 3' end of the spacer sequence. The design of a gRNA of this invention may be based on a Type I, Type II, Type III, Type IV, Type V, or Type VI CRISPR-Cas system.

In some embodiments, a Cas12a gRNA may comprise, from 5' to 3', a repeat sequence (full length or portion thereof ("handle")); e.g., pseudoknot-like structure) and a spacer sequence.

In some embodiments, a guide nucleic acid may comprise more than one repeat sequence-spacer sequence (e.g., 2, 3, 4, 5, 6, 7, 8, 9, 10, or more repeat-spacer sequences) (e.g.,

repeat-spacer-repeat, e.g., repeat-spacer-repeat-spacer-repeat-spacer-repeat-spacer-repeat-spacer, and the like). The guide nucleic acids of this invention are synthetic, human-made, and not found in nature. A gRNA can be quite long and may be used as an aptamer (like in the MS2 recruitment strategy) or other RNA structures hanging off the spacer.

5 A "repeat sequence" as used herein, refers to, for example, any repeat sequence of a wild-type CRISPR Cas locus (e.g., a Cas9 locus, a Cas12a locus, a C2c1 locus, etc.) or a repeat sequence of a synthetic crRNA that is functional with the CRISPR-Cas effector protein encoded by the nucleic acid constructs of the invention. A repeat sequence useful with this invention can be any known or later identified repeat sequence of a CRISPR-Cas locus (e.g., Type I, Type II, 10 Type III, Type IV, Type V or Type VI) or it can be a synthetic repeat designed to function in a Type I, II, III, IV, V or VI CRISPR-Cas system. A repeat sequence may comprise a hairpin structure and/or a stem loop structure. In some embodiments, a repeat sequence may form a pseudoknot-like structure at its 5' end (i.e., "handle"). Thus, in some embodiments, a repeat sequence can be identical to or substantially identical to a repeat sequence from wild-type Type I 15 CRISPR-Cas loci, Type II, CRISPR-Cas loci, Type III, CRISPR-Cas loci, Type IV CRISPR-Cas loci, Type V CRISPR-Cas loci and/or Type VI CRISPR-Cas loci. A repeat sequence from a wild-type CRISPR-Cas locus may be determined through established algorithms, such as using the CRISPRfinder offered through CRISPRdb (see, Grissa et al. *Nucleic Acids Res.* 35(Web Server issue):W52-7). In some embodiments, a repeat sequence or portion thereof is linked at 20 its 3' end to the 5' end of a spacer sequence, thereby forming a repeat-spacer sequence (e.g., guide nucleic acid, guide RNA/DNA, crRNA, crDNA).

In some embodiments, a repeat sequence comprises, consists essentially of, or consists of at least 10 nucleotides depending on the particular repeat and whether the guide nucleic acid comprising the repeat is processed or unprocessed (e.g., about 10, 11, 12, 13, 14, 15, 16, 17, 18, 25 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50 to 100 or more nucleotides, or any range or value therein). In some embodiments, a repeat sequence comprises, consists essentially of, or consists of about 10 to about 20, about 10 to about 30, about 10 to about 45, about 10 to about 50, about 15 to about 30, about 15 to about 40, about 15 to about 45, about 15 to about 50, about 20 to about 30, about 20 30 to about 40, about 20 to about 50, about 30 to about 40, about 40 to about 80, about 50 to about 100 or more nucleotides.

A repeat sequence linked to the 5' end of a spacer sequence can comprise a portion of a repeat sequence (e.g., 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35 or more contiguous nucleotides of a wild type repeat

sequence). In some embodiments, a portion of a repeat sequence linked to the 5' end of a spacer sequence can be about five to about ten consecutive nucleotides in length (e.g., about 5, 6, 7, 8, 9, 10 nucleotides) and have at least 90% sequence identity (e.g., at least about 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or more) to the same region (e.g., 5' end) of a wild type  
5 CRISPR Cas repeat nucleotide sequence. In some embodiments, a portion of a repeat sequence may comprise a pseudoknot-like structure at its 5' end (e.g., "handle").

A "spacer sequence" as used herein is a nucleotide sequence that is complementary to portion of a target nucleic acid (e.g., target DNA) (e.g., protospacer). In some embodiments, the spacer sequences is complementary to a portion of consecutive nucleotides of a *HY5* gene,  
10 wherein the *HY5* gene (a) comprises a sequence having at least 80% sequence identity (e.g., at least about 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 99 or 100% sequence identity) to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO: 73**; (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NO:75-105**, optionally at least 80% sequence identity to any one of **SEQ ID**  
15 **NOs:79-81, 83-85, 87-89, 91-93, 95-97, 99-101, or 103-105**; (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74** and/or, (d) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, optionally wherein the sequence identity may be at least 85% or at least 90% or it may be at least 95%, optionally the  
20 sequence identity may be at least 100%. A spacer sequence can be fully complementary or substantially complementary (e.g., at least about 70% complementary (e.g., about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%)) to a target nucleic acid. In some embodiments, the spacer sequence can have one, two, three, four, or five  
25 mismatches as compared to the target nucleic acid, which mismatches can be contiguous or noncontiguous. In some embodiments, the spacer sequence can have 70% complementarity to a target nucleic acid. In other embodiments, the spacer nucleotide sequence can have 80% complementarity to a target nucleic acid. In still other embodiments, the spacer nucleotide sequence can have 85%, 90%, 95%, 96%, 97%, 98%, 99% or 99.5% complementarity, and the  
30 like, to the target nucleic acid (protospacer). In some embodiments, the spacer sequence is 100% complementary to the target nucleic acid. A spacer sequence may have a length from about 15 nucleotides to about 30 nucleotides (e.g., 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, or 30 nucleotides, or any range or value therein). Thus, in some embodiments, a spacer sequence may have complete complementarity or substantial complementarity over a



region of a target nucleic acid (e.g., protospacer) that is at least about 15 nucleotides to about 30 nucleotides in length. In some embodiments, the spacer is about 20 nucleotides in length. In some embodiments, the spacer is about 21, 22, or 23 nucleotides in length. In some embodiments, a spacer sequence may comprise any one of the sequences of **SEQ ID NOs:107-113**, or any combination thereof.

In some embodiments, the 5' region of a spacer sequence of a guide nucleic acid may be identical to a target DNA, while the 3' region of the spacer may be substantially complementary to the target DNA (such as a spacer of a Type V CRISPR-Cas system), or the 3' region of a spacer sequence of a guide nucleic acid may be identical to a target DNA, while the 5' region of the spacer may be substantially complementary to the target DNA (such as a spacer of a Type II CRISPR-Cas system), and therefore, the overall complementarity of the spacer sequence to the target DNA may be less than 100%. Thus, for example, in a guide for a Type V CRISPR-Cas system, the first 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 nucleotides in the 5' region (i.e., seed region) of, for example, a 20 nucleotide spacer sequence may be 100% complementary to the target DNA, while the remaining nucleotides in the 3' region of the spacer sequence are substantially complementary (e.g., at least about 70% complementary) to the target DNA. In some embodiments, the first 1 to 8 nucleotides (e.g., the first 1, 2, 3, 4, 5, 6, 7, 8, nucleotides, and any range therein) of the 5' end of the spacer sequence may be 100% complementary to the target DNA, while the remaining nucleotides in the 3' region of the spacer sequence are substantially complementary (e.g., at least about 50% complementary (e.g., 50%, 55%, 60%, 65%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or more)) to the target DNA.

As a further example, in a guide for a Type II CRISPR-Cas system, the first 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 nucleotides in the 3' region (i.e., seed region) of, for example, a 20 nucleotide spacer sequence may be 100% complementary to the target DNA, while the remaining nucleotides in the 5' region of the spacer sequence are substantially complementary (e.g., at least about 70% complementary) to the target DNA. In some embodiments, the first 1 to 10 nucleotides (e.g., the first 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 nucleotides, and any range therein) of the 3' end of the spacer sequence may be 100% complementary to the target DNA, while the remaining nucleotides in the 5' region of the spacer sequence are substantially complementary (e.g., at least about 50% complementary (e.g., at least about 50%, 55%, 60%, 65%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%,

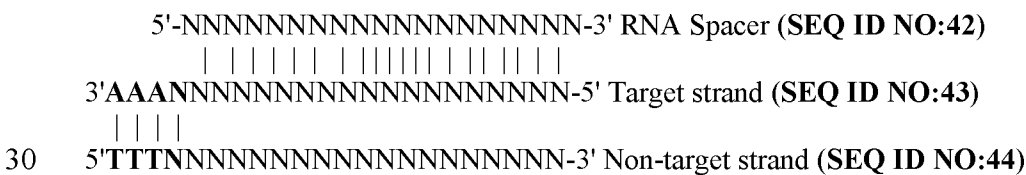
88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or more or any range or value therein)) to the target DNA.

In some embodiments, a seed region of a spacer may be about 8 to about 10 nucleotides in length, about 5 to about 6 nucleotides in length, or about 6 nucleotides in length.

5 As used herein, a "target nucleic acid", "target DNA," "target nucleotide sequence," "target region," or a "target region in the genome" refers to a region of a plant's genome that is fully complementary (100% complementary) or substantially complementary (e.g., at least 70% complementary (e.g., 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 10 98%, 99%, or more)) to a spacer sequence in a guide nucleic acid of this invention. A target region useful for a CRISPR-Cas system may be located immediately 3' (e.g., a Type V CRISPR-Cas system) or immediately 5' (e.g., a Type II CRISPR-Cas system) to a PAM sequence in the genome of the organism (e.g., a plant genome). A target region may be selected from any region of at least 15 consecutive nucleotides (e.g., 16, 17, 18, 19, 20, 21, 22, 23, 24, 15 25, 26, 27, 28, 29, 30 nucleotides, and the like) located immediately adjacent to a PAM sequence.

