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(54) **METHOD FOR IMPROVING PLANT GROWTH WITH A TRNA SYNTHETASE GENE THAT ACTIVATES TOR**

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C12N 15/82 (2006.01)

(52) **U.S. Cl.**
CPC ***C12N 9/93*** (2013.01); ***C12N 15/8216*** (2013.01); ***C12N 15/8262*** (2013.01); *C12Y 601/01022* (2013.01)

(57) **ABSTRACT**

The present invention provides compositions and methods for increasing the growth, growth rate, and/or yield of a plant that lacks root nodules by engineering it to overexpress the protein asparaginyl-tRNA synthetase 1 (NARS1). Specifically, the present invention provides plants that are engineered to overexpress NARS1, seeds produced by said plants, and methods of generating and growing said plants.

Specification includes a Sequence Listing.

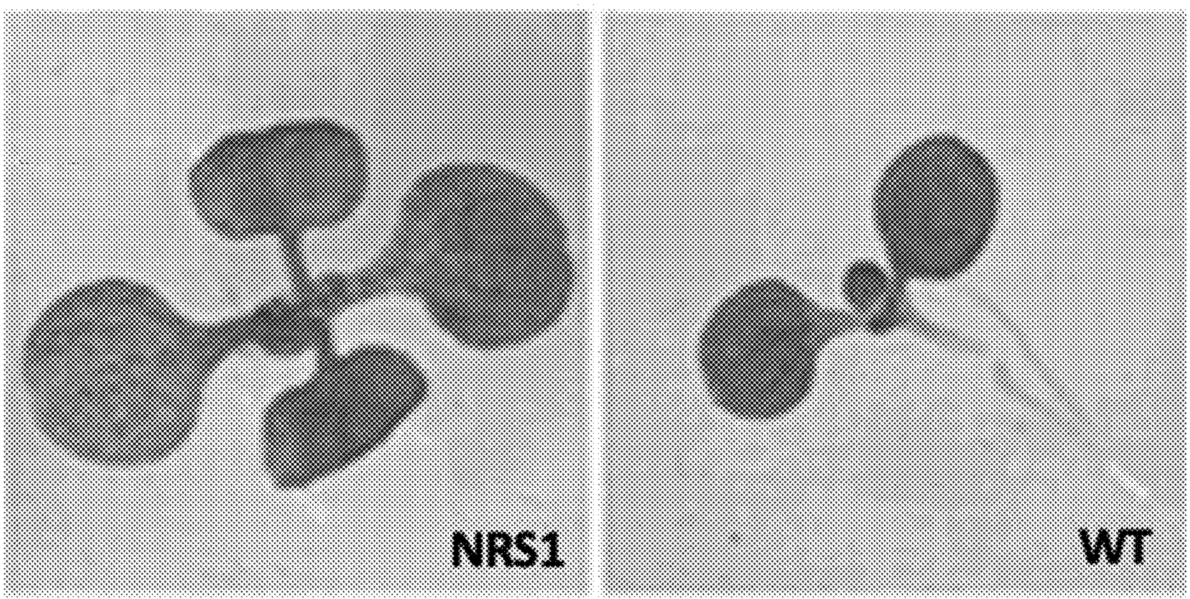

FIG. 1A

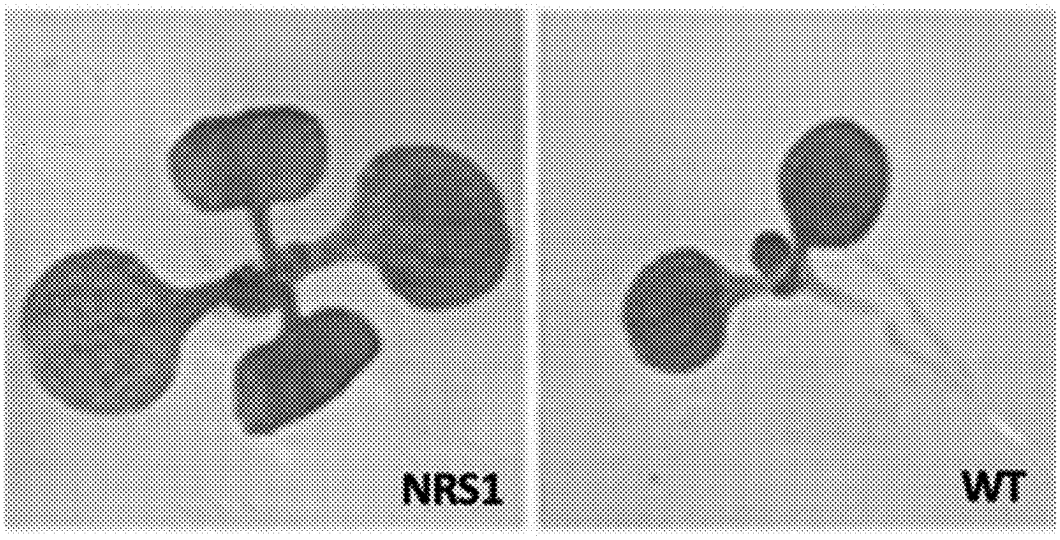
NRS1
WT

FIG. 1B

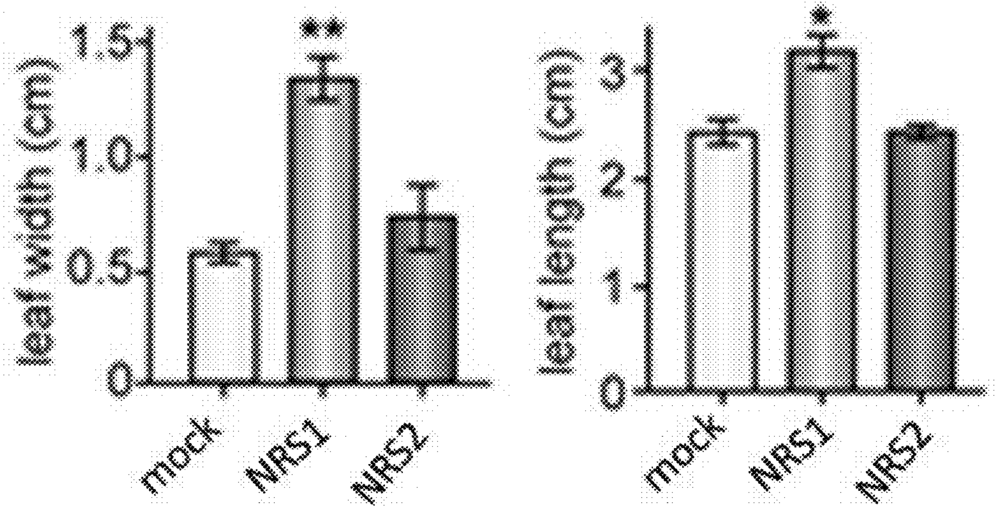
leaf width (cm)
1.5
1.0
0.5
0
**
mock
NRS1
NRS2
leaf length (cm)
3
2
1
0
*
mock
NRS1
NRS2

FIG. 2

Col-0

35S:PRO: NRS1: pEG1

FIG. 3

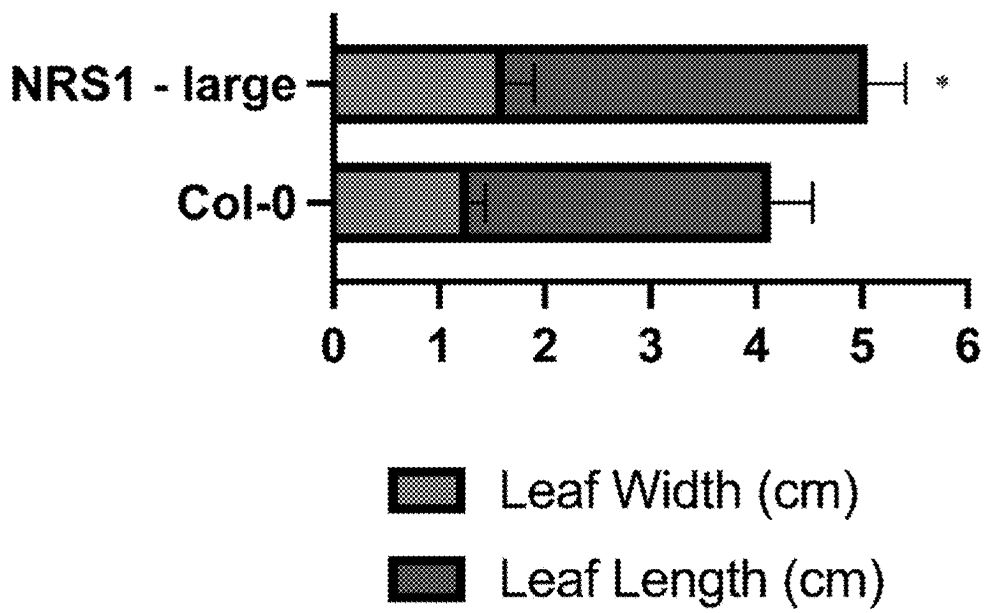

FIG. 4

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ref|NP_200479.1|1-572        1 MADEIVPPATQLAAVSLENDGSTVQRAQFSNRVLIRTILDRPDGGAGLAGQTVRIGGWVK  60
id|mod_ref-XP_009120180      1 MGGDIPPPVDQLAAVSLTSDSSTVQKARFSDRVRIQSILGRPDGGAGLAGQKVRISGWVK  60
id|mod_ref-XP_016682506      1 --DQALPPSDQLAAMNLTD---TVEKHAFSDRVLIRSIVGRPDGGAGLAGQRVRAGGWVK  55
id|mod_ref-XP_003633115      1 MAEDSVPPVDQMALATLNQDVSTFVKAQFSDRHLIRTILSRPDGGAGLAGQTVKVGGWVK  60
ref|XP_008368472.2|1-58      1 MADDSTPPSAQLAAVTLSNDAS-VLKAEFSDRVLIRSIISRPDGGSGLAGQKIRIGGWVK  59
ref|XP_006475374.1|1-56      1 MADNELP-VDQVATMQLNDDA--VQRHQFSDRVLIKSILTRPDGGAGLAGRQVRVGGWVK  57
id|mod_ref-XP_003525303      1 MADASSPPTDQLAAITLDE----VPKANFSDRVPIRSIISRTDGGSGLAGRKARVGGWVK  56
id|mod_ref-XP_025700817      1 ---------AEQLAATTINDDG-IVPKAEFSDRVPINSIISRADGGSGLAGKKARVGGWVK  51
ref|XP_034211989.1|1-56      1 MADDLT---AQLATATLDNNAS-VQRAQFSNRVPIKSIILRPDGGSGLAGKLARVGGWVK  56
ref|XP_023754320.1|1-56      1 MADNGVLPVDKLTI------SETVKEAEFSQRVLVRSILGRPDGGAGLAGQTLKVGGWVK  54
ref|XP_004294177.1|1-58      1 MADDAAQLGSQLAETAL-NESVSDLKAEFSQRVPIKSIISRPDGGSAPAGQKVRVGGWVK  59
id|mod_ref-XP_006351366      1 MSVDSAPPVEKLTL------SDAVEEAKFSQRVPIRSIVGRPDGGAGLAGNVVKIGGWVK  54
id|mod_ref-NP_001304912      1 MSVDSAPPVEKLTLTDV------VEEARFSQRVPICSIVGRTDGGAGLAGNVVKIGGWVK  54
id|mod_ref-XP_015821402      1 ----------------------------TDRTRIRSILA---DGAARAGERVVVGGWVK  28
id|mod_ref-XP_020114036      1 -GDESAPPTSEMAEPSLQ-------------RTLIKSIVSSSAAAAALVGRRVVVGGWVK  46
id|mod_ref-XP_044318137      1 ----------------------------TSRTRIRAILD---AGDAMAGERVVVGGWVK  28
id|mod_ref-NP_001398552      1 -----------------------------------------------------VVVGGWVR   8
id|mod_ref-XP_010672177      1 --------------------------APPSKRIPIRSILNRSDGGASLAGQRVTVRGWVR  34

