



Office de la Propriété
Intellectuelle
du Canada

Un organisme
d'Industrie Canada

Canadian
Intellectual Property
Office

An agency of
Industry Canada

CA 2669875 C 2017/01/03

(11)(21) **2 669 875**

(12) **BREVET CANADIEN
CANADIAN PATENT**

(13) **C**

(86) **Date de dépôt PCT/PCT Filing Date:** 2007/11/14
(87) **Date publication PCT/PCT Publication Date:** 2008/05/22
(45) **Date de délivrance/Issue Date:** 2017/01/03
(85) **Entrée phase nationale/National Entry:** 2009/05/14
(86) **N° demande PCT/PCT Application No.:** US 2007/084714
(87) **N° publication PCT/PCT Publication No.:** 2008/061156
(30) **Priorité/Priority:** 2006/11/15 (US60/866,055)

(51) **Cl.Int./Int.Cl. C12N 15/82** (2006.01),
C07K 14/415 (2006.01), **A01H 5/00** (2006.01)

(72) **Inventeurs/Inventors:**
DAVIES, JOHN P., US;
NG, HEIN TSOENG (MEDARD), US;
WAGNER, D. RY, US

(73) **Propriétaire/Owner:**
AGRIGENETICS, INC., US

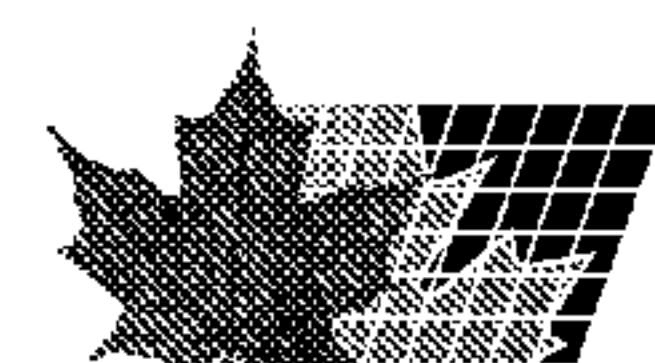
(74) **Agent:** GOUDREAU GAGE DUBUC

(54) **Titre : GENERATION DE VEGETAUX A TENEUR MODIFIEE EN PROTEINES, EN FIBRES OU EN HUILE**

(54) **Title: GENERATION OF PLANTS WITH ALTERED PROTEIN, FIBER, OR OIL CONTENT**

(57) **Abrégé/Abstract:**

The present invention is directed to plants that display an improved oil quantity phenotype or an improved meal quality phenotype due to altered expression of an IMQ nucleic acid. The invention is further directed to methods of generating plants with an improved oil quantity phenotype or improved meal quality phenotype.



(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau



(43) International Publication Date
22 May 2008 (22.05.2008)

PCT

(10) International Publication Number
WO 2008/061156 A3

(51) International Patent Classification:

C12N 15/82 (2006.01) *A01H 5/00* (2006.01)
C07K 14/415 (2006.01)

(21) International Application Number:

PCT/US2007/084714

(22) International Filing Date:

14 November 2007 (14.11.2007)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

60/866,055 15 November 2006 (15.11.2006) US

(71) **Applicant** (*for all designated States except US*): **AGRI-GENETICS, INC.** [US/US]; 9330 Zionsville Road, Indianapolis, IN 46268-1053 (US).

(72) Inventors; and

(75) **Inventors/Applicants** (*for US only*): **DAVIES, John, P.** [US/US]; 4617 S.E. Madison Street, Portland, OR 97215 (US). **NG, Hein Tsoeng (medard)** [—/US]; 3038 Amberfield Trail, Charlottesville, VA 22911 (US). **WAGNER, D., Ry** [US/US]; 85281 Ridgeway Road, Pleasant Hill, OR 97455 (US).

(74) **Agent**: **CARLSON, Anne**; Klarquist Sparkman, LLP, One World Trade Center, Suite 1600, 121 SW Salmon Street, Portland, OR 97204 (US).

(81) **Designated States** (*unless otherwise indicated, for every kind of national protection available*): AE, AG, AL, AM,

AT, AU, AZ, BA, BB, BG, BH, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IS, JP, KE, KG, KM, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, SV, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) **Designated States** (*unless otherwise indicated, for every kind of regional protection available*): ARIPO (BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IS, IT, LT, LU, LV, MC, MT, NL, PL, PT, RO, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Declaration under Rule 4.17:

— *of inventorship (Rule 4.17(iv))*

Published:

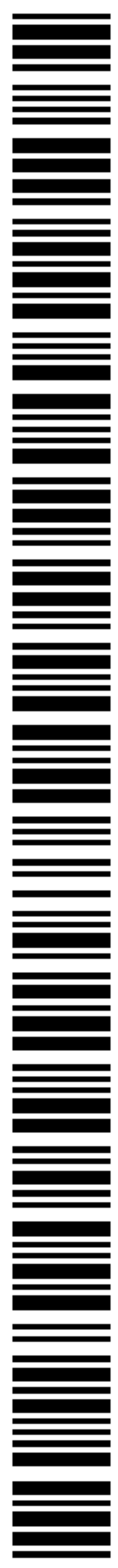
— *with international search report*
— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*
— *with sequence listing part of description published separately in electronic form and available upon request from the International Bureau*

(88) **Date of publication of the international search report:**

8 January 2009

(54) **Title**: GENERATION OF PLANTS WITH ALTERED PROTEIN, FIBER, OR OIL CONTENT

(57) **Abstract**: The present invention is directed to plants that display an improved oil quantity phenotype or an improved meal quality phenotype due to altered expression of an IMQ nucleic acid. The invention is further directed to methods of generating plants with an improved oil quantity phenotype or improved meal quality phenotype.



WO 2008/061156 A3

GENERATION OF PLANTS WITH ALTERED PROTEIN, FIBER, OR OIL CONTENT

5 FIELD OF THE DISCLOSURE

The present disclosure is related to transgenic plants with altered oil, protein, and/or fiber content, as well as methods of making plants having altered oil, protein, and/or fiber content and producing oil from such plants.

10 BACKGROUND

The ability to manipulate the composition of crop seeds, particularly the content and composition of seed oil and protein, as well as the available metabolizable energy (“AME”) in the seed meal in livestock, has important applications in the agricultural industries, relating both to processed food oils and to animal feeds. Seeds of agricultural crops contain a variety of valuable constituents, including oil, protein and starch. Industrial processing can separate some or all of these constituents for individual sale in specific applications. For instance, nearly 60% of the U.S. soybean crop is crushed by the soy processing industry. Soy processing yields purified oil, which is sold at high value, while the remaining seed meal is sold for livestock feed (U.S. Soybean Board, 2001 Soy Stats). Canola seed is also crushed to produce oil and the co-product canola meal (Canola Council of Canada). Canola meal contains a high percentage of protein and a good balance of amino acids but because it has a high fiber and phytate content, it is not readily digested by livestock (Slominski, B.A., *et al.*, 1999 Proceedings of the 10th International Rapeseed Congress, Canberra, Australia) and has a lower value than soybean meal.

Over 55% of the corn produced in the U.S. is used as animal feed (Iowa Corn Growers Association). The value of the corn is directly related to its ability to be

digested by livestock. Thus, it is desirable to maximize both oil content of seeds and the AME of meal. For processed oilseeds such as soy and canola, increasing the absolute oil content of the seed will increase the value of such grains, while increasing the AME of meal will increase its value. For processed corn, either an increase or a decrease in oil content may be desired, depending on how the other major constituents are to be used. Decreasing oil may improve the quality of isolated starch by reducing undesired flavors associated with oil oxidation. Alternatively, when the starch is used for ethanol production, where flavor is unimportant, increasing oil content may increase overall value.

In many feed grains, such as corn and wheat, it is desirable to increase seed oil content, because oil has higher energy content than other seed constituents such as carbohydrate. Oilseed processing, like most grain processing businesses, is a capital-intensive business; thus small shifts in the distribution of products from the low valued components to the high value oil component can have substantial economic impacts for grain processors. In addition, increasing the AME of meal by adjusting seed protein and fiber content and composition, without decreasing seed oil content, can increase the value of animal feed.

Biotechnological manipulation of oils has been shown to provide compositional alteration and improvement of oil yield. Compositional alterations include high oleic acid soybean and corn oil (U.S. Patent Nos. 6,229,033 and 6,248,939), and laurate-containing seeds (U.S. Patent No. 5,639,790), among others. Work in compositional alteration has predominantly focused on processed oilseeds, but has been readily extendable to non-oilseed crops, including corn. While there is considerable interest in increasing oil content, the only currently practiced biotechnology in this area is High-Oil Corn (HOC) technology (DuPont, U.S. Patent No. 5,704,160). HOC employs high oil pollinators developed by classical selection breeding along with elite (male-sterile) hybrid females in a production system referred to as TopCross. The TopCross High Oil system raises harvested grain oil content in maize from about 3.5% to about 7%, improving the energy content of the grain.

While it has been fruitful, the HOC production system has inherent limitations. First, the system of having a low percentage of pollinators responsible

for an entire field's seed set contains inherent risks, particularly in drought years. Second, oil content in current HOC fields has plateaued at about 9% oil. Finally, high-oil corn is not primarily a biochemical change, but rather an anatomical mutant (increased embryo size) that has the indirect result of increasing oil content. For
5 these reasons, an alternative high oil strategy, particularly one that derives from an altered biochemical output, would be especially valuable.

Manipulation of seed composition has identified several components that improve the nutritive quality, digestibility, and AME in seed meal. Increasing the lysine content in canola and soybean (Falco *et al.*, 1995 *Bio/Technology* 13:577-582)
10 increases the availability of this essential amino acid and decreases the need for nutritional supplements. Soybean varieties with increased seed protein were shown to contain considerably more metabolizable energy than conventional varieties (Edwards *et al.*, 1999, *Poultry Sci.* 79:525-527). Decreasing the phytate content of corn seed has been shown to increase the bioavailability of amino acids in animal
15 feeds (Douglas *et al.*, 2000, *Poultry Sci.* 79:1586-1591) and decreasing oligosaccharide content in soybean meal increases the metabolizable energy in the meal (Parsons *et al.*, 2000, *Poultry Sci.* 79:1127-1131).

Soybean and canola are the most obvious target crops for the processed oil and seed meal markets since both crops are crushed for oil and the remaining meal
20 sold for animal feed. A large body of commercial work (*e.g.*, U.S. Patent No. 5,952,544; PCT Application No. WO9411516) demonstrates that *Arabidopsis* is an excellent model for oil metabolism in these crops. Biochemical screens of seed oil composition have identified *Arabidopsis* genes for many critical biosynthetic enzymes and have led to identification of agronomically important gene orthologs.
25 For instance, screens using chemically mutagenized populations have identified lipid mutants whose seeds display altered fatty acid composition (Lemieux *et al.*, 1990, *Theor. Appl. Genet.* 80, 234-240; James and Dooner, 1990, *Theor. Appl. Genet.* 80, 241-245). T-DNA mutagenesis screens (Feldmann *et al.*, 1989, *Science* 243: 1351-1354) that detected altered fatty acid composition identified the omega 3 desaturase
30 (*FAD3*) and delta-12 desaturase (*FAD2*) genes (U.S. Pat No 5952544; Yadav *et al.*, 1993, *Plant Physiol.* 103, 467-476; Okuley *et al.*, 1994, *Plant Cell* 6(1):147-158). A screen which focused on oil content rather than oil quality, analyzed chemically-

induced mutants for wrinkled seeds or altered seed density, from which altered seed oil content was inferred (Focks and Benning, 1998, *Plant Physiol.* 118:91-101).

Another screen, designed to identify enzymes involved in production of very long chain fatty acids, identified a mutation in the gene encoding a diacylglycerol acyltransferase (DGAT) as being responsible for reduced triacyl glycerol
5 accumulation in seeds (Katavic V *et al.*, 1995, *Plant Physiol.* 108(1):399-409). It was further shown that seed-specific over-expression of the DGAT cDNA was associated with increased seed oil content (Jako *et al.*, 2001, *Plant Physiol.* 126(2):861-74). *Arabidopsis* is also a model for understanding the
10 accumulation of seed components that affect meal quality. For example, *Arabidopsis* contains albumin and globulin seed storage proteins found in many dicotyledonous plants including canola and soybean (Shewry 1995, *Plant Cell* 7:945-956). The biochemical pathways for synthesizing components of fiber, such as cellulose and lignin, are conserved within the vascular plants, and mutants of
15 *Arabidopsis* affecting these components have been isolated (reviewed in Chapel and Carpita 1998, *Current Opinion in Plant Biology* 1:179-185).

Activation tagging in plants refers to a method of generating random mutations by insertion of a heterologous nucleic acid construct comprising regulatory sequences (*e.g.*, an enhancer) into a plant genome. The regulatory
20 sequences can act to enhance transcription of one or more native plant genes; accordingly, activation tagging is a fruitful method for generating gain-of-function, generally dominant mutants (see, *e.g.*, Hayashi *et al.*, 1992, *Science* 258: 1350-1353; Weigel D *et al.*, 2000, *Plant Physiology*, 122:1003-1013). The inserted construct provides a molecular tag for rapid identification of the native plant whose mis-
25 expression causes the mutant phenotype. Activation tagging may also cause loss-of-function phenotypes. The insertion may result in disruption of a native plant gene, in which case the phenotype is generally recessive.

Activation tagging has been used in various species, including tobacco and *Arabidopsis*, to identify many different kinds of mutant phenotypes and the genes
30 associated with these phenotypes (Wilson *et al.*, 1996, *Plant Cell* 8: 659-671; Schaffer *et al.*, 1998, *Cell* 93: 1219-1229; Fridborg *et al.*, 1999, *Plant Cell* 11: 1019-1032; Kardailsky *et al.*, 1999, *Science* 286: 1962-1965; and Christensen S *et al.*,

1998, 9th *International Conference on Arabidopsis Research*, Univ. of Wisconsin-Madison, June 24-28, Abstract 165).

SUMMARY

5 Provided herein are transgenic plants having an Improved Seed Quality phenotype. Transgenic plants with an Improved Seed Quality phenotype may include an improved oil quantity and/or an improved meal quality. Transgenic plants with improved meal quality have an Improved Meal Quality (IMQ) phenotype and transgenic plants with improved oil quantity have an Improved Oil Quantity
10 (IOQ) phenotype. The IMQ phenotype in a transgenic plant may include altered protein and/or fiber content in any part of the transgenic plant, for example in the seeds. The IOQ phenotype in a transgenic plant may include altered oil content in any part of the transgenic plant, for example in the seeds. In particular embodiments, a transgenic plant may include an IOQ phenotype and/or an IMQ
15 phenotype. In some embodiments of a transgenic plant, the IMQ phenotype may be an increase in protein content in the seed and/or a decrease in the fiber content of the seed. In other embodiments of a transgenic plant, the IOQ phenotype is an increase in the oil content of the seed (a high oil phenotype). Also provided is seed meal derived from the seeds of transgenic plants, wherein the seeds have altered protein
20 content and/or altered fiber content. Further provided is oil derived from the seeds of transgenic plants, wherein the seeds have altered oil content. Any of these changes can lead to an increase in the AME from the seed or seed meal from transgenic plants, relative to control, non-transgenic, or wild-type plants. Also provided herein is meal, feed, or food produced from any part of the transgenic plant
25 with an IMQ phenotype and/or IOQ phenotype.

 In certain embodiments, the disclosed transgenic plants comprise a transformation vector comprising an IMQ nucleotide sequence that encodes or is complementary to a sequence that encodes an "IMQ" polypeptide. In particular
30 embodiments, expression of an IMQ polypeptide in a transgenic plant causes an altered oil content, an altered protein content, and/or an altered fiber content in the transgenic plant. In preferred embodiments, the transgenic plant is selected from the group consisting of plants of the *Brassica* species, including canola and rapeseed,

soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat and rice. Also provided is a method of producing oil or seed meal, comprising growing the transgenic plant and recovering oil and/or seed meal from said plant. The disclosure further provides feed, meal, grain, or seed
5 comprising a nucleic acid sequence that encodes an IMQ polypeptide. The disclosure also provides feed, meal, grain, or seed comprising the IMQ polypeptide, or an ortholog thereof.

Examples of the disclosed transgenic plant are produced by a method that comprises introducing into progenitor cells of the plant a plant transformation vector
10 comprising an IMQ nucleotide sequence that encodes, or is complementary to a sequence that encodes, an IMQ polypeptide, and growing the transformed progenitor cells to produce a transgenic plant, wherein the IMQ polynucleotide sequence is expressed, causing an IOQ phenotype and/or an IMQ phenotype in the transgenic plant. In some specific, non-limiting examples, the method produces transgenic
15 plants wherein expression of the IMQ polypeptide causes a high (increased) oil, high (increased) protein, and/or low (decreased) fiber phenotype in the transgenic plant, relative to control, non-transgenic, or wild-type plants.

Additional methods are disclosed herein of generating a plant having an IMQ and/or an IOQ phenotype, wherein a plant is identified that has an allele in its IMQ
20 nucleic acid sequence that results in an IMQ phenotype and/or an IOQ phenotype, compared to plants lacking the allele. The plant can generate progeny, wherein the progeny inherit the allele and have an IMQ phenotype and/or an IOQ phenotype. In some embodiments of the method, the method employs candidate gene/QTL methodology or TILLING methodology.

25 Also provided herein is a transgenic plant cell having an IMQ phenotype and/or an IOQ phenotype. The transgenic plant cell comprises a transformation vector comprising an IMQ nucleotide sequence that encodes or is complementary to a sequence that encodes an IMQ polypeptide. In preferred embodiments, the transgenic plant cell is selected from the group consisting of plants of the *Brassica*
30 species, including canola and rapeseed, soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat and rice. In other embodiments, the plant cell is a seed, pollen, propagule, or embryo cell. The

disclosure also provides plant cells from a plant that is the direct progeny or the indirect progeny of a plant grown from the progenitor cells.

In one aspect, the present invention relates to a transgenic plant cell comprising a plant transformation vector comprising a nucleotide sequence that
5 encodes an improved meal quality (IMQ) polypeptide comprising the amino acid sequence of SEQ ID NO: 82, or an IMQ polypeptide comprising an amino acid sequence at least 80% identical overall to SEQ ID NO: 82, wherein the transgenic plant cell is of a transgenic plant that has an improved meal quality phenotype, relative to control plants.

10 In another aspect, the present invention relates to a meal, feed, or food produced from the seed of a plant comprising the above mentioned transgenic plant cell.