A "protospacer sequence" refers to the target double stranded DNA and specifically to the portion of the target DNA (e.g., or target region in the genome) that is fully or substantially complementary (and hybridizes) to the spacer sequence of the CRISPR repeat-spacer sequences 20 (e.g., guide nucleic acids, CRISPR arrays, crRNAs).

In the case of Type V CRISPR-Cas (e.g., Cas12a) systems and Type II CRISPR-Cas (Cas9) systems, the protospacer sequence is flanked by (e.g., immediately adjacent to) a protospacer adjacent motif (PAM). For Type IV CRISPR-Cas systems, the PAM is located at the 5' end on the non-target strand and at the 3' end of the target strand (see below, as an 25 example).



In the case of Type II CRISPR-Cas (e.g., Cas9) systems, the PAM is located immediately 3' of the target region. The PAM for Type I CRISPR-Cas systems is located 5' of the target strand. There is no known PAM for Type III CRISPR-Cas systems. Makarova et al. 35 describes the nomenclature for all the classes, types, and subtypes of CRISPR systems (*Nature*

*Reviews Microbiology* 13:722–736 (2015)). Guide structures and PAMs are described in by R. Barrangou (*Genome Biol.* 16:247 (2015)).

Canonical Cas12a PAMs are T rich. In some embodiments, a canonical Cas12a PAM sequence may be 5'-TTN, 5'-TTTN, or 5'-TTTTV. In some embodiments, canonical Cas9 (e.g.,  
5 *S. pyogenes*) PAMs may be 5'-NGG-3'. In some embodiments, non-canonical PAMs may be used but may be less efficient.

Additional PAM sequences may be determined by those skilled in the art through established experimental and computational approaches. Thus, for example, experimental approaches include targeting a sequence flanked by all possible nucleotide sequences and  
10 identifying sequence members that do not undergo targeting, such as through the transformation of target plasmid DNA (Esvelt et al. 2013. *Nat. Methods* 10:1116-1121; Jiang et al. 2013. *Nat. Biotechnol.* 31:233-239). In some aspects, a computational approach can include performing BLAST searches of natural spacers to identify the original target DNA sequences in  
15 bacteriophages or plasmids and aligning these sequences to determine conserved sequences adjacent to the target sequence (Briner and Barrangou. 2014. *Appl. Environ. Microbiol.* 80:994-1001; Mojica et al. 2009. *Microbiology* 155:733-740).

In some embodiments, the present invention provides expression cassettes and/or vectors comprising the nucleic acid constructs of the invention (e.g., one or more components of an editing system of the invention). In some embodiments, expression cassettes and/or vectors  
20 comprising the nucleic acid constructs of the invention and/or one or more guide nucleic acids may be provided. In some embodiments, a nucleic acid construct of the invention encoding a base editor (e.g., a construct comprising a CRISPR-Cas effector protein and a deaminase domain (e.g., a fusion protein)) or the components for base editing (e.g., a CRISPR-Cas effector protein fused to a peptide tag or an affinity polypeptide, a deaminase domain fused to a peptide tag or an  
25 affinity polypeptide, and/or a UGI fused to a peptide tag or an affinity polypeptide), may be comprised on the same or on a separate expression cassette or vector from that comprising the one or more guide nucleic acids. When the nucleic acid construct encoding a base editor or the components for base editing is/are comprised on separate expression cassette(s) or vector(s) from that comprising the guide nucleic acid, a target nucleic acid may be contacted with (e.g.,  
30 provided with) the expression cassette(s) or vector(s) encoding the base editor or components for base editing in any order from one another and the guide nucleic acid, e.g., prior to, concurrently with, or after the expression cassette comprising the guide nucleic acid is provided (e.g., contacted with the target nucleic acid).

Fusion proteins of the invention may comprise sequence-specific nucleic acid binding domains, CRISPR-Cas polypeptides, and/or deaminase domains fused to peptide tags or affinity polypeptides that interact with the peptide tags, as known in the art, for use in recruiting the deaminase to the target nucleic acid. Methods of recruiting may also comprise guide nucleic acids linked to RNA recruiting motifs and deaminases fused to affinity polypeptides capable of interacting with RNA recruiting motifs, thereby recruiting the deaminase to the target nucleic acid. Alternatively, chemical interactions may be used to recruit polypeptides (e.g., deaminases) to a target nucleic acid.

A peptide tag (e.g., epitope) useful with this invention may include, but is not limited to, a GCN4 peptide tag (e.g., Sun-Tag), a c-Myc affinity tag, an HA affinity tag, a His affinity tag, an S affinity tag, a methionine-His affinity tag, an RGD-His affinity tag, an octapeptide sold under the tradename FLAG®, a strep tag or strep tag II, a V5 tag, and/or a VSV-G epitope. In some embodiments, a peptide tag may also include phosphorylated tyrosines in specific sequence contexts recognized by SH2 domains, characteristic consensus sequences containing phosphoserines recognized by 14-3-3 proteins, proline rich peptide motifs recognized by SH3 domains, PDZ protein interaction domains or the PDZ signal sequences, and an AGO hook motif from plants. Peptide tags are disclosed in WO 2018/136783 and U.S. Patent Application Publication No. 2017/0219596, which are incorporated by reference for their disclosures of peptide tags. Any epitope that may be linked to a polypeptide and for which there is a corresponding affinity polypeptide that may be linked to another polypeptide may be used with this invention as a peptide tag. A peptide tag may comprise or be present in one copy or in 2 or more copies of the peptide tag (e.g., multimerized peptide tag or multimerized epitope) (e.g., about 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 9, 20, 21, 22, 23, 24, or 25 or more peptide tags). When multimerized, the peptide tags may be fused directly to one another or they may be linked to one another via one or more amino acids (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20 or more amino acids, optionally about 3 to about 10, about 4 to about 10, about 5 to about 10, about 5 to about 15, or about 5 to about 20 amino acids, and the like, and any value or range therein. In some embodiments, an affinity polypeptide that interacts with/binds to a peptide tag may be an antibody. In some embodiments, the antibody may be a scFv antibody. In some embodiments, an affinity polypeptide that binds to a peptide tag may be synthetic (e.g., evolved for affinity interaction) including, but not limited to, an affibody, an anticalin, a monobody and/or a DARPin (see, e.g., Sha et al., *Protein Sci.* 26(5):910-924 (2017)); Gilbreth (*Curr Opin Struc Biol* 22(4):413-420 (2013)), U.S. Patent No. 9,982,053, each of which are incorporated by reference in their entireties for the teachings relevant to affibodies,

anticalins, monobodies and/or DARPins. Example peptide tag sequences and their affinity polypeptides include, but are not limited to, the amino acid sequences of **SEQ ID NOs:45-47**.

In some embodiments, a guide nucleic acid may be linked to an RNA recruiting motif, and a polypeptide to be recruited (e.g., a deaminase) may be fused to an affinity polypeptide that  
5 binds to the RNA recruiting motif, wherein the guide binds to the target nucleic acid and the RNA recruiting motif binds to the affinity polypeptide, thereby recruiting the polypeptide to the guide and contacting the target nucleic acid with the polypeptide (e.g., deaminase). In some  
10 RNA recruiting motifs and their affinity polypeptides include, but are not limited to, the sequences of **SEQ ID NOs:48-58**.

In some embodiments, a polypeptide fused to an affinity polypeptide may be a reverse transcriptase and the guide nucleic acid may be an extended guide nucleic acid linked to an  
15 RNA recruiting motif. In some embodiments, an RNA recruiting motif may be located on the 3' end of the extended portion of an extended guide nucleic acid (e.g., 5'-3', repeat-spacer-extended portion (RT template-primer binding site)-RNA recruiting motif). In some  
20 embodiments, an RNA recruiting motif may be embedded in the extended portion.

In some embodiments of the invention, an extended guide RNA and/or guide RNA may be linked to one or to two or more RNA recruiting motifs (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 or  
25 more motifs; e.g., at least 10 to about 25 motifs), optionally wherein the two or more RNA recruiting motifs may be the same RNA recruiting motif or different RNA recruiting motifs. In some embodiments, an RNA recruiting motif and corresponding affinity polypeptide may include, but is not limited, to a telomerase Ku binding motif (e.g., Ku binding hairpin) and the  
30 corresponding affinity polypeptide Ku (e.g., Ku heterodimer), a telomerase Sm7 binding motif and the corresponding affinity polypeptide Sm7, an MS2 phage operator stem-loop and the corresponding affinity polypeptide MS2 Coat Protein (MCP), a PP7 phage operator stem-loop and the corresponding affinity polypeptide PP7 Coat Protein (PCP), an SfMu phage Com stem-loop and the corresponding affinity polypeptide Com RNA binding protein, a PUF binding site (PBS) and the affinity polypeptide Pumilio/fem-3 mRNA binding factor (PUF), and/or a  
synthetic RNA-aptamer and the aptamer ligand as the corresponding affinity polypeptide. In  
some embodiments, the RNA recruiting motif and corresponding affinity polypeptide may be an MS2 phage operator stem-loop and the affinity polypeptide MS2 Coat Protein (MCP). In some  
embodiments, the RNA recruiting motif and corresponding affinity polypeptide may be a PUF binding site (PBS) and the affinity polypeptide Pumilio/fem-3 mRNA binding factor (PUF).

In some embodiments, the components for recruiting polypeptides and nucleic acids may those that function through chemical interactions that may include, but are not limited to, rapamycin-inducible dimerization of FRB – FKBP; Biotin-streptavidin; SNAP tag; Halo tag; CLIP tag; DmrA-DmrC heterodimer induced by a compound; bifunctional ligand (e.g., fusion of  
5 two protein-binding chemicals together, e.g., dihydrofolate reductase (DHFR).