id|Query_844971-572         61 SGRDQGKRTFSFLAVNDGSCPANLQVMVDPSLYDVSNLVATGTCVTVDGVLKVPPKGKGT 120
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id|mod_ref-XP_009120180     61 TGREQGKGAFAFLEVNDGSCPANLQVMVDASVSDLSKLIATGTCVTVDGCLKLPPEGKGT 120
id|mod_ref-XP_016682506     56 TGREQGKGTFAFLELNDGSCPANLQVIVDAGLAVLSKLVATGTCVVVDGILKVPPEG--T 113
id|mod_ref-XP_003633115     61 TGREQGKGSFAFLELNDGSCPANLQVIVDAAVAPLGQLVQTGTCVHVEGLLKVPPEG--T 118
ref|XP_008368472.2|1-58     60 TGRKADKDAFAFLELNDGSCASNLQVIVKADKYDLGQLVQTGTCLSVDGVLKLPPDGK-- 117
ref|XP_006475374.1|1-56     58 TGREQGKGSFAFLEVNDGSCPANLQVIVDKDVADLGQLVPTGTCVYVEGMLKNPPEG--T 115
id|mod_ref-XP_003525303     57 TGRKADKDAFAFLEINDGSCAGNLQVIVEAALGELGQLVPTGTCVVVDGHLKLPPAG--T 114
id|mod_ref-XP_025700817     52 TGRKADKDAFAFLELNDGSCPGNLQVIVEAGLVELSQVVPTGTCVVLDGELKLPPEG--A 109
ref|XP_034211989.1|1-56     57 TGRKADKDAFAFLELNDGSCSGNLQVIVEADKGDLGQLVLTGTSLVVDGVLKLPPDGK-- 114
ref|XP_023754320.1|1-56     55 KGREQGKGSFAFLELNDGSCTANLQLIIYSDVAPLGQFTPTGTSLHVEGVLQMPPADKGG 114
ref|XP_004294177.1|1-58     60 TGRKADKDAFAFLELNDGSCPGNLQVIVDADKGDLGQLVPTGTCVVVEGELKLPPAG--V 117
id|mod_ref-XP_006351366     55 TGREQGKGSFAFLEVNDGSCPGNLQVIVDASVHKLGDLVLTGTCVHVEGELKVPPEG--A 112
id|mod_ref-NP_001304912     55 TGREQGKGSFAFLEVNDGSCPGNLQVIVDASVHKLGDLVLTGTCVHVEGELKVPPEG--A 112
id|mod_ref-XP_015821402     29 TGREQGKGTFAFLELNDGSCASNLQVLVDAAVHPLAPLTATGTSVLVEGELKKPPEG--A  86
id|mod_ref-XP_020114036     47 TGREQGKGTFAFLELNDGSCLANLQVIVDAKVHPLSELVPTGTCVLVEGELKKPPEG--T 104
id|mod_ref-XP_044318137     29 TGRQQGKGEFAFLEVNDGSCQQNLQVMVDKDVHPLASLTHTGTSVLVEGVLKKPPAE--A  86
id|mod_ref-NP_001398552      9 TGREQGKGSFAFLELSDGSCAATLQAIVDAAVHPLARLTATGTSVLVEGVIKEPPEG--T  66
id|mod_ref-XP_010672177     35 TGRKGNKGALGFIELNDGSCCANLQVIVESSVGELSQLLSPGSCVSLSGELKSPPQSGND  94

id|Query_844971-572        121 QQQIELNVVKVIDVGTVDASKYPLPKTKLTLETLRDVLHLRSRTNSISAVARIRNALAFA 180
ref|NP_200479.1|1-572      121 QQQIELNVVKVIDVGTVDASKYPLPKTKLTLETLRDVLHLRSRTNSISAVARIRNALAFA 180
id|mod_ref-XP_009120180    121 KQKIELSVVEVVDVGTVDPGTYPIPKTKLTLERLREFLHLRARTNSISAIARIRHALAIA 180
id|mod_ref-XP_016682506    114 RQKIELRVEKVVSVGEVDPAKYPIPKTKLTLEFLRDHLHLRARTNTIAAIARIRNALAFA 173
id|mod_ref-XP_003633115    119 KQRVELRVEKVHHVGPVDPAKYPLPKTRLTLEFLRDFVHFRPRTNTISAVARIRNALAYA 178
ref|XP_008368472.2|1-58    118 KQKVELDVEKVIHVGPVDPAKYPLPKTRLTLEFLREHVHLRSRTNTISAVTRIRNALAFA 177
ref|XP_006475374.1|1-56    116 KQKIELRVQKVVDVGMVDPAKYPIPKTKLTLEFLRDRIPFRPRTNTIAAVARIRNALAYA 175
id|mod_ref-XP_003525303    115 RQKIELRADKVLHVGPVDPAKYPLPKMRLTLEFLRDFVHLRSRTNTISAVARIRNALAYA 174
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ref|XP_034211989.1|1-56    115 RQKVELRVEKVIHVGLVDPSKYPLPKTRLPLEFLRDYVHLRSRTNTISAVLRIRDALAYA 174
ref|XP_023754320.1|1-56    115 KQSIELKVSKVLDVGAADPAKYPLPKTRLTLEFLRDYVHLRPRTNTISAIARIRNALAYA 174
ref|XP_004294177.1|1-58    118 KQKVELRVEEVLHLGPVDPLKYPLHKGKISLERLRDVVHLRPRTNTISAVARIRDQLAYA 177
id|mod_ref-XP_006351366    113 KQKVELRVQKVISVGTVDAAKYPLPKTKLTLEFLRDVVHLRSRTNTISAVARIRNALAYA 172
id|mod_ref-NP_001304912    113 KQKVELRVQKVISVGTVDAAKYPLPKTKLTLEFLRDVVHLRSRTNTISAVARIRNALAYA 172
id|mod_ref-XP_015821402     87 KQRVELRVDRVIEVGEVDPAAYPLPKTKLTLENLRDVVHLRSRTNTIGAVARIRHQLACA 146
id|mod_ref-XP_020114036    105 KQRIELRVDKVLALGPVD-SKYPLPKTRLTLENLREVVHLRARTNTISAVARIRDELAYG 163
id|mod_ref-XP_044318137     87 KQRIELKVERVIELGEVDAAAYPLPKTKITLETLRDFVHLRARTNTIGAVARIRHQLAYA 146
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FIG. 4 (continued)

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ref|NP_200479.1|1-572        181 THSFFQEHSFLYIHTPIITTSDCEGAGEMFQATTLINYTERLEQDLIQNPPPTEADVEAA 240
tr|mod_ref-XP_000120180      181 THTFFDEEGFLYVQTPIITTSDCEGAGEMFQVTTLISTTEKLERELIENPPPTEADVEAA 240
tr|mod_ref-XP_016682505      174 THSFFQEHNFLYVHTPLTTSDCEGAGEMFQVTTLISSSEKLEKELIKNPPPSEVDIEAA 233
tr|mod_ref-XP_003633115      179 THTFFQNHGFLYIHTPIITTSDCEGAGEMFQVTTLISDAEKVEKELIKNPPPSEADIEAA 238
ref|XP_008368472.2|1-58      178 THSFFHKHGFLYVQTPIITTSDCEGAGEMFQVTTLFSSAEKLEKDLIENPPPSEADIEAA 237
ref|XP_006478374.1|1-56      176 THTFLQKQGFLYIHTPIITTSDCEGAGEMFQVTTLISDAQKLEKELIKNPPPSEADIEAA 235
tr|mod_ref-XP_003525302      175 THTFFNKEGFLYVHTPIVTTSDCEGAGEMFQVTTLLSEAERLEKELLQNPPPSEADVEAA 234
tr|mod_ref-XP_025700817      170 THTFFNKHGFLYVHTPIITTSDCEGAGEMFQVTTLLSEAERLEKELIHNPPPTEAEVEAA 229
ref|XP_034217989.1|1-56      175 THTFFHQHGFRYVQTPIITTSDCEGAGEMFQVTTLINEAEKLEKDLIENPP-SEADIEAA 233
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tr|Query_844971-572          241 RLIVIERQNVVAELKAAKASKEAITAAVAELKIAKETFAHIDERSRLRPGLPKKDQNIDY 300
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tr|mod_ref-XP_003633115      239 KALVKEKQEAVAQLKSAKASKQEITASVAELNKAKENLSRLEERSKLKPGIPQKDQKIDY 298
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tr|mod_ref-NP_001398856      187 KVLVKEKQDVVAQLKAAKASKQEISTAVDELNRAKEIVSKLEERFKLKPGIPRKDQSIAF 246
tr|mod_ref-XP_010672177      215 KLVVEEKQEVLKKLKAAEASKEEIKAAVQVLMKAKQDLAKLEERAKLQPGIPKKDQKVDY 274

tr|Query_844971-572          301 SKDFFGRQAFLTVSGQLQVETYACALSNVYTFGPTFRAENSHTSRHLAEFWMVEPEIAFA 360
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tr|mod_ref-XP_000120180      301 SQDFFGRQAFLTVSGQLQVETYACGLSDVYTFGPTFRAENSHTSRHLAEFWMVEPELAFA 360
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tr|mod_ref-XP_003633115      299 SQDFFARQAFLTVSGQLQVETYACAVSSVYTFGPTFRAEHSHTSRHLAEFWMVEPEIAFA 358
ref|XP_008368472.2|1-58      298 TQDFFARQAFLTVSGQLQVESYACALSSVYTFGPTFRAENSHTSRHLAEFWMVEPEIAFA 357
ref|XP_006478374.1|1-56      296 TQDFFARQAFLTVSGQLQVETYACAVSNVYTFGPTFRAEHSHTSRHLAEFWMVEPEMAFS 355
tr|mod_ref-XP_003525302      295 GKDFFARQAFLTVSGQLQVESYACALSSVYTFGPTFRAENSHTSRHLAEFWMVEPEIAFA 354
tr|mod_ref-XP_025700817      290 AKDFFARQAFLTVSGQLQVESYACALSSVYTFGPTFRAENSHTSRHLAEFWMVEPELAFA 349
ref|XP_034217989.1|1-56      294 TQDFFGRQAFLTVSGQLQVESYACSLSTVYTFGPTFRAENSHTSRHLAEFWMVEPEIAFA 353
ref|XP_023754320.1|1-58      295 SQDFFARQAFLTVSGQLQVETFACALSSVYTFGPTFRAEHSHTSRHLAEFWMVEPEIAFA 354
ref|XP_004294177.1|1-58      298 TQDFFARQAFLTVSGQLQVESYACAISSVYTFGPTFRAENSHTSRHLAEFWMVEPELAFA 357
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tr|mod_ref-XP_044318137      267 ENDFFKRQAFLTVSGQLQVETFACALSSVYTFGPTFRAENSHTSRHLAEFWMVEPEIAFA 326
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FIG. 4 (continued)

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ref|NP_200479.1|1-572      361 DLEDDMNCAEAYVKYMCNMLLEKCYADMELMAKNFDSGCIDRLKLVASTPFGRITYTKAI 420
tr|mod_ref-XP_009120180    361 DLEDDMNCAEAYVKYMCNMLLEKRYDDMELMAKNFDKGCIDRLKLVASTPFGRLTYTEAI 420
tr|mod_ref-XP_016682509    354 DLQDDMNCAEAYVKYMCNMLLDKCLDDMLFMAKSYDKGCIDRLRMVASVPFVRISYTEAV 413
tr|mod_ref-XP_003633115    359 DLKDDMNCAEAYVKFLCQMLLDNCIDDMEFMAKNFDKGCIDRLRMVASTPFERISYTEAI 418
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ref|XP_008475374.1|1-56    356 DLKDDMNCAEAYVKFMCQMLLDHCFDDMEFMAKNYDKSCINRLRMVASTPFERITYTEAV 415
tr|mod_ref-XP_003525302    355 ELKDDMNCAEAYVKFMCQMLLDNCLEDMEFMADKFDKGCIDRLKLVASTPFIRVTYTEAV 414
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ref|XP_034211989.1|1-56    354 ELEDDMNCAEAYVKYLCQSLLDNCREDMEFMADKIDKSCIDRLTMVAKTPFVRITYTEAV 413
ref|XP_023754320.1|1-56    355 DIEDDMKCAEAYVRFMCQMLLDNCLDDMEFIAEKFDEHAINRLKMVASTNFVRLTYTEAV 414
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tr|mod_ref-XP_044318137    327 NLQDDMNCAERYVQYLCKMLLKHCREDMEFMVKHVDKTAIERLELVSSTPFERISYTKAV 386
tr|mod_ref-NP_001308552    307 NLQDDMNCAEKYVQYLCKMLLDHCQEDMEFMVKNYDKSAIERLELVSSTPFVRISYTKAV 366
tr|mod_ref-XP_010672177    335 DLEDDMNCAEAYIKFMCQMLLDRCLDDMVYIAKLFDNTAIDRLKMVASTAFVRITYTEAV 394