In another aspect, the present invention relates to a method of producing meal, the method comprising growing a transgenic plant comprising the above
15 mentioned transgenic plant cell, and recovering meal from the plant, thereby producing meal.

In another aspect, the present invention relates to a method of producing an improved meal quality phenotype in a plant, the method comprising: (a) introducing
20 into progenitor cells of the plant a plant transformation vector comprising a nucleotide sequence that encodes an improved meal quality (IMQ) polypeptide comprising the amino acid sequence of SEQ ID NO: 82, or an IMQ polypeptide comprising an amino acid sequence at least 80% identical overall to SEQ ID NO: 82; and (b) growing the transformed progenitor cells to produce a transgenic plant, wherein the nucleotide sequence is expressed, and the transgenic plant exhibits an
25 improved meal quality phenotype relative to control plants, thereby producing the improved meal quality phenotype in the plant.

In another aspect, the present invention relates to a transgenic plant cell of a plant obtained by the above mentioned method.

In another aspect, the present invention relates to a method of generating a
30 plant having an improved meal quality phenotype, the method comprising: (i) identifying a plant as comprising an allele of a gene, wherein the allele encodes a

polypeptide comprising an amino acid sequence at least 80% identical overall to SEQ ID NO: 82, wherein the allele is overexpressed relative to a control plant, and wherein the allele results in improved meal quality phenotype, compared to plants lacking the allele; and (ii) generating progeny of the identified plant, wherein the generated progeny inherit the allele and have the improved meal quality phenotype relative to control plants.

In another aspect, the present invention relates to a feed, meal, or food comprising a polypeptide encoded by the nucleic acid sequence of SEQ ID NO: 81, or a nucleic acid sequence at least 80% identical overall to SEQ ID NO: 81.

In another aspect, the present invention relates to a feed, meal, or food comprising a polypeptide comprising the amino acid sequence of SEQ ID NO: 82, or an IMQ polypeptide comprising an amino acid sequence at least 80% identical overall to SEQ ID NO: 82.

In another aspect, the present invention relates to a method of producing an improved meal quality phenotype in a plant, said method comprising:

(a) introducing into progenitor cells of the plant a plant transformation vector comprising a nucleotide sequence that encodes an improved meal quality (IMQ) polypeptide comprising the amino acid sequence of SEQ ID NO: 82, or an IMQ polypeptide comprising an amino acid sequence at least 80% identical to the full length of SEQ ID NO: 82; and

(b) growing the transformed progenitor cells to produce a transgenic plant, wherein the nucleotide sequence is expressed; and

(c) measuring the improved meal quality phenotype in the transgenic plant, wherein the transgenic plant exhibits an improved meal quality phenotype relative to control plants lacking the plant transformation vector, thereby producing the improved meal quality phenotype in the plant.

In another aspect, the present invention relates to a transgenic plant cell of a plant obtained by the above-mentioned method, wherein the cell is of a *Brassica* species.

In another aspect, the present invention relates to a transgenic plant cell of a plant obtained by the above-mentioned method, wherein the cell is a canola,

rapeseed, soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat, or rice cell.

In another aspect, the present invention relates to a transgenic plant cell of a *Brassica* species which is of a direct or indirect progeny of a plant obtained by the
5 above-mentioned method, wherein the cells of the direct or indirect progeny of the plant comprise the plant transformation vector.

In another aspect, the present invention relates to a transgenic plant cell which is a canola, rapeseed, soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat, or rice cell and which is of a direct or
10 indirect progeny of a plant obtained by the above-mentioned method, wherein the cells of the direct or indirect progeny of the plant comprise the plant transformation vector.

In another aspect, the present invention relates to a method of generating a plant having an improved meal quality phenotype, said method comprising:

15 (i) identifying a plant as comprising an allele of a gene, wherein the allele encodes a polypeptide comprising an amino acid sequence at least 80% identical to the full length of SEQ ID NO: 82, wherein the allele is overexpressed relative to a control plant, and wherein the allele results in improved meal quality phenotype, compared to plants lacking the allele; and

20 (ii) generating progeny of said identified plant, wherein the generated progeny inherit the allele; and

(iii) measuring the improved meal quality phenotype in the progeny, wherein the progeny have the improved meal quality phenotype relative to control plants lacking the allele.

25 In another aspect, the present invention relates to a feed, meal, or food comprising a plant transformation vector comprising the nucleic acid sequence of SEQ ID NO: 81, or a nucleic acid sequence at least 80% identical to the full length of SEQ ID NO: 81, wherein the feed, meal, or food has an improved meal quality phenotype relative to a control feed, meal, or food that does not comprise the plant
30 transformation vector.

In another aspect, the present invention relates to a feed, meal, or food comprising a plant transformation vector comprising a nucleotide sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO: 82, or a polypeptide comprising an amino acid sequence at least 80% identical to the full length of SEQ ID NO: 82, wherein the feed, meal, or food has an improved meal quality phenotype relative to a control feed, meal, or food that does not comprise the plant transformation vector.

DETAILED DESCRIPTION

10 Terms

Unless otherwise indicated, all technical and scientific terms used herein have the same meaning as they would to one skilled in the art of the present disclosure. Practitioners are particularly directed to Sambrook *et al.* (*Molecular Cloning: A Laboratory Manual* (Second Edition), Cold Spring Harbor Press, Plainview, N.Y., 1989) and Ausubel FM *et al.* (*Current Protocols in Molecular Biology*, John Wiley & Sons, New York, N.Y., 1993) for definitions and terms of the art. It is to be understood that this disclosure is not limited to the particular methodology, protocols, and reagents described, as these may vary.

As used herein, the term “IMQ phenotype” refers to plants, or any part of a plant (for example, seeds, or meal produced from seeds), with an altered protein and/or fiber content (phenotype). As provided herein, altered protein and/or fiber content includes either an increased or decreased level of protein and/or fiber content in plants, seeds or seed meal. Any combination of these changes can lead to an IMQ phenotype. For example, in one specific non-limiting example, an IMQ phenotype can refer to increased protein and decreased fiber content. In another specific non-limiting example, an IMQ phenotype can refer to unchanged protein and decreased fiber content. In yet another specific non-limiting example, an IMQ phenotype can refer to increased protein and unchanged fiber content. It is also provided that any combination of these changes can lead to an increase in the AME (available metabolizable energy) from the seed or meal generated from the seed. An IMQ phenotype also includes an improved seed quality (ISQ) phenotype or an improved seed meal quality phenotype.

As used herein, the term “IOQ phenotype” refers to plants, or any part of a plant (for example, seeds), with an altered oil content (phenotype). As provided herein, altered oil content includes an increased, for example a high, oil content in
5 plants or seeds. In some embodiments, a transgenic plant can express both an IOQ phenotype and an IMQ phenotype. In specific, non-limiting examples, a transgenic

plant having a combination of an IOQ phenotype and an IMQ phenotype can lead to an increase in the AME (available metabolizable energy) from the seed or meal generated from the seed. An IOQ phenotype also includes an improved seed quality (ISQ) phenotype.

5 As used herein, the term “available metabolizable energy” (AME) refers to the amount of energy in the feed that is able to be extracted by digestion in an animal and is correlated with the amount of digestible protein and oil available in animal meal. AME is determined by estimating the amount of energy in the feed prior to feeding and measuring the amount of energy in the excreta of the animal
10 following consumption of the feed. In one specific, non-limiting example, a transgenic plant with an increase in AME includes transgenic plants with altered seed protein and/or fiber content and without a decrease in seed oil content (seed oil content remains unchanged or is increased), resulting in an increase in the value of animal feed derived from the seed.

15 As used herein, the term “content” refers to the type and relative amount of, for instance, a seed or seed meal component.

 As used herein, the term “fiber” refers to non-digestible components of the plant seed including cellular components such as cellulose, hemicellulose, pectin, lignin, and phenolics.

20 As used herein, the term “meal” refers to seed components remaining following the extraction of oil from the seed. Examples of components of meal include protein and fiber.

 As used herein, the term “vector” refers to a nucleic acid construct designed for transfer between different host cells. An “expression vector” refers to a vector
25 that has the ability to incorporate and express heterologous DNA fragments in a foreign cell. Many prokaryotic and eukaryotic expression vectors are commercially available. Selection of appropriate expression vectors is within the knowledge of those having skill in the art.

 A “heterologous” nucleic acid construct or sequence has a portion of the
30 sequence that is not native to the plant cell in which it is expressed. Heterologous, with respect to a control sequence refers to a control sequence (*i.e.* promoter or enhancer) that does not function in nature to regulate the same gene the expression

of which it is currently regulating. Generally, heterologous nucleic acid sequences are not endogenous to the cell or part of the genome in which they are present, and have been added to the cell by infection, transfection, microinjection, electroporation, or the like. A “heterologous” nucleic acid construct may contain a control sequence/DNA coding sequence combination that is the same as, or different from, a control sequence/DNA coding sequence combination found in the native plant. Specific, non-limiting examples of a heterologous nucleic acid sequence include an IMQ nucleic acid sequence, or a fragment, derivative (variant), or ortholog thereof.

10 As used herein, the term “gene” means the segment of DNA involved in producing a polypeptide chain, which may or may not include regions preceding and following the coding region, *e.g.* 5' untranslated (5' UTR) or “leader” sequences and 3' UTR or “trailer” sequences, as well as intervening sequences (introns) between individual coding segments (exons) and non-transcribed regulatory sequences.

15 As used herein, “recombinant” includes reference to a cell or vector, that has been modified by the introduction of a heterologous nucleic acid sequence or that the cell is derived from a cell so modified. Thus, for example, recombinant cells express genes that are not found in identical form within the native (non-recombinant) form of the cell or express native genes that are otherwise abnormally expressed, under expressed, or not expressed at all as a result of deliberate human intervention.

As used herein, the term “gene expression” refers to the process by which a polypeptide is produced based on the nucleic acid sequence of a gene. The process includes both transcription and translation; accordingly, “expression” may refer to either a polynucleotide or polypeptide sequence, or both. Sometimes, expression of a polynucleotide sequence will not lead to protein translation. “Over-expression” refers to increased expression of a polynucleotide and/or polypeptide sequence relative to its expression in a wild-type (or other reference [*e.g.*, non-transgenic]) plant and may relate to a naturally-occurring or non-naturally occurring sequence.

25 “Ectopic expression” refers to expression at a time, place, and/or increased level that does not naturally occur in the non-altered or wild-type plant. “Under-expression” refers to decreased expression of a polynucleotide and/or polypeptide sequence,

30

generally of an endogenous gene, relative to its expression in a wild-type plant. The terms “mis-expression” and “altered expression” encompass over-expression, under-expression, and ectopic expression.

The term “introduced” in the context of inserting a nucleic acid sequence
5 into a cell, includes “transfection,” “transformation,” and “transduction” and includes reference to the incorporation of a nucleic acid sequence into a eukaryotic or prokaryotic cell where the nucleic acid sequence may be incorporated into the genome of the cell (for example, chromosome, plasmid, plastid, or mitochondrial DNA), converted into an autonomous replicon, or transiently expressed (for
10 example, transfected mRNA).

As used herein, a “plant cell” refers to any cell derived from a plant, including cells from undifferentiated tissue (*e.g.*, callus), as well as from plant seeds, pollen, propagules, and embryos.

As used herein, the terms “native” and “wild-type” relative to a given plant
15 trait or phenotype refers to the form in which that trait or phenotype is found in the same variety of plant in nature. In one embodiment, a wild-type plant is also a control plant. In another embodiment, a wild-type plant is a non-transgenic plant.

As used herein, the term “modified” regarding a plant trait, refers to a change in the phenotype of a transgenic plant (for example, a transgenic plant with any
20 combination of an altered oil content, an altered protein content, and/or an altered fiber content) in any part of the transgenic plant, for example the seeds, relative to a similar non-transgenic plant. As used herein, the term “altered” refers to either an increase or a decrease of a plant trait or phenotype (for example, oil content, protein content, and/or fiber content) in a transgenic plant, relative to a similar non-
25 transgenic plant. In one specific, non-limiting example, a transgenic plant with a modified trait includes a plant with an increased oil content, increased protein content, and/or decreased fiber content relative to a similar non-transgenic plant. In another specific, non-limiting example, a transgenic plant with a modified trait includes unchanged oil content, increased protein content, and/or decreased fiber
30 content relative to a similar non-transgenic plant. In yet another specific, non-limiting example, a transgenic plant with a modified trait includes an increased oil content, increased protein content, and/or unchanged fiber content relative to a

similar non-transgenic plant. Specific, non-limiting examples of a change in phenotype include an IMQ phenotype or an IOQ phenotype.

An “interesting phenotype (trait)” with reference to a transgenic plant refers to an observable or measurable phenotype demonstrated by a T1 and/or subsequent generation plant, which is not displayed by the corresponding non-transgenic plant (i.e., a genotypically similar plant that has been raised or assayed under similar conditions). An interesting phenotype may represent an improvement in the plant (for example, increased oil content, increased protein content, and/or decreased fiber content in seeds of the plant) or may provide a means to produce improvements in other plants. An “improvement” is a feature that may enhance the utility of a plant species or variety by providing the plant with a unique and/or novel phenotype or quality. Such transgenic plants may have an improved phenotype, such as an IMQ phenotype or an IOQ phenotype.

The phrase “altered oil content phenotype” refers to a measurable phenotype of a genetically modified (transgenic) plant, where the plant displays a statistically significant increase or decrease in overall oil content (i.e., the percentage of seed mass that is oil), as compared to the similar, but non-modified (non-transgenic) plant. A high oil phenotype refers to an increase in overall oil content. The phrase “altered protein content phenotype” refers to measurable phenotype of a genetically modified plant, where the plant displays a statistically significant increase or decrease in overall protein content (i.e., the percentage of seed mass that is protein), as compared to the similar, but non-modified plant. A high protein phenotype refers to an increase in overall protein content. The phrase “altered fiber content phenotype” refers to measurable phenotype of a genetically modified plant, where the plant displays a statistically significant increase or decrease in overall fiber content (i.e., the percentage of seed mass that is fiber), as compared to the similar, but non-modified plant. A low fiber phenotype refers to decrease in overall fiber content.

As used herein, a “mutant” polynucleotide sequence or gene differs from the corresponding wild-type polynucleotide sequence or gene either in terms of sequence or expression, where the difference contributes to a modified or altered plant phenotype or trait. Relative to a plant or plant line, the term “mutant” refers to

a plant or plant line which has a modified or altered plant phenotype or trait, where the modified or altered phenotype or trait is associated with the modified or altered expression of a wild-type polynucleotide sequence or gene.

As used herein, the term “T1” refers to the generation of plants from the seed
5 of T0 plants. The T1 generation is the first set of transformed plants that can be selected by application of a selection agent, *e.g.*, an antibiotic or herbicide, for which the transgenic plant contains the corresponding resistance gene. The term “T2” refers to the generation of plants by self-fertilization of the flowers of T1 plants, previously selected as being transgenic. T3 plants are generated from T2 plants, etc.
10 As used herein, the “direct progeny” of a given plant derives from the seed (or, sometimes, other tissue) of that plant and is in the immediately subsequent generation; for instance, for a given lineage, a T2 plant is the direct progeny of a T1 plant. The “indirect progeny” of a given plant derives from the seed (or other tissue) of the direct progeny of that plant, or from the seed (or other tissue) of subsequent
15 generations in that lineage; for instance, a T3 plant is the indirect progeny of a T1 plant.

As used herein, the term “plant part” includes any plant organ or tissue, including, without limitation, seeds, embryos, meristematic regions, callus tissue, leaves, roots, shoots, gametophytes, sporophytes, pollen, and microspores. Plant
20 cells can be obtained from any plant organ or tissue and cultures prepared therefrom. Provided herein is a transgenic plant cell having an IMQ phenotype and/or an IOQ phenotype. The transgenic plant cell comprises a transformation vector comprising an IMQ nucleotide sequence that encodes or is complementary to a sequence that encodes an IMQ polypeptide. In preferred embodiments, the transgenic plant cell is
25 selected from the group consisting of plants of the *Brassica* species, including canola and rapeseed, soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat and rice. In other embodiments, the plant cell is a seed, pollen, propagule, or embryo cell. The disclosure also provides plant cells from a plant that is the direct progeny or the indirect progeny of a plant
30 grown from said progenitor cells. The class of plants which can be used in the methods of the present invention is generally as broad as the class of higher plants

amenable to transformation techniques, including both monocotyledonous and dicotyledonous plants.

As used herein, “transgenic plant” includes a plant that comprises within its genome a heterologous polynucleotide. The heterologous polynucleotide can be either stably integrated into the genome, or can be extra-chromosomal. Preferably, the polynucleotide of the present invention is stably integrated into the genome such that the polynucleotide is passed on to successive generations. A plant cell, tissue, organ, or plant into which the heterologous polynucleotides have been introduced is considered “transformed,” “transfected,” or “transgenic.” Direct and indirect progeny of transformed plants or plant cells that also contain the heterologous polynucleotide are also considered transgenic.

Disclosed herein are transgenic plants having an Improved Seed Quality phenotype. Transgenic plants with an Improved Seed Quality phenotype may include an improved oil quantity and/or an improved meal quality. Transgenic plants with improved meal quality have an IMQ phenotype and transgenic plants with improved oil quantity have an IOQ phenotype. The IMQ phenotype in a transgenic plant may include altered protein and/or fiber content in any part of the transgenic plant, for example in the seeds. The IOQ phenotype in a transgenic plant may include altered oil content in any part of the transgenic plant, for example in the seeds. In particular embodiments, a transgenic plant may include an IOQ phenotype and/or an IMQ phenotype. In some embodiments of a transgenic plant, the IMQ phenotype may be an increase in protein content in the seed and/or a decrease in the fiber content of the seed. In other embodiments of a transgenic plant, the IOQ phenotype is an increase in the oil content of the seed (a high oil phenotype). Also provided is seed meal derived from the seeds of transgenic plants, wherein the seeds have altered protein content and/or altered fiber content. Further provided is oil derived from the seeds of transgenic plants, wherein the seeds have altered oil content. Any of these changes can lead to an increase in the AME from the seed or seed meal from transgenic plants, relative to control, non-transgenic, or wild-type plants. Also provided herein is meal, feed, or food produced from any part of the transgenic plant with an IMQ phenotype and/or IOQ phenotype.