In some embodiments, the nucleic acid constructs, expression cassettes or vectors of the invention that are optimized for expression in a plant may be about 70% to 100% identical (e.g., about 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78%, 79%, 80%, 81%, 82%, 83%, 84%,  
10 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.5% or 100%) to the nucleic acid constructs, expression cassettes or vectors comprising the same polynucleotide(s) but which have not been codon optimized for expression in a plant.

Further provided herein are cells comprising one or more polynucleotides, guide nucleic acids, nucleic acid constructs, expression cassettes or vectors of the invention.

The nucleic acid constructs of the invention (e.g., a construct comprising a sequence  
15 specific nucleic acid binding domain, a CRISPR-Cas effector domain, a deaminase domain, reverse transcriptase (RT), RT template and/or a guide nucleic acid, etc) and expression cassettes/vectors comprising the same may be used as an editing system of this invention for modifying target nucleic acids and/or their expression.

A target nucleic acid of any plant or plant part (or groupings of plants, for example, into  
20 a genus or higher order classification) may be modified (e.g., mutated, e.g., base edited, cleaved, nicked, etc.) using the polypeptides, polynucleotides, ribonucleoproteins (RNPs), nucleic acid constructs, expression cassettes, and/or vectors of the invention including an angiosperm, a gymnosperm, a monocot, a dicot, a C3, C4, CAM plant, a bryophyte, a fern and/or fern ally, a microalgae, and/or a macroalgae. A plant and/or plant part that may be modified as described  
25 herein may be a plant and/or plant part of any plant species/variety/cultivar. In some embodiments, a plant that may be modified as described herein is a monocot. In some embodiments, a plant that may be modified as described herein is a dicot.

The term "plant part," as used herein, includes but is not limited to reproductive tissues  
(e.g., petals, sepals, stamens, pistils, receptacles, anthers, pollen, flowers, fruits, flower bud,  
30 ovules, seeds, embryos, nuts, kernels, ears, cobs and husks); vegetative tissues (e.g., petioles, stems, roots, root hairs, root tips, pith, coleoptiles, stalks, shoots, branches, bark, apical meristem, axillary bud, cotyledon, hypocotyls, and leaves); vascular tissues (e.g., phloem and xylem); specialized cells such as epidermal cells, parenchyma cells, chlorenchyma cells, schlerenchyma cells, stomates, guard cells, cuticle, mesophyll cells; callus tissue; and cuttings.

The term "plant part" also includes plant cells, including plant cells that are intact in plants and/or parts of plants, plant protoplasts, plant tissues, plant organs, plant cell tissue cultures, plant calli, plant clumps, and the like. As used herein, "shoot" refers to the above ground parts including the leaves and stems. As used herein, the term "tissue culture" encompasses cultures  
5 of tissue, cells, protoplasts, and callus.

As used herein, "plant cell" refers to a structural and physiological unit of the plant, which typically comprise a cell wall but also includes protoplasts. A plant cell of the present invention can be in the form of an isolated single cell or can be a cultured cell or can be a part of a higher-organized unit such as, for example, a plant tissue (including callus) or a plant organ.  
10 In some embodiments, a plant cell can be an algal cell. A "protoplast" is an isolated plant cell without a cell wall or with only parts of the cell wall. Thus, in some embodiments of the invention, a transgenic cell comprising a nucleic acid molecule and/or nucleotide sequence of the invention is a cell of any plant or plant part including, but not limited to, a root cell, a leaf cell, a tissue culture cell, a seed cell, a flower cell, a fruit cell, a pollen cell, and the like. In  
15 some aspects of the invention, the plant part can be a plant germplasm. In some aspects, a plant cell can be non-propagating plant cell that does not regenerate into a plant.

"Plant cell culture" means cultures of plant units such as, for example, protoplasts, cell culture cells, cells in plant tissues, pollen, pollen tubes, ovules, embryo sacs, zygotes, and embryos at various stages of development.

As used herein, a "plant organ" is a distinct and visibly structured and differentiated part of a plant such as a root, stem, leaf, flower bud, or embryo.  
20

"Plant tissue" as used herein means a group of plant cells organized into a structural and functional unit. Any tissue of a plant *in planta* or in culture is included. This term includes, but is not limited to, whole plants, plant organs, plant seeds, tissue culture and any groups of plant  
25 cells organized into structural and/or functional units. The use of this term in conjunction with, or in the absence of, any specific type of plant tissue as listed above or otherwise embraced by this definition is not intended to be exclusive of any other type of plant tissue.

In some embodiments of the invention, a transgenic tissue culture or transgenic plant cell culture is provided, wherein the transgenic tissue or cell culture comprises a nucleic acid  
30 molecule/nucleotide sequence of the invention. In some embodiments, transgenes may be eliminated from a plant developed from the transgenic tissue or cell by breeding of the transgenic plant with a non-transgenic plant and selecting among the progeny for the plants comprising the desired gene edit and not the transgenes used in producing the edit.

Any plant comprising an endogenous *HY5* gene, wherein the *HY5* gene is capable of regulating a Shade Avoidance Response (SAR) in the plant may be modified as described herein to reduce/attenuate or eliminate SAR in the plant. In some embodiments, a plant may be a monocot. In some embodiments, a plant may be a dicot.

5 Non-limiting examples of plants that may be modified as described herein may include turf grasses (e.g., bluegrass, bentgrass, ryegrass, fescue), feather reed grass, tufted hair grass, miscanthus, arundo, switchgrass, vegetable crops, including artichokes, kohlrabi, arugula, leeks, asparagus, lettuce (e.g., head, leaf, romaine), malanga, melons (e.g., muskmelon, watermelon, crenshaw, honeydew, cantaloupe), cole crops (e.g., brussels sprouts, cabbage, cauliflower, 10 broccoli, collards, kale, chinese cabbage, bok choy), cardoni, carrots, napa, okra, onions, celery, parsley, chick peas, parsnips, chicory, peppers, potatoes, cucurbits (e.g., marrow, cucumber, zucchini, squash, pumpkin, honeydew melon, watermelon, cantaloupe), radishes, dry bulb onions, rutabaga, eggplant, salsify, escarole, shallots, endive, garlic, spinach, green onions, squash, greens, beet (sugar beet and fodder beet), sweet potatoes, chard, horseradish, tomatoes, 15 turnips, and spices; a fruit crop such as apples, apricots, cherries, nectarines, peaches, pears, plums, prunes, cherry, quince, fig, nuts (e.g., chestnuts, pecans, pistachios, hazelnuts, pistachios, peanuts, walnuts, macadamia nuts, almonds, and the like), citrus (e.g., clementine, kumquat, orange, grapefruit, tangerine, mandarin, lemon, lime, and the like), blueberries, black raspberries, boysenberries, cranberries, currants, gooseberries, loganberries, raspberries, 20 strawberries, blackberries, grapes (wine and table), avocados, bananas, kiwi, persimmons, pomegranate, pineapple, tropical fruits, pomes, melon, mango, papaya, and lychee, a field crop plant such as clover, alfalfa, timothy, evening primrose, meadow foam, corn/maize (field, sweet, popcorn), hops, jojoba, buckwheat, safflower, quinoa, wheat, rice, barley, rye, millet, sorghum, oats, triticale, sorghum, tobacco, kapok, a leguminous plant (beans (e.g., green and dried), 25 lentils, peas, soybeans), an oil plant (rape, canola, mustard, poppy, olive, sunflower, coconut, castor oil plant, cocoa bean, groundnut, oil palm), duckweed, *Arabidopsis*, a fiber plant (cotton, flax, hemp, jute), *Cannabis* (e.g., *Cannabis sativa*, *Cannabis indica*, and *Cannabis ruderalis*), lauraceae (cinnamon, camphor), or a plant such as coffee, sugar cane, tea, and natural rubber plants; and/or a bedding plant such as a flowering plant, a cactus, a succulent and/or an 30 ornamental plant (e.g., roses, tulips, violets), as well as trees such as forest trees (broad-leaved trees and evergreens, such as conifers; e.g., elm, ash, oak, maple, fir, spruce, cedar, pine, birch, cypress, eucalyptus, willow), as well as shrubs and other nursery stock. In some embodiments, the nucleic acid constructs of the invention and/or expression cassettes and/or vectors encoding the same may be used to modify maize, soybean, wheat, canola, rice, tomato, pepper, or



sunflower, and the like.

In some embodiments, a plant that may be modified as described herein may include, but is not limited to, corn, soy, canola, wheat, rice, cotton, sugarcane, sugar beet, barley, oats, alfalfa, sunflower, safflower, oil palm, sesame, coconut, tobacco, potato, sweet potato, cassava,  
 5 coffee, apple, plum, apricot, peach, cherry, pear, fig, banana, citrus, cocoa, avocado, olive, almond, walnut, strawberry, watermelon, pepper, grape, tomato, cucumber, blackberry, raspberry, black raspberry or a *Brassica* spp. (e.g., *B. napus*, *B. oleraceae*, *B. rapa*, *B. juncea*, and/or *B. nigra*).

In some embodiments, a plant that may be modified as described herein is a corn plant  
 10 (i.e., maize, *Zea mays*). In some embodiments, a plant that may be modified as described herein is a wheat plant (e.g., *Triticum aestivum*, *T. durum*, and/or *T. compactum*).

The invention will now be described with reference to the following examples. It should be appreciated that these examples are not intended to limit the scope of the claims to the invention but rather are intended to be exemplary of certain embodiments. Any variations in the  
 15 exemplified methods that occur to the skilled artisan are intended to fall within the scope of the invention.