tr|Query_849971-572        421 ELLEEAVAKGKEFDNNVEWGIDLASEHERYLTEVLFQKPLIVYNYPKGIKAFYMRLNDDE 480
ref|NP_200479.1|1-572      421 ELLEEAVAKGKEFDNNVEWGIDLASEHERYLTEVLFQKPLIVYNYPKGIKAFYMRLNDDE 480
tr|mod_ref-XP_009120180    421 KILEEAVAKGKKFDNDVEWGIDLASEHERYLTEVVFQKPLIVYNYPKGIKAFYMRLNDDG 480
tr|mod_ref-XP_016682509    414 ELLEEAVRGGKKFENEVKWGIDLASEHERYLTEVKFQKPLIVYNYPKGIKAFYMRLNDDL 473
tr|mod_ref-XP_003633115    419 KLLEEAVKKGKKFENKVEWGIDLASEHERFLTEVLFKKPVIVHDYPKGIKAFYMRLNDDM 478
ref|XP_008388472.2|1-568   418 DLLIDAVKKGKKFENNVEWGIDLASEHERYLTEVLFQKPVIVYNYPKGIKAFYMRLNDDL 477
ref|XP_008475374.1|1-56    416 ELLEVAVKEGKHFENKVEWGIDLASEHERYLTEVKFQKPVIVYNYPKGIKAFYMRLNDDL 475
tr|mod_ref-XP_003525302    415 EILEDAVKNGKKFENEVKWGIDLASEHERYLTEVKFQKPVIVYNYPKDIKAFYMRLNDDL 474
tr|mod_ref-XP_025700817    410 EILEEAVKNGKKFDNEVKWGIDLASEHERYLTEVKFQKPVIVYNYPKGIKAFYMRLNDDS 469
ref|XP_034211989.1|1-56    414 ELLTEAVKNGKKFENNVEWGIDLASEHERYLTEVRFQKPVIVYNYPKGIKAFYMRLNDDS 473
ref|XP_023754320.1|1-56    415 TILEEAVSKGHGFENKVEWGIDLASEHEKYLTETKFESPVIVYNYPKGIKAFYMKVNPDN 474
ref|XP_004294177.1|1-56    418 ELLIDAVKNGKKFENPVEWGIDLASEHERFLTEVKFQKPVIVYNYPKGIKAFYMRLNDDN 477
tr|mod_ref-XP_008351366    413 DILEVA-AKVKKFENKVEWGIDLASEHERYLTEEHFKAPVIVFNYPKGIKAFYMKVNEDN 471
tr|mod_ref-NP_001304915    413 DILEVA-AKVKKFENKVEWGIDLASEHERYLTEEHFKAPVIVFNYPKGIKAFYMKVNEDN 471
tr|mod_ref-XP_015631402    387 ELLKNVT--DKKFENKVEWGIDLASEHERYLTEVIFKKPVIVYNYPKEIKAFYMRLNDDQ 444
tr|mod_ref-XP_020114036    404 GLLKNVT--DKEFENKVEWGIDLASEHERYLTEVIFKKPVIVYNYPKGIKAFYMRLNDDQ 461
tr|mod_ref-XP_044318137    387 EILE---GVDKKFENKVEWGIDLASEHERYLTEVIFKKPVIVYNYPKGIKAFYMRLNDDQ 443
tr|mod_ref-NP_001308552    367 ELLKNVT--DKKFDNKVEWGIDLASEHERYLTEDIFKKPVIVYNYPKGIKAFYMRLNDDD 424
tr|mod_ref-XP_010672177    395 TLLEQV--KDKKFENKVEWGSDLASEHERYLTEVIFKKPVIVYNYPKGIKAFYMRLNDDG 452

tr|Query_849971-572        481 KTVAAMDVLVPKVGELIGGSQREERYDVIKKRIEEMGLPIEPYEWYLDLRRYGTVKHCGF 540
ref|NP_200479.1|1-572      481 KTVAAMDVLVPKVGELIGGSQREERYDVIKKRIEEMGLPIEPYEWYLDLRRYGTVKHCGF 540
tr|mod_ref-XP_009120180    481 KTVAAMDVLVPKVGELIGGSQREERIDVIMERLEEIGLPVEPYEWYLDLRRYGTAKHSGF 540
tr|mod_ref-XP_016682509    474 KTVAAMDVLVPKVGELIGGSQREERYEVIRERILEVGLPLEPYEWYLDLRRYGTVKHCGF 533
tr|mod_ref-XP_003633115    479 KTVAAMDVLVPKVGELIGGSQREERYEVIEKRILEMGLPLEPYEWYLDLRRYGTVKHCGF 538
ref|XP_008388472.2|1-568   478 KTVAAMDVLVPKVGELIGGSQREERYDIIMSRIEEMGLPAEPYEWYLDLRRFGTVKHCGF 537
ref|XP_008475374.1|1-56    476 KTVAAMDVLVPKVGELIGGSQREERYDVIKSRIEDMGLPLEPYEWYLDLRRFGTVKHSGF 535
tr|mod_ref-XP_003525302    475 KTVAAMDVLVPKVGELIGGSQREERYDVIQTRIKEMGLPVEPYEWYLDLRRYGTVKHAGF 534
tr|mod_ref-XP_025700817    470 KTVAAMDVLVPKVGELIGGSQREERYDVIVQRIKEMGLPLEPYEWYLDLRRYGTVKHSGF 529
ref|XP_034211989.1|1-56    474 KTVAAMDVLVPKVGELIGGSQREERYDVIVSRIKEMGLPLEPYEWYLDLRRYGTVKHCGF 533
ref|XP_023754320.1|1-56    475 KTVAAMDVLVPKVGELIGGSQREENYEVIKERILEMGLPLEPYEWYLDLRRYGTVKHSGF 534
ref|XP_004294177.1|1-56    478 KTVAAMDLLVPKVGELIGGSQREERYDVIHSRIAEMGLPIEPYEWYLDLRRFGTAKHSGF 537
tr|mod_ref-XP_008351366    472 KTVAAMDLLVPKVGELIGGSQREENYEVLRSRILDMGLPLEPYEWYLDLRRYGTVKHSGF 531
tr|mod_ref-NP_001304915    472 KTVAAMDLLVPKVGELIGGSQREENYEVLRSRILDLGLPLEPYEWYLDLRRYGTVKHSGF 531
tr|mod_ref-XP_015631402    445 KTVAAMDVLVPKVGELVGGSQREERLDLLKTRIGDAGLPLEPYEWYLDLRRFGSVKHSGF 504
tr|mod_ref-XP_020114036    462 KTVAAMDVLVPKVGELIGGSQREERIDVLKSRILESGLPLEPYEWYLDLRRYGTVKHCGF 521
tr|mod_ref-XP_044318137    444 KTVAAMDVLVPKVGELIGGSQREERLDILKQRILDAGLPLEPYEWYLDLRRFGSVKHSGF 503
tr|mod_ref-NP_001308552    425 KTVAAMDVLVPKVGELIGGSQREERLDVLKQRILDAGLPLEPYEWYLDLRRFGSVKHSGF 484
tr|mod_ref-XP_010672177    453 KTVAAMDVLVPKVGELVGGSQREERYDVIVKRIVEMGLPLEHYKWYLDLRRYGTAIHAGF 512
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FIG. 4 (continued)

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tr|Query_8449?/1-572       541 GLGFERMILFATGLDNIRDVIPFPRYPGKADL 572
ref|NP_000479.1|/1-572     541 GLGFERMILFATGLDNIRDVIPFPRYPGKADL 572
tr|mod_ref-XP_00912018     541 GLGFERMVLFATGMDNIRDVIPFPRYPGRADL 572
tr|mod_ref-XP_01666250     534 GLGFERMILFATGIENIRDVIPFPRYPGRADL 565
tr|mod_ref-XP_00303311     539 GLGFERMILFATGIDNIRDVIPFPRYPGRADL 570
ref|XP_008088472.2|/1-56   538 GLGFERMVLFATGIDNIRDVIPFPRYPGRADL 569
ref|XP_006475374.1|/1-56   536 GLGFERMILFATGIDNIRDVIPFPRYPGRADL 567
tr|mod_ref-XP_00352530     535 GLGFERMILFATGLENIRDVIPFPRYPGRADL 566
tr|mod_ref-XP_02570081     530 GLGFERMILFATGLENIRDVIPFPRYPGRADL 561
ref|XP_034211989.1|/1-56   534 GLGFERMVLFATGIDNIRDVIPFPRYPGRADL 565
ref|XP_023754320.1|/1-56   535 GLGFERMILFATGIDNIRDVIPFPRFPGRADL 566
ref|XP_004294177.1|/1-56   538 GLGFERMVLFATGMDNIRDVIPFPRFPGRADL 569
tr|mod_ref-XP_00835130     532 GLGFERMILFATGIENIRDVIPFPRYPGRADL 563
tr|mod_ref-NP_00130491     532 GLGFERMILFATGIENIRDVIPFPRYPGRADL 563
tr|mod_ref-XP_01582140     505 GLGFERMILFATGLENIRDVIPFPRYPGRADL 536
tr|mod_ref-XP_02011403     522 GLGFERMIMFATGLDNIRDVIPFPRYPGKADL 553
tr|mod_ref-XP_04431813     504 GLGFERMILFATGLENIRDVIPFPRYPGRADL 535
tr|mod_ref-NP_00139855     485 GLGFERMILFATGMENIRDVIPFPRYPGRADL 516
tr|mod_ref-XP_01067217     513 GLGFERMLLFATGIDNIRDVIPFPRYPGRADL 544
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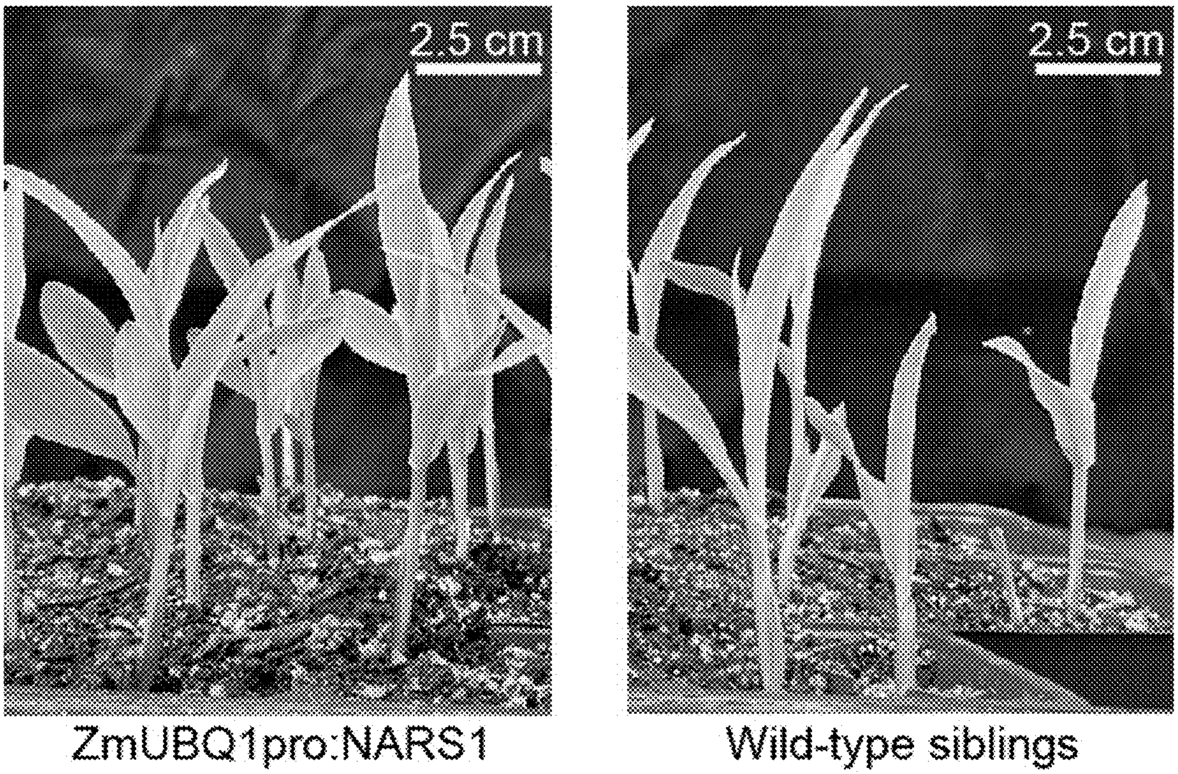
FIG. 5



METHOD FOR IMPROVING PLANT GROWTH WITH A TRNA SYNTHETASE GENE THAT ACTIVATES TOR

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Application No. 63/510,280, filed Jun. 26, 2023, the contents of which are incorporated by reference in their entireties.