In certain embodiments, the disclosed transgenic plants comprise a transformation vector comprising an IMQ nucleotide sequence that encodes or is complementary to a sequence that encodes an “IMQ” polypeptide. In particular embodiments, expression of an IMQ polypeptide in a transgenic plant causes an altered oil content, an altered protein content, and/or an altered fiber content in the transgenic plant. In preferred embodiments, the transgenic plant is selected from the group consisting of plants of the *Brassica* species, including canola and rapeseed, soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat and rice. Also provided is a method of producing oil or seed meal, comprising growing the transgenic plant and recovering oil and/or seed meal from said plant. The disclosure further provides feed, meal, grain, or seed comprising a nucleic acid sequence that encodes an IMQ polypeptide. The disclosure also provides feed, meal, grain, or seed comprising the IMQ polypeptide, or an ortholog thereof.

Various methods for the introduction of a desired polynucleotide sequence encoding the desired protein into plant cells are available and known to those of skill in the art and include, but are not limited to: (1) physical methods such as microinjection, electroporation, and microprojectile mediated delivery (biolistics or gene gun technology); (2) virus mediated delivery methods; and (3) *Agrobacterium*-mediated transformation methods (see, for example, WO 2007/053482 and WO 2005/107437).

The most commonly used methods for transformation of plant cells are the *Agrobacterium*-mediated DNA transfer process and the biolistics or microprojectile bombardment mediated process (*i.e.*, the gene gun). Typically, nuclear transformation is desired but where it is desirable to specifically transform plastids, such as chloroplasts or amyloplasts, plant plastids may be transformed utilizing a microprojectile-mediated delivery of the desired polynucleotide.

Agrobacterium-mediated transformation is achieved through the use of a genetically engineered soil bacterium belonging to the genus *Agrobacterium*. A number of wild-type and disarmed strains of *Agrobacterium tumefaciens* and *Agrobacterium rhizogenes* harboring Ti or Ri plasmids can be used for gene transfer into plants. Gene transfer is done via the transfer of a specific DNA known as “T-

DNA” that can be genetically engineered to carry any desired piece of DNA into many plant species.

Agrobacterium-mediated genetic transformation of plants involves several steps. The first step, in which the virulent *Agrobacterium* and plant cells are first brought into contact with each other, is generally called “inoculation.” Following the inoculation, the *Agrobacterium* and plant cells/tissues are permitted to be grown together for a period of several hours to several days or more under conditions suitable for growth and T-DNA transfer. This step is termed “co-culture.” Following co-culture and T-DNA delivery, the plant cells are treated with bactericidal or bacteriostatic agents to kill the *Agrobacterium* remaining in contact with the explant and/or in the vessel containing the explant. If this is done in the absence of any selective agents to promote preferential growth of transgenic versus non-transgenic plant cells, then this is typically referred to as the “delay” step. If done in the presence of selective pressure favoring transgenic plant cells, then it is referred to as a “selection” step. When a “delay” is used, it is typically followed by one or more “selection” steps.

With respect to microprojectile bombardment (U.S. Patent No. 5,550,318; U.S. Patent No. 5,538,880, U.S. Patent No. 5,610,042; and PCT Publication WO 95/06128), particles are coated with nucleic acids and delivered into cells by a propelling force. Exemplary particles include those comprised of tungsten, platinum, and preferably, gold.

An illustrative embodiment of a method for delivering DNA into plant cells by acceleration is the Biolistics Particle Delivery System (BioRad, Hercules, CA), which can be used to propel particles coated with DNA or cells through a screen, such as a stainless steel or Nytex screen, onto a filter surface covered with monocot plant cells cultured in suspension.

Microprojectile bombardment techniques are widely applicable, and may be used to transform virtually any plant species. Examples of species that have been transformed by microprojectile bombardment include monocot species such as maize (PCT Publication No. WO 95/06128), barley, wheat (U.S. Patent No. 5,563,055, incorporated herein by reference in its entirety), rice, oat, rye, sugarcane,

and sorghum, as well as a number of dicots including tobacco, soybean (U.S. Patent No. 5,322,783), sunflower, peanut, cotton, tomato, and legumes in general (U.S. Patent No. 5,563,055).

To select or score for transformed plant cells regardless of transformation methodology, the DNA introduced into the cell contains a gene that functions in a regenerative plant tissue to produce a compound that confers upon the plant tissue resistance to an otherwise toxic compound. Genes of interest for use as a selectable, screenable, or scorable marker would include but are not limited to GUS, green fluorescent protein (GFP), luciferase (LUX), antibiotic or herbicide tolerance genes. Examples of antibiotic resistance genes include the penicillins, kanamycin (and neomycin, G418, bleomycin), methotrexate (and trimethoprim), chloramphenicol, and tetracycline. Polynucleotide molecules encoding proteins involved in herbicide tolerance are known in the art, and include, but are not limited to a polynucleotide molecule encoding 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) described in U.S. Patent No. 5,627,061, U.S. Patent No 5,633,435, and U.S. Patent No 6,040,497 and *aroA* described in U.S. Patent No. 5,094,945 for glyphosate tolerance; a polynucleotide molecule encoding bromoxynil nitrilase (Bxn) described in U.S. Patent No. 4,810,648 for Bromoxynil tolerance; a polynucleotide molecule encoding phytoene desaturase (*crtI*) described in Misawa *et al.*, (*Plant J.* 4:833-840, 1993) and Misawa *et al.*, (*Plant J.* 6:481-489, 1994) for norflurazon tolerance; a polynucleotide molecule encoding acetohydroxyacid synthase (AHAS, also known as ALS) described in Sathasiivan *et al.* (*Nucl. Acids Res.* 18:2188-2193, 1990) for tolerance to sulfonyleurea herbicides; and the bar gene described in DeBlock, *et al.*, (*EMBO J.* 6:2513-2519, 1987) for glufosinate and bialaphos tolerance.

The regeneration, development, and cultivation of plants from various transformed explants are well documented in the art. This regeneration and growth process typically includes the steps of selecting transformed cells and culturing those individualized cells through the usual stages of embryonic development through the rooted plantlet stage. Transgenic embryos and seeds are similarly regenerated. The resulting transgenic rooted shoots are thereafter planted in an appropriate plant growth medium such as soil. Cells that survive the exposure to the selective agent,

or cells that have been scored positive in a screening assay, may be cultured in media that supports regeneration of plants. Developing plantlets are transferred to soil less plant growth mix, and hardened off, prior to transfer to a greenhouse or growth chamber for maturation.

5 The present invention can be used with any transformable cell or tissue. By transformable as used herein is meant a cell or tissue that is capable of further propagation to give rise to a plant. Those of skill in the art recognize that a number of plant cells or tissues are transformable in which after insertion of exogenous DNA and appropriate culture conditions the plant cells or tissues can form into a
10 differentiated plant. Tissue suitable for these purposes can include but is not limited to immature embryos, scutellar tissue, suspension cell cultures, immature inflorescence, shoot meristem, nodal explants, callus tissue, hypocotyl tissue, cotyledons, roots, and leaves.

Any suitable plant culture medium can be used. Examples of suitable media
15 would include but are not limited to MS-based media (Murashige and Skoog, *Physiol. Plant*, 15:473-497, 1962) or N6-based media (Chu *et al.*, *Scientia Sinica* 18:659, 1975) supplemented with additional plant growth regulators including but not limited to auxins, cytokinins, ABA, and gibberellins. Those of skill in the art are familiar with the variety of tissue culture media, which when supplemented
20 appropriately, support plant tissue growth and development and are suitable for plant transformation and regeneration. These tissue culture media can either be purchased as a commercial preparation, or custom prepared and modified. Those of skill in the art are aware that media and media supplements such as nutrients and growth
25 regulators for use in transformation and regeneration and other culture conditions such as light intensity during incubation, pH, and incubation temperatures that can be optimized for the particular variety of interest.

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

Identification of Plants with an Improved Oil Quantity Phenotype and/or Improved Meal Quality Phenotype

An *Arabidopsis* activation tagging screen (ACTTAG) was used to identify the association between 1) ACTTAG plant lines with an altered protein, fiber and/or oil content (phenotype, for example, see columns 4, 5 and 6, respectively, of Table 1, below) and 2) the nucleic acid sequences identified in column 3 of Tables 2 and 3, wherein each nucleic acid sequence is provided with a gene alias or an IMQ designation (IMQ#; see column 1 in Tables 1, 2, and 3). Briefly, and as further described in the Examples, a large number of *Arabidopsis* plants were mutated with the pSKI015 vector, which comprises a T-DNA from the Ti plasmid of *Agrobacterium tumefaciens*, a viral enhancer element, and a selectable marker gene (Weigel *et al.*, 2000, *Plant Physiology*, 122:1003-1013). When the T-DNA inserts into the genome of transformed plants, the enhancer element can cause up-regulation of genes in the vicinity, generally within about nine kilobases (kb) of the enhancers. T1 plants were exposed to the selective agent in order to specifically recover transformed plants that expressed the selectable marker and therefore harbored T-DNA insertions. T1 plants were allowed to grow to maturity, self-fertilize and produce seed. T2 seed was harvested, labeled and stored. To amplify the seed stocks, about eighteen T2 were sown in soil and, after germination, exposed to the selective agent to recover transformed T2 plants. T3 seed from these plants was harvested and pooled. Oil, protein and fiber content of the seed were estimated using Near Infrared Spectroscopy (NIR) as described in the Examples.

Quantitative determination of fatty acid (FA) content (column 7, Table 1) in T2 seeds was performed using the following methods. A sample of 15 to 20 T2 seeds from each line tested. This sample generally contained plants with homozygous insertions, no insertions, and hemizygous insertions in a standard 1:1:2 ratios. The seed sample was massed on UMT-2 ultra-microbalance (Mettler-Toledo Co., Ohio, USA) and then transferred to a glass extraction vial. Lipids were extracted from the seeds and trans-esterified in 500 ul 2.5% H₂SO₄ in MeOH for 3 hours at 80 °C, following the method of Browse *et al.* (Biochem J 235:25-31, 1986) with modifications. A known amount of heptadecanoic acid was included in the reaction as an internal standard. 750 ul of water and 400 ul of hexane were added to

each vial, which was then shaken vigorously and allowed to phase separate. Reaction vials were loaded directly onto gas chromatography (GC) for analysis and the upper hexane phase was sampled by the autosampler. Gas chromatography with Flame Ionization detection was used to separate and quantify the fatty acid methyl esters. Agilent 6890 Plus GC's were used for separation with Agilent Innnowax columns (30m x 0.25mm ID, 250um film thickness). The carrier gas was Hydrogen at a constant flow of 2.5 ml/ minute. 1ul of sample was injected in splitless mode (inlet temperature 220°C, Purge flow 15ml/min at 1 minute). The oven was programmed for an initial temperature of 105°C, initial time 0.5 minutes, followed by a ramp of 60°C per minute to 175°C, a 40°C /minute ramp to 260°C with a final hold time of 2 minutes. Detection was by Flame Ionization (Temperature 275°C, Fuel flow 30.0 ml/min, Oxidizer 400.0 ml/min). Instrument control and data collection and analysis were monitored using the Millennium Chromatography Management System (Version 3.2, Waters Corporation, Milford, MA). Peaks were initially identified by comparison with standards. Integration and quantification were performed automatically, but all analyses were subsequently examined manually to verify correct peak identification and acceptable signal to noise ratio before inclusion of the derived results in the study.

The association of an IMQ nucleic acid sequence with an IMQ phenotype or an IOQ phenotype was discovered by analysis of the genomic DNA sequence flanking the T-DNA insertion in the ACTTAG line identified in column 3 of Table 1. An ACTTAG line is a family of plants derived from a single plant that was transformed with a T-DNA element containing four tandem copies of the CaMV 35S enhancers. Accordingly, the disclosed IMQ nucleic acid sequences and/or polypeptides may be employed in the development of transgenic plants having an improved seed quality phenotype, including an IMQ phenotype and/or an IOQ phenotype. IMQ nucleic acid sequences may be used in the generation of transgenic plants, such as oilseed crops, that provide improved oil yield from oilseed processing and result in an increase in the quantity of oil recovered from seeds of the transgenic plant. IMQ nucleic acid sequences may also be used in the generation of transgenic plants, such as feed grain crops, that provide an IMQ phenotype resulting in increased energy for animal feeding, for example, seeds or seed meal with an

altered protein and/or fiber content, resulting in an increase in AME. IMQ nucleic acid sequences may further be used to increase the oil content of specialty oil crops, in order to augment yield and/or recovery of desired unusual fatty acids. Transgenic plants that have been genetically modified to express IMQ polypeptides can be used
 5 in the production of seeds, wherein the transgenic plants are grown, and oil and seed meal are obtained from plant parts (*e.g.* seed) using standard methods.

IMQ Nucleic Acids and Polypeptides

The IMQ designation for each of the IMQ nucleic acid sequences discovered
 10 in the activation tagging screen described herein are listed in column 1 of Tables 1-3, below. The disclosed IMQ polypeptides are listed in column 5 of Table 2 and column 4 of Table 3. As used herein, the term “IMQ polypeptide” refers to any polypeptide that when expressed in a plant causes an IMQ phenotype and/or an IOQ phenotype in any part of the plant, for example the seeds. In one embodiment, an
 15 IMQ polypeptide refers to a full-length IMQ protein, or a fragment, derivative (variant), or ortholog thereof that is “functionally active,” such that the protein fragment, derivative, or ortholog exhibits one or more of the functional activities associated with one or more of the disclosed full-length IMQ polypeptides, for example, the amino acid sequences provided in the GenBank entry referenced in
 20 column 5 of Table 2, which correspond to the amino acid sequences set forth as SEQ ID NO: 2, SEQ ID NO: 4, SEQ ID NO: 6, SEQ ID NO: 8, SEQ ID NO: 10, SEQ ID NO: 12, SEQ ID NO: 14, SEQ ID NO: 16, SEQ ID NO: 18, SEQ ID NO: 20, SEQ ID NO: 22, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 28, SEQ ID NO: 30, SEQ ID NO: 32, SEQ ID NO: 34, SEQ ID NO: 36, SEQ ID NO: 38, SEQ ID NO:
 25 40, SEQ ID NO: 42, SEQ ID NO: 44, SEQ ID NO: 46, SEQ ID NO: 48, SEQ ID NO: 50, SEQ ID NO: 52, SEQ ID NO: 54, SEQ ID NO: 56, SEQ ID NO: 58, SEQ ID NO: 60, SEQ ID NO: 62, SEQ ID NO: 64, SEQ ID NO: 66, SEQ ID NO: 68, SEQ ID NO: 70, SEQ ID NO: 72, SEQ ID NO: 74, SEQ ID NO: 76, SEQ ID NO: 78, SEQ ID NO: 80, SEQ ID NO: 82, SEQ ID NO: 84, SEQ ID NO: 86, SEQ ID
 30 NO: 88, SEQ ID NO: 90, SEQ ID NO: 92, SEQ ID NO: 94, SEQ ID NO: 96, SEQ ID NO: 98, or SEQ ID NO: 100, or an ortholog thereof. In one preferred embodiment, a functionally active IMQ polypeptide causes an IMQ phenotype

and/or an IOQ phenotype in a transgenic plant. In another embodiment, a functionally active IMQ polypeptide causes an altered oil, protein, and/or fiber content phenotype (for example, an altered seed meal content phenotype) when mis-expressed in a plant. In other preferred embodiments, mis-expression of the IMQ polypeptide causes a high oil (such as, increased oil), high protein (such as, increased protein), and/or low fiber (such as, decreased fiber) phenotype in a plant. In another embodiment, mis-expression of the IMQ polypeptide causes an improved AME of meal. In yet another embodiment, a functionally active IMQ polypeptide can rescue defective (including deficient) endogenous IMQ activity when expressed in a plant or in plant cells; the rescuing polypeptide may be from the same or from a different species as the species with the defective polypeptide activity. The disclosure also provides feed, meal, grain, food, or seed comprising the IMQ polypeptide, or a fragment, derivative (variant), or ortholog thereof.

In another embodiment, a functionally active fragment of a full length IMQ polypeptide (for example, a functionally active fragment of a native polypeptide having the amino acid sequence set forth as SEQ ID NO: 2, SEQ ID NO: 4, SEQ ID NO: 6, SEQ ID NO: 8, SEQ ID NO: 10, SEQ ID NO: 12, SEQ ID NO: 14, SEQ ID NO: 16, SEQ ID NO: 18, SEQ ID NO: 20, SEQ ID NO: 22, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 28, SEQ ID NO: 30, SEQ ID NO: 32, SEQ ID NO: 34, SEQ ID NO: 36, SEQ ID NO: 38, SEQ ID NO: 40, SEQ ID NO: 42, SEQ ID NO: 44, SEQ ID NO: 46, SEQ ID NO: 48, SEQ ID NO: 50, SEQ ID NO: 52, SEQ ID NO: 54, SEQ ID NO: 56, SEQ ID NO: 58, SEQ ID NO: 60, SEQ ID NO: 62, SEQ ID NO: 64, SEQ ID NO: 66, SEQ ID NO: 68, SEQ ID NO: 70, SEQ ID NO: 72, SEQ ID NO: 74, SEQ ID NO: 76, SEQ ID NO: 78, SEQ ID NO: 80, SEQ ID NO: 82, SEQ ID NO: 84, SEQ ID NO: 86, SEQ ID NO: 88, SEQ ID NO: 90, SEQ ID NO: 92, SEQ ID NO: 94, SEQ ID NO: 96, SEQ ID NO: 98, or SEQ ID NO: 100, or a naturally occurring ortholog thereof) retains one or more of the biological properties associated with the full-length IMQ polypeptide, such as signaling activity, binding activity, catalytic activity, or cellular or extra-cellular localizing activity. An IMQ fragment preferably comprises an IMQ domain, such as a C- or N-terminal or catalytic domain, among others, and preferably comprises at least 10, preferably at least 20, more preferably at least 25, and most preferably at least 50

contiguous amino acids of an IMQ protein. Functional domains of IMQ genes are listed in column 8 of Table 2 and can be identified using the PFAM program (Bateman A *et al.*, 1999, *Nucleic Acids Res.* 27:260-262) or INTERPRO (Mulder *et al.*, 2003, *Nucleic Acids Res.* 31, 315-318) program. Functionally active variants of full-length IMQ polypeptides, or fragments thereof, include polypeptides with amino acid insertions, deletions, or substitutions that retain one or more of the biological properties associated with the full-length IMQ polypeptide. In some cases, variants are generated that change the post-translational processing of an IMQ polypeptide. For instance, variants may have altered protein transport or protein localization characteristics, or altered protein half-life, compared to the native polypeptide.