**EXAMPLES**

**Example 1: Design of the genomic editing construct for *HY5***

A strategy for generating edits in the *HY5* gene of *Zea mays* (e.g., Zm00001d015743  
 20 (SEQ ID NO:72) was developed. In one strategy, the constructs were developed for generating a deletion at the N-terminus of the HY5 polypeptide. To generate a range of alleles, multiple CRISPR guide nucleic acids comprising spacers (SEQ ID NOs:107-113)(see Table 1) having complementarity to targets within the *HY5* gene were designed and placed into a construct. Lines carrying edits in the *HY5* gene were screened and those that showed about 10% of the  
 25 sequencing reads having edits in the targeted gene were advanced to the next generation.

**Table 1.** Example spacers for targeting *HY5* gene

Spacer	SEQ ID NO:
PWsp1759 - AGCAGCGAGAGGTCGTCCAG	107
PWsp1761- GCAGCGAGAGGTCGTCCAGC	108
PWsp1763 - CGTGGTGCCCGGAGCTGGAC	109
PWsp1760 - TGTCACGTGGTGCCCGGAG	110
PWsp1762 - GCACTCTCCTTATCTCATCG	111
PWsp1757 - GGCCAGCTCCGGCACTCTC	112
PWsp1758 - GGCAACTCCAGGCCAGCTC	113

**Table 2.** Vector region 1 for deletion (pWISE5210)

Sequence	SEQ ID NO:
PWsp1759 - AGCAGCGAGAGGTCGTCCAG	107
PWsp1761 - GCAGCGAGAGGTCGTCCAGC	108
PWsp1763 - CGTGGTGCCCGGAGCTGGAC	109
PWsp1760 - TGTCCACGTGGTGCCCGGAG	110

**Table 3.** Vector region 2 for deletion (pWISE5212)

Sequence	SEQ ID NO:
PWsp1762 - GCACTCTCCTTATCTCATCG	111
PWsp1757 - GGCCAGCTCCGGCACTCTC	112
PWsp1758 - GGCAACTCCAGGCCAGCTC	113

**Table 4.** Vector deletion between region 1 and 2 (pWISE5211)

Sequence	SEQ ID NO:
PWsp1759 - AGCAGCGAGAGGTCGTCCAG	107
PWsp1761 - GCAGCGAGAGGTCGTCCAGC	108
PWsp1763 - CGTGGTGCCCGGAGCTGGAC	109
PWsp1760 - TGTCCACGTGGTGCCCGGAG	110
PWsp1762 - GCACTCTCCTTATCTCATCG	111
PWsp1757 - GGCCAGCTCCGGCACTCTC	112
PWsp1758 - GGCAACTCCAGGCCAGCTC	113

5

**Example 2: Edited alleles**

Edited alleles of the *HY5* gene Zm00001d015743 were generated as described in Example 1 and are further described Table 5.

**Table 5.** Edited alleles

Allele name	Edit description	Notes
Allele A (SEQ ID NO:114)	3 bp deletion (CCA) starting at position 2159 of SEQ ID NO:72	In frame deletion which leads to deletion of the amino acid “S” at position 16 (of SEQ ID NO:74), and the conversion of the amino acid “S” at position 18 (of SEQ ID NO:74) to amino acid “C”, giving rise to the amino acid sequence (SEQ ID NO:115)
Allele B (SEQ ID NO:116)	6 bp deletion (AGCTCC) starting at position 2161 of SEQ ID NO:72	In frame deletion of the amino acids “SS” at position 16-17 of SEQ ID NO:74 giving rise to the amino acid sequence (SEQ ID NO:117)
Allele C (SEQ ID NO:118)	6 bp deletion (GTCCAG) starting at position 2157 of SEQ ID NO:72	In frame deletion of the amino acids “SS” at position 16-17 of SEQ ID NO:74 giving rise

		to the amino acid sequence (SEQ ID NO:117)
Allele D (SEQ ID NO:119)	8 bp deletion (GTCCAGCT) starting at position 2157 of SEQ ID NO:72	Out of frame mutation leading to an early stop codon, (SEQ ID NO:120)
Allele E (SEQ ID NO:121)	9 bp deletion (GTCCAGCTC) starting at position 2157 of SEQ ID NO:72	In frame deletion of the amino acids "SSS" at position 16-18 of SEQ ID NO:74 giving rise to the amino acid sequence (SEQ ID NO:122)
Allele F (SEQ ID NO:123)	18 bp deletion (CGAGAGGTCGTCCAGCTC; SEQ ID NO:141) starting at position 2148 of SEQ ID NO:72	In frame deletion of the amino acids "SSSERS" (SEQ ID NO:142) at position 11-16 of SEQ ID NO:74 giving rise to the amino acid sequence (SEQ ID NO:124)
Allele G (SEQ ID NO:125)	34 bp deletion (AGGTCGTCCAGCTCCGGGCAC CACGTGGACATGG; SEQ ID NO:143) starting at position 2152 of SEQ ID NO:72	Out of frame mutation leading to an early stop codon, (SEQ ID NO:126)
Allele H (SEQ ID NO:127)	6 bp deletion (GCTCCG) starting at position 2162 of SEQ ID NO:72	In frame deletion which replaces the amino acids "SSG" at position 18-20 of SEQ ID NO:74 with the amino acid "R" giving rise to the amino acid sequence (SEQ ID NO:128)

### Example 3: Shade avoidance assay

Corn seeds were sown directly into soil and placed in the greenhouse for 4 days to allow seeds to germinate. On day 5, the flats of germinating seeds were transferred to the shade avoidance tents placed in the growth chamber set for 16h light/8h dark, 28°C day/23°C night. The control tent received light in the red:far red ratio of 1-1.3. The shade simulation tent received light in the red:far red ratio of 0.2-0.22. Both tents were set to maintain the light intensity of 250-300  $\mu\text{mol}/\text{m}^2/\text{s}$  for 16 hours, followed by 8-hour dark period. The temperature in the tents was set at 78F/72F during the light/dark cycle, respectively. Once the seedlings reach the V2 stage, the seedling were imaged to measure V1 sheath height, V2 sheath height.

### Example 4: Shade avoidance response of edited alleles A through H

Plants containing the edited alleles of *HY5* which are described in **Example 2** were evaluated for changes in shade avoidance as described in **Example 3**. The observations are summarized in **Table 6** and **Table 7** and demonstrate that the edited alleles of *HY5* have altered the plants shade avoidance response that may affect plant yield. The positive control line is a  
5 corn plant previously known to have a decreased shade avoidance response.

**Table 6.** V1 sheath height (mm)

Genotype	Treatment	Mean	Standard Deviation	Number
Wild type control	Control	49.00167	3.790986	30
Wild type control	Shade	56.28987	4.318820	23
Homozygous Allele F	Control	42.36737	5.741881	19
Homozygous Allele F	Shade	47.88506	5.187126	18
Heterozygous Allele C; Heterozygous Allele D; no wild type allele present	Control	46.79500	1.277030	7
Heterozygous Allele C; Heterozygous Allele D; no wild type allele present	Shade	54.69067	1.075833	6
Homozygous Allele E	Control	52.06700	5.387904	24
Homozygous Allele E	Shade	56.62653	3.772613	15
Homozygous Allele A	Control	47.63300	5.189810	19
Homozygous Allele A	Shade	54.02508	4.771172	13
Heterozygous Allele B; Heterozygous Allele A; no wild type allele present	Control	49.64621	7.641583	28
Heterozygous Allele B; Heterozygous Allele A; no wild type allele present	Shade	58.12200	3.622392	25
Heterozygous Allele G; Heterozygous Allele H; no wild type allele present	Control	51.29679	5.563238	24
Heterozygous Allele G; Heterozygous Allele H; no wild type allele present	Shade	55.77104	5.180882	24
Positive control line	Control	56.65744	5.081385	25
Positive control line	Shade	59.96335	4.231810	26

**Table 7.** V2 sheath height (mm)

Genotype	Treatment	Mean	Standard Deviation	Number
Wild type control	Control	95.26062	5.747566	29
Wild type control	Shade	103.72875	5.992237	24
Homozygous Allele F	Control	87.10021	12.302576	19

Homozygous Allele F	Shade	101.78160	4.758528	15
Heterozygous Allele C; Heterozygous Allele D; no wild type allele present	Control	89.08780	6.431332	10
Heterozygous Allele C; Heterozygous Allele D; no wild type allele present	Shade	102.55588	10.928794	8
Homozygous Allele E	Control	98.61325	9.944611	24
Homozygous Allele E	Shade	105.00046	4.696020	13
Homozygous Allele A	Control	93.63659	5.831212	17
Homozygous Allele A	Shade	104.10180	9.581910	15
Heterozygous Allele B; Heterozygous Allele A; no wild type allele present	Control	99.63904	8.245581	28
Heterozygous Allele B; Heterozygous Allele A; no wild type allele present	Shade	110.97040	4.510844	25
Heterozygous Allele G; Heterozygous Allele H; no wild type allele present	Control	97.77758	6.702013	24
Heterozygous Allele G; Heterozygous Allele H; no wild type allele present	Shade	107.30237	6.066834	24
Positive control line	Control	102.18076	7.144315	25
Positive control line	Shade	108.38642	5.705616	24

**Example 5: Edited alleles pWISE5211**

5 Edited alleles of the *HY5* gene Zm00001d015743 were generated as described in Example 1 and Table 4 using the vector pWISE5211. The edited alleles are further described in Table 8.

Table 8. pWISE5211 edited alleles

Allele Name	Edit Description	Notes
Allele I (SEQ ID NO:129)	357 bp deletion (SEQ ID NO:144) starting at position 2157 of SEQ ID NO:72	In frame deletion that removes the first 40 amino acids of SEQ ID NO:74 and replaces them with the amino acids “MQEQAASSRPSSSERS” (SEQ ID NO:145) to generate the amino acid sequence (SEQ ID NO:130)

**Example 6: Edited alleles pWISE5212**

10 Edited alleles of the *HY5* gene Zm00001d015743 were generated as described in Example 1 and Table 4 using the vector pWISE5212. The edited alleles are further described in Table 9.