STATEMENT REGARDING FEDERALLY SPONSORED RESEARCH

[0002] This invention was made with government support under grant numbers OD023072 and GM145814 awarded by the National Institutes of Health. The government has certain rights in the invention.

SEQUENCE LISTING STATEMENT

[0003] This application includes a sequence listing in XML format titled "960296.04519_ST26.xml", which is 43,231 bytes in size and was created on Jun. 24, 2024. The sequence listing is electronically submitted with this application via Patent Center and is incorporated herein by reference in its entirety.

BACKGROUND

[0004] Plants are an essential resource. We rely on them for food, water, medicine, clean air, habitat, shelter, and fuel. Climate change is expected to have a serious negative impact on our ability to grow plants. Genetic methods to improve crop plant growth rates and yields are urgently needed as demands for agricultural products (bioenergy/biomass, food, feedstock, etc.) increase while fertilizer resources and available arable land decrease. Accordingly, there is a need in the art for more efficient methods of growing plants.

SUMMARY

[0005] The present invention provides compositions and methods for increasing the growth, growth rate, and/or yield of a plant that lacks root nodules by engineering it to overexpress the protein asparaginyl-tRNA synthetase 1 (NARS1).

[0006] In a first aspect, the present invention provides plants that lack root nodules and are engineered to overexpress a NARS1 protein.

[0007] In a second aspect, the present invention provides seeds produced by the plants described herein.

[0008] In a third aspect, the present invention provides methods of generating a plant that overexpresses NARS1. The methods comprise: (a) introducing a construct comprising a heterologous promoter operably linked to a polynucleotide encoding a NARS1 protein into a plant cell; and (b) growing the plant cell into a plant.

[0009] In a fourth aspect, the present invention provides methods of growing a plant that overexpresses NARS1. The methods comprise (a) planting a seed described herein; and (b) growing the seed into a plant.

BRIEF DESCRIPTION OF THE DRAWINGS

[0010] FIGS. **1**A-**1**B show a comparison of transgenic *Arabidopsis thaliana* plants that overexpress NARS1 to wild-type control plants after 7 days of growth. FIG. **1**A shows representative images of the transgenic *Arabidopsis thaliana* seedlings (left) and the wild-type seedlings (right). FIG. **1**B is two bar graphs showing the leaf width (left) and leaf length (right) of leaf 6 of wild-type control plants ("mock"), plants that overexpress NARS1 ("NRS1"), and plants that overexpress NARS2 ("NRS2") after 7 days of growth. Leaf width and length were compared via t test (** p<10-5; * p<10-2; n≥24).

[0011] FIG. **2** shows a comparison of transgenic *Arabidopsis thaliana* plants that overexpress NARS1 to wild-type control plants after 4 weeks of growth. Representative images of Col-0 wild-type plants (top) and plants transformed with 35S:PRO:NRS1:pEG103 (bottom) are shown.

[0012] FIG. **3** is a bar graph comparing the leaf width and length of transgenic *Arabidopsis thaliana* plants that overexpress NARS1 to that of wild-type (Col-0) control plants. The length and width of leaf 6 was measured and compared via t test (** p<10-5; * p<10-2; n≥24).

[0013] FIG. **4** is an alignment of NARS1 protein sequences from 18 different plant species. The identity of each sequence is detailed in Table 1, below.

TABLE 1

NARS1 protein sequences aligned in FIG. 4

Row in Alignment	SEQ ID NO:	Organism	Reference Genome
1	2	Query sequence	NR
2	2	*Arabidopsis thaliana*	NP_200479.1
3	3	*Brassica rapa*	XP_009120180.1
4	4	*Gossypium hirsutum*	XP_016682509.1
5	5	*Vitis vinifera*	XP_003633115.1
6	6	*Malus domestica*	XP_008368472.2
7	7	*Citrus sinensis*	XP_006475374.1
8	8	*Glycine max*	XP_003525302.1
9	9	*Arachis hypogaea*	XP_025700817.1
10	10	*Prunus dulcis*	XP_034211989.1
11	11	*Lactuca sativa*	XP_023754320.1
12	12	*Fragaria vesca* subsp. *vesca*	XP_004294177.1
13	13	*Solanum tuberosum*	XP_006351366.1
14	14	*Solanum lycopersicum*	NP_001304912.1
15	15	*Oryza sativa*Japonica Group	XP_015621402.1
16	16	*Ananas comosus*	XP_020114036.1
17	17	*Triticum aestivum*	XP_044318137.1
18	18	*Zea mays*	NP_001398552.1
19	19	*Beta vulgaris* subsp. *vulgaris*	XP_010672177.2

[0014] FIG. **5** shows representative images of T1 generation (family A) transgenic maize plants that overexpress NARS1 (left) to wild-type control plants (right) two weeks after germination.

DETAILED DESCRIPTION

[0015] The present invention provides plants that lack root nodules and are engineered to overexpress an asparaginyl-tRNA synthetase 1 (NARS1) protein, seeds produced by said plants, and methods of generating and growing said plants.

[0016] NARS1 is a cytosolic asparaginyl-tRNA synthetase, i.e., an enzyme that catalyzes the attachment of asparagine to its cognate tRNA for use in protein translation. In the Examples, the inventor demonstrates that both transgenic *Arabidopsis thaliana* plants (Example 1) and transgenic maize plants (Example 2) that overexpress the *Arabidopsis thaliana* NARS1 protein grow larger and faster than wild-type controls. These results suggest that NARS1 overexpression may be used to increase plant growth, plant growth rate, and/or yield.

[0017] In a previous study, researchers showed that overexpressing NARS1 in the legume *Lotus corniculatus* increased plant biomass and height in a single transgenic line (Yano et al., Plant Root 9:6-14, 2014). However, the researchers concluded that the increased biomass observed in this legume was due to increased nitrogen fixation due to an increased number of root nodules in these plants. Additionally, the same group later showed that overexpressing NARS1 in soybean increased plant height and bushiness in a single transgenic line (Arifin et al., Plant Biotechnol (Tokyo) 36 (4): 233-240, 2019). Thus, NARS1 overexpression was not expected to have a positive effect on growth or yield in plants lacking root nodules, and the inventor's discovery that increased NARS1 expression results in increased growth in *Arabidopsis thaliana* and maize was surprising.

Plants:

[0018] In a first aspect, the present invention provides plants that lack root nodules and overexpress an asparaginyl-tRNA synthetase 1 (NARS1) protein. The plants may be engineered to overexpress NARS1 relative to a control plant using any method known to those of skill in the art.

[0019] The terms "protein," "polypeptide," and "peptide" are used interchangeably herein to refer to a polymer of amino acids, i.e., a series of amino acid residues connected by peptide bonds between the alpha-amino and carboxy groups of adjacent residues. Proteins may be modified (e.g., via acetylation, glycosylation, etc.) and may include amino acid analogs.

[0020] A used herein, a protein is "overexpressed" in a plant if it is expressed at higher levels than it is expressed in a control plant. For example, a plant that overexpresses a protein may express that protein at levels that are at least 25%, at least 50%, at least 2-times, at least 3-times, at least 4-times, at least 5-times, at least 6-times, at least 7-times, at least 8-times, at least 9-times, at least 10-times, at least 20-times, at least 30-times, at least 40-times, or at least 100-times higher than levels found in a control plant.

[0021] As used herein, a "control plant" is a comparable plant (i.e., a plant of the same species, variety, age, etc.) that was grown under substantially similar conditions but that was not engineered to overexpress a NARS1 protein. Plants that are grown in "substantially similar conditions" are grown in similar locations and soil conditions, are planted with similar timing, are subjected to similar treatments and abiotic stresses, and the like.

[0022] In some embodiments, the plant comprises a construct comprising a heterologous promoter operably linked to a polynucleotide encoding the NARS1 protein. As used herein, the term "construct" refers a to recombinant polynucleotide, i.e., a synthetic polynucleotide that was formed by combining at least two polynucleotide components from different sources. For example, a construct may comprise the coding region of one gene operably linked to a promoter that is (1) associated with another gene found within the same genome, (2) from the genome of a different species, or (3) synthetic. Constructs include plasmids, viral constructs, and transposon-based constructs. Constructs can be generated using conventional recombinant DNA methods. In some embodiments, the construct comprises multiple different NARS1-enocoding polynucleotides and/or multiple copies of a single NARS1-enocoding polynucleotide.

[0023] As used herein, the term "promoter" refers to a DNA sequence that defines where transcription of a polynucleotide begins. RNA polymerase and the necessary transcription factors bind to the promoter to initiate transcription. Promoters are typically located directly upstream (i.e., at the 5' end) of the transcription start site. However, a promoter may also be located at the 3' end, within a coding region, or within an intron of a gene that it regulates. Promoters may be derived in their entirety from an endogenous or heterologous gene, may be composed of elements derived from multiple regulatory sequences found in nature, or may comprise synthetic DNA. A promoter is "operably linked" to a polynucleotide if the promoter is positioned such that it can affect transcription of said polynucleotide. The promoters used in the constructs of the present invention are "heterologous", meaning that they are not naturally associated with the NARS1-enocoding polynucleotide to which they are operably linked.

[0024] In preferred embodiments, the promoter is a constitutive promoter. A "constitutive promoter" is a promoter that drives transcription of a polynucleotide in most cell types of an organism at most times. In Example 1, the inventor generates transgenic *Arabidopsis thaliana* plants that comprise a construct comprising a cauliflower mosaic virus (CaMV) 35S promoter/enhancer sequence (SEQ ID NO: 26), which comprises the CaMV 35S promoter of SEQ ID NO: 27, operably linked to a NARS1-encoding polynucleotide. In Example 2, the inventor generates transgenic maize plants that comprise a construct comprising the *Zea mays* ubiquitin 1 (UBQ1) promoter (SEQ ID NO: 24) operably linked to a NARS1-encoding polynucleotide. Both the CaMV 35S promoter and the maize UBQ1 promoter are considered strong constitutive promoters for transgene expression in plants. Thus, in certain embodiments, the promoter is a CaMV 35S promoter or a maize UBQ1 promoter.

[0025] In some embodiments, the promoter is an "inducible promoter," i.e., a promoter that allows for controlled expression of a gene under particular conditions or in the presence of a particular molecule (e.g., tetracycline, dexamethasone). For example, the use of a starvation response promoter would allow for NARS1 overexpression only when nutrients are limiting, and the use of a drought-repressive promoter would allow for NARS1 overexpression only in the presence of a drought response.

[0026] Examples of other suitable promoters that may be used in the constructs of the present invention include, without limitation, other ubiquitin promoters, RuBisCO small subunit 1 (RbcS1) promoters, actin promoters, *Agrobacterium* octopine synthase (OCS) promoters, mannopine synthase (MAS) promoters, and Cestrum yellow leaf curling virus (CmYLCV) promoters.

[0027] The terms "polynucleotide," "nucleic acid," and "oligonucleotide" are used interchangeably to refer a polymer of DNA or RNA. A polynucleotide may be single-stranded or double-stranded and may represent the sense or the antisense strand. A polynucleotide may be synthesized or obtained from a natural source. A polynucleotide may contain natural, non-natural, or altered nucleotides, as well as natural, non-natural, or altered internucleotide linkages. The constructs of the present invention comprise a polynucleotide that encodes a NARS1 protein (i.e., a "NARS1-encoding polynucleotide").

[0028] In the Examples, the inventor generated both transgenic *Arabidopsis thaliana* plants that comprise a construct encoding an endogenous NARS1 protein and transgenic maize plants that comprise a construct encoding a heterologous NARS1 protein (i.e., the same *Arabidopsis thaliana* NARS1 protein). Thus, the overexpressed NARS1 protein may be either an endogenous NARS1 protein (i.e., a NARS1 protein that is natively expressed by the plant) or a heterologous NARS1 protein (i.e., a NARS1 protein that is not natively expressed by the plant).