As used herein, the term "IMQ nucleic acid" refers to any polynucleotide that when expressed in a plant causes an IMQ phenotype and/or an IOQ phenotype in any part of the plant, for example the seeds. In one embodiment, an IMQ polynucleotide encompasses nucleic acids with the sequence provided in or complementary to the GenBank entry referenced in column 3 of Table 2, which correspond to nucleic acid sequences set forth as SEQ ID NO: 1, SEQ ID NO: 3, SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 9, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15, SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, SEQ ID NO: 25, SEQ ID NO: 27, SEQ ID NO: 29, SEQ ID NO: 31, SEQ ID NO: 33, SEQ ID NO: 35, SEQ ID NO: 37, SEQ ID NO: 39, SEQ ID NO: 41, SEQ ID NO: 43, SEQ ID NO: 45, SEQ ID NO: 47, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 53, SEQ ID NO: 55, SEQ ID NO: 57, SEQ ID NO: 59, SEQ ID NO: 61, SEQ ID NO: 63, SEQ ID NO: 65, SEQ ID NO: 67, SEQ ID NO: 69, SEQ ID NO: 71, SEQ ID NO: 73, SEQ ID NO: 75, SEQ ID NO: 77, SEQ ID NO: 79, SEQ ID NO: 81, SEQ ID NO: 83, SEQ ID NO: 85, SEQ ID NO: 87, SEQ ID NO: 89, SEQ ID NO: 91, SEQ ID NO: 93, SEQ ID NO: 95, SEQ ID NO: 97, or SEQ ID NO: 99, as well as functionally active fragments, derivatives, or orthologs thereof. An IMQ nucleic acid of this disclosure may be DNA, derived from genomic DNA or cDNA, or RNA. Genomic sequences of the genes listed in Table 2 are known and available in public databases such as GenBank.

In one embodiment, a functionally active IMQ nucleic acid encodes or is complementary to a nucleic acid that encodes a functionally active IMQ

polypeptide. A functionally active IMQ nucleic acid also includes genomic DNA that serves as a template for a primary RNA transcript (*i.e.*, an mRNA precursor) that requires processing, such as splicing, before encoding the functionally active IMQ polypeptide. An IMQ nucleic acid can include other non-coding sequences, which may or may not be transcribed; such sequences include 5' and 3' UTRs, polyadenylation signals and regulatory sequences that control gene expression, among others, as are known in the art. Some polypeptides require processing events, such as proteolytic cleavage, covalent modification, etc., in order to become fully active. Accordingly, functionally active nucleic acids may encode the mature or the pre-processed IMQ polypeptide, or an intermediate form. An IMQ polynucleotide can also include heterologous coding sequences, for example, sequences that encode a marker included to facilitate the purification of the fused polypeptide, or a transformation marker. In another embodiment, a functionally active IMQ nucleic acid is capable of being used in the generation of loss-of-function IMQ phenotypes, for instance, via antisense suppression, co-suppression, etc. The disclosure also provides feed, meal, grain, food, or seed comprising a nucleic acid sequence that encodes an IMQ polypeptide.

In one preferred embodiment, an IMQ nucleic acid used in the disclosed methods comprises a nucleic acid sequence that encodes, or is complementary to a sequence that encodes, an IMQ polypeptide having at least 50%, 60%, 70%, 75%, 80%, 85%, 90%, 95%, 97%, 98%, or 99% sequence identity to a disclosed IMQ polypeptide sequence, for example the amino acid sequence set forth as SEQ ID NO: 2, SEQ ID NO: 4, SEQ ID NO: 6, SEQ ID NO: 8, SEQ ID NO: 10, SEQ ID NO: 12, SEQ ID NO: 14, SEQ ID NO: 16, SEQ ID NO: 18, SEQ ID NO: 20, SEQ ID NO: 22, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 28, SEQ ID NO: 30, SEQ ID NO: 32, SEQ ID NO: 34, SEQ ID NO: 36, SEQ ID NO: 38, SEQ ID NO: 40, SEQ ID NO: 42, SEQ ID NO: 44, SEQ ID NO: 46, SEQ ID NO: 48, SEQ ID NO: 50, SEQ ID NO: 52, SEQ ID NO: 54, SEQ ID NO: 56, SEQ ID NO: 58, SEQ ID NO: 60, SEQ ID NO: 62, SEQ ID NO: 64, SEQ ID NO: 66, SEQ ID NO: 68, SEQ ID NO: 70, SEQ ID NO: 72, SEQ ID NO: 74, SEQ ID NO: 76, SEQ ID NO: 78, SEQ ID NO: 80, SEQ ID NO: 82, SEQ ID NO: 84, SEQ ID NO: 86, SEQ ID

NO: 88, SEQ ID NO: 90, SEQ ID NO: 92, SEQ ID NO: 94, SEQ ID NO: 96, SEQ ID NO: 98, or SEQ ID NO: 100.

In another embodiment, an IMQ polypeptide comprises a polypeptide sequence with at least 50% or 60% identity to a disclosed IMQ polypeptide sequence (for example, the amino acid sequence set forth as SEQ ID NO: 2, SEQ ID NO: 4, SEQ ID NO: 6, SEQ ID NO: 8, SEQ ID NO: 10, SEQ ID NO: 12, SEQ ID NO: 14, SEQ ID NO: 16, SEQ ID NO: 18, SEQ ID NO: 20, SEQ ID NO: 22, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 28, SEQ ID NO: 30, SEQ ID NO: 32, SEQ ID NO: 34, SEQ ID NO: 36, SEQ ID NO: 38, SEQ ID NO: 40, SEQ ID NO: 42, SEQ ID NO: 44, SEQ ID NO: 46, SEQ ID NO: 48, SEQ ID NO: 50, SEQ ID NO: 52, SEQ ID NO: 54, SEQ ID NO: 56, SEQ ID NO: 58, SEQ ID NO: 60, SEQ ID NO: 62, SEQ ID NO: 64, SEQ ID NO: 66, SEQ ID NO: 68, SEQ ID NO: 70, SEQ ID NO: 72, SEQ ID NO: 74, SEQ ID NO: 76, SEQ ID NO: 78, SEQ ID NO: 80, SEQ ID NO: 82, SEQ ID NO: 84, SEQ ID NO: 86, SEQ ID NO: 88, SEQ ID NO: 90, SEQ ID NO: 92, SEQ ID NO: 94, SEQ ID NO: 96, SEQ ID NO: 98, or SEQ ID NO: 100) and may have at least 70%, 80%, 85%, 90%, 95%, 97%, 98%, or 99% sequence identity to a disclosed IMQ polypeptide sequence. In a further embodiment, an IMQ polypeptide comprises 50%, 60%, 70%, 80%, 85%, 90%, 95%, 97%, 98%, or 99% sequence identity to a disclosed IMQ polypeptide sequence, and may include a conserved protein domain of the IMQ polypeptide (such as the protein domain(s) listed in column 8 of Table 2). In another embodiment, an IMQ polypeptide comprises a polypeptide sequence with at least 50%, 60%, 70%, 80%, 85%, 90%, 95%, 97%, 98%, or 99% sequence identity to a functionally active fragment of the polypeptide referenced in column 5 of Table 2. In yet another embodiment, an IMQ polypeptide comprises a polypeptide sequence with at least 50%, 60 %, 70%, 80%, 90%, 95%, 97%, 98%, or 99% identity to the polypeptide sequence of the GenBank entry referenced in column 5 of Table 2 over its entire length and comprises a conserved protein domain(s) listed in column 8 of Table 2.

In another aspect, an IMQ polynucleotide sequence is at least 50% to 60% identical over its entire length to a disclosed IMQ nucleic acid sequence, such as the nucleic acid sequence set forth as SEQ ID NO: 1, SEQ ID NO: 3, SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 9, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15,

SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, SEQ ID NO: 25, SEQ ID NO: 27, SEQ ID NO: 29, SEQ ID NO: 31, SEQ ID NO: 33, SEQ ID NO: 35, SEQ ID NO: 37, SEQ ID NO: 39, SEQ ID NO: 41, SEQ ID NO: 43, SEQ ID NO: 45, SEQ ID NO: 47, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 53, SEQ ID NO: 55, SEQ ID NO: 57, SEQ ID NO: 59, SEQ ID NO: 61, SEQ ID NO: 63, SEQ ID NO: 65, SEQ ID NO: 67, SEQ ID NO: 69, SEQ ID NO: 71, SEQ ID NO: 73, SEQ ID NO: 75, SEQ ID NO: 77, SEQ ID NO: 79, SEQ ID NO: 81, SEQ ID NO: 83, SEQ ID NO: 85, SEQ ID NO: 87, SEQ ID NO: 89, SEQ ID NO: 91, SEQ ID NO: 93, SEQ ID NO: 95, SEQ ID NO: 97, or SEQ ID NO: 99, or nucleic acid sequences that are complementary to such an IMQ sequence, and may comprise at least 70%, 80%, 85%, 90%, 95%, 97%, 98%, or 99% sequence identity to the disclosed IMQ sequence, or a functionally active fragment thereof, or complementary sequences. In another embodiment, a disclosed IMQ nucleic acid comprises a nucleic acid sequence as shown in SEQ ID NO: 1, SEQ ID NO: 3, SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 9, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15, SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, SEQ ID NO: 25, SEQ ID NO: 27, SEQ ID NO: 29, SEQ ID NO: 31, SEQ ID NO: 33, SEQ ID NO: 35, SEQ ID NO: 37, SEQ ID NO: 39, SEQ ID NO: 41, SEQ ID NO: 43, SEQ ID NO: 45, SEQ ID NO: 47, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 53, SEQ ID NO: 55, SEQ ID NO: 57, SEQ ID NO: 59, SEQ ID NO: 61, SEQ ID NO: 63, SEQ ID NO: 65, SEQ ID NO: 67, SEQ ID NO: 69, SEQ ID NO: 71, SEQ ID NO: 73, SEQ ID NO: 75, SEQ ID NO: 77, SEQ ID NO: 79, SEQ ID NO: 81, SEQ ID NO: 83, SEQ ID NO: 85, SEQ ID NO: 87, SEQ ID NO: 89, SEQ ID NO: 91, SEQ ID NO: 93, SEQ ID NO: 95, SEQ ID NO: 97, or SEQ ID NO: 99,, or nucleic acid sequences that are complementary to such an IMQ sequence, and nucleic acid sequences that have substantial sequence homology to a such IMQ sequences. As used herein, the phrase “substantial sequence homology” refers to those nucleic acid sequences that have slight or inconsequential sequence variations from such IMQ sequences, *i.e.*, the sequences function in substantially the same manner and encode an IMQ polypeptide.

As used herein, “percent (%) sequence identity” with respect to a specified subject sequence, or a specified portion thereof, is defined as the percentage of

nucleotides or amino acids in an identified sequence identical with the nucleotides or amino acids in the subject sequence (or specified portion thereof), after aligning the sequences and introducing gaps, if necessary to achieve the maximum percent sequence identity, as generated by the program WU-BLAST-2.0a19 (Altschul *et al.*,
5 *J. Mol. Biol.*, 1990, 215:403-410) with search parameters set to default values. The HSP S and HSP S2 parameters are dynamic values and are established by the program itself depending upon the composition of the particular sequence and composition of the particular database against which the sequence of interest is being searched. A “percent (%) identity value” is determined by the number of
10 matching identical nucleotides or amino acids divided by the sequence length for which the percent identity is being reported. “Percent (%) amino acid sequence similarity” is determined by performing the same calculation as for determining % amino acid sequence identity, but including conservative amino acid substitutions in addition to identical amino acids in the computation. A conservative amino acid
15 substitution is one in which an amino acid is substituted for another amino acid having similar properties such that the folding or activity of the protein is not significantly affected. Aromatic amino acids that can be substituted for each other are phenylalanine, tryptophan, and tyrosine; interchangeable hydrophobic amino acids are leucine, isoleucine, methionine, and valine; interchangeable polar amino
20 acids are glutamine and asparagine; interchangeable basic amino acids are arginine, lysine and histidine; interchangeable acidic amino acids are aspartic acid and glutamic acid; and interchangeable small amino acids are alanine, serine, threonine, cysteine and glycine.

Derivative nucleic acid molecules of the subject nucleic acid molecules
25 include sequences that selectively hybridize to the disclosed IMQ nucleic acid sequences (for example, the nucleic acid sequence set forth as SEQ ID NO: 1, SEQ ID NO: 3, SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 9, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15, SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, SEQ ID NO: 25, SEQ ID NO: 27, SEQ ID NO: 29, SEQ ID NO: 31,
30 SEQ ID NO: 33, SEQ ID NO: 35, SEQ ID NO: 37, SEQ ID NO: 39, SEQ ID NO: 41, SEQ ID NO: 43, SEQ ID NO: 45, SEQ ID NO: 47, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 53, SEQ ID NO: 55, SEQ ID NO: 57, SEQ ID NO: 59, SEQ

ID NO: 61, SEQ ID NO: 63, SEQ ID NO: 65, SEQ ID NO: 67, SEQ ID NO: 69, SEQ ID NO: 71, SEQ ID NO: 73, SEQ ID NO: 75, SEQ ID NO: 77, SEQ ID NO: 79, SEQ ID NO: 81, SEQ ID NO: 83, SEQ ID NO: 85, SEQ ID NO: 87, SEQ ID NO: 89, SEQ ID NO: 91, SEQ ID NO: 93, SEQ ID NO: 95, SEQ ID NO: 97, or
5 SEQ ID NO: 99). The stringency of hybridization can be controlled by temperature, ionic strength, pH, and the presence of denaturing agents such as formamide during hybridization and washing. Conditions routinely used are well known (see, *e.g.*, *Current Protocol in Molecular Biology*, Vol. 1, Chap. 2.10, John Wiley & Sons, Publishers (1994); Sambrook *et al.*, 1989, *Molecular Cloning: A Laboratory Manual*
10 (Second Edition), Cold Spring Harbor Press, Plainview, N.Y.,).

In some embodiments, a nucleic acid molecule of the disclosure is capable of hybridizing to a nucleic acid molecule containing the disclosed nucleotide sequence under stringent hybridization conditions that are: prehybridization of filters containing nucleic acid for 8 hours to overnight at 65° C in a solution comprising 6X
15 single strength citrate (SSC) (1X SSC is 0.15 M NaCl, 0.015 M Na citrate; pH 7.0), 5X Denhardt's solution, 0.05% sodium pyrophosphate and 100 µg/ml herring sperm DNA; hybridization for 18-20 hours at 65° C in a solution containing 6X SSC, 1X Denhardt's solution, 100 µg/ml yeast tRNA and 0.05% sodium pyrophosphate; and washing of filters at 65° C for 1 h in a solution containing 0.1X SSC and 0.1% SDS
20 (sodium dodecyl sulfate). In other embodiments, moderately stringent hybridization conditions are used that are: pretreatment of filters containing nucleic acid for 6 h at 40° C in a solution containing 35% formamide, 5X SSC, 50 mM Tris-HCl (pH 7.5), 5 mM EDTA, 0.1% PVP, 0.1% Ficoll, 1% BSA, and 500 µg/ml denatured salmon sperm DNA; hybridization for 18-20 h at 40° C in a solution containing 35%
25 formamide, 5X SSC, 50 mM Tris-HCl (pH 7.5), 5 mM EDTA, 0.02% PVP, 0.02% Ficoll, 0.2% BSA, 100 µg/ml salmon sperm DNA, and 10% (wt/vol) dextran sulfate; followed by washing twice for 1 hour at 55° C in a solution containing 2X SSC and 0.1% SDS. Alternatively, low stringency conditions can be used that comprise: incubation for 8 hours to overnight at 37° C in a solution comprising 20%
30 formamide, 5 x SSC, 50 mM sodium phosphate (pH 7.6), 5X Denhardt's solution, 10% dextran sulfate, and 20 µg/ml denatured sheared salmon sperm DNA;

hybridization in the same buffer for 18 to 20 hours; and washing of filters in 1 x SSC at about 37° C for 1 hour.

As a result of the degeneracy of the genetic code, a number of polynucleotide sequences encoding an IMQ polypeptide can be produced. For example, codons may
5 be selected to increase the rate at which expression of the polypeptide occurs in a particular host species, in accordance with the optimum codon usage dictated by the particular host organism (see, *e.g.*, Nakamura *et al.*, 1999, *Nucleic Acids Res.* 27:292). Such sequence variants may be used in the methods disclosed herein.

The disclosed methods may use orthologs of a disclosed *Arabidopsis* IMQ
10 nucleic acid sequence. Representative putative orthologs of each of the disclosed *Arabidopsis* IMQ genes are identified in column 3 of Table 3, below. Methods of identifying the orthologs in other plant species are known in the art. In general, orthologs in different species retain the same function, due to presence of one or more protein motifs and/or 3-dimensional structures. In evolution, when a gene
15 duplication event follows speciation, a single gene in one species, such as *Arabidopsis*, may correspond to multiple genes (paralogs) in another. As used herein, the term “orthologs” encompasses paralogs. When sequence data is available for a particular plant species, orthologs are generally identified by sequence homology analysis, such as BLAST analysis, usually using protein bait
20 sequences. Sequences are assigned as a potential ortholog if the best hit sequence from the forward BLAST result retrieves the original query sequence in the reverse BLAST (Huynen MA and Bork P, 1998, *Proc. Natl. Acad. Sci.*, 95:5849-5856; Huynen MA *et al.*, 2000, *Genome Research*, 10:1204-1210).