Table 9. pWISE5212 edited alleles

Allele Name	Edit Description	Notes
Allele J (SEQ ID NO:131)	Compound deletion of 6 bp (ACGATG) starting at position 2495 of SEQ ID NO:72; and deletion of 5 bp (AGCTG) starting at position 2519 of SEQ ID NO:72	In frame deletion of “DD” at position 34-35 of SEQ ID NO:74 followed by an out of frame mutation leading to an early stop codon (SEQ ID NO:132)
Allele K (SEQ ID NO:133)	Compound deletion of 3 bp (CGA) starting at position 2493 of SEQ ID NO:72; and deletion of 13 bp (AGGAGAGTGCCGG; SEQ ID NO:146) starting at position 2506 of SEQ ID NO:72	Out of frame mutation leading to an early stop codon (SEQ ID NO:134)
Allele L (SEQ ID NO:135)	21 bp deletion (GCCGGAGCTGGGCCTG GAGTT; SEQ ID NO:147) starting at position 2514 of SEQ ID NO:72	In frame deletion of amino acids “PELGLEL” (SEQ ID NO:148) at position 41-47 of SEQ ID NO:74 giving rise the amino acid sequence (SEQ ID NO:136)
Allele M (SEQ ID NO:137)	36 bp deletion (GGAGAGCGACGATGAG ATAAGGAGAGTGCCGGA GCT; SEQ ID NO:149) starting at position 2487 of SEQ ID NO:72	In frame deletion of amino acids “ESDDEIRRVPPEL” (SEQ ID NO:150) at position 32-43 of SEQ ID NO:74 giving rise to the amino acid sequence (SEQ ID NO:138)
Allele N (SEQ ID NO:139)	8 bp deletion (CGGAGCTG) starting at position 2516 of SEQ ID NO:72	Out of frame mutation leading to an early stop codon (SEQ ID NO:140)

**Example 7: Shade avoidance response of edited alleles I through N**

5 Plants containing the edited alleles of *HY5* which are described in **Example 5** and **Example 6** were evaluated for changes in shade avoidance as described in **Example 3**. The observations are summarized in **Table 10** for sheath height.

10 An analysis of the data collected was generated by calculating the difference in sheath height between the control tent and the shade tent for an individual and this value is represented in Table 11 as the “deltaV shade” and was calculated at both the V1 and V2 growth stage. Additionally, the value “%deltaV” was calculated as the deltaV shade value as a percentage of the V sheath height in the control tent and was also calculated at both the V1 and V2 growth stage.

These observations demonstrate that the edited alleles of *HY5* have altered the plants shade avoidance response that may affect plant yield. The positive control line is a corn plant previously known to have a decreased shade avoidance response.

5 **Table 10.** V1 and V2 sheath height

Genotype	V1 sheath height (control)	V2 sheath height (control)	#of plants (control)	V1 sheath height (shade)	V2 sheath height (shade)	#of plants (shade)
WT control	44.56	89.84	25	49.63	94.72	32
Positive control for shade	44.26	88.95	13	50.16	98.44	15
Homozygous Allele I	38.53	79.32	20	42.23	80.25	25
Homozygous Allele J	41.47	91.33	11	46.51	98.02	19
Homozygous Allele K	42.70	88.37	28	50.38	96.35	31
Homozygous Allele L	48.48	91.52	27	51.05	97.53	31
Homozygous Allele M	39.58	82.21	16	47.39	94.49	23
Homozygous Allele N	39.77	80.29	24	45.17	90.33	25

**Table 11.** Shade analysis

Genotype	deltaV1 shade	deltaV2 shade	%deltaV1	%deltaV2
WT control	<b>5.07</b>	<b>4.88</b>	<b>11.38</b>	<b>5.43</b>
Positive control for shade	5.89	9.50	13.31	10.68
homozygous Allele I	3.70	0.93	9.60	1.18
Homozygous Allele J	5.03	6.69	12.14	7.32
Homozygous Allele K	7.67	7.98	17.97	9.03
Homozygous Allele L	2.57	6.00	5.30	6.56
Homozygous Allele M	7.81	12.28	19.73	14.94
Homozygous Allele N	5.40	10.03	13.57	12.49

10 In some jurisdictions, products obtained exclusively by essentially biological processes are excluded from patent protection. Accordingly, the claimed plants, plant parts and cells and their progeny can be defined as directed only to those plants, plant parts and cells and their progeny which are obtained by technical intervention (regardless of any further propagation through crossing and selection). An embodiment of the invention is directed at plants, or plant  
15 parts or progeny produced or obtainable using gene editing technology by introducing through

stable or transient transformation an RNA-specific CRISPR/Cas system directed against or targeting an *HY5* nucleotide sequence, or one or more polynucleotide sequence(s) encoding said RNA-specific CRISPR/Cas system into the plant or the plant part. Alternatively, the subject matter excluded from patentability may be disclaimed. An embodiment of the invention is

5 directed at plants, part of plants or progeny thereof comprising the alterations of the *HY5* gene as elsewhere herein described, provided that the plants, parts or plants or progeny are not obtained exclusively through essentially biological processes, wherein essentially biological processes are processes for the production of plants or animals if they consist entirely of natural phenomena such as crossing or selection.

10 The foregoing is illustrative of the present invention and is not to be construed as limiting thereof. The invention is defined by the following claims, with equivalents of the claims to be included therein.



**WHAT IS CLAIMED IS:**

1. A plant or plant part thereof comprising at least one mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene encoding a HY5 transcription factor.  
5
2. The plant or plant part thereof of claim 1, wherein the HY5 transcription factor is capable of regulating response to illumination in the plant (e.g., shade avoidance response (SAR)).
- 10 3. The plant or plant part thereof of claim 1 or claim 2, wherein the at least one mutation is in the N-terminal region of the endogenous *HY5* transcription factor.
4. The plant or plant part thereof of any one of claims 1-3, wherein the HY5 transcription factor comprises a Constitutive Photomorphogenic 1 (COP1) binding site and the at least one  
15 mutation is in the COP1 binding site of the HY5 transcription factor encoded by the *HY5* gene.
5. The plant or plant part thereof of any one of claims 1-4, wherein the at least one mutation results in a HY5 transcription factor having no or reduced COP1 binding, optionally wherein the binding is reduced by about 30% to about 100% as compared to a HY5 transcription factor that  
20 is devoid of the at least one mutation.
6. The plant or plant part thereof of any one of claims 1-5, wherein the at least one mutation is in a region of the endogenous *HY5* transcription factor having at least 80% sequence identity to **SEQ ID NO:106**.  
25
7. The plant or plant part thereof of any one of the preceding claims, wherein the endogenous *HY5* gene:
  - (a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;
  - 30 (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;
  - (c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

(d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

8. The plant or plant part thereof of any one of the preceding claims, wherein the at least  
5 one mutation is a substitution, a deletion and/or an insertion.
9. The plant or plant thereof of any one of the preceding claims, where the mutation is an  
in-frame deletion, an out-of-frame deletion, an in-frame insertion, or an out-of-frame insertion.
- 10 10. The plant or plant part thereof of claim 8 or claim 9, wherein the deletion or insertion in the  
endogenous *HY5* gene is a deletion or an insertion of 1 to about 100 base pairs.
11. The plant or plant part thereof of any one of claims 1-10, wherein the at least one  
mutation in an endogenous gene encoding a HY5 polypeptide is an out-of-frame deletion or an  
15 in-frame deletion, optionally wherein the at least one mutation is an in-frame deletion.
12. The plant or plant part thereof of any one of claims 1-8, wherein the at least one mutation  
comprises a base substitution to an A, a T, a G, or a C.
- 20 13. The plant or plant part thereof of any one of the preceding claims, wherein the at least  
one mutation in an endogenous gene encoding a HY5 polypeptide is a dominant allele or  
dominant-negative allele.
14. The plant or part thereof of any one of the preceding claims, wherein the at least one  
25 mutation is a non-natural mutation.
15. The plant or plant part thereof of any one of the preceding claims, wherein the plant is a  
monocot or dicot.
- 30 16. The plant or plant part thereof of any one of the preceding claims, wherein the plant is  
planted at an increased density without a decrease in plant yield on a per plant basis.
17. The plant or plant part thereof of claim 16, wherein the planting density is increased by  
about 5% to about 75% without a decrease in plant yield on a per plant basis.

18. The plant or plant part thereof of any one of the preceding claims, wherein the plant is corn, soy, canola, wheat, rice, cotton, sugarcane, sugar beet, barley, oats, alfalfa, sunflower, safflower, oil palm, sesame, coconut, tobacco, potato, sweet potato, cassava, coffee, apple, plum,  
5 apricot, peach, cherry, pear, fig, banana, citrus, cocoa, avocado, olive, almond, walnut, strawberry, watermelon, pepper, grape, tomato, cucumber, blackberry, raspberry, black raspberry or a *Brassica* spp.
19. The plant or plant part thereof of any one of the preceding claims, wherein the plant is a  
10 corn plant.
20. The plant or plant part thereof of any one of the preceding claims, wherein the at least one mutation in the endogenous *Elongated Hypocotyl5 (HY5)* gene results in a nucleotide sequence having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated HY5 polypeptide  
15 having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.
21. A plant cell comprising an editing system comprising:  
20 (a) a CRISPR-Cas associated effector protein; and  
(b) a guide acid (gRNA, gDNA, crRNA, crDNA) having a spacer sequence with complementarity to an endogenous target gene encoding an Elongated Hypocotyl5 (HY5) transcription factor.
22. The plant cell of claim 21, wherein the endogenous target gene encoding a HY5  
25 transcription factor polypeptide  
(a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;  
(b) comprises a region having at least 80% sequence identity to any one of the nucleotide  
30 sequences of **SEQ ID NOs:75-105**;  
(c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or  
(d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