[0029] The plants of the present invention are engineered to overexpress a NARS1 protein. The term "engineered" is used herein to refer to plants that have been altered by the hand of man. Those of skill in the art are aware of multiple methods for engineering a plant to overexpress a particular protein. For example, a plant may be engineered to overexpress a NARS1 protein by introducing a NARS1-encoding polynucleotide into the plant using well-known recombinant or molecular biology techniques. In embodiments in which the NARS1 protein is an endogenous protein, the NARS1-encoding polynucleotide may comprise one or more extra copy of an endogenous polynucleotide (i.e., a polynucleotide that is natively found in the plant) that encodes NARS1. The endogenous NARS1-encoding polypeptide may optionally be altered to include synonymous mutations and/or additional sequences (e.g., adaptor sequences, a sequence encoding a reporter molecule or a protein tag), e.g., to increase protein expression or to increase the case of cloning or protein detection. In embodiments in which the NARS1 protein is a heterologous protein, the NARS1-encoding polynucleotide comprises a heterologous polynucleotide (i.e., a polynucleotide that is not natively found in the plant) that encodes NARS1. In some embodiments, the engineered plants are genetically modified. For example, in some embodiments, the NARS1-encoding polynucleotide is integrated into the genome of the plant (e.g., using an *Agrobacterium* vector, viral vector, nuclease, or transposase). In these embodiments, the NARS1-encoding polynucleotide may be either inserted randomly into the genome or inserted into a specific location (e.g., via homologous recombination). In other embodiments, the NARS1-encoding polynucleotide remains extrachromosomal (i.e., as part of an extrachromosomal plasmid).

[0030] Alternatively, a plant may be engineered to overexpress a NARS1 protein by upregulating the expression of an endogenous NARS1 gene. As used herein, a gene is "upregulated" if it has manipulated such that it is transcribed at higher levels than it would be in the absence of said manipulation. For example, an upregulated gene may be transcribed at levels that are at least 25%, at least 50%, at least 2-times, at least 3-times, at least 4-times, at least 5-times, at least 6-times, at least 7-times, at least 8-times, at least 9-times, at least 10-times, at least 20-times, at least 30-times, at least 40-times, or at least 100-times higher than the levels at which it would be transcribed in the absence of manipulation. In another alternative, the transcript may be made more stable such that additional NARS1 is generated as compared to a control plant. Upregulation can be accomplished by inserting a regulatory element into the genome such that it is operably linked to a target gene. Examples of suitable regulatory elements that can be used to upregulate a target gene include, without limitation, promoters, enhancers, and insulators. Alternatively, upregulation can be accomplished using CRISPR-mediated transcriptional activation (CRISPRa). In CRISPRa, a modified nuclease-dead form of Cas9 (dCas9) to which an activator domain has been attached is targeted to a target gene using guide RNAs and used to transcriptionally activate the target gene.

[0031] The plants of the present invention overexpress NARS1 in at least one tissue. The plants may express NARS1 in one, two, three, four, five, six, or more different tissues. For example, the plants may overexpress NARS1 in developing seeds to enhance seed size or yield, overexpress NARS1 in developing fruits to enhance fruit size or yield, overexpress NARS1 in leaves to potentially enhance photosynthesis, carbon sequestration, and/or leaf growth, or overexpress NARS1 in roots to enhance root growth and/or nutrient uptake. Alternatively, the plants may overexpress NARS1 in every tissue or substantially every tissue. Further, the plants may either overexpress NARS1 at one or more particular stages of development or may overexpress NARS1 throughout all developmental stages.

[0032] The term "plant" can refer to a plant at any stage of development or to any part of a plant, including a plant cutting, a plant cell, a plant cell culture, a plant organ, a plant tissue, a plant seed, or a plantlet. In some embodiments, the plant is selected from the group consisting of maize, tobacco, hemp, rice, canola, potato, wheat, cotton, and sugar beet. As is noted in the Examples, the inventor has generated both *Arabidopsis thaliana* and maize plants that overexpress the *Arabidopsis thaliana* NARS1 protein (SEQ ID NO: 2). Thus, in some embodiments, the plants are *Arabidopsis thaliana* or maize.

[0033] The plants of the present invention lack root nodules. A "root nodule" is a swelling on the root of a plant that allows the plant to house nitrogen-fixing *rhizobia* bacteria. (Note: Root nodules are primarily found in legumes, but are also found in Actinorhizal plants (e.g., alder and bayberry) and plants of the genus *Parasponia*.) Thus, the plants of the present invention are not capable of forming a symbiotic relationship with *rhizobia* that results in independent nitrogen fixation. In at least some embodiments, the plants are non-leguminous. The term "non-leguminous" refers to plants that do not belong to the Fabaceae family (i.e., plants that are not a legume).

[0034] The plants of the present invention may overexpress any plant-derived NARS1 protein. NARS1 is well conserved in plants, as is demonstrated in FIG. **4**, which is an alignment of NARS1 protein sequences from 18 different plants (i.e., SEQ ID NOs: 2-19). Within the "superrosids" group of eudicots (which includes canola, cotton, grape, apple, citrus, soybean, peanut, almond, lettuce, and strawberry), NARS1 is about 70% identical at the amino acid level based on global alignments of the full-length protein (100% of NARS1 aligned). Within monocots (which include rice, wheat, maize, and banana), NARS1 is 70% identical at the amino acid level with local alignments (>90% of NARS1 aligned) or ~63% identical based on an end-to-end alignment. The N-terminal 50 amino acids, which have no assigned or predicted function, do not align entirely. Within the "superasterids" group of eudicots (which includes sugar beets, potato, and tomato), NARS1 is >63% identical at the amino acid level based on global alignments of full-length NARS1 (100% of NARS1 aligned). As an outgroup, human cytosolic NARS1 is only 36% identical over 46% of the protein and 16.6% identical across the whole protein. Thus, in some embodiments, the NARS1 protein is at least 90% identical to an amino acid sequence selected from SEQ ID NOs: 2-19 (see Table 1). In the Examples, the inventor generated transgenic plants that overexpress the *Arabidopsis thaliana* NARS1 protein of SEQ ID NO: 2. Thus, in certain embodiments, the NARS1 protein is at least 90% identical to SEQ ID NO: 2. In some embodiments, the NARS1 protein comprises or consists of SEQ ID NO: 2. Seeds:

[0035] In a second aspect, the present invention provides seeds produced by the plants described herein. A "seed" is an embryonic plant enclosed in a protective outer covering. The seeds provided herein are engineered such that they will develop into plants that overexpress NARS1 in at least one tissue.

Methods of Generating Plants that Overexpress NARS1:

[0036] In a third aspect, the present invention provides methods of generating a plant that lacks root nodules and overexpresses NARS1. The methods comprise: (a) introducing a construct comprising a heterologous promoter operably linked to a polynucleotide encoding a NARS1 protein into a plant cell; and (b) growing the plant cell into a plant.

[0037] The term "plant cell" refers to any cell of a plant. A plant cell is the basic structural and functional unit of plants. A plant cell comprises a protoplast and a cell wall. A plant cell can be in the form of an isolated single cell, part of an aggregate of cells, or part of a higher order structure or plant. The plant cells used with the present invention are part of or are derived from plants that lack root nodules.

[0038] As used herein, "introducing" describes a process by which exogenous polynucleotides are introduced into a recipient cell. Suitable introduction methods include, without limitation, *Agrobacterium*-mediated transformation, transposition-based plant transformation, the floral dip method, bacteriophage or viral infection, electroporation, heat shock, lipofection, microinjection, vacuum-infiltration, and particle bombardment.

[0039] In the Examples, the inventor utilized *Agrobacterium*-mediated transformation to introduce NARS1-encoding polynucleotides into plants. Thus, in preferred embodiments, the polynucleotides are introduced via *Agrobacterium*-mediated transformation. In this method, the NARS1-encoding polynucleotide is delivered into plant cells as part of a binary *Agrobacterium* vector, in which it is flanked by two transfer DNA (T-DNA) border repeat sequences. Prior to transformation into plant cells, this binary vector is co-transformed into *Agrobacterium tumefaciens* along with a second vector that is referred to as a vir helper plasmid. The vir helper plasmid encodes components necessary for integration of the region flanked by the T-DNA border repeat sequences into the genome of plant cells. Thus, when the binary vector and the vir helper plasmid are both present in the same *Agrobacterium* cell, proteins encoded by the vir helper plasmid act in trans on the T-DNA border repeat sequences to mediate processing, secretion, and host genome integration of the intervening transgene. Genome insertion occurs without any significant bias with respect to insertion site sequence.

[0040] In the present methods, NARS1-encoding constructs are introduced into a plant cell. Suitable constructs and parts thereof (i.e., heterologous promoters and NARS1-encoding polynucleotides) for use in these methods are described above, in the section titled "Plants."

[0041] As used herein, "growing" describes a process in which suitable conditions (i.e., light, soil, water, nutrients, temperature) for plant growth are established and maintained.

[0042] Any type of plant that lacks root nodules may be generated using the present methods. In some embodiments, the plant is selected from the group consisting of maize, tobacco, hemp, rice, canola, potato, wheat, cotton, and sugar beet. In some embodiments, the plant is *Arabidopsis thaliana* or maize.

Methods of Growing Plants that Overexpress NARS1:

[0043] In a fourth aspect, the present invention provides methods of growing a plant that lacks root nodules and overexpresses NARS1. The methods comprise (a) planting a seed described herein; and (b) growing the seed into a plant. As used herein, "planting" describes a process in which a seed in placed in soil or another suitable growth medium (e.g., peat moss, coconut coir, vermiculite, perlite, sand, polymer-based gels).

[0044] In the Examples, the inventor demonstrates that transgenic *Arabidopsis thaliana* and maize plants that overexpress NARS1 grow larger and faster than wild-type control plants. Thus, in some embodiments, the plant grown using the present methods has an increased growth rate and/or yield as compared to a control plant.

[0045] The term "growth rate" describes the rate at which the size of a plant increases. Growth rate can be assessed, for example, by measuring the leaf area, leaf width, leaf length, leaf number, stem length, rosette diameter, root length, seed number, seed weight, height, biomass, or volume of a plant over time.

[0046] The term "yield" describes the amount of harvestable (i.e., useful) material produced by a plant. Examples of harvestable plant materials include, without limitation, flowers, pollen, seedlings, tubers, leaves, stems, fruit, seeds, and roots.

[0047] The present disclosure is not limited to the specific details of construction, arrangement of components, or method steps set forth herein. The compositions and methods disclosed herein are capable of being made, practiced, used, carried out and/or formed in various ways that will be apparent to one of skill in the art in light of the disclosure that follows. The phraseology and terminology used herein is for the purpose of description only and should not be regarded as limiting to the scope of the claims. Ordinal indicators, such as first, second, and third, as used in the description and the claims to refer to various structures or method steps, are not meant to be construed to indicate any specific structures or steps, or any particular order or configuration to such structures or steps. All methods described herein can be performed in any suitable order unless otherwise indicated herein or otherwise clearly contradicted by context. The use of any and all examples, or exemplary language (e.g., "such as") provided herein, is intended merely to facilitate the disclosure and does not imply any limitation on the scope of the disclosure unless otherwise claimed. No language in the specification, and no structures shown in the drawings, should be construed as indicating that any non-claimed element is essential to the practice of the disclosed subject matter. The use herein of the terms "including," "comprising," or "having," and variations thereof, is meant to encompass the elements listed thereafter and equivalents thereof, as well as additional elements. Embodiments recited as "including," "comprising," or "having" certain elements are also contemplated as "consisting essentially of" and "consisting of" those certain elements.

[0048] Recitation of ranges of values herein are merely intended to serve as a shorthand method of referring individually to each separate value falling within the range, unless otherwise indicated herein, and each separate value is incorporated into the specification as if it were individually recited herein. For example, if a concentration range is stated as 1% to 50%, it is intended that values such as 2% to 40%, 10% to 30%, or 1% to 3%, etc., are expressly enumerated in this specification. These are only examples of what is specifically intended, and all possible combinations of numerical values between and including the lowest value and the highest value enumerated are to be considered to be expressly stated in this disclosure. Use of the word "about" to describe a particular recited amount or range of amounts is meant to indicate that values very near to the recited amount are included in that amount, such as values that could or naturally would be accounted for due to manufacturing tolerances, instrument and human error in forming measurements, and the like. All percentages referring to amounts are by weight unless indicated otherwise.

[0049] No admission is made that any reference, including any non-patent or patent document cited in this specification, constitutes prior art. In particular, it will be understood that, unless otherwise stated, reference to any document herein does not constitute an admission that any of these documents form part of the common general knowledge in the art in the United States or in any other country. Any discussion of the references states what their authors assert, and the applicant reserves the right to challenge the accuracy and pertinence of any of the documents cited herein. All references cited herein are fully incorporated by reference, unless explicitly indicated otherwise. The present disclosure shall control in the event there are any disparities between any definitions and/or description found in the cited references.

[0050] The following examples are meant only to be illustrative and are not meant as limitations on the scope of the invention or of the appended claims.

EXAMPLES

Example 1

[0051] In the following example, the inventor demonstrates that transgenic *Arabidopsis thaliana* plants that overexpress NARS1 grow larger and faster than wild-type controls.