Programs for multiple sequence alignment, such as CLUSTAL (Thompson
25 JD *et al.*, 1994, *Nucleic Acids Res.* 22:4673-4680) may be used to highlight conserved regions and/or residues of orthologous proteins and to generate phylogenetic trees. In a phylogenetic tree representing multiple homologous sequences from diverse species (*e.g.*, retrieved through BLAST analysis), orthologous sequences from two species generally appear closest on the tree with
30 respect to all other sequences from these two species. Structural threading or other analysis of protein folding (*e.g.*, using software by ProCeryon, Biosciences, Salzburg, Austria) may also identify potential orthologs. Nucleic acid hybridization

methods may also be used to find orthologous genes and are preferred when sequence data are not available. Degenerate PCR and screening of cDNA or genomic DNA libraries are common methods for finding related gene sequences and are well known in the art (see, *e.g.*, Sambrook, 1989, *Molecular Cloning: A Laboratory Manual* (Second Edition), Cold Spring Harbor Press, Plainview, N.Y.; Dieffenbach and Dveksler, 1995, *PCR Primer: A Laboratory Manual*, Cold Spring Harbor Laboratory Press, NY). For instance, methods for generating a cDNA library from the plant species of interest and probing the library with partially homologous gene probes are described in Sambrook *et al.* A highly conserved portion of the *Arabidopsis* IMQ coding sequence may be used as a probe. IMQ ortholog nucleic acids may hybridize to the nucleic acid of SEQ ID NO: 1, SEQ ID NO: 3, SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 9, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15, SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, SEQ ID NO: 25, SEQ ID NO: 27, SEQ ID NO: 29, SEQ ID NO: 31, SEQ ID NO: 33, SEQ ID NO: 35, SEQ ID NO: 37, SEQ ID NO: 39, SEQ ID NO: 41, SEQ ID NO: 43, SEQ ID NO: 45, SEQ ID NO: 47, SEQ ID NO: 49, SEQ ID NO: 51, SEQ ID NO: 53, SEQ ID NO: 55, SEQ ID NO: 57, SEQ ID NO: 59, SEQ ID NO: 61, SEQ ID NO: 63, SEQ ID NO: 65, SEQ ID NO: 67, SEQ ID NO: 69, SEQ ID NO: 71, SEQ ID NO: 73, SEQ ID NO: 75, SEQ ID NO: 77, SEQ ID NO: 79, SEQ ID NO: 81, SEQ ID NO: 83, SEQ ID NO: 85, SEQ ID NO: 87, SEQ ID NO: 89, SEQ ID NO: 91, SEQ ID NO: 93, SEQ ID NO: 95, SEQ ID NO: 97, or SEQ ID NO: 99 under high, moderate, or low stringency conditions. After amplification or isolation of a segment of a putative ortholog, that segment may be cloned and sequenced by standard techniques and utilized as a probe to isolate a complete cDNA or genomic DNA clone.

Alternatively, it is possible to initiate an EST project to generate a database of sequence information for the plant species of interest. In another approach, antibodies that specifically bind known IMQ polypeptides are used for ortholog isolation (see, *e.g.*, Harlow and Lane, 1988, 1999, *Antibodies: A Laboratory Manual*, Cold Spring Harbor Laboratory Press, New York). Western blot analysis can determine that an IMQ ortholog (*i.e.*, a protein orthologous to a disclosed IMQ polypeptide) is present in a crude extract of a particular plant species. When

reactivity is observed, the sequence encoding the candidate ortholog may be isolated by screening expression libraries representing the particular plant species.

Expression libraries can be constructed in a variety of commercially available vectors, including lambda gt11, as described in Sambrook, *et al.*, 1989. Once the
5 candidate ortholog(s) are identified by any of these means, candidate orthologous sequence are used as bait (the “query”) for the reverse BLAST against sequences from *Arabidopsis* or other species in which IMQ nucleic acid and/or polypeptide sequences have been identified.

IMQ nucleic acids and polypeptides may be obtained using any available
10 method. For instance, techniques for isolating cDNA or genomic DNA sequences of interest by screening DNA libraries or by using polymerase chain reaction (PCR), as previously described, are well known in the art. Alternatively, nucleic acid sequence may be synthesized. Any known method, such as site directed mutagenesis (Kunkel
TA *et al.*, 1991, *Methods Enzymol.* 204:125-39), may be used to introduce desired
15 changes into a cloned nucleic acid.

In general, the methods disclosed herein involve incorporating the desired form of the IMQ nucleic acid into a plant expression vector for transformation of plant cells, and the IMQ polypeptide is expressed in the host plant. Transformed plants and plant cells expressing an IMQ polypeptide express an IMQ phenotype
20 and/or an IOQ phenotype and, in one specific, non-limiting example, may have high (increased) oil, high (increased) protein, and/or low (decreased) fiber content.

An “isolated” IMQ nucleic acid molecule is other than in the form or setting in which it is found in nature, and is identified and separated from least one contaminant nucleic acid molecule with which it is ordinarily associated in the
25 natural source of the IMQ nucleic acid. However, an isolated IMQ nucleic acid molecule includes IMQ nucleic acid molecules contained in cells that ordinarily express IMQ where, for example, the nucleic acid molecule is in a chromosomal location different from that of natural cells.

**Generation of Genetically Modified Plants with an Improved Oil Quantity
Phenotype and/or an Improved Meal Quality Phenotype**

The disclosed IMQ nucleic acids and polypeptides may be used in the generation of transgenic plants having a modified or altered oil, protein, and/or fiber content phenotype. As used herein, an “altered oil content (phenotype)” may refer to altered oil content in any part of the plant. In a preferred embodiment, altered expression of the IMQ gene in a plant is used to generate plants with a high oil content (phenotype). As used herein, an “altered protein content (phenotype)” may refer to altered protein content in any part of the plant. In a preferred embodiment, altered expression of the IMQ gene in a plant is used to generate plants with a high (or increased) protein content (phenotype). As used herein, an “altered fiber content (phenotype)” may refer to altered fiber content in any part of the plant. In a preferred embodiment, altered expression of the IMQ gene in a plant is used to generate plants with a low (or decreased) fiber content (phenotype). The altered oil, protein, and/or fiber content is often observed in seeds. Examples of a transgenic plant include plants comprising a plant transformation vector with a nucleotide sequence that encodes or is complementary to a sequence that encodes an IMQ polypeptide having the amino acid sequence as set forth in SEQ ID NO: 2, SEQ ID NO: 4, SEQ ID NO: 6, SEQ ID NO: 8, SEQ ID NO: 10, SEQ ID NO: 12, SEQ ID NO: 14, SEQ ID NO: 16, SEQ ID NO: 18, SEQ ID NO: 20, SEQ ID NO: 22, SEQ ID NO: 24, SEQ ID NO: 26, SEQ ID NO: 28, SEQ ID NO: 30, SEQ ID NO: 32, SEQ ID NO: 34, SEQ ID NO: 36, SEQ ID NO: 38, SEQ ID NO: 40, SEQ ID NO: 42, SEQ ID NO: 44, SEQ ID NO: 46, SEQ ID NO: 48, SEQ ID NO: 50, SEQ ID NO: 52, SEQ ID NO: 54, SEQ ID NO: 56, SEQ ID NO: 58, SEQ ID NO: 60, SEQ ID NO: 62, SEQ ID NO: 64, SEQ ID NO: 66, SEQ ID NO: 68, SEQ ID NO: 70, SEQ ID NO: 72, SEQ ID NO: 74, SEQ ID NO: 76, SEQ ID NO: 78, SEQ ID NO: 80, SEQ ID NO: 82, SEQ ID NO: 84, SEQ ID NO: 86, SEQ ID NO: 88, SEQ ID NO: 90, SEQ ID NO: 92, SEQ ID NO: 94, SEQ ID NO: 96, SEQ ID NO: 98, or SEQ ID NO: 100, or an ortholog thereof.

Transgenic plants, such as corn, soybean and canola containing the disclosed nucleic acid sequences, can be used in the production of vegetable oil and meal. Vegetable oil is used in a variety of food products, while meal from seed is used as

an animal feed. After harvesting seed from transgenic plants, the seed is cleaned to remove plant stalks and other material and then flaked in roller mills to break the hulls. The crushed seed is heated to 75-100° C to denature hydrolytic enzymes, lyse the unbroken oil containing cells, and allow small oil droplets to coalesce. Most of the oil is then removed (and can be recovered) by pressing the seed material in a screw press. The remaining oil is removed from the presscake by extraction with and organic solvents, such as hexane. The solvent is removed from the meal by heating it to approximately 100° C. After drying, the meal is then granulated to a consistent form. The meal, containing the protein, digestible carbohydrate, and fiber of the seed, may be mixed with other materials prior to being used as an animal feed.

The methods described herein for generating transgenic plants are generally applicable to all plants. Although activation tagging and gene identification is carried out in *Arabidopsis*, the IMQ nucleic acid sequence (or an ortholog, variant or fragment thereof) may be expressed in any type of plant. In a preferred embodiment, oil-producing plants produce and store triacylglycerol in specific organs, primarily in seeds. Such species include soybean (*Glycine max*), rapeseed and canola (including *Brassica napus*, *B. campestris*), sunflower (*Helianthus annuus*), cotton (*Gossypium hirsutum*), corn (*Zea mays*), cocoa (*Theobroma cacao*), safflower (*Carthamus tinctorius*), oil palm (*Elaeis guineensis*), coconut palm (*Cocos nucifera*), flax (*Linum usitatissimum*), castor (*Ricinus communis*), and peanut (*Arachis hypogaea*), as well as wheat, rice and oat. Fruit- and vegetable-bearing plants, grain-producing plants, nut-producing plants, rapid cycling *Brassica* species, alfalfa (*Medicago sativa*), tobacco (*Nicotiana*), turfgrass (*Poaceae* family), other forage crops, and wild species may also be a source of unique fatty acids. In other embodiments, any plant expressing the IMQ nucleic acid sequence can also express increased protein and/or decreased fiber content in a specific plant part or organ, such as in seeds.

The skilled artisan will recognize that a wide variety of transformation techniques exist in the art, and new techniques are continually becoming available. Any technique that is suitable for the target host plant can be employed within the scope of the present invention. For example, the constructs can be introduced in a variety of forms including, but not limited to, as a strand of DNA, in a plasmid, or in

an artificial chromosome. The introduction of the constructs into the target plant cells can be accomplished by a variety of techniques, including, but not limited to, *Agrobacterium*-mediated transformation, electroporation, microinjection, microprojectile bombardment, calcium-phosphate-DNA co-precipitation, or liposome-mediated transformation of a heterologous nucleic acid. The transformation of the plant is preferably permanent, *i.e.* by integration of the introduced expression constructs into the host plant genome, so that the introduced constructs are passed onto successive plant generations. Depending upon the intended use, a heterologous nucleic acid construct comprising an IMQ polynucleotide may encode the entire protein or a biologically active portion thereof.

In one embodiment, binary Ti-based vector systems may be used to transfer polynucleotides. Standard *Agrobacterium* binary vectors are known to those of skill in the art, and many are commercially available (*e.g.*, pBI121 Clontech Laboratories, Palo Alto, CA). A construct or vector may include a plant promoter to express the nucleic acid molecule of choice. In a preferred embodiment, the promoter is a plant promoter.

The optimal procedure for transformation of plants with *Agrobacterium* vectors will vary with the type of plant being transformed. Exemplary methods for *Agrobacterium*-mediated transformation include transformation of explants of hypocotyl, shoot tip, stem or leaf tissue, derived from sterile seedlings and/or plantlets. Such transformed plants may be reproduced sexually, or by cell or tissue culture. *Agrobacterium* transformation has been previously described for a large number of different types of plants and methods for such transformation may be found in the scientific literature. Of particular relevance are methods to transform commercially important crops, such as plants of the *Brassica* species, including canola and rapeseed, (De Block *et al.*, 1989, *Plant Physiol.*, 91:694-701), sunflower (Everett *et al.*, 1987, *Bio/Technology*, 5:1201), soybean (Christou *et al.*, 1989, *Proc. Natl. Acad. Sci USA*, 86:7500-7504; Kline *et al.*, 1987, *Nature*, 327:70), wheat, rice and oat.

Expression (including transcription and translation) of an IMQ nucleic acid sequence may be regulated with respect to the level of expression, the tissue type(s) where expression takes place and/or developmental stage of expression. A number

of heterologous regulatory sequences (e.g., promoters and enhancers) are available for controlling the expression of an IMQ nucleic acid. These include constitutive, inducible and regulatable promoters, as well as promoters and enhancers that control expression in a tissue- or temporal-specific manner. Exemplary constitutive

5 promoters include the raspberry E4 promoter (U.S. Patent Nos. 5,783,393 and 5,783,394), the nopaline synthase (NOS) promoter (Ebert *et al.*, *Proc. Natl. Acad. Sci. (U.S.A.)* 84:5745-5749, 1987), the octopine synthase (OCS) promoter (which is carried on tumor-inducing plasmids of *Agrobacterium tumefaciens*), the caulimovirus promoters such as the cauliflower mosaic virus (CaMV) 19S promoter

10 (Lawton *et al.*, *Plant Mol. Biol.* 9:315-324, 1987) and the CaMV 35S promoter (Odell *et al.*, *Nature* 313:810-812, 1985 and Jones JD *et al.*, 1992, *Transgenic Res.*, 1:285-297), the figwort mosaic virus 35S-promoter (U.S. Patent No. 5,378,619), the light-inducible promoter from the small subunit of ribulose-1,5-bis-phosphate carboxylase (ssRUBISCO), the Adh promoter (Walker *et al.*, *Proc. Natl. Acad. Sci. (U.S.A.)* 84:6624-6628, 1987), the sucrose synthase promoter (Yang *et al.*, *Proc. Natl. Acad. Sci. (U.S.A.)* 87:4144-4148, 1990), the R gene complex promoter (Chandler *et al.*, *The Plant Cell* 1:1175-1183, 1989), the chlorophyll a/b binding protein gene promoter, the CsVMV promoter (Verdaguer B *et al.*, 1998, *Plant Mol Biol.*, 37:1055-1067), and the melon actin promoter (published PCT application

15 WO0056863). Exemplary tissue-specific promoters include the tomato E4 and E8 promoters (U.S. Patent No. 5,859,330) and the tomato 2AII gene promoter (Van Haaren MJJ *et al.*, 1993, *Plant Mol Bio.*, 21:625-640).

In one preferred embodiment, expression of the IMQ nucleic acid sequence is under control of regulatory sequences from genes whose expression is associated

25 with early seed and/or embryo development. Indeed, in a preferred embodiment, the promoter used is a seed-enhanced promoter. Examples of such promoters include the 5' regulatory regions from such genes as napin (Kridl *et al.*, *Seed Sci. Res.* 1:209:219, 1991), globulin (Belanger and Kriz, *Genet.*, 129: 863-872, 1991, GenBank Accession No. L22295), gamma zein Z 27 (Lopes *et al.*, *Mol Gen Genet.*, 247:603-613, 1995), L3 oleosin promoter (U.S. Patent No. 6,433,252), phaseolin (Bustos *et al.*, *Plant Cell*, 1(9):839-853, 1989), arcelin5 (U.S. Application No. 2003/0046727), a soybean 7S promoter, a 7S α promoter (U.S. Application No.

30

2003/0093828), the soybean 7S α ' beta conglycinin promoter, a 7S α ' promoter (Beachy *et al.*, *EMBO J.*, 4:3047, 1985; Schuler *et al.*, *Nucleic Acid Res.*, 10(24):8225-8244, 1982), soybean trypsin inhibitor (Riggs *et al.*, *Plant Cell* 1(6):609-621, 1989), ACP (Baerson *et al.*, *Plant Mol. Biol.*, 22(2):255-267, 1993),
5 stearyl-ACP desaturase (Slocombe *et al.*, *Plant Physiol.* 104(4):167-176, 1994), soybean α ' subunit of β -conglycinin (Chen *et al.*, *Proc. Natl. Acad. Sci.* 83:8560-8564, 1986), *Vicia faba* USP (P-Vf.Usp, SEQ ID NO: 1, 2, and 3 in (U.S. Application No. 2003/229918) and *Zea mays* L3 oleosin promoter (Hong *et al.*, *Plant Mol. Biol.*, 34(3):549-555, 1997). Also included are the zeins, which are a
10 group of storage proteins found in corn endosperm. Genomic clones for zein genes have been isolated (Pedersen *et al.*, *Cell*, 29:1015-1026, 1982; and Russell *et al.*, *Transgenic Res.* 6(2):157-168) and the promoters from these clones, including the 15 kD, 16 kD, 19 kD, 22 kD, 27 kD and genes, could also be used. Other promoters known to function, for example, in corn include the promoters for the following
15 genes: *waxy*, *Brittle*, *Shrunken 2*, Branching enzymes I and II, starch synthases, debranching enzymes, oleosins, glutelins and sucrose synthases. Legume genes whose promoters are associated with early seed and embryo development include *V. faba legumin* (Baumlein *et al.*, 1991, *Mol. Gen. Genet.* 225:121-8; Baumlein *et al.*, 1992, *Plant J.* 2:233-9), *V. faba usp* (Fiedler *et al.*, 1993, *Plant Mol. Biol.* 22:669-
20 79), pea *convicilin* (Bown *et al.*, 1988, *Biochem. J.* 251:717-26), pea *lectin* (dePater *et al.*, 1993, *Plant Cell* 5:877-86), *P. vulgaris beta phaseolin* (Bustos *et al.*, 1991, *EMBO J.* 10:1469-79), *P. vulgaris DLEC2* and *PHS [beta]* (Bobb *et al.*, 1997, *Nucleic Acids Res.* 25:641-7), and soybean *beta-Conglycinin*, 7S storage protein (Chamberland *et al.*, 1992, *Plant Mol. Biol.* 19:937-49).
25 Cereal genes whose promoters are associated with early seed and embryo development include rice *glutelin* ("GluA-3," Yoshihara and Takaiwa, 1996, *Plant Cell Physiol.* 37:107-11; "GluB-1," Takaiwa *et al.*, 1996, *Plant Mol. Biol.* 30:1207-21; Washida *et al.*, 1999, *Plant Mol. Biol.* 40:1-12; "Gt3," Leisy *et al.*, 1990, *Plant Mol. Biol.* 14:41-50), rice *prolamin* (Zhou & Fan, 1993, *Transgenic Res.* 2:141-6),
30 wheat *prolamin* (Hammond-Kosack *et al.*, 1993, *EMBO J.* 12:545-54), maize *zein* (Z4, Matzke *et al.*, 1990, *Plant Mol. Biol.* 14:323-32), and barley *B-hordeins* (Entwistle *et al.*, 1991, *Plant Mol. Biol.* 17:1217-31).