23. The plant cell of claim 21 or claim 22, wherein the guide nucleic acid comprises a nucleotide sequence of any one of **SEQ ID NOs:107-113**.
- 5 24. The plant cell of any one of claims 21-23, wherein the plant cell is cell from corn, soy, canola, wheat, rice, cotton, sugarcane, sugar beet, barley, oats, alfalfa, sunflower, safflower, oil palm, sesame, coconut, tobacco, potato, sweet potato, cassava, coffee, apple, plum, apricot, peach, cherry, pear, fig, banana, citrus, cocoa, avocado, olive, almond, walnut, strawberry, watermelon, pepper, grape, tomato, cucumber, blackberry, raspberry, black raspberry or a  
10 *Brassica* spp.
25. The plant cell of any one of claims 21-24, wherein the plant cell is a corn cell.
26. A plant cell comprising at least one mutation in an endogenous *Elongated Hypocotyl5*  
15 (*HY5*) gene,  
wherein the at least one mutation is a substitution, insertion and/or a deletion that is introduced into the endogenous *HY5* gene using an editing system that comprises a nucleic acid binding domain that binds to a target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene:  
20 (a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;  
(b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;  
(c) encodes a polypeptide comprising a sequence having at least 80% sequence identity  
25 to the amino acid sequence of **SEQ ID NO:74**; and/or  
(d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.
27. The plant cell of claim 26, wherein the nucleic acid binding domain of the editing system  
30 is from a polynucleotide-guided endonuclease, a CRISPR-Cas endonuclease (e.g., CRISPR-Cas effector protein), a zinc finger nuclease, a transcription activator-like effector nuclease (TALEN) and/or an Argonaute protein.

28. The plant cell of claim 26 or claim 27, wherein the at least one mutation is a substitution, a deletion and/or an insertion.
29. The plant cell of any one of claims 26-28, wherein the at least one mutation is a deletion  
5 in the region of the *HY5* gene encoding the COP1 binding site, optionally wherein the encoded region comprises a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.
30. The plant cell of any one of claims 26-29, wherein the deletion or insertion in the  
10 endogenous *HY5* gene is a deletion or insertion of 1 to about 100 base pairs.
31. The plant cell of any one of claims 26-30, wherein the deletion is an in-frame deletion or an out-of-frame deletion.
- 15 32. The plant cell of any one of claims 26-31, wherein the at least one mutation is a non-natural mutation.
33. The plant cell of any one of claims 26-32, wherein the plant cell is cell from corn, soy, canola, wheat, rice, cotton, sugarcane, sugar beet, barley, oats, alfalfa, sunflower, safflower, oil  
20 palm, sesame, coconut, tobacco, potato, sweet potato, cassava, coffee, apple, plum, apricot, peach, cherry, pear, fig, banana, citrus, cocoa, avocado, olive, almond, walnut, strawberry, watermelon, pepper, grape, tomato, cucumber, blackberry, raspberry, black raspberry or a Brassica spp.
- 25 34. The plant cell of any one of claims 26-33, wherein the plant cell is a corn cell.
35. The plant cell of any one of claims 26-34, wherein the at least one mutation in the endogenous *Elongated Hypocotyl5 (HY5)* gene results in a nucleotide sequence having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129,**  
30 **131, 133, 135, 137, or 139** and/or encodes a mutated HY5 polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

36. A plant regenerated from the plant part of any one of claims 1-20 or from the plant cell of any one of claims 21-35.
37. The plant of any one of claims 1-20 or 36, wherein the plant comprises an attenuated  
5 Shade Avoidance Response.
38. The plant of claim 36 or claim 37, wherein the plant is corn.
39. A method of providing a plurality of plants having increased yield when each plant of the  
10 plurality of plants is planted in close proximity to one another, the method comprising planting two or more plants of any one of claims 1-20 or 36-38 in close proximity to one another, thereby providing a plurality of plants having increased yield as compared to a plurality of control plants planted in close proximity to one another.
- 15 40. A method of producing/breeding a transgene-free genome-edited (e.g., base-edited) plant, comprising:  
(a) crossing the plant of any one of claims 1-20 or 36-38 with a transgene-free plant, thereby introducing the mutation into the plant that is transgene-free; and  
(b) selecting a progeny plant that comprises the mutation but is transgene-free, thereby  
20 producing a transgene-free genome-edited (e.g., base-edited) plant.
41. A method of creating a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene in a plant, comprising:  
(a) targeting a gene editing system to a portion of the *HY5* gene that comprises a  
25 nucleotide sequence having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; and  
(b) selecting a plant that comprises a modification located in a region of the *HY5* gene having at least 80% identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**.
- 30 42. A method of generating variation in an *Elongated Hypocotyl5 (HY5)* gene, comprising:  
introducing an editing system into a plant cell, wherein the editing system is targeted to a region of a *HY5* gene that encodes a HY5 transcription factor, and  
contacting the region of the *HY5* gene with the editing system, thereby introducing a mutation into the *HY5* gene and generating variation in the *HY5* gene of the plant cell.

43. The method of claim 42, wherein the *HY5* gene comprises a nucleotide sequence having at least 80% sequence identity to any one of **SEQ ID NO:72** or **SEQ ID NO:73**, and/or encodes an amino acid sequence having at least 80% sequence identity to **SEQ ID NO:74**.

5

44. The method of claim 42 or claim 43, wherein the region of the *HY5* gene that is targeted comprises at least 80% sequence identity to a nucleotide sequence of any one of **SEQ ID NOs:75-105**.

10 45. The method of any one of claims 42-44, wherein contacting the region of the endogenous *HY5* gene in the plant cell with the editing system produces a plant cell comprising in its genome an edited endogenous *HY5* gene, the method further comprising (a) regenerating a plant from the plant cell; (b) selfing the plant to produce progeny plants (E1); (c) assaying the progeny plants of (b) for reduced shade avoidance response (SAR)/shade avoidance syndrome (SAS); and (d)  
15 selecting the progeny plants exhibiting reduced shade avoidance response (SAR)/shade avoidance syndrome (SAS) as compared to a control plant devoid of the mutation.

46. The method of claim 45, further comprising (e) selfing the selected progeny plants of (d) to produce progeny plants (E2); (f) assaying the progeny plants of (e) for reduced shade  
20 avoidance response (SAR)/shade avoidance syndrome (SAS); and (g) selecting the progeny plants exhibiting reduced shade avoidance response (SAR)/shade avoidance syndrome (SAS) as compared to a control plant, optionally repeating (e) through (g) one or more additional times.

47. A method of detecting a mutant *Elongated Hypocotyl5 (HY5)* gene (a mutation in an  
25 endogenous *HY5* gene) in a plant comprising detecting in the genome of the plant a *HY5* gene having at least one mutation in a region having at least 80% sequence identity to a nucleotide sequence of any one of **SEQ ID NOs: 75-105**.

48. The method of claim 47, wherein the mutant *HY5* gene that is detected comprises a  
30 nucleic acid sequence having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated *HY5* polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

49. A method for editing a specific site in the genome of a plant cell, the method comprising cleaving, in a site-specific manner, a target site within an endogenous *Elongated Hypocotyl5* (*HY5*) gene in the plant cell, the endogenous *HY5* gene:

(a) comprising a sequence having at least 80% sequence identity to the nucleotide  
5 sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

(b) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

(c) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

10 (d) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby generating an edit in the endogenous *HY5* gene of the plant cell.

50. The method of claim 49, wherein the *HY5* gene encodes a HY5 transcription factor that comprises a COP1 binding region and the edit results in a mutation in the COP1 binding region  
15 of the HY5 transcription factor.

51. The method of claim 49 or claim 50, wherein the edit results in a HY5 transcription factor having no or reduced COP1 binding, optionally wherein the binding is reduced by about 30% to about 100% as compared to a control plant that is devoid of the edit.  
20

52. The method of any one of claims 49-51, wherein the edit is in a region of the HY5 transcription factor having at least 80% sequence identity to **SEQ ID NO:106**.

53. The method of any one of claims 49-52, wherein the edit in the endogenous *HY5* gene results in the endogenous *HY5* gene having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated HY5 polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.  
25

54. The method of any one of claims 49-53, further comprising regenerating a plant from the plant cell comprising the edit in the endogenous *HY5* gene to produce a plant comprising the edit in its endogenous *HY5* gene.  
30



55. The method of claim 54, wherein the plant comprising the edit in its endogenous *HY5* gene has an attenuated Shade Avoidance Response compared to a control plant devoid of the edit.

5 56. A method for making a plant, comprising:

(a) contacting a population of plant cells that comprise an endogenous gene encoding an Elongated Hypocotyl5 (HY5) transcription factor with a nuclease targeted to the endogenous gene, wherein the nuclease is linked to a nucleic acid binding domain that binds to a target site within the endogenous gene, the endogenous gene

10 (i) comprising a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

(ii) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

15 (iii) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

(iv) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**;

20 (b) selecting a plant cell from the population of plant cells in which the endogenous gene encoding a HY5 transcription factor has been mutated, thereby producing a plant cell comprising a mutation in the endogenous gene; and

(c) growing the selected plant cell into a plant comprising the mutation in the endogenous gene encoding a HY5 transcription factor, optionally wherein the mutation reduces or eliminates the ability of the HY5 transcription factor to bind a Constitutive Photomorphogenic 1 (COP1) polypeptide.

25

57. A method for reducing/suppressing a Shade Avoidance Response in a plant, comprising:

(a) contacting a plant cell comprising an endogenous *Elongated Hypocotyl5 (HY5)* gene with a nuclease targeted to the endogenous gene, wherein the nuclease is linked to a nucleic acid binding domain that binds to a target site within the endogenous gene, the endogenous gene:

30 (i) comprising a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

(ii) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

(iii) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

(iv) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby producing a plant cell comprising a mutation in the endogenous *HY5* gene; and

(b) growing the plant cell into a plant, thereby reducing/suppressing the Shade Avoidance Response in the plant.