BACKGROUND

[0052] Target of rapamycin (TOR) is a highly conserved serine/threonine protein kinase that coordinates growth and development with nutritional status in eukaryotes. TOR regulates key pathways such as nucleotide biosynthesis, ribosome biogenesis, and leaf initiation. TOR is well-studied in humans because is it dysregulated in many human diseases, including diabetes and cancer. In animals and fungi, many of the upstream mechanisms that activate TOR are known, but almost nothing is known about how TOR activity is regulated in plants.

[0053] Asparaginyl-tRNA synthetase 1 (NARS1; official names: EMB2755 and SYNC1; gene ID: At5g56680) was identified as a potential regulator of TOR in plants in a forward genetic screen for defective embryonic cell-cell (plasmodesmatal) trafficking that repeatedly identified mutants defective in TOR signaling (Brunkard et al., Proc. Natl. Acad. Sci. U.S.A 117 (9): 5049-5058, 2020; Kim et al., Development 129 (5): 1261-1272, 2002). NARS1 mutants have stunted growth and defective TOR signaling. RNA sequencing revealed that NARS1 mutants exhibit several signatures of TOR inactivation, including significant repression of ribosomal protein genes and induction of proteolytic and catabolism-related genes (Busche et al., Plant Cell 33 (5): 1615-1632, 2021; Xiong et al., Nature 496 (7444): 181-186, 2013). Further, NARS1 mutants and knockdowns also show significantly less phosphorylation of S6K-T449 (orthologous to human S6K-T389), a residue that is uniquely phosphorylated by TOR kinase in plants. Thus, NARS1 activates TOR in plants and represents the first proposed amino acid sensor in plants.

Materials and Methods:

[0054] RNA was extracted from *Arabidopsis thaliana* Col-0 and used as a template for RT-PCR with SuperScript III and Phusion, following the manufacturer's instructions. The NARS1 coding sequence was amplified from this RNA using the forward primer caccATGGCTGATGAGATTGTG (SEQ ID NO: 20) and the reverse primer AAGATCAGCTTTTCCAGGATAGCG (SEQ ID NO: 21), which were synthesized by IDT DNA. The forward primer included a CACC overhang for directional cloning into D-TOPO pENTR vectors. The reverse primer was designed to exclude the final stop codon from the NARS1 coding sequence to allow translational readthrough to C-terminal epitope tags. The PCR product size was verified by agarose gel electrophoresis and was purified using a gel extraction kit (NEB Monarch kit).

[0055] The product was subcloned into pENTR using the D-TOPO pENTR kit (Invitrogen) and used to transform chemically competent *E. coli* cells (genotype DH10B) using kanamycin selection. Resistant colonies were screened for by colony PCR and positive transformants were used in minipreps to isolate plasmid (NEB Monarch kit). Purified plasmid was sequenced using Sanger sequencing. This plasmid was digested with EcoRV and recombined with pEarleyGate 103 using Gateway LR Clonase (for full plasmid information, see abrc.osu.edu/stocks/number/CD3-685 [abrc.osu.edu]). pEarleyGate 103 includes a C-terminal GFP tag and a CaMV 35S promoter. Insertion into pEarleyGate 103 was validated using Sanger sequencing. The resulting plasmid is referred to herein as 35S: PRO: NRS1: pEG103.

[0056] 35S: PRO: NRS1: pEG103 was introduced into *Agrobacterium tumefaciens* (genotype GV3101) and used to transform *Arabidopsis thaliana* inflorescences following the standard floral dip protocol. Positive transformants were screened for herbicide resistance, NARS1 insertions were validated by PCR, and NARS1 overexpression was validated by fluorescence microscopy. Dwarf transgenic lines showed an insertion but no GFP fluorescence, indicating gene silencing, whereas large transgenic lines showed strong GFP fluorescence in leaves.

Results:

[0057] To test the effects of constitutively overexpressing NARS1 in plants, the *Arabidopsis thaliana* NARS1 coding sequence (SEQ ID NO: 1, which encodes the NARS1 protein of SEQ ID NO: 2) was cloned into pEarleyGate 103 and was used to transform *Arabidopsis thaliana* ecotype Col-O using an *Agrobacterium* vector (GV3101) and the floral dip method. Several stable transgenic *Arabidopsis thaliana* lines that overexpress NARS1 fused to a GFP reporter were generated. Consistently, NARS1 transgenic plants with visible GFP fluorescence (which indicates successful overexpression of the NARS1 protein) were found to grow larger and faster than wild-type controls (FIG. **1**A). On average, NARS1 overexpression significantly increased the leaf width and length compared to wild-type controls (Col-0) (FIG. **2** and FIG. **3**). This phenotype is similar to TOR overexpression, which causes increased organ and cell size. Yet, overexpressing another annotated asparaginyl tRNA synthetase protein, i.e., NARS2, had no effect on plant growth or TOR activity (FIG. **1**B). This indicates that this effect is specific to NARS1 and not a general property of asparaginyl tRNA synthetases. These results suggest that NARS1 overexpression could be used in agriculture to improve plant growth and yield.

Example 2

[0058] In the following example, the inventor demonstrates that transgenic maize plants that overexpress NARS1 grow larger and faster than wild-type controls.

Materials and Methods:

Cloning $ZmUbi1_{PRO}$:NARS1

[0059] The *Arabidopsis thaliana* NARS1 coding sequence was subcloned into a plasmid such that it was flanked by the promoter and terminator from the *Zea mays* ubiquitin 1 (UBQ1) gene to drive high, consistent levels of NARS1 expression throughout maize development. Specifically, an open reading frame (ORF) encoding the *Arabidopsis thaliana* NARS1 protein was synthesized de novo using GenSmart to optimize codons for expression in maize. Adaptors were added to the 5' and 3' ends of the ORF to facilitate GoldenGate cloning. The 5' adaptor sequence is GGTCTCTA and the 3' adaptor sequence is GCTTTGAGACC (SEQ ID NO: 22). Both adaptor sequences include Bsal recognition sites. The sequence of the resulting construct is provided as SEQ ID NO: 23.

[0060] This construct was subcloned, using a GoldenGate cloning strategy, into the T-DNA of a binary vector. The resulting vector includes the *Zea mays* Ubiquitin 1 (ZmUbi1) promoter (SEQ ID NO: 24), which includes a 5' leader sequence with an intron, upstream of the NARS1 coding sequence. The resulting vector also includes the ZmUbi1 terminator (SEQ ID NO: 25), which includes a 3' untranslated region, downstream of the NARS1 coding sequence. The resulting binary vector is referred to herein as $ZmUbi1_{PRO}$:NARS1.

Maize Transformation

[0061] Maize cells of inbred genotype LH244 were transformed using *Agrobacterium tumefaciens* carrying the $ZmUbi1_{PRO}$:NARS1 binary vector, which also includes a gene that confers resistance to the herbicide glufosinate. Transformants were selected for glufosinate resistance and cultured under conditions to induce plant regeneration. Mature transformants were self-pollinated to generate T1 seeds for further analysis.

Growth Assays

[0062] T1 seeds from two independently transformed parents were sown on wet potting soil and grown in a greenhouse in Madison, WI with ~16 h daylength for two weeks. The transgenic plants were grown alongside (i.e., in the same flat) untransformed LH244 sibling plants. All seeds germinated at similar times. Two weeks after germination, plants were photographed and analyzed for differences in growth and vigor.

Image Analysis

[0063] All images were analyzed using ImageJ.

Results:

[0064] T1 $ZmUbi1_{PRO}$:NARS1 plants from two independent parents (family A and family B) were compared to wild-type sibling controls two weeks after germination in the greenhouse on potting soil mix (FIG. **5**). Under these conditions, the average height of wild-type LH244 plants was 6.8 cm (n=7, standard deviation 3.4 cm), the average height of family A $ZmUbi1_{PRO}$:NARS1 plants was 8.9 cm (n=9, standard deviation 2.1 cm), and the average height of family B $ZmUbi1_{PRO}$:NARS1 plants was 8.6 cm (n=5, standard deviation 2.1 cm). Therefore, the $ZmUbi1_{PRO}$:NARS1 plants from family A were 30.6% taller than wild-type plants, and the $ZmUbi1_{PRO}$:NARS1 plants from family B were 26.6% taller than wild-type plants, on average. Transgenic $ZmUbi1_{PRO}$:NARS1 plants also exhibited less variability in height, with standard deviations that were 61.5% lower than the standard deviations observed for wild-type plants. Transgenic $ZmUbi1_{PRO}$:NARS1 plants from both families also qualitatively appeared to have larger, greener, and more vigorous shoots than the wild-type plants.

SEQUENCE LISTING

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                        mol_type = genomic DNA
                        organism = Arabidopsis thaliana
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cgacctgacg gcggagctgg actcgccggt caaacggttc gaatcggtgg ttgggtaaaa  180
tccggaagag atcaagggaa aagaacgttt tctttccttg cggttaacga tggatcttgt  240
ccagcgaatc ttcaggttat ggtggatccg tctctctatg atgtctctaa cttggttgct  300
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cgttctagaa ctaactcgat ctcagctgtt gcaagaattc gtaatgctct tgcttttgcg  540
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SGRDQGKRTF SFLAVNDGSC PANLQVMVDP SLYDVSNLVA TGTCVTVDGV LKVPPKGKGT  120
QQQIELNVVK VIDVGTVDAS KYPLPKTKLT LETLRDVLHL RSRTNSISAV ARIRNALAFA  180
THSFFQEHSF LYIHTPIITT SDCEGAGEMF QATTLINYTE RLEQDLIDNP PPTEADVEAA  240
RLIVIERGNV VAELKAAKAS KEAITAAVAE LKIAKETFAH IDERSRLRPG LPKKDGNIDY  300
SKDFFGRQAF LTVSGQLQVE TYACALSNVY TFGPTFRAEN SHTSRHLAEF WMVEPEIAFA  360
DLEDDMNCAE AYVKYMCNWL LEKCYADMEL MAKNFDSGCI DRLKLVASTP FGRITYTKAI  420
ELLEEAVAKG KEFDNNVEWG IDLASEHERY LTEVLFQKPL IVYNYPKGIK AFYMRLNDDE  480
KTVAAMDVLV PKVGELIGGS QREERYDVIK KRIEEMGLPI EPYEWYLDLR RYGTVKHCGF  540
GLGFERMILF ATGLDNIRDV IPFPRYPGKA DL                                572

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TGREQGKGAF AFLEVNDGSC PANLQVMVDA SVSDLSKLIA TGTCVTVDGC LKLPPEGKGT  120
KQKIELSVVE VVDVGTVDPG TYPIPKTKLT LERLREFLHL RARTNSISAI ARIRHALAIA  180
THTFFDEEGF LYVQTPIITT SDCEGAGEMF QVTTLISTTE KLERELIENP PPTEADVEAA  240
RVVVKERGEA VAELKAAKAS KEAILASVAE LNDAKANLAA TEARSRLKPG LPKVDGKIDY  300
SQDFFGRQAF LTVSGQLQVE TYACGLSDVY TFGPTFRAEN SHTSRHLAEF WMVEPELAFA  360
DLEDDMNCAE AYVKYMCKWL LEKRYDDMEL MAKNFDKGCI DRLKLVASTP FGRLTYTEAI  420
KILEEAVAKG KKFDNDVEWG IDLASEHERY LTEVVFQKPL IVYNYPKGIK AFYMRLNDDG  480
KTVAAMDVLV PKVGELIGGS QREERIDVIM ERLEEIGLPV EPYEWYLDLR RYGTAKHSGF  540
GLGFERMVLF ATGMDNIRDV IPFPRYPGRA DL                                572

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HNFLYVHTPI LTTSDCEGAG EMFQVTTLIS ESEKLEKELI KNPPPSEVDI EAARQVVSER  240
GEAVKQLKAA KASKSEITAS VAELNKAKEN LSKLEERSKL KPGIPKKDGK IDYTQDFFAR  300
QAFLTVSGQL QVETYACAVS NVYTFGPTFR AEHSHTSRHL AEFWMVEPEI AFADLQDDMN  360
CAEAYVKYMC NWLLDKCLDD MLFMAKSYDK GCIDRLRMVA SVPFVRISYT EAVELLEEAV  420
RGGKKFENEV KWGIDLASEH ERYLTEVKFQ KPLIVYNYPK GIKAFYMRLN DDLKTVAAMD  480
VLVPKVGELI GGSQREERYE VIRERILEVG LPLEPYEWYL DLRRYGTVKH CGFGLGFERM  540
ILFATGIENI RDVIPFPRYP GRADL                                        565