Other genes whose promoters are associated with early seed and embryo development include oil palm GLO7A (7S globulin, Morcillo *et al.*, 2001, *Physiol. Plant* 112:233-243), *Brassica napus napin*, 2S storage protein, and napA gene (Josefsson *et al.*, 1987, *J. Biol. Chem.* 262:12196-201; Stalberg *et al.*, 1993, *Plant Mol. Biol.* 1993 23:671-83; Ellerstrom *et al.*, 1996, *Plant Mol. Biol.* 32:1019-27), *Brassica napus oleosin* (Keddie *et al.*, 1994, *Plant Mol. Biol.* 24:327-40), *Arabidopsis oleosin* (Plant *et al.*, 1994, *Plant Mol. Biol.* 25:193-205), *Arabidopsis* FAE1 (Rossak *et al.*, 2001, *Plant Mol. Biol.* 46:717-25), *Canavalia gladiata* conA (Yamamoto *et al.*, 1995, *Plant Mol. Biol.* 27:729-41), and *Catharanthus roseus* strictosidine synthase (Str, Ouwerkerk and Memelink, 1999, *Mol. Gen. Genet.* 261:635-43). In another preferred embodiment, regulatory sequences from genes expressed during oil biosynthesis are used (see, *e.g.*, U.S. Patent No. 5,952, 544). Alternative promoters are from plant storage protein genes (Bevan *et al.*, 1993, *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* 342:209-15). Additional promoters that may be utilized are described, for example, in U.S. Patent Nos. 5,378,619; 5,391,725; 5,428,147; 5,447,858; 5,608,144; 5,608,144; 5,614,399; 5,633,441; 5,633,435; and 4,633,436.

In yet another aspect, in some cases it may be desirable to inhibit the expression of the endogenous IMQ nucleic acid sequence in a host cell. Exemplary methods for practicing this aspect of the invention include, but are not limited to antisense suppression (Smith, *et al.*, 1988, *Nature*, 334:724-726; van der Krol *et al.*, 1988, *BioTechniques*, 6:958-976); co-suppression (Napoli, *et al.*, 1990, *Plant Cell*, 2:279-289); ribozymes (PCT Publication WO 97/10328); and combinations of sense and antisense (Waterhouse, *et al.*, 1998, *Proc. Natl. Acad. Sci. USA*, 95:13959-13964). Methods for the suppression of endogenous sequences in a host cell typically employ the transcription or transcription and translation of at least a portion of the sequence to be suppressed. Such sequences may be homologous to coding as well as non-coding regions of the endogenous sequence. Antisense inhibition may use the entire cDNA sequence (Sheehy *et al.*, 1988, *Proc. Natl. Acad. Sci. USA*, 85:8805-8809), a partial cDNA sequence including fragments of 5' coding sequence, (Cannon *et al.*, 1990, *Plant Mol. Biol.*, 15:39-47), or 3' non-coding sequences (Ch'ng *et al.*, 1989, *Proc. Natl. Acad. Sci. USA*, 86:10006-10010).

Cosuppression techniques may use the entire cDNA sequence (Napoli *et al.*, 1990, *Plant Cell*, 2:279-289; van der Krol *et al.*, 1990, *Plant Cell*, 2:291-299), or a partial cDNA sequence (Smith *et al.*, 1990, *Mol. Gen. Genetics*, 224:477-481).

Standard molecular and genetic tests may be performed to further analyze the association between a nucleic acid sequence and an observed phenotype. Exemplary techniques are described below.

1. DNA/RNA analysis

The stage- and tissue-specific gene expression patterns in mutant versus wild-type lines may be determined, for instance, by *in situ* hybridization. Analysis of the methylation status of the gene, especially flanking regulatory regions, may be performed. Other suitable techniques include over-expression, ectopic expression, expression in other plant species and gene knock-out (reverse genetics, targeted knock-out, viral induced gene silencing (VIGS; see, Baulcombe D, 1999, *Arch. Virol. Suppl.* 15:189-201).

In a preferred application expression profiling, generally by microarray analysis, is used to simultaneously measure differences or induced changes in the expression of many different genes. Techniques for microarray analysis are well known in the art (Schena M *et al.*, *Science* 1995 270:467-470; Baldwin D *et al.*, 1999, *Cur. Opin. Plant Biol.* 2(2):96-103; Dangond F, *Physiol Genomics* (2000) 2:53-58; van Hal NL *et al.*, *J Biotechnol.* (2000) 78:271-280; Richmond T and Somerville S, *Curr. Opin. Plant Biol.* 2000 3:108-116). Expression profiling of individual tagged lines may be performed. Such analysis can identify other genes that are coordinately regulated as a consequence of the over-expression of the gene of interest, which may help to place an unknown gene in a particular pathway.

2. Gene Product Analysis

Analysis of gene products may include recombinant protein expression, antisera production, immunolocalization, biochemical assays for catalytic or other activity, analysis of phosphorylation status, and analysis of interaction with other proteins via yeast two-hybrid assays.

3. Pathway Analysis

Pathway analysis may include placing a gene or gene product within a particular biochemical, metabolic or signaling pathway based on its mis-expression

phenotype or by sequence homology with related genes. Alternatively, analysis may comprise genetic crosses with wild-type lines and other mutant lines (creating double mutants) to order the gene in a pathway, or determining the effect of a mutation on expression of downstream “reporter” genes in a pathway.

5

Generation of Mutated Plants with an Improved Oil Quantity Phenotype and/or Improved Meal Quality Phenotype

Additional methods are disclosed herein of generating a plant having an IMQ and/or an IOQ phenotype, wherein a plant is identified that has an allele in its IMQ
10 nucleic acid sequence that results in an IMQ phenotype and/or an IOQ phenotype, compared to plants lacking the allele. The plant can generate progeny, wherein the progeny inherit the allele and have an IMQ phenotype and/or an IOQ phenotype. For example, provided herein is a method of identifying plants that have mutations in the endogenous IMQ nucleic acid sequence that confer an IMQ phenotype and/or
15 an IOQ phenotype and generating progeny of these plants with an IMQ and/or IOQ phenotype that are not genetically modified. In some embodiments, the plants have an IMQ phenotype with an altered protein and/or fiber content or seed meal content, or an IOQ phenotype, with an altered oil content.

In one method, called “TILLING” (for targeting induced local lesions in
20 genomes), mutations are induced in the seed of a plant of interest, for example, using EMS (ethylmethane sulfonate) treatment. The resulting plants are grown and self-fertilized, and the progeny are used to prepare DNA samples. PCR amplification and sequencing of the IMQ nucleic acid sequence is used to identify whether a mutated plant has a mutation in the IMQ nucleic acid sequence. Plants having IMQ
25 mutations may then be tested for altered oil, protein, and/or fiber content, or alternatively, plants may be tested for altered oil, protein, and/or fiber content, and then PCR amplification and sequencing of the IMQ nucleic acid sequence is used to determine whether a plant having altered oil, protein, and/or fiber content has a mutated IMQ nucleic acid sequence. TILLING can identify mutations that may alter
30 the expression of specific genes or the activity of proteins encoded by these genes (see Colbert *et al.*, 2001, *Plant Physiol.* 126:480-484; McCallum *et al.*, 2000, *Nature Biotechnology* 18:455-457).

In another method, a candidate gene/Quantitative Trait Locus (QTLs) approach can be used in a marker-assisted breeding program to identify alleles of or mutations in the IMQ nucleic acid sequence or orthologs of the IMQ nucleic acid sequence that may confer altered oil, protein, and/or fiber content (see Bert *et al.*,
 5 *Theor Appl Genet.*, 2003 Jun;107(1):181-9; and Lionneton *et al.*, *Genome*, 2002 Dec;45(6):1203-15). Thus, in a further aspect of the disclosure, an IMQ nucleic acid is used to identify whether a plant having altered oil, protein, and/or fiber content has a mutation an endogenous IMQ nucleic acid sequence or has a particular allele that causes altered oil, protein, and/or fiber content.

10 While the disclosure has been described with reference to specific methods and embodiments, it will be appreciated that various modifications and changes may be made without departing from the disclosure. All publications cited herein are referenced for the purpose of describing and disclosing compositions and methodologies that might be used in connection with the disclosure.

15

EXAMPLES

EXAMPLE 1

Generation of Plants with an IMQ Phenotype and/or an IOQ Phenotype by Transformation with an Activation Tagging Construct

20 This Example describes the generation of transgenic plants with altered oil, protein, and/or fiber content.

Mutants were generated using the activation tagging "ACTTAG" vector, pSKI015 (GI#6537289; Weigel D *et al.*, 2000, *Plant Physiology*, 122:1003-1013). Standard methods were used for the generation of *Arabidopsis* transgenic plants, and
 25 were essentially as described in published application PCT WO0183697. Briefly, T0 *Arabidopsis* (Col-0) plants were transformed with *Agrobacterium* carrying the pSKI015 vector, which comprises T-DNA derived from the *Agrobacterium* Ti plasmid, an herbicide resistance selectable marker gene, and the 4X CaMV 35S enhancer element. Transgenic plants were selected at the T1 generation based on
 30 herbicide resistance. T2 seed (from T1 plants) was harvested and sown in soil. T2

plants were exposed to the herbicide to kill plants lacking the ACTTAG vector. T2 plants were grown to maturity, allowed to self-fertilize and set seed. T3 seed (from the T2 plants) was harvested in bulk for each line.

T3 seed was analyzed by Near Infrared Spectroscopy (NIR) at the time of
5 harvest. NIR spectra were captured using a Bruker 22 near infrared spectrometer. Bruker Software was used to estimate total seed oil, total seed protein and total seed fiber content using data from NIR analysis and reference methods according to the manufacturer's instructions. Oil content predicting calibrations were developed following the general method of AOCS Procedure Am1-92, Official Methods and
10 Recommended Practices of the American Oil Chemists Society, 5th Ed., AOCS, Champaign, Ill. A NIR protein content predicting calibration was developed using total nitrogen content data of seed samples following the general method of Dumas Procedure AOAC 968.06 (Official Methods of Analysis of AOAC International 17th Edition AOAC, Gaithersburg, MD). A fiber content predicting calibration was
15 developed by measuring crude fiber content in a set of seed samples. Fiber content of in a known mass of seed was determined using the method of Honig and Rackis, (1979, *J. Agri. Food Chem.*, 27: 1262-1266). Digestible protein content of in a known mass of seed was determined by quantifying the individual amino acids liberated by an acid hydrolysis Steine and Moore (1958, *Anal. Chem.*, 30:1185-
20 1190). The quantification was performed by the Amino Quant (Agilent). The undigested protein remaining associated with the non digestible fraction is measured by the same method described for the whole seed homogenate. Digestible protein content is determined by subtracting the amount of undigested protein associated with the non digestible fraction from the total amount of protein in the seed sample.

25 Seed oil, protein, digestible protein and fiber values in 82,274 lines were determined by NIR spectroscopy and normalized to allow comparison of seed component values in plants grown at different times. Oil, protein and fiber values were normalized by calculating the average oil, protein and fiber values in seed from all plants planted on the same day (including a large number of other ACTTAG
30 plants, including control, wild-type, or non-transgenic plants). The seed components for each line was expressed as a "percent relative value" which was calculated by dividing the component value for each line with the average component value for all

lines planted on the same day (which should approximate the value in control, wild-type, or non-transgenic plants). The “percent relative protein” and “percent relative fiber” were calculated similarly.

Inverse PCR was used to recover genomic DNA flanking the T-DNA insertion. The PCR product was subjected to sequence analysis and placed on the genome using a basic BLASTN search and/or a search of the *Arabidopsis* Information Resource (TAIR) database (available at the publicly available website). Promoters within 9 kb of the enhancers in the ACTTAG element are considered to be within “activation space.” Genes with T-DNA inserts within coding sequences were not considered to be within “activation space.” The ACTTAG lines with the above average oil and protein values, and below average fiber values were identified and are listed in column 3 of Table 1.

Table 1.

1. Gene alias	2. Tair	3. ACTTAG Line	4. Relative Seed Protein Content	5. Relative Seed Fiber Content	6. Relative Seed Oil Content	7. GC FA
IMQ34.3	At2g31480	W000137133	135.45%	89.55%	82.65%	
IMQ34.4	At2g31490	W000137133	135.45%	89.55%	82.65%	
IMQ35.1	At2g34030	W000041983	122.86%	92.93%	80.44%	
IMQ35.2	At2g34040	W000041983	122.86%	92.93%	80.44%	
IMQ35.2	At2g34040	W000041983	122.86%	92.93%	80.44%	
IMQ35.3	At2g34050	W000041983	122.86%	92.93%	80.44%	
IMQ35.4	At2g34060	W000041983	122.86%	92.93%	80.44%	
IMQ36.1	At2g34400	W000146178	109.53%	93.15%	94.98%	
IMQ36.2	At2g34410	W000146178	109.53%	93.15%	94.98%	
IMQ36.2	At2g34410	W000146178	109.53%	93.15%	94.98%	
IMQ36.2	At2g34410	W000146178	109.53%	93.15%	94.98%	
IMQ36.3	At2g34420	W000146178	109.53%	93.15%	94.98%	
IMQ36.3	At2g34420	W000146178	109.53%	93.15%	94.98%	
IMQ36.4	At2g34430	W000146178	109.53%	93.15%	94.98%	
IMQ36.5	At2g34440	W000146178	109.53%	93.15%	94.98%	
IMQ36.6	At2g34450	W000146178	109.53%	93.15%	94.98%	
IMQ36.6	At2g34450	W000146178	109.53%	93.15%	94.98%	
IMQ37.1	At3g03800	W000168536	105.07%	88.55%	98.42%	
IMQ37.2	At3g03810	W000168536	105.07%	88.55%	98.42%	
IMQ37.3	At3g03820	W000168536	105.07%	88.55%	98.42%	
IMQ37.4	At3g03830	W000168536	105.07%	88.55%	98.42%	
IMQ37.5	At3g03840	W000168536	105.07%	88.55%	98.42%	
IMQ37.6	At3g03847	W000168536	105.07%	88.55%	98.42%	97.87%
IMQ37.7	At3g03850	W000168536	105.07%	88.55%	98.42%	
IMQ38.1	At3g07100	W000182714	101.56%	90.13%	99.68%	

1. Gene alias	2. Tair	3. ACTTAG Line	4. Relative Seed Protein Content	5. Relative Seed Fiber Content	6. Relative Seed Oil Content	7. GC FA
IMQ38.2	At3g07110	W000182714	101.56%	90.13%	99.68%	
IMQ38.2	At3g07110	W000182714	101.56%	90.13%	99.68%	
IMQ38.3	At3g07120	W000182714	101.56%	90.13%	99.68%	
IMQ38.4	At3g07130	W000182714	101.56%	90.13%	99.68%	
IMQ38.5	At3g07140	W000182714	101.56%	90.13%	99.68%	
IMQ38.5	At3g07140	W000182714	101.56%	90.13%	99.68%	
IMQ38.6	At3g07150	W000182714	101.56%	90.13%	99.68%	
IMQ39.1	At3g15480	W000190425	151.85%	106.23%	57.69%	
IMQ39.2	At3g15490	W000190425	151.85%	106.23%	57.69%	69.66%
IMQ39.3	At3g15500	W000190425	151.85%	106.23%	57.69%	
IMQ40.1	At3g16580	W000085366	119.68%	89.13%	84.56%	99.63%
IMQ40.2	At3g16590	W000085366	119.68%	89.13%	84.56%	
IMQ40.3	At3g16600	W000085366	119.68%	89.13%	84.56%	
IMQ40.4	At3g16610	W000085366	119.68%	89.13%	84.56%	
IMQ40.5	At3g16620	W000085366	119.68%	89.13%	84.56%	
IMQ41.1	At3g17640	W000192636	101.54%	86.92%	102.79%	107.27%
IMQ41.2	At3g17650	W000192636	101.54%	86.92%	102.79%	
IMQ41.3	At3g17660	W000192636	101.54%	86.92%	102.79%	
IMQ41.4	At3g17670	W000192636	101.54%	86.92%	102.79%	
IMQ41.5	At3g17680	W000192636	101.54%	86.92%	102.79%	107.27%
IMQ41.5	At3g17680	W000192636	101.54%	86.92%	102.79%	
IMQ41.6	At3g17690	W000192636	101.54%	86.92%	102.79%	107.27%
IMQ42.1	At3g19850	W000093675	112.46%	94.21%	95.25%	
IMQ42.2	At3g19860	W000093675	112.46%	94.21%	95.25%	
IMQ42.2	At3g19860	W000093675	112.46%	94.21%	95.25%	

Table 2.