58. A method for producing a plant or part thereof comprising at least one cell having a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene, the method comprising contacting a target site within the endogenous *HY5* gene in the plant or plant part with a nuclease comprising a cleavage domain and a nucleic acid binding domain, wherein the nucleic acid binding domain of the nuclease binds to a target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene:

(a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

(b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

(c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

(d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby producing a plant or part thereof comprising at least one cell having a mutation in the endogenous *HY5* gene.

59. A method of producing a plant or part thereof comprising a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene that produces a mutated HY5 transcription factor having reduced binding by a Constitutive Photomorphogenic 1 (COP1) polypeptide, the method comprising contacting a target site within the endogenous *HY5* gene in the plant or plant part with a nuclease comprising a cleavage domain and a nucleic acid binding domain, wherein the nucleic acid binding domain of the nuclease binds to a target site within the endogenous *HY5* gene, wherein the endogenous *HY5* gene:

(a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

(b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

(c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

5 (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby producing a plant or part thereof having a mutation in the endogenous *HY5* gene that produces a mutated HY5 transcription factor having reduced binding by the COP1 polypeptide.

10 60. The method of any one of claims 56-59, wherein the nuclease cleaves the endogenous *HY5* gene, thereby introducing the mutation into the endogenous *HY5* gene, optionally wherein the mutation is introduced into a region of the *HY5* gene that encodes a COP1 binding region of a HY5 transcription factor, optionally wherein the COP1 binding region is located in the N-terminus of the encoded HY5 transcription factor.

15 61. The method of any one of claims 56-60, wherein the mutation is a non-natural mutation.

62. The method of any one of claims 56-61, wherein the mutation is a base substitution, a base insertion and/or a base deletion.

20 63. The method of any one of claims 56-62, wherein the mutation is a dominant mutation or a dominant-negative.

25 64. The method of any one of claims 56-63, wherein the mutation is an in-frame or an out-of-frame insertion or an in-frame or an out-of-frame deletion.

65. The method of any one of claims 56-64, wherein the mutation comprises a point mutation.

30 66. The method of any one of claims 56-65, wherein the mutation is a deletion of one base pair to about 100 base pairs.

67. The method of any one of claims 56-66, wherein the nuclease is a zinc finger nuclease, transcription activator-like effector nucleases (TALEN), endonuclease (e.g., FokI) or a CRISPR-Cas effector protein.

5 68. The method of any one of claims 42-46, 50-55, or 59-67, wherein the HY5 transcription factor is capable of regulating response to illumination in the plant (e.g., shade avoidance response (SAR)).

10 69. The method of any one of claims 42-46, 50-55, or 59-67, wherein the HY5 transcription factor comprises a COP1 binding region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

15 70. The method of any one of claims 56-69, wherein the plant that is produced comprises a mutated endogenous *HY5* gene having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated HY5 polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138**.

20 71. The method of any one of claims 56-70, wherein the plant that is produced exhibits a reduced Shade Avoidance Response as compared to a control plant devoid of the mutation.

25 72. The method of claim 71, wherein the plant having a reduced Shade Avoidance Response comprises at least one of the following phenotypes of increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants comprising a reduced Shade Avoidance Response as compared to a plant that does not comprise a reduced Shade Avoidance Response that is planted in close proximity with one or  
30 more plants that are devoid of the mutation and do not exhibit a reduced Shade Avoidance Response.

73. A method for modifying an endogenous *Elongated Hypocotyl5 (HY5)* gene in a corn plant or part thereof for reducing Shade Avoidance Response in the corn plant or part thereof,

the method comprising modifying a target site within the endogenous *HY5* gene in the corn plant or a part thereof, wherein the endogenous *HY5* gene:

(a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

5 (b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

(c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

10 (d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, thereby modifying the endogenous *HY5* gene and reducing Shade Avoidance Response in the corn plant or part thereof.

74. The method of claim 73, wherein the target site is within a region of the *HY5* gene having at least 80% sequence identity to a nucleotide sequence of any one of **SEQ ID NOs:75-105**.

75. A plant produced by any one of the methods of 36 or 56-75.

20 76. The plant of claim 75, wherein the plant comprises a mutated *HY5* gene, wherein the mutated *HY5* gene has at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated *HY5* polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138** as described herein.

25 77. The plant of claim 75 or claim 76, wherein the plant exhibits a reduced Shade Avoidance Response.

30 78. A guide nucleic acid that binds to a target site within an endogenous *HY5* gene, the endogenous *HY5* gene comprising a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**; comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**; encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

79. A guide nucleic acid that binds to a target site within an endogenous *HY5* gene, wherein the target site is in a region of the endogenous *HY5* gene having at least 80% sequence identity to any one of **SEQ ID NOs:75-105**.

5

80. The guide nucleic acid of claim 78 or claim 79 wherein the guide nucleic acid comprises a spacer having the nucleotide sequence of any one of **SEQ ID NOs:107-113**.

81. A system comprising the guide nucleic acid of any one of claims 78-80, and a CRISPR-Cas effector protein that associates with the guide nucleic acid.

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82. The system of claim 81, further comprising a tracr nucleic acid that associates with the guide nucleic acid and a CRISPR-Cas effector protein, optionally wherein the tracr nucleic acid and the guide nucleic acid are covalently linked.

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83. A gene editing system comprising a CRISPR-Cas effector protein in association with a guide nucleic acid, wherein the guide nucleic acid comprises a spacer sequence that binds to an *Elongated Hypocotyl5 (HY5)* gene.

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84. The gene editing system of claim 83, wherein the *HY5* gene:

(a) comprises a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

(b) comprises a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

25

(c) encodes a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

(d) encodes a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

30

85. The gene editing system of claim 83 or claim 84, wherein the guide nucleic acid comprises a spacer sequence having a nucleotide sequence of any one of **SEQ ID NOs:107-113**.

86. The gene editing system of any one of claims 83-85, further comprising a tracr nucleic acid that associates with the guide nucleic acid and a CRISPR-Cas effector protein, optionally wherein the tracr nucleic acid and the guide nucleic acid are covalently linked.

5 87. A complex comprising a CRISPR-Cas effector protein comprising a cleavage domain and a guide nucleic acid, wherein the guide nucleic acid binds to a target site within an *Elongated Hypocotyl5 (HY5)* gene, the *HY5* gene:

(a) comprising a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

10 (b) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

(c) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

15 (d) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**, wherein the cleavage domain cleaves a target strand in the *HY5* gene.

88. An expression cassette comprising (a) a polynucleotide encoding CRISPR-Cas effector protein comprising a cleavage domain and (b) a guide nucleic acid that binds to a target site within an *Elongated Hypocotyl5 (HY5)* gene, wherein the guide nucleic acid comprises a spacer  
20 sequence that is complementary to and binds to the target site within the *HY5* gene, the *HY5* gene:

(a) comprising a sequence having at least 80% sequence identity to the nucleotide sequence of **SEQ ID NO:72** or **SEQ ID NO:73**;

25 (b) comprising a region having at least 80% sequence identity to any one of the nucleotide sequences of **SEQ ID NOs:75-105**;

(c) encoding a polypeptide comprising a sequence having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:74**; and/or

(d) encoding a region having at least 80% sequence identity to the amino acid sequence of **SEQ ID NO:106**.

30 89. A nucleic acid encoding an *Elongated Hypocotyl5 (HY5)* transcription factor having a mutated COP1 binding region, optionally wherein the mutation is located in a nucleic acid having at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated *HY5* polypeptide having at

least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138.**

90. A nucleic acid comprising a mutated endogenous *Elongated Hypocotyl5 (HY5)* gene,  
5 wherein the mutated endogenous *HY5* gene has at least 90% sequence identity to any one of **SEQ ID NOs:114, 116, 118, 119, 121, 123, 125, 127, 129, 131, 133, 135, 137, or 139** and/or encodes a mutated HY5 polypeptide having at least 90% sequence identity to any one of **SEQ ID NOs:115, 117, 120, 122, 124, 126, 128, 130, 132, 134, 136, or 138.**
- 10 91. A plant or plant part thereof comprising the nucleic acid of claim 89 or claim 90.
92. A corn plant or plant part thereof comprising the nucleic acid of claim 89 or claim 90, optionally wherein the corn plant comprises a short stature/semi-dwarf phenotype.
- 15 93. A corn plant or plant part thereof comprising at least one mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene having the gene identification number (gene ID) of Zm00001d015743.
94. The plant of claim 91 or the corn plant of claim 92 or claim 93, wherein the plant or corn  
20 plant exhibits a reduced Shade Avoidance Response as compared to a plant devoid of the mutation.
95. The plant or corn plant of claim 94, further comprising increased yield, increased upright growth, decreased height, decreased shoot:root ratio, decreased leaf length, increased  
25 mechanical strength of stems, reduced lodging rate, delayed senescence, substantially no change in flowering time, increased photosynthesis efficiency and grain filling, and/or enhanced defense responses against pathogens and herbivores when planted in close proximity with one or more plants comprising a reduced Shade Avoidance Response as compared to a plant that does not  
30 comprise a reduced Shade Avoidance Response that is planted in close proximity with one or more plants that are devoid of the mutation and do not exhibit a reduced Shade Avoidance Response.