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RVELRVEKVH HVGPVDPAKY PLPKTRLTLE FLRDFVHFRP RTNTISAVAR IRNALAYATH  180
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LVKEKGEAVA QLKSAKASKG EITASVAELN KAKENLSRLE ERSKLKPGIP QKDGKIDYSQ  300
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LEEAVKKDKK FENKVEWGID LASEHERFLT EVLFKKPVIV HDYPKGIKAF YMRLNDDMKT  480
VAAMDVLVPK VGELIGGSQR EERYEVIEKR ILEMGLPLEP YEWYLDLRRY GTVKHCGFGL  540
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FFHKHGFLYV QTPIITTSDC EGAGEMFQVT TLFSEAEKLE KDLIENPPPS EADIEAAKLI  240
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EQGKGSFAFL EVNDGSCPAN LQVIVDKDVA DLGQLVPTGT CVYVEGMLKN PPEGTKQKIE  120
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MNCAEAYVKF MCDWLLDHCF DDMEFMAKNY DKSCINRLRM VASTPFERIT YTEAVELLEV  420
AVKEGKHFEN KVEWGIDLAS EHERYLTEVK FQKPVIVYNY PKGIKAFYMR LNDDLKTVAA  480
MDVLVPKVGE LIGGSQREER YDVIKSRIED MGLPLEPYEW YLDLRRFGTV KHSGFGLGFE  540
RMILFATGID NIRDVIPFPR YPGRADL                                     567

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VKNGKKFENE VKWGIDLASE HERYLTEVKF QKPVIVYNYP KDIKAFYMRL NDDLKTVAAM  480
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MILFATGLEN IRDVIPFPRY PGRADL                                      566

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                       organism = Prunus dulcis
SEQUENCE: 10
MADDLTAQLA TATLDNNASV QRAQFSNRVP IKSIILRPDG GSGLAGKLAR VGGWVKTGRK  60
ADKDAFAFLE LNDGSCSGNL QVIVEADKGD LGQLVLTGTS LVVDGVLKLP PDGKRQKVEL  120
RVEKVIHVGL VDPSKYPLPK TRLPLEFLRD YVHLRSRTNT ISAVLRIRDA LAYATHTFFH  180
DHGFRYVQTP IITTSDCEGA GEMFQVTTLI NEAEKLEKDL IENPPSEADI EAAKLIAKER  240
GIDVAQLKSA KASKQEIGAA VVELQKAKDN LVKLEERSKL QPGIPRKDGK IDYTQDFFGR  300
QAFLTVSGQL QVESYACSLS TVYTFGPTFR AENSHTSRHL AEFWMVEPEI AFAELEDDMN  360
CAEAYVKYLC QSLLDNCRED MEFMADKIDK SCIDRLTMVA KTPFVRITYT EAVELLTEAV  420
KNGKKFENHV EWGIDLASEH ERYLTEVRFQ KPVIVYNYPK GIKAFYMRLN DDSKTVAAMD  480
VLVPKVGELI GGSQREERYD VIVSRIKEMG LPLEPYEWYL DLRRYGTVKH CGFGLGFERM  540
VLFATGIDNI RDVIPFPRYP GRADL                                       565

SEQ ID NO: 11          moltype = AA  length = 566
FEATURE                Location/Qualifiers
source                 1..566
                       mol_type = protein
                       organism = Lactuca sativa
SEQUENCE: 11
MADNGVLPVD KLTISETVKE AEFSQRVLVR SILGRPDGGA GLAGQTLKVG GWVKKGREQG  60
KGSFAFLELN DGSCTANLQL IIYSDVAPLG QFTPTGTSLH VEGVLQMPPA DKQGKQSIEL  120
KVSKVLDVGA ADPAKYPLPK TRLTLEFLRD YVHLRPRTNT ISAIARIRNA LAYATHTFFQ  180
KHGFLYVHTP IITTSDCEGA GEMFQVTTLI NDSEKLEKEL LKNPPPSQED VDAARAAVKE  240
KGGIVAKLKS DKADKSAITV AVAELTKAKE TLSKIEERFN QKPGIPKKDG KVDYSQDFFA  300
RQAFLTVSGQ LQVETFACAL SSVYTFGPTF RAEHSHTSRH LAEFWMVEPE IAFADIEDDM  360
KCAEAYVRFM CQWLLDNCLD DMEFIAEKFD EHAINRLKMV ASTNFVRLTY TEAVTILEEA  420
VSKGHQFENK VEWGIDLASE HEKYLTETKF ESPVIVYNYP KGIKAFYMKV NPDNKTVAAM  480
DVLVPKVGEL IGGSQREENY EVIKERILEM GLPLEPYEWY LDLRRYGTVK HSGFGLGFER  540
MILFATGIDN IRDVIPFPRF PGRADL                                      566

SEQ ID NO: 12          moltype = AA  length = 569
FEATURE                Location/Qualifiers
source                 1..569
                       mol_type = protein
                       organism = Fragaria vesca
SEQUENCE: 12
MADDAAQLGS QLAETALNES VSDLKAEFSD RVPIKSIISR PDGGSAFAGQ KVRVGGWVKT  60
GRKADKDAFA FLELNDGSCP GNLQVIVQAD KGDLGQLVPT GTCVVVEGEL KLPPAGVKQK  120
VELRVEEVLH LGPVDPLKYP LHKGKISLER LRDVVHLRPR TNTISAVARI RDDLAYATHT  180
FFRKHGFRYV HTPIITTSDC EGAGEMFQVT TLISEGERLE RELIKNPPPS EADVEAANLI  240
VTEKGDAVSQ LKSAKASKDE IGAAVAELKR AKENVLKLQE RAKLQPGIPK KDGKVDYTQD  300
FFARQAFLTV SGQLQVESYA CAISSVYTFG PTFRAENSHT SRHLAEFWMV EPELAFAELK  360
DDMNCAEAYV KFLCKWLLDN CYDDMEFFSR QYDKTCIERL RMVASTPFER ITYTEAVELL  420
IDAVKNGKKF ENPVEWGIDL ASEHERFLTE VKFQKPVIVY NYPKGIKAFY MRLNDDNKTV  480
AAMDVLVPKV GELIGGSQRE ERYDVIHSRI AEMDLPIEPY EWYLDLRRFG TAKHSGFGLG  540
FERMVLFATG MDNIRDVIPF PRFPGRADL                                   569

SEQ ID NO: 13          moltype = AA  length = 563
FEATURE                Location/Qualifiers
source                 1..563
                       mol_type = protein
                       organism = Solanum tuberosum
SEQUENCE: 13
MSVDSAPPVE KLTLSDAVEE AKFSQRVPIR SIVGRPDGGA GLAGNVVKIG GWVKTGREQG  60
KGSFAFLEVN DGSCPGNLQV IVDASVHKLG DLVLTGTCVH VEGELKVPPE GAKQKVELRV  120
QKVISVGTVD AAKYPLPKTK LTLEFLRDVV HLRSRTNTIS AVARIRNALA YATHTFFQNN  180
GFLYIHTPII TTSDCEGAGE MFQVTSLISE AEKLEKELKE NPAPSESDIQ AAEQLAKEKG  240
EVVAQLKASK ASNKKISAAV AELQRAKENL LKLQERSRLS AGIPKKDGKI DYSEDFFGRQ  300
AFLTVSGQLQ VETYACALSS VYTFGPTFRA EQSHTTRHLA EFWMVEPEIA FADIQDDMNC  360
AEAYVKFLCQ WLLDHCLDDM EFMTKFVDKD ALSRLRMVAS SNCHRITYTE AIDILEVAAK  420
VKKFENKVEW GIDLASEHER YLTEEHFKAP VIVFNYPKGI KAFYMKVNED NKTVAAMDLL  480
VPKVGELIGG SQREENYEVL RSRILDMGLP LEPYEWYLDL RRYGTVKHSG FGLGFERMIL  540
FATGIENIRD VIPFPRYPGR ADL                                         563
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SEQ ID NO: 14           moltype = AA  length = 563
FEATURE                 Location/Qualifiers
source                  1..563
                        mol_type = protein
                        organism = Solanum lycopersicum
SEQUENCE: 14
MSVDSAPPVE KLTLTDVVEE ARFSQRVPIC SIVGRTDGGA GLAGNVVKIG GWVKTGREQG  60
KGSFAFLEVN DGSCPGNLQV IVDASVHKLG DLVLTGTCVH VEGELKVPPE GAKQKVELRV  120
QKVISVGTVD AAKYPLPKTK LTLEFLRDVV HLRSRTNTIS AVARIRNALA YATHTFFQNN  180
GFLYIHTPII TTSDCEGAGE MFQVTSLISE AEKLEKELKE NPAPSESDTQ AAEKLAKEKG  240
EVVAQLKASK ASNKKISAAV AELQRAKENL LKLQERSRLS AGIPKKDGKI DYSEDFFGRQ  300
AFLTVSGQLQ VETYACALSS VYTFGPTFRA EQSHTTRHLA EFWMVEPEIA FADIQDDMNC  360
AEAYVKFLCQ WLLDHCLDDM EFMTKFVDKD ALSRLRMVAS SNCHRITYTE AIDILEVAAK  420
VKKFENKVEW GIDLASEHER YLTEEHFKAP VIVFNYPKGI KAFYMKVNED NKTVAAMDLL  480
VPKVGELIGG SQREENYEVL RSRILDLGLP LEPYEWYLDL RRYGTVKHSG FGLGFERMIL  540
FATGIENIRD VIPFPRYPGR ADL                                          563

SEQ ID NO: 15           moltype = AA  length = 536
FEATURE                 Location/Qualifiers
source                  1..536
                        mol_type = protein
                        organism = Oryza sativa
SEQUENCE: 15
TDRTRIRSIL ADGAARAGER VVVGGWVKTG REQGKGTFAF LELNDGSCAS NLQVLVDAAV  60
HPLAPLTATG TSVLVEGELK KPPEGAKQRV ELRVDRVIEV GEVDPAAYPL PKTKLTLENL  120
RDVVHLRSRT NTIGAVARIR HQLACATHRF FDENGFLYVH TPIITTSDCE GAGEMFQVTT  180
LFSHAEKVEK ELKENPAPSE SDIEAARVVV KEKGDAVAQL KAAKASKQEI TAAVAELNKA  240
KENVSRLEER SKLKPGIPRD DGTVAYENDF FKRQAFLTVS GQLQVETYAC ALSSVYTFGP  300
TFRAENSHTS RHLAEFWMVE PEIAFANLQD DMNCAERYVQ YLCKWLLEHC REDMEFMVKN  360
YDKTAIERLE LVSSTPFQRI SYTKAVELLK NVTDKKFENK VEWGIDLASE HERYLTEVIF  420
KKPVIVYNYP KEIKAFYMRL NDDQKTVAAM DVLVPKVGEL VGGSQREERL DLLKTRIQDA  480
GLPLEPYEWY LDLRRFGSVK HSGFGLGFER MILFATGLEN IRDVIPFPRY PGRADL      536

SEQ ID NO: 16           moltype = AA  length = 553
FEATURE                 Location/Qualifiers
source                  1..553
                        mol_type = protein
                        organism = Ananas comosus
SEQUENCE: 16
GDESAPPTSE MAEPSLQRTL IKSIVSSSAA AAALVGRRVV VGGWVKTGRE QGKGTFAFLE  60
LNDGSCLANL QVIVDAKVHP LSELVPTGTC VLVEGELKKP PEGTKQRIEL RVDKVLALGP  120
VDSKYPLPKT RLTLENLREV VHLRARTNTI SAVARIRDEL AYGTHTFFRK NGFRYVHTPI  180
ITTSDCEGAG EMFQVTTLFS LAEKTEKELK QNPPPSESEV EAAKVLVKEK GEAVAQLKSS  240
KASKEEISAS VAELTKAKEN LSRLEERFKL KPGIPRKDGG IAFENDFFKR QAFLTVSGQL  300
QVETYACALG NVYTFGPTFR AENSHTSRHL AEFWMVEPEM AFANLQDDMN YAESYVKFLC  360
QWLLDHCLED MEFMVKNYDK TAIERLKLVA SVPFEHISYT KAVQLLKNVT DKEFENKVEW  420
GIDLASEHER YLTEVIFKKP VIVYNYPKGI KAFYMRLNDD QKTVAAMDVL VPKVGELIGG  480
SQREERIDVL KSRILESGLP LEPYEWYLDL RRYGTVKHCG FGLGFERMIM FATGLDNIRD  540
VIPFPRYPGK ADL                                                     553

SEQ ID NO: 17           moltype = AA  length = 535
FEATURE                 Location/Qualifiers
source                  1..535
                        mol_type = protein
                        organism = Triticum aestivum
SEQUENCE: 17
TSRTRIRAIL DAGDAMAGER VVVGGWVKTG RQQGKGEFAF LEVNDGSCQG NLQVMVDKDV  60
HPLASLTHTG TSVLVEGVLK KPPAEAKQRI ELKVERVIEL GEVDAAAYPL PKTKITLETL  120
RDFVHLRART NTIGAVARIR HQLAYATHSF FDENGFLYIH TPIITTSDCE GAGEMFQVTA  180
LFSQAEKVEK ELKENPAPSE ADVEAAKLVV KEKGDAVAQL KAAKASKQEI TAAVSVLTKA  240
KENVLRVEER SKLKPGLPKD DGKIAFENDF FKRQAFLTVS GQLQVETYAC ALSNVYTFGP  300
TFRAENSHTS RHLAEFWMVE PEIAFANLQD DMNCAERYVQ YLCKWLLKHC REDMEFMVKH  360
VDKTAIERLE LVSSTPFERI SYTKAVEILE GVDKKFENKV EWGIDLASEH ERYLTEVIFK  420
KPVIVYNYPK GIKAFYMRLN DDQKTVAAMD VLVPKVGELI GGSQREERLD ILKQRILDAD  480
LPLEPYEWYL DLRRFGSVKH SGFGLGFERM ILFATGLENI RDVIPFPRYP GRADL       535

SEQ ID NO: 18           moltype = AA  length = 516
FEATURE                 Location/Qualifiers
source                  1..516
                        mol_type = protein
                        organism = Zea mays
SEQUENCE: 18
VVVGGWVRTG REQGKGSFAF LELSDGSCAA TLQAIVDAAV HPLARLTATG TSVLVEGVIK  60
EPPEGTKQNV ELKVSRVLEV GEVDAAVYPL PKVKLTLEKL RDVVHLRSRT NTIGAVARIR  120
HQLAYASHRF FDENGFLYVH TPIITTSDCE GAGEMFQVTT LFSQAEKTEK ELRENPKPSD  180
SEIEAAKVLV KEKGDVVAQL KAAKASKQEI STAVDELNRA KEIVSKLEER FKLKPGIPRK  240
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DGSIAFENDF FKRQAFLTVS GQLQVETYAC ALSSVYTFGP TFRAENSHTS RHLAEFWMVE  300
PEIAFANLQD DMNCAEKYVQ YLCKWLLDHC QEDMEFMVKN YDKSAIERLE LVSSTPFVRI  360
SYTKAVELLK NVTDKKFDNK VEWGIDLASE HERYLTEDIF KKPVIVYNYP KGIKAFYMRL  420
NDDDKTVAAM DVLVPKVGEL IGGSQREERL DVLKQRILDA GLPLEPYEWY LDLRRFGSVK  480
HSGFGLGFER MILFATGMEN IRDVIPFPRY PGRADL                           516

SEQ ID NO: 19          moltype = AA  length = 544
FEATURE                Location/Qualifiers
source                 1..544
                       mol_type = protein
                       organism = Beta vulgaris
SEQUENCE: 19
APFSKRIPIR SILNRSDGGA SLAGQRVTVR GWVRTGRKQN KGALGFIELN DGSCCANLQV  60
IVESSVCELS QLLSPGSCVS LSGELKSPPQ SGNDKQKIEL FADHVIEVVP VDPTEYPLPK  120
TRIITEYLRD FVHLRPRTSS FAAVARIRSE LAYATHTFFR DNGFINVHTP IITTIDCEGA  180
GETFQVTTLI KDAEKLEQEL LENPAPSEAD VEAAKLVVEE KGEVLKKLKA AEASKEEIKA  240
AVGVLMKAKG DLAKLEERAK LQPGIPKKDG KVDYAHDFFA RQAFLTVSGQ LQAESFACSL  300
GSVYTFGPTF RAEHSHTSRH LAEFWMVEPE IAFADLEDDM NCAEAYIKFM CQWLLDRCLD  360
DMVVIAKLFD NTAIDRLKMV ASTAFVRITY TEAVTLLEQV KDKKFENKVE WGSDLASEHE  420
RYLTEVIFKK PVIVYNYPKG IKAFYMRLND DGKTVAAMDV LVPKVGELVG GSQREERYDV  480
IVKRIVEMGL PLEHYKWYLD LRRYGTAIHA GFGLGFERML LFATGIDNIR DVIPFPRYPG  540
RADL                                                              544

SEQ ID NO: 20          moltype = DNA  length = 22
FEATURE                Location/Qualifiers
source                 1..22
                       mol_type = other DNA
                       organism = synthetic construct
SEQUENCE: 20
caccatggct gatgagattg tg                                          22

SEQ ID NO: 21          moltype = DNA  length = 24
FEATURE                Location/Qualifiers
source                 1..24
                       mol_type = other DNA
                       organism = synthetic construct
SEQUENCE: 21
aagatcagct tttccaggat agcg                                        24

SEQ ID NO: 22          moltype = DNA  length = 11
FEATURE                Location/Qualifiers
source                 1..11
                       mol_type = other DNA
                       organism = synthetic construct
SEQUENCE: 22
gctttgagac c                                                      11

SEQ ID NO: 23          moltype = DNA  length = 2014
FEATURE                Location/Qualifiers
source                 1..2014
                       mol_type = other DNA
                       organism = synthetic construct
misc_feature           1..8
                       note = 5' adaptor
misc_feature           2004..2014
                       note = 3' adaptor
SEQUENCE: 23
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ccggtggggg gtccgggggt tcggcgtgga gccacccgca gtttgaaaaa ggtggcggga  180
gcggaggcgg cagcggcggg tccgcctggt cacatcctca gttcgagaaa tcaggctctg  240
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gcaagaggac attttctttc ctggcggtga atgacggaag ttgtcctgct aatctgcagg  540
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ccgtcgatgg tgtgcttaag gttccgccta aaggcaaggg gacccaacag cagatcgagc  660
ttaatgttgt caaagtgatt gatgttggaa cggtggacgc ctcaaagtat cctctgccta  720
agaccaagct gacgttagag actctgaggg acgtgctcca cctaaggagc aggactaata  780
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aggagcacag tttcctgtac attcatacgc cgatcatcac cacctcggac tgcgagggtg  900
cgggtgagat gtttcaagcg accactctta tcaactacac cgagcggctg gagcaagatc  960
ttatagacaa ccccccgccg actgaagcag acgtggaggc agcgagactg atcgtcattg  1020
agagaggaaa cgtggtggct gaattgaaag cagcaaaggc gagcaaggag gctatcacgg  1080
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tctccaatgt gtacactttc ggcccgacgt tccgcgcaga aaattcccat acatcgcgcc  1320
acctggccga attctggatg gtcgagcctg aaattgcctt tgctgatttg gaagatgaca  1380
tgaactgcgc ggaggcttac gtgaagtaca tgtgcaactg gttgctagag aagtgctatg  1440
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ccaaaggcat caaggcgttc tacatgcgcc tgaatgatga tgagaaaaca gtcgccgcca  1740
tggacgtcct cgtcccaaaa gttggcgaac tcataggtgg aagccagaga gaggagcgct  1800
atgatgtcat caaaaagaga attgaagaga tgggcctacc catagaacca tacgagtggt  1860
acctggatct caggaggtat ggaacagtca agcactgcgg ctttggattg ggctttgagc  1920
gcatgatttt gtttgcaacg ggcctcgaca acatacggga tgttatccca ttcccgcggt  1980
accctggcaa agctgacctg tgagctttga gacc                              2014

SEQ ID NO: 24           moltype = DNA  length = 2014
FEATURE                 Location/Qualifiers
source                  1..2014
                        mol_type = genomic DNA
                        organism = Zea mays
SEQUENCE: 24
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cagtttatct atctttatac atatatttaa actttactct acgaataata taatctatag  180
tactacaata atatcagtgt tttagagaat catataaatg aacagttaga catggtctaa  240
aggacaattg agtattttga caacaggact ctacagtttt atctttttag tgtgcatgtg  300
ttctcctttt tttttgcaaa tagcttcacc tatataatac ttcatccatt ttattagtac  360
atccatttag ggtttagggt taatggtttt tatagactaa tttttttagt acatctattt  420
tattctattt tagcctctaa attaagaaaa ctaaaactct attttagttt ttttatttaa  480
taatttagat ataaaataga ataaaataaa gtgactaaaa attaaacaaa taccctttaa  540
gaaattaaaa aaactaagga aacattttttc ttgtttcgag tagataatgc cagcctgtta  600
aacgccgtcg acgagtctaa cggacaccaa ccagcgaacc agcagcgtcg cgtcgggcca  660
agcgaagcag acggcacggc atctctgtcg ctgcctctgg acccctctcg agagttccgc  720
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tgagccggca cggcaggcgg cctcctcctc ctctcacggc accggcagct acgggggatt  840
cctttcccac cgctccttcg ctttcccttc ctcgcccgcc gtaataaata gacaccccct  900
ccacaccctc tttccccaac ctcgtgttgt tcggagcgca cacacacaca accagatctc  960
ccccaaatcc acccgtcggc acctccgctt caaggtacgc cgctcgtcct cccccccccc  1020
cctctctacc ttctctagat cggcgttccg gtccatggtt agggcccggt agttctactt  1080
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cggatgcgac ctgtacgtca gacacgttct gattgctaac ttgccagtgt ttctctttgg  1200
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tggatctgta tgtgtgtgcc atacatattc atagttacga attgaagatg atggatggaa  1500
atatcgatct aggataggta tacatgttga tgcgggtttt actgatgcat atacagagat  1560
gctttttgtt cgcttggttg tgatgatgtg gtgtggttgg gcggtcgttc attcgttcta  1620
gatcggagta gaatactgtt tcaaactacc tggtgtattt attaattttg gaactgtatg  1680
tgtgtgtcat acatcttcat agttacgagt ttaagatgga tggaaatatc gatctaggat  1740
aggtatacat gttgatgtgg gttttactga tgcatataca tgatggcata tgcagcatct  1800
attcatatgc tctaaccttg agtacctatc tattataata aacaagtatg ttttataatt  1860
attttgatct tgatatactt ggatgatggc atatgcagca gctatatgtg gattttttta  1920
gccctgcctt catacgctat ttatttgctt ggtactgttt cttttgtcga tgctcaccct  1980
gttgtttggt gttacttctg cagaatgaga gacc                              2014

SEQ ID NO: 25           moltype = DNA  length = 932
FEATURE                 Location/Qualifiers
source                  1..932
                        mol_type = genomic DNA
                        organism = Zea mays
SEQUENCE: 25
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tttaataatg gaccggttgt gttgtgtgtg cgtactaccc agaactatga caaatcatga  180
ataagtttga tgtttgaaat taaagcctgt gctcattatg ttctgtcttt cagttgtctc  240
ctaatatttg cctgcaggta ctggctatct accgtttctt acttaggagg tgtttgaatg  300
cactaaaact aatagttagt ggctaaaatt agttaaaaca tccaaacacc atagctaata  360
gttgaactat tagctatttt tggaaaatta gttaatagtg aggtagttat ttgttagcta  420
gctaattcaa ctaacaattt ttagccaact aacaattagt ttcagtgcat tcaaacaccc  480
ccttaatgtt aacgtggttc tatctaccga ctcctaatat atggttgatt gttcggtttg  540
ttgctatgct attgggttct gattgctgct agttcttgct gaatccagaa gttctcgtag  600
tatagctcag attcatatta tttatttgag tgataagtga tccaggttat tactatgtta  660
gctaggtttt ttttacaagg ataaattatc tgtgatcata attcttatga aagctttatg  720
tttcctggag gcagtggcat gcaatgcatg acagcaactt gatcacacca gctgaggtag  780
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atacggtaac aaggttctta aatctgttca ccaaatcatt ggagaacaca catacacatt  840
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cttttggcca tgagtcgtga ccgcttgaga cc                                932

SEQ ID NO: 26          moltype = DNA  length = 1341
FEATURE                Location/Qualifiers
source                 1..1341
                       mol_type = genomic DNA
                       organism = Cauliflower mosaic virus
SEQUENCE: 26
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cagaatcggg tattcaacac cctcatatca actactacgt tgtgtataac ggtccacatg  120
ccggtatata cgatgactgg ggttgtacaa aggcggcaac aaacggcgtt cccggagttg  180
cacacaagaa atttgccact attacagagg caagagcagc agctgacgcg tacacaacaa  240
gtcagcaaac agacaggttg aacttcatcc ccaaaggaga agctcaactc aagcccaaga  300
gctttgctaa ggccctaaca agcccaccaa