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. SEQ ID NO	5. Polypeptide seq. GI#	6. SEQ ID NO	7. Putative biochemical function/protein name	8. Conserved protein domain
IMQ34.3	At2g31480	gi 42569515	SEQ ID NO: 1	gi 42569516	SEQ ID NO: 2	unknown protein	
IMQ34.4	At2g31490	gi 18402681	SEQ ID NO: 3	gi 18402682	SEQ ID NO: 4	unknown protein	
IMQ35.1	At2g34030	gi 42569600	SEQ ID NO: 5	gi 42569601	SEQ ID NO: 6	calcium ion binding	IPR002048 Calcium-binding EF-hand
IMQ35.2	At2g34040	gi 30685905	SEQ ID NO: 7	gi 30685906	SEQ ID NO: 8	unknown protein	IPR008383 Apoptosis inhibitory 5
IMQ35.2	At2g34040	gi 30685899	SEQ ID NO: 9	gi 18403429	SEQ ID NO: 10	unknown protein	IPR008383 Apoptosis inhibitory 5
IMQ35.3	At2g34050	gi 30685908	SEQ ID NO: 11	gi 18403431	SEQ ID NO: 12	unknown protein	IPR010591 ATP11

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. SEQ ID NO	5. Polypeptide seq. GI#	6. SEQ ID NO	7. Putative biochemical function/protein name	8. Conserved protein domain
IMQ35.4	At2g34060	gi 30685912	SEQ ID NO: 13	gi 15226205	SEQ ID NO: 14	peroxidase	IPR002016 Haem peroxidase, plant/fungal/bacterial; IPR000823 Plant peroxidase
IMQ36.1	At2g34400	gi 18403542	SEQ ID NO: 15	gi 15226301	SEQ ID NO: 16	unknown protein	IPR002885 Pentatricopeptide repeat; IPR008940 Protein prenyltransferase
IMQ36.2	At2g34410	gi 79324292	SEQ ID NO: 17	gi 79324293	SEQ ID NO: 18	O-acetyltransferase	IPR012419 Cas1p-like
IMQ36.2	At2g34410	gi 79324284	SEQ ID NO: 19	gi 79324285	SEQ ID NO: 20	O-acetyltransferase	IPR012419 Cas1p-like
IMQ36.2	At2g34410	gi 42569608	SEQ ID NO: 21	gi 42569609	SEQ ID NO: 22	O-acetyltransferase	IPR012419 Cas1p-like
IMQ36.3	At2g34420	gi 30686050	SEQ ID NO: 23	gi 30686051	SEQ ID NO: 24	LHB1B2	IPR001344 Chlorophyll A-B binding protein
IMQ36.3	At2g34420	gi 30686047	SEQ ID NO: 25	gi 18403546	SEQ ID NO: 26	LHB1B2	IPR001344 Chlorophyll A-B binding protein
IMQ36.4	At2g34430	gi 30686053	SEQ ID NO: 27	gi 18403549	SEQ ID NO: 28	LHB1B1; chlorophyll binding	IPR001344 Chlorophyll A-B binding protein
IMQ36.5	At2g34440	gi 18403551	SEQ ID NO: 29	gi 15226309	SEQ ID NO: 30	transcription factor	IPR002100 Transcription factor, MADS-box
IMQ36.6	At2g34450	gi 79324302	SEQ ID NO: 31	gi 79324303	SEQ ID NO: 32	unknown protein	IPR000910 HMG1/2 (high mobility group) box
IMQ36.6	At2g34450	gi 30686062	SEQ ID NO: 33	gi 18403553	SEQ ID NO: 34	transcription factor	IPR000910 HMG1/2 (high mobility group) box
IMQ37.1	At3g03800	gi 18396673	SEQ ID NO: 35	gi 15228637	SEQ ID NO: 36	SYP131; t-SNARE	IPR000727 Target SNARE coiled-coil region; IPR006011 Syntaxin, N-terminal; IPR006012 Syntaxin/epimorphin family
IMQ37.2	At3g03810	gi 30678945	SEQ ID NO: 37	gi 30678946	SEQ ID NO: 38	unknown protein	IPR004348 Hypothetical plant protein
IMQ37.3	At3g03820	gi 18396679	SEQ ID NO: 39	gi 15228639	SEQ ID NO: 40	unknown protein	IPR003676 Auxin responsive SAUR protein
IMQ37.4	At3g03830	gi 30678954	SEQ ID NO: 41	gi 15228640	SEQ ID NO: 42	unknown protein	IPR003676 Auxin responsive SAUR protein
IMQ37.5	At3g03840	gi 42563470	SEQ ID NO: 43	gi 15228641	SEQ ID NO: 44	unknown protein	IPR003676 Auxin responsive SAUR protein

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. SEQ ID NO	5. Polypeptide seq. GI#	6. SEQ ID NO	7. Putative biochemical function/protein name	8. Conserved protein domain
IMQ37.6	At3g03847	gi 22330828	SEQ ID NO: 45	gi 22330829	SEQ ID NO: 46	unknown protein	IPR003676 Auxin responsive SAUR protein
IMQ37.7	At3g03850	gi 30678958	SEQ ID NO: 47	gi 30678959	SEQ ID NO: 48	unknown protein	
IMQ38.1	At3g07100	gi 30680128	SEQ ID NO: 49	gi 30680129	SEQ ID NO: 50	protein binding / transporter	IPR006896 Sec23/Sec24 trunk region; IPR006900 Sec23/Sec24 helical region; IPR006895 Zinc finger, Sec23/Sec24-type; IPR006706 Extensin-like region; IPR007123 Gelsolin region
IMQ38.2	At3g07110	gi 79313148	SEQ ID NO: 51	gi 79313149	SEQ ID NO: 52	structural constituent of ribosome	IPR005822 Ribosomal protein L13; IPR005755 Ribosomal protein L13, archaea and eukaryotic form; IPR005823 Ribosomal protein L13, bacterial and organelle form
IMQ38.2	At3g07110	gi 30680131	SEQ ID NO: 53	gi 15231394	SEQ ID NO: 54	structural constituent of ribosome	IPR005822 Ribosomal protein L13; IPR005755 Ribosomal protein L13, archaea and eukaryotic form; IPR005823 Ribosomal protein L13, bacterial and organelle form
IMQ38.3	At3g07120	gi 30680134	SEQ ID NO: 55	gi 15231396	SEQ ID NO: 56	protein binding / ubiquitin-protein ligase/ zinc ion binding	IPR001841 Zinc finger, RING-type
IMQ38.4	At3g07130	gi 42563571	SEQ ID NO: 57	gi 15231398	SEQ ID NO: 58	hydrolase/ protein serine/threonine phosphatase	IPR004843 Metallophosphoesterase
IMQ38.5	At3g07140	gi 42570456	SEQ ID NO: 59	gi 30680143	SEQ ID NO: 60	GPI-anchor transamidase	IPR007245 Gpi16 subunit, GPI transamidase component
IMQ38.5	At3g07140	gi 30680141	SEQ ID NO: 61	gi 18397852	SEQ ID NO: 62	GPI-anchor transamidase	IPR007245 Gpi16 subunit, GPI transamidase component

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. SEQ ID NO	5. Polypeptide seq. GI#	6. SEQ ID NO	7. Putative biochemical function/protein name	8. Conserved protein domain
IMQ38.6	At3g07150	gi 18397855	SEQ ID NO: 63	gi 15231401	SEQ ID NO: 64	unknown protein	
IMQ39.1	At3g15480	gi 30683621	SEQ ID NO: 65	gi 18400781	SEQ ID NO: 66	unknown protein	IPR009606 Protein of unknown function DUF1218
IMQ39.2	At3g15490	gi 42564187	SEQ ID NO: 67	gi 42564188	SEQ ID NO: 68	unknown protein	IPR005061 Protein of unknown function DUF292, eukaryotic
IMQ39.3	At3g15500	gi 30683631	SEQ ID NO: 69	gi 15232604	SEQ ID NO: 70	ATNAC3; transcription factor	IPR003441 No apical meristem (NAM) protein
IMQ40.1	At3g16580	gi 30684147	SEQ ID NO: 71	gi 18401179	SEQ ID NO: 72	unknown protein	IPR001810 Cyclin-like F-box; IPR006527 F-box protein interaction domain
IMQ40.2	At3g16590	gi 18401181	SEQ ID NO: 73	gi 15228255	SEQ ID NO: 74	unknown protein	IPR006527 F-box protein interaction domain; IPR001810 Cyclin-like F-box
IMQ40.3	At3g16600	gi 18401188	SEQ ID NO: 75	gi 15228256	SEQ ID NO: 76	ATP binding / ATP-dependent helicase/ DNA binding / helicase/ nucleic acid binding / protein binding / ubiquitin-protein ligase/ zinc ion binding	IPR000330 SNF2-related; IPR001410 DEAD/DEAH box helicase; IPR001650 Helicase, C-terminal
IMQ40.4	At3g16610	gi 18401191	SEQ ID NO: 77	gi 15228257	SEQ ID NO: 78	unknown protein	IPR001841 Zinc finger, RING-type; IPR002885 Pentatricopeptide repeat
IMQ40.5	At3g16620	gi 30684161	SEQ ID NO: 79	gi 15228272	SEQ ID NO: 80	ATTOC120; protein translocase	IPR006703 AIG1; IPR002917 GTP-binding protein, HSR1-related; IPR005688 Chloroplast protein import component Toc34; IPR005690 Chloroplast protein import component Toc86/159
IMQ41.1	At3g17640	gi 18401587	SEQ ID NO: 81	gi 15229088	SEQ ID NO: 82	protein binding	IPR001611 Leucine-rich repeat; IPR007090 Leucine-rich repeat, plant specific

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. SEQ ID NO	5. Polypeptide seq. GI#	6. SEQ ID NO	7. Putative biochemical function/protein name	8. Conserved protein domain
IMQ41.2	At3g17650	gi 18401589	SEQ ID NO: 83	gi 18401590	SEQ ID NO: 84	PDE321; oligopeptide transporter	IPR004813 Oligopeptide transporter OPT superfamily; IPR004814 Oligopeptide transporter OPT; IPR004648 Tetrapeptide transporter, OPT1/isp4
IMQ41.3	At3g17660	gi 18401594	SEQ ID NO: 85	gi 15229090	SEQ ID NO: 86	DNA binding	IPR001164 Arf GTPase activating protein
IMQ41.4	At3g17670	gi 18401598	SEQ ID NO: 87	gi 15229091	SEQ ID NO: 88	unknown protein	IPR001440 TPR repeat
IMQ41.5	At3g17680	gi 79313276	SEQ ID NO: 89	gi 79313277	SEQ ID NO: 90	unknown protein	IPR011684 KIP1-like
IMQ41.5	At3g17680	gi 30684630	SEQ ID NO: 91	gi 30684631	SEQ ID NO: 92	unknown protein	IPR011684 KIP1-like
IMQ41.6	At3g17690	gi 30684635	SEQ ID NO: 93	gi 15229093	SEQ ID NO: 94	ATCNGC19; calmodulin binding / cyclic nucleotide binding / ion channel	IPR005821 Ion transport protein; IPR000595 Cyclic nucleotide-binding
IMQ42.1	At3g19850	gi 42565033	SEQ ID NO: 95	gi 42565034	SEQ ID NO: 96	protein binding / signal transducer	IPR004249 NPH3; IPR000210 BTB/POZ
IMQ42.2	At3g19860	gi 79313298	SEQ ID NO: 97	gi 79313299	SEQ ID NO: 98	DNA binding	IPR001092 Basic helix-loop-helix dimerisation region bHLH; IPR003106 Leucine zipper, homeobox-associated
IMQ42.2	At3g19860	gi 30685522	SEQ ID NO: 99	gi 15230975	SEQ ID NO: 100	DNA binding / transcription factor	IPR001092 Basic helix-loop-helix dimerisation region bHLH; IPR003106 Leucine zipper, homeobox-associated

Table 3.

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. Polypeptide seq. GI#	5. Orthologous Genes: Nucleic Acid/Polypeptide seq. GI#		
				Nucleic Acid GI#	Polypeptide GI#	Species
IMQ34.3	At2g31480	gi 42569515	gi 42569516			
IMQ34.4	At2g31490	gi 18402681	gi 18402682	gi 15146360	gi 57899156	Oryza sativa (japonica cultivar-group)
				gi 34910867	gi 34910868	Oryza sativa (japonica cultivar-group)
				gi 34909561	gi 34909562	Oryza sativa (japonica cultivar-group)
IMQ35.1	At2g34030	gi 42569600	gi 42569601	gi 30690570	gi 15218787	Arabidopsis thaliana
				gi 18403422	gi 15226191	Arabidopsis thaliana
				gi 30695447	gi 30695448	Arabidopsis thaliana
IMQ35.2	At2g34040	gi 30685905	gi 30685906	gi 30685899	gi 18403429	Arabidopsis thaliana

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. Polypeptide seq. GI#	5. Orthologous Genes: Nucleic Acid/Polypeptide seq. GI#		
				Nucleic Acid GI#	Polypeptide GI#	Species
				gi 42562386	gi 42562387	Arabidopsis thaliana
				gi 50907460	gi 50907461	Oryza sativa (japonica cultivar-group)
				gi 66526466	gi 66526467	Apis mellifera
IMQ35.2	At2g34040	gi 30685899	gi 18403429	gi 30685905	gi 30685906	Arabidopsis thaliana
				gi 42562386	gi 42562387	Arabidopsis thaliana
				gi 50907460	gi 50907461	Oryza sativa (japonica cultivar-group)
				gi 66526466	gi 66526467	Apis mellifera
IMQ35.3	At2g34050	gi 30685908	gi 18403431	gi 50907446	gi 50907447	Oryza sativa (japonica cultivar-group)
				gi 66814293	gi 66814294	Dictyostelium discoideum
				gi 50751555	gi 50751556	Gallus gallus
IMQ35.4	At2g34060	gi 30685912	gi 15226205	gi 50918940	gi 50918941	Oryza sativa (japonica cultivar-group)
				gi 55701112	gi 55701113	Oryza sativa (japonica cultivar-group)
				gi 537318	gi 537319	Medicago sativa
IMQ36.1	At2g34400	gi 18403542	gi 15226301	gi 18398256	gi 15231970	Arabidopsis thaliana
				gi 18399739	gi 15230593	Arabidopsis thaliana
				gi 18420159	gi 15233645	Arabidopsis thaliana
IMQ36.2	At2g34410	gi 79324292	gi 79324293	gi 79324284	gi 79324285	Arabidopsis thaliana
				gi 42569608	gi 42569609	Arabidopsis thaliana
				gi 30694937	gi 18422663	Arabidopsis thaliana
				gi 42562401	gi 42562402	Arabidopsis thaliana
				gi 20161482	gi 55297065	Oryza sativa (japonica cultivar-group)
IMQ36.2	At2g34410	gi 79324284	gi 79324285	gi 79324292	gi 79324293	Arabidopsis thaliana
				gi 42569608	gi 42569609	Arabidopsis thaliana
				gi 30694937	gi 18422663	Arabidopsis thaliana
				gi 42562401	gi 42562402	Arabidopsis thaliana
				gi 20161482	gi 55297065	Oryza sativa (japonica cultivar-group)
IMQ36.2	At2g34410	gi 42569608	gi 42569609	gi 79324292	gi 79324293	Arabidopsis thaliana
				gi 79324284	gi 79324285	Arabidopsis thaliana
				gi 30694937	gi 18422663	Arabidopsis thaliana
				gi 42562401	gi 42562402	Arabidopsis thaliana
				gi 20161482	gi 55297065	Oryza sativa (japonica cultivar-group)
IMQ36.3	At2g34420	gi 30686050	gi 30686051	gi 30686047	gi 18403546	Arabidopsis thaliana
				gi 30690951	gi 15220615	Arabidopsis thaliana
				gi 30690948	gi 18397288	Arabidopsis thaliana
				gi 30690947	gi 18397286	Arabidopsis thaliana
IMQ36.3	At2g34420	gi 30686047	gi 18403546	gi 30690951	gi 15220615	Arabidopsis thaliana
				gi 30690948	gi 18397288	Arabidopsis thaliana
				gi 30690947	gi 18397286	Arabidopsis thaliana
				gi 31323255	gi 31323256	Brassica oleracea
IMQ36.4	At2g34430	gi 30686053	gi 18403549	gi 21137	gi 21138	Sinapis alba
				gi 18266038	gi 18266039	Brassica oleracea
				gi 30690951	gi 15220615	Arabidopsis thaliana
IMQ36.5	At2g34440	gi 18403551	gi 15226309	gi 18397662	gi 15230767	Arabidopsis thaliana
				gi 30682570	gi 30682571	Arabidopsis thaliana
				gi 18424355	gi 15239333	Arabidopsis thaliana
IMQ36.6	At2g34450	gi 79324302	gi 79324303	gi 30686062	gi 18403553	Arabidopsis thaliana
				gi 20161570	gi 56202161	Oryza sativa (japonica cultivar-group)
				gi 63020535	gi 63020536	Cucumis sativus
				gi 34910913	gi 34910914	Oryza sativa (japonica cultivar-group)
IMQ36.6	At2g34450	gi 30686062	gi 18403553	gi 20161570	gi 56202161	Oryza sativa (japonica cultivar-group)
				gi 63020535	gi 63020536	Cucumis sativus
				gi 42562198	gi 18394900	Arabidopsis thaliana
IMQ37.1	At3g03800	gi 18396673	gi 15228637	gi 18415700	gi 18415701	Arabidopsis thaliana
				gi 51963279	gi 51963280	Oryza sativa (japonica cultivar-group)

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. Polypeptide seq. GI#	5. Orthologous Genes: Nucleic Acid/Polypeptide seq. GI#		
				Nucleic Acid GI#	Polypeptide GI#	Species
				gi 50934370	gi 50934371	Oryza sativa (japonica cultivar-group)
				gi 55741415	gi 55741416	Oryza sativa (japonica cultivar-group)
IMQ37.2	At3g03810	gi 30678945	gi 30678946	gi 46063406	gi 50878393	Oryza sativa (japonica cultivar-group)
				gi 30689847	gi 30689848	Arabidopsis thaliana
				gi 30679041	gi 22329335	Arabidopsis thaliana
IMQ37.3	At3g03820	gi 18396679	gi 15228639	gi 42563470	gi 15228641	Arabidopsis thaliana
				gi 30678958	gi 30678959	Arabidopsis thaliana
				gi 30678954	gi 15228640	Arabidopsis thaliana
IMQ37.4	At3g03830	gi 30678954	gi 15228640	gi 30678958	gi 30678959	Arabidopsis thaliana
				gi 18396679	gi 15228639	Arabidopsis thaliana
				gi 42563470	gi 15228641	Arabidopsis thaliana
IMQ37.5	At3g03840	gi 42563470	gi 15228641	gi 18396679	gi 15228639	Arabidopsis thaliana
				gi 30678958	gi 30678959	Arabidopsis thaliana
				gi 30678954	gi 15228640	Arabidopsis thaliana
IMQ37.6	At3g03847	gi 22330828	gi 22330829	gi 42567926	gi 15238721	Arabidopsis thaliana
				gi 42567924	gi 15238716	Arabidopsis thaliana
				gi 30686621	gi 15238715	Arabidopsis thaliana
IMQ37.7	At3g03850	gi 30678958	gi 30678959	gi 30678954	gi 15228640	Arabidopsis thaliana
				gi 42563470	gi 15228641	Arabidopsis thaliana
				gi 18396679	gi 15228639	Arabidopsis thaliana
IMQ38.1	At3g07100	gi 30680128	gi 30680129	gi 50921574	gi 50921575	Oryza sativa (japonica cultivar-group)
				gi 74199102	gi 74199103	Mus musculus
				gi 31340743	gi 30424898	Mus musculus
IMQ38.2	At3g07110	gi 79313148	gi 79313149	gi 30680131	gi 15231394	Arabidopsis thaliana
				gi 30695535	gi 15239698	Arabidopsis thaliana
				gi 30687674	gi 15230197	Arabidopsis thaliana
				gi 42566745	gi 15235617	Arabidopsis thaliana
IMQ38.2	At3g07110	gi 30680131	gi 15231394	gi 30695535	gi 15239698	Arabidopsis thaliana
				gi 30687674	gi 15230197	Arabidopsis thaliana
				gi 42566745	gi 15235617	Arabidopsis thaliana
IMQ38.3	At3g07120	gi 30680134	gi 15231396	gi 42565172	gi 15230243	Arabidopsis thaliana
				gi 42570141	gi 42570142	Arabidopsis thaliana
				gi 18413925	gi 15235589	Arabidopsis thaliana
IMQ38.4	At3g07130	gi 42563571	gi 15231398	gi 13925770	gi 13925771	Glycine max
				gi 62177682	gi 62177683	Medicago truncatula
				gi 50920094	gi 50920095	Oryza sativa (japonica cultivar-group)
IMQ38.5	At3g07140	gi 42570456	gi 30680143	gi 30680141	gi 18397852	Arabidopsis thaliana
				gi 77548247	gi 77550830	Oryza sativa (japonica cultivar-group)
				gi 73992512	gi 73992513	Canis familiaris
IMQ38.5	At3g07140	gi 30680141	gi 18397852	gi 42570456	gi 30680143	Arabidopsis thaliana
				gi 77548247	gi 77550830	Oryza sativa (japonica cultivar-group)
				gi 73992512	gi 73992513	Canis familiaris
IMQ38.6	At3g07150	gi 18397855	gi 15231401	gi 19386744	gi 57900451	Oryza sativa (japonica cultivar-group)
				gi 34907011	gi 34907012	Oryza sativa (japonica cultivar-group)
IMQ39.1	At3g15480	gi 30683621	gi 18400781	gi 42562716	gi 18404242	Arabidopsis thaliana
				gi 38016520	gi 38016521	Gossypium barbadense
				gi 50936920	gi 50936921	Oryza sativa (japonica cultivar-group)
IMQ39.2	At3g15490	gi 42564187	gi 42564188	gi 18397647	gi 15226014	Arabidopsis thaliana
				gi 50918206	gi 50918207	Oryza sativa (japonica cultivar-group)
				gi 30683233	gi 15222251	Arabidopsis thaliana
IMQ39.3	At3g15500	gi 30683631	gi 15232604	gi 42562715	gi 15219112	Arabidopsis thaliana
				gi 66394515	gi 66394516	Glycine max