96. A guide nucleic acid that binds to a target nucleic acid within an endogenous *Elongated Hypocotyl5 (HY5)* gene having the gene identification number (gene ID) (Maize Genetics and Genomics Database (Maize GDB)) of Zm00001d015743.
- 5 97. A method of producing a plant comprising a mutation in an endogenous *Elongated Hypocotyl5 (HY5)* gene and at least one polynucleotide of interest, the method comprising  
crossing a first plant, which is the plant of any one of claims 1-20, 36-38, 75-77, or 91-95, with a second plant that comprises the at least one polynucleotide of interest to produce progeny plants; and  
10 selecting progeny plants comprising the mutation in the *HY5* gene and the at least one polynucleotide of interest, thereby producing the plant comprising a mutation in an endogenous *HY5* gene and at least one polynucleotide of interest.
98. A method of producing a plant comprising a mutation in an endogenous *HY5* gene and at  
15 least one polynucleotide of interest, the method comprising  
introducing at least one polynucleotide of interest into a plant of any one of claims 1-20, 36-38, 75-77, or 91-95, thereby producing a plant comprising a mutation in a *HY5* gene and at least one polynucleotide of interest.
- 20 99. The method of any one of claim 97 or claim 98, wherein the polynucleotide of interest is a polynucleotide that confers herbicide tolerance, insect resistance, disease resistance, increased yield, increased nutrient use efficiency or abiotic stress resistance.
100. A method of producing a plant comprising a mutation in an endogenous *HY5* gene and  
25 exhibiting a phenotype of improved yield traits, improved plant architecture and/or improved defense traits, comprising  
crossing a first plant, which is the plant of any one of claims 1-20, 36-38, 75-77, or 91-95, with a second plant that exhibits a phenotype of improved yield traits, improved plant architecture and/or improved defense traits; and  
30 selecting progeny plants comprising the mutation in the *HY5* gene and a phenotype of improved yield traits, improved plant architecture and/or improved defense traits, thereby producing the plant comprising a mutation in an endogenous *HY5* gene and exhibiting a phenotype of improved yield traits, improved plant architecture and/or improved defense traits as compared to a control plant.

101. A method of controlling weeds in a container (e.g., pot, or seed tray and the like), a growth chamber, a greenhouse, a field, a recreational area, a lawn, or on a roadside, comprising applying an herbicide to one or more (a plurality) plants of any one of claims 1-20, 36-38, 75-77, or 91-95 growing in a container, a growth chamber, a greenhouse, a field, a recreational area, a lawn, or on a roadside, thereby controlling the weeds in the container, the growth chamber, the greenhouse, the field, the recreational area, the lawn, or on the roadside in which the one or more plants are growing.
102. A method of reducing insect predation on a plant, comprising applying an insecticide to one or more plants of any one of claims 1-20, 36-38, 75-77, or 91-95, thereby reducing insect predation on the one or more plants.
103. A method of reducing fungal disease on a plant, comprising applying a fungicide to one or more plants of any one of claims 1-20, 36-38, 75-77, or 91-95, thereby reducing fungal disease on the one or more plants.
104. The method of claim 102 or claim 103, wherein the one or more plants are growing in a container, a growth chamber, a greenhouse, a field, a recreational area, a lawn, or on a roadside.

# INTERNATIONAL SEARCH REPORT

International application No  
**PCT/US2023/069039**

**A. CLASSIFICATION OF SUBJECT MATTER**  
**INV. C07K14/415 C12N9/22 C12N15/82**  
**ADD.**

According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**  
 Minimum documentation searched (classification system followed by classification symbols)  
**C07K C12N**

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)  
**EPO-Internal, BIOSIS, Sequence Search, EMBASE, WPI Data**

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
<b>X</b>	<p><b>QIU ZHENGKUN ET AL: "Identification of Candidate HY5-Dependent and -Independent Regulators of Anthocyanin Biosynthesis in Tomato", PLANT AND CELL PHSIOLOGY, vol. 60, no. 3, 1 March 2019 (2019-03-01), pages 643-656, XP093083629, UK</b></p> <p><b>ISSN: 0032-0781, DOI: 10.1093/pcp/pcy236</b></p> <p><b>Retrieved from the Internet:</b></p> <p><b>URL: <a href="https://academic.oup.com/pcp/article-pdf/60/3/643/28012139/pcy236.pdf">https://academic.oup.com/pcp/article-pdf/60/3/643/28012139/pcy236.pdf</a></b></p> <p><b>the whole document</b></p> <p style="text-align: center;">----- -/--</p>	<p><b>1-3, 8-15, 18, 21, 24, 42, 68, 71, 72, 75, 77, 83</b></p>

Further documents are listed in the continuation of Box C.       See patent family annex.

\* Special categories of cited documents :

<p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier application or patent but published on or after the international filing date</p> <p>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p>	<p>"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone</p> <p>"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art</p> <p>"&amp;" document member of the same patent family</p>
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Date of the actual completion of the international search	Date of mailing of the international search report
<b>21 September 2023</b>	<b>04/10/2023</b>

Name and mailing address of the ISA/ European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Authorized officer  <p style="text-align: center;"><b>Kania, Thomas</b></p>
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# INTERNATIONAL SEARCH REPORT

International application No.

PCT/US2023/069039

## Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing:
  - a.  forming part of the international application as filed.
  - b.  furnished subsequent to the international filing date for the purposes of international search (Rule 13*ter*.1(a)).  
 accompanied by a statement to the effect that the sequence listing does not go beyond the disclosure in the international application as filed.
2.  With regard to any nucleotide and/or amino acid sequence disclosed in the international application, this report has been established to the extent that a meaningful search could be carried out without a WIPO Standard ST.26 compliant sequence listing.
3. Additional comments:

## INTERNATIONAL SEARCH REPORT

International application No

PCT/US2023/069039

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>CN 113 789 334 A (UNIV ZHEJIANG ET AL.) 14 December 2021 (2021-12-14)</p> <p>the whole document</p> <p>-----</p>	<p>1-3, 8-15, 18, 21, 24, 42, 68, 71, 72, 75, 77, 83</p>
X	<p>WO 2011/031680 A1 (MENDEL BIOTECHNOLOGY INC [US]; KHANNA RAJNISH [US] ET AL.) 17 March 2011 (2011-03-17) SEQ ID NO: 12 has 93% sequence identity to present SEQ ID NO: 114</p> <p>-----</p>	<p>90, 91</p>
Y	<p>WO 2021/155084 A1 (PAIRWISE PLANTS SERVICES INC [US]) 5 August 2021 (2021-08-05) the whole document</p> <p>-----</p>	<p>1-104</p>
Y	<p>BHATNAGAR AKANKSHA ET AL: "HY5-COP1: the central module of light signaling pathway", JOURNAL OF PLANT BIOCHEMISTRY AND BIOTECHNOLOGY, vol. 29, no. 4, 16 October 2020 (2020-10-16), pages 590-610, XP037312753, ISSN: 0971-7811, DOI: 10.1007/S13562-020-00623-3 the whole document</p> <p>-----</p>	<p>1-104</p>
Y	<p>HUAI JUNLING ET AL: "Functional analysis of ZmCOP1 and ZmHY5 reveals conserved light signaling mechanism in maize and Arabidopsis", PHYSIOLOGIA PLANTARUM., vol. 169, no. 3, 1 July 2020 (2020-07-01), pages 369-379, XP093083649, DK ISSN: 0031-9317, DOI: 10.1111/pp1.13099 Retrieved from the Internet: URL:https://onlinelibrary.wiley.com/doi/full-xml/10.1111/pp1.13099&gt; the whole document</p> <p>-----</p>	<p>1-104</p>
Y	<p>SREERAMIAH N. GANGAPPA ET AL.: "The Multifaceted Roles of HY5 in Plant Growth and Development", MOLECULAR PLANT, vol. 9, no. 10, 1 October 2016 (2016-10-01), pages 1353-1365, XP055515827, ISSN: 1674-2052, DOI: 10.1016/j.molp.2016.07.002 the whole document</p> <p>-----</p>	<p>1-104</p>

## INTERNATIONAL SEARCH REPORT

International application No  
PCT/US2023/069039

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>ZHANG QIANQIAN ET AL: "GT Factor ZmGT-3b Is Associated With Regulation of Photosynthesis and Defense Response to Fusarium graminearum Infection in Maize Seedling", FRONTIERS IN PLANT SCIENCE, vol. 12, 18 November 2021 (2021-11-18), XP93083648, DOI: 10.3389/fpls.2021.724133 page 16, left-hand column, last paragraph - page 16, right-hand column, paragraph 1 -----</p>	1-104
A	<p>WO 03/020888 A2 (MONSANTO TECHNOLOGY LLC [US]) 13 March 2003 (2003-03-13) -----</p>	1-104
A	<p>FERNÁNDEZ-MILMANDA GUADALUPE L ET AL: "Shade Avoidance: Expanding the Color and Hormone Palette", TRENDS IN PLANT SCIENCE, ELSEVIER, AMSTERDAM, NL, vol. 26, no. 5, 16 January 2021 (2021-01-16), pages 509-523, XP086536746, ISSN: 1360-1385, DOI: 10.1016/J.TPLANTS.2020.12.006 [retrieved on 2021-01-16] -----</p>	1-104

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No <b>PCT/US2023/069039</b>
----------------------------------------------------------

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
<b>CN 113789334</b>	<b>A</b>	<b>14-12-2021</b>	<b>NONE</b>
<hr style="border-top: 1px dashed black;"/>			
<b>WO 2011031680</b>	<b>A1</b>	<b>17-03-2011</b>	<b>AR 078349 A1 02-11-2011</b>
			<b>US 2012210456 A1 16-08-2012</b>
			<b>WO 2011031680 A1 17-03-2011</b>
<hr style="border-top: 1px dashed black;"/>			
<b>WO 2021155084</b>	<b>A1</b>	<b>05-08-2021</b>	<b>AR 121242 A1 04-05-2022</b>
			<b>BR 112022012417 A2 30-08-2022</b>
			<b>CA 3165291 A1 05-08-2021</b>
			<b>CN 115335392 A 11-11-2022</b>
			<b>EP 4097125 A1 07-12-2022</b>
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