1. Gene alias	2. Tair	3. Nucleic Acid seq. GI#	4. Polypeptide seq. GI#	5. Orthologous Genes: Nucleic Acid/Polypeptide seq. GI#		
				Nucleic Acid GI#	Polypeptide GI#	Species
				gi 6175245	gi 6175246	Lycopersicon esculentum
IMQ40.1	At3g16580	gi 30684147	gi 18401179	gi 42569040	gi 15225992	Arabidopsis thaliana
				gi 18398432	gi 15223198	Arabidopsis thaliana
				gi 18401705	gi 18401706	Arabidopsis thaliana
IMQ40.2	At3g16590	gi 18401181	gi 15228255	gi 18391359	gi 15221240	Arabidopsis thaliana
				gi 30684644	gi 18401609	Arabidopsis thaliana
				gi 18391361	gi 15221246	Arabidopsis thaliana
IMQ40.3	At3g16600	gi 18401188	gi 15228256	gi 18403060	gi 18403061	Arabidopsis thaliana
				gi 18402451	gi 15231009	Arabidopsis thaliana
				gi 51964729	gi 51964730	Oryza sativa (japonica cultivar-group)
IMQ40.4	At3g16610	gi 18401191	gi 15228257	gi 50941690	gi 50941691	Oryza sativa (japonica cultivar-group)
				gi 18483227	gi 18483237	Sorghum bicolor
				gi 34906109	gi 34906110	Oryza sativa (japonica cultivar-group)
				gi 28564706	gi 57899529	Oryza sativa (japonica cultivar-group)
IMQ40.5	At3g16620	gi 30684161	gi 15228272	gi 30679717	gi 15227268	Arabidopsis thaliana
				gi 37536637	gi 37536638	Oryza sativa (japonica cultivar-group)
				gi 44662984	gi 44662985	Physcomitrella patens
IMQ41.1	At3g17640	gi 18401587	gi 15229088	gi 50931366	gi 50931367	Oryza sativa (japonica cultivar-group)
				gi 51339056	gi 55733924	Oryza sativa (japonica cultivar-group)
				gi 30678565	gi 30678566	Arabidopsis thaliana
IMQ41.2	At3g17650	gi 18401589	gi 18401590	gi 30694308	gi 18402162	Arabidopsis thaliana
				gi 42562981	gi 15218799	Arabidopsis thaliana
				gi 74267415	gi 74267416	Oryza sativa (japonica cultivar-group)
IMQ41.3	At3g17660	gi 18401594	gi 15229090	gi 30696440	gi 18423615	Arabidopsis thaliana
				gi 50510177	gi 50510178	Oryza sativa (japonica cultivar-group)
				gi 60596222	gi 76152990	Schistosoma japonicum
IMQ41.4	At3g17670	gi 18401598	gi 15229091	gi 53749471	gi 53749491	Solanum demissum
				gi 50929586	gi 50929587	Oryza sativa (japonica cultivar-group)
				gi 50928358	gi 50928359	Oryza sativa (japonica cultivar-group)
IMQ41.5	At3g17680	gi 79313276	gi 79313277	gi 30684630	gi 30684631	Arabidopsis thaliana
				gi 22330106	gi 22330107	Arabidopsis thaliana
				gi 53749471	gi 53749473	Solanum demissum
				gi 50916296	gi 50916297	Oryza sativa (japonica cultivar-group)
IMQ41.5	At3g17680	gi 30684630	gi 30684631	gi 22330106	gi 22330107	Arabidopsis thaliana
				gi 53749471	gi 53749473	Solanum demissum
				gi 50916296	gi 50916297	Oryza sativa (japonica cultivar-group)
IMQ41.6	At3g17690	gi 30684635	gi 15229093	gi 42564623	gi 18401606	Arabidopsis thaliana
				gi 50913256	gi 50913257	Oryza sativa (japonica cultivar-group)
				gi 51091189	gi 51091194	Oryza sativa (japonica cultivar-group)
IMQ42.1	At3g19850	gi 42565033	gi 42565034	gi 22330149	gi 22330150	Arabidopsis thaliana
				gi 50929590	gi 50929591	Oryza sativa (japonica cultivar-group)
				gi 50938220	gi 50938221	Oryza sativa (japonica cultivar-group)
IMQ42.2	At3g19860	gi 79313298	gi 79313299	gi 30685522	gi 15230975	Arabidopsis thaliana
				gi 51536177	gi 51536178	Oryza sativa (japonica cultivar-group)
				gi 30690567	gi 30690568	Arabidopsis thaliana
				gi 22329203	gi 22329204	Arabidopsis thaliana
IMQ42.2	At3g19860	gi 30685522	gi 15230975	gi 51536177	gi 51536178	Oryza sativa (japonica cultivar-group)
				gi 22329203	gi 22329204	Arabidopsis thaliana
				gi 30690567	gi 30690568	Arabidopsis thaliana

EXAMPLE 2

Analysis of the *Arabidopsis* IMQ Sequence

Sequence analyses were performed with BLAST (Altschul *et al.*, 1990, *J. Mol. Biol.* 215:403-410), PFAM (Bateman *et al.*, 1999, *Nucleic Acids Res.* 27:260-
5 262), INTERPRO (Mulder *et al.* 2003 *Nucleic Acids Res.* 31, 315-318.), PSORT (Nakai K, and Horton P, 1999, *Trends Biochem. Sci.* 24:34-6), and/or CLUSTAL (Thompson JD *et al.*, 1994, *Nucleic Acids Res.* 22:4673-4680). Conserved domains for each protein are listed in column 8 of Table 2.

10 EXAMPLE 3

To test whether over-expression of the genes in Tables 1 and 2 alter the seed composition phenotype, protein, digestible protein, oil and fiber content in seeds from transgenic plants expressing these genes was compared with protein, digestible protein, oil and fiber content in seeds from non-transgenic control plants. To do this,
15 the genes were cloned into plant transformation vectors behind the strong constitutive CsVMV promoter and the seed specific PRU promoter. These constructs were transformed into *Arabidopsis* plants using the floral dip method. The plant transformation vector contains a gene, which provides resistance to a toxic compound, and serves as a selectable marker. Seed from the transformed plants
20 were plated on agar medium containing the toxic compound. After 7 days, transgenic plants were identified as healthy green plants and transplanted to soil. Non-transgenic control plants were germinated on agar medium, allowed to grow for 7 days and then transplanted to soil. Transgenic seedlings and non-transgenic control plants were transplanted to two inch pots that were placed in random
25 positions in a 10 inch by 20 inch tray. The plants were grown to maturity, allowed to self-fertilize and set seed. Seed was harvested from each plant and its oil content estimated by Near Infrared (NIR) Spectroscopy using methods previously described. The effect of each construct on seed composition was examined in at least two experiments.

30 Table 4 lists constructs tested for causing a significant increase in oil, protein, digestible protein or a significant decrease in fiber were identified by a two-way Analysis of Variance (ANOVA) test at a p-value ≤ 0.05 . The ANOVA p-values

for Protein, Oil, Digestible Protein and Fiber are listed in columns 4-7, respectively. Those with a significant p-value are listed in bold. The Average values for Protein, Oil, Digestible Protein and Fiber are listed in columns 8-11, respectively and were calculated by averaging the average values determined for the transgenic plants in each experiment.

Table 4.

1. Gene	2. TAIR	3. Construct	4. ANOVA Protein	5. ANOVA Oil	6. ANOVA Digestible Protein	7. ANOVA Fiber	8. Protein	9. Oil	10. Digestible Protein	11. Fiber
IMQ37.5	At3g03840	CsVMV::At3g03840	0.433	0.415	0.033	0.197	101.7%	98.7%	102.4%	98.7%
IMQ37.6	At3g03847	CsVMV::At3g03847	0.998	0.503	0.634	0.864	100.2%	99.4%	100.5%	99.8%
IMQ37.6	At3g03847	Pru::At3g03847	0.007	0.002	0.115	0.547	104.6%	94.4%	101.2%	100.6%
IMQ39.1	At3g15480	Pru::At3g15480	0.521	0.450	0.026	0.011	100.9%	100.4%	101.4%	97.6%
IMQ39.2	At3g15490	CsVMV::At3g15490	0.192	0.463	0.176	0.035	103.1%	99.1%	101.7%	97.8%
IMQ39.2	At3g15490	Pru::At3g15490	0.125	0.551	<0.0001	<0.0001	100.9%	99.4%	101.4%	97.8%
IMQ40.1	At3g16580	CsVMV::At3g16580	0.436	0.321	0.050	0.173	101.3%	99.0%	101.6%	98.9%
IMQ40.1	At3g16580	Pru::At3g16580	0.269	0.972	0.005	0.003	101.3%	99.7%	101.7%	97.4%
IMQ40.3	At3g16600	Pru::At3g16600	0.019	0.019	0.220	0.136	102.8%	97.0%	99.5%	101.5%
IMQ41.1	At3g17640	CsVMV::At3g17640	0.991	0.671	0.864	0.725	99.9%	99.5%	99.8%	99.8%
IMQ41.1	At3g17640	Pru::At3g17640	0.024	0.006	0.041	0.003	97.1%	105.0%	101.3%	96.8%
IMQ41.5	At3g17680	CsVMV::At3g17680	0.567	0.873	0.803	0.290	101.3%	100.2%	100.3%	98.9%
IMQ41.5	At3g17680	Pru::At3g17680	0.569	0.252	0.009	0.004	99.3%	101.8%	102.1%	96.7%
IMQ41.6	At3g17690	CsVMV::At3g17690	0.388	0.856	0.060	0.039	102.7%	99.8%	102.4%	97.3%
IMQ41.6	At3g17690	Pru::At3g17690	0.263	0.726	0.003	0.009	102.0%	99.2%	103.0%	96.5%

EXAMPLE 4

To test whether over-expression of the genes identified in Tables 1-4 alter the seed composition phenotype, protein, digestible protein, oil, and fiber content in seeds from transgenic plants expressing these genes is compared with protein, digestible protein, oil and fiber content in seeds from non-transgenic control plants. Any one of the genes identified in Tables 1-4 is used to transform *Brassica napus* (canola). To do this, the genes are cloned into plant transformation vectors behind the strong constitutive CsVMV promoter and the seed specific phaseolin promoter. These constructs (which include a gene encoding a selection agent) are transformed into canola plants.

Transformation of canola is accomplished via *Agrobacterium*-mediated transformation. Seeds are surface-sterilized with 10% commercial bleach for 10 minutes and rinsed 3 times with sterile distilled water. The seeds are then placed on

one half concentration of MS basal medium (Murashige and Skoog, *Physiol. Plant.* 15:473-497, 1962) and maintained under growth regime set at 25° C, and a photoperiod of 16 hrs light/8 hrs dark.

Hypocotyl segments (3-5 mm) are excised from 5 - 7 day old seedlings and placed on callus induction medium K1D1 (MS medium with 1 mg/l kinetin and 1 mg/l 2,4-D) for 3 days as pre-treatment. The segments are then transferred into a petri plate, treated with *Agrobacterium* Z707S or LBA4404 strain containing pDAB721. The *Agrobacterium* is grown overnight at 28° C in the dark on a shaker at 150 rpm and subsequently re-suspended in the culture medium.

After 30 minute treatment of the hypocotyl segments with *Agrobacterium*, these are placed back on the callus induction medium for 3 days. Following co-cultivation, the segments are placed on K1D1TC (callus induction medium containing 250 mg/l Carbenicillin and 300 mg/l Timentin) for one week of recovery. Alternately, the segments are placed directly on selection medium K1D1H1 (above medium with 1 mg/l selection agent, for example an herbicide). Carbenicillin and Timentin are antibiotics used to kill the *Agrobacterium*. The selection agent is used to allow the growth of the transformed cells.

Callus samples from independent events are tested by PCR. All the samples tested are positive for the presence of the transformed gene, whereas the non-transformed controls are negative. Callus samples are confirmed to express the appropriate protein as determined by ELISA.

Callused hypocotyl segments are then placed on B3Z1H1 (MS medium, 3 mg/l benzylamino purine, 1 mg/l Zeatin, 0.5 gm/l MES [2-(N-morpholino) ethane sulfonic acid], 5 mg/l silver nitrate, 1 mg/l selection agent, Carbenicillin and Timentin) shoot regeneration medium. After shoots start to regenerate (approximately 3 weeks), hypocotyl segments along with the shoots are transferred to B3Z1H3 medium (MS medium, 3 mg/l benzylamino purine, 1 mg/l Zeatin, 0.5 gm/l MES [2-(N-morpholino) ethane sulfonic acid], 5 mg/l silver nitrate, 3 mg/l selection agent, Carbenicillin and Timentin) for 3 weeks.

Shoots are excised from the hypocotyl segments and transferred to shoot elongation medium MESH10 (MS, 0.5 gm/l MES, 10 mg/l selection agent, Carbenicillin, Timentin) for 2-4 weeks. The elongated shoots are cultured for root

induction on MSI.1 (MS with 0.1 mg/l Indolebutyric acid). Once the plants have a well established root system, these are transplanted into soil. The plants are acclimated under controlled environmental conditions in the Conviron for 1-2 weeks before transfer to the greenhouse. The transformed T0 plants self-pollinate in the greenhouse to obtain T1 seed. Transgenic plants are selected at the T1 generation based on resistance to a selection agent. T2 seed (from T1 plants) is harvested and sown in soil. T2 plants are grown to maturity, allowed to self-fertilize and set seed. T3 seed (from the T2 plants) is harvested in bulk for each line. Seed oil, protein, digestible protein, and fiber values are measured as discussed in Example 1.

IT IS CLAIMED:

1. A method of producing an improved meal quality phenotype in a plant, said method comprising:

(a) introducing into progenitor cells of the plant a plant transformation vector comprising a nucleotide sequence that encodes an improved meal quality (IMQ) polypeptide comprising the amino acid sequence of SEQ ID NO: 82, or an IMQ polypeptide comprising an amino acid sequence at least 80% identical to the full length of SEQ ID NO: 82; and

(b) growing the transformed progenitor cells to produce a transgenic plant, wherein the nucleotide sequence is expressed; and

(c) measuring the improved meal quality phenotype in the transgenic plant, wherein the transgenic plant exhibits an improved meal quality phenotype relative to control plants lacking the plant transformation vector, thereby producing the improved meal quality phenotype in the plant.

2. The method of claim 1, wherein the IMQ polypeptide comprises the amino acid sequence of SEQ ID NO: 82.

3. A transgenic plant cell of a plant obtained by the method of claim 1 or 2, wherein the cell is of a *Brassica* species.

4. A transgenic plant cell of a plant obtained by the method of claim 1 or 2, wherein the cell is a canola, rapeseed, soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat, or rice cell.

5. The transgenic plant cell of claim 4, wherein the plant cell is a canola cell.

6. The transgenic plant cell of claim 4, wherein the plant cell is a rapeseed cell.

7. A transgenic plant cell of a *Brassica* species which is of a direct or indirect progeny of a plant obtained by the method of claim 1 or claim 2, wherein the cells of the direct or indirect progeny of the plant comprise the plant transformation vector.
8. A transgenic plant cell which is a canola, rapeseed, soy, corn, sunflower, cotton, cocoa, safflower, oil palm, coconut palm, flax, castor, peanut, wheat, oat, or rice cell and which is of a direct or indirect progeny of a plant obtained by the method of claim 1 or claim 2, wherein the cells of the direct or indirect progeny of the plant comprise the plant transformation vector.
9. The transgenic plant cell of claim 8, wherein the transgenic plant cell is a canola cell.
10. The transgenic plant cell of claim 8, wherein the transgenic plant cell is a rapeseed cell.
11. A method of generating a plant having an improved meal quality phenotype, said method comprising:
 - (i) identifying a plant as comprising an allele of a gene, wherein the allele encodes a polypeptide comprising an amino acid sequence at least 80% identical to the full length of SEQ ID NO: 82, wherein the allele is overexpressed relative to a control plant, and wherein the allele results in improved meal quality phenotype, compared to plants lacking the allele; and
 - (ii) generating progeny of said identified plant, wherein the generated progeny inherit the allele; and
 - (iii) measuring the improved meal quality phenotype in the progeny, wherein the progeny have the improved meal quality phenotype relative to control plants lacking the allele.
12. The method of claim 11 that employs candidate gene/QTL methodology.
13. The method of claim 11 that employs TILLING methodology.

14. A feed, meal, or food comprising a plant transformation vector comprising the nucleic acid sequence of SEQ ID NO: 81, or a nucleic acid sequence at least 80% identical to the full length of SEQ ID NO: 81, wherein the feed, meal, or food has an improved meal quality phenotype relative to a control feed, meal, or food that does not comprise the plant transformation vector.

15. The feed, meal, or food of claim 14, wherein the plant transformation vector comprises the nucleic acid sequence of SEQ ID NO: 81.

16. A feed, meal, or food comprising a plant transformation vector comprising a nucleotide sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO: 82, or a polypeptide comprising an amino acid sequence at least 80% identical to the full length of SEQ ID NO: 82, wherein the feed, meal, or food has an improved meal quality phenotype relative to a control feed, meal, or food that does not comprise the plant transformation vector.

17. The feed, meal, or food of claim 16, wherein the plant transformation vector comprises a nucleotide sequence encoding a polypeptide comprising the amino acid sequence of SEQ ID NO: 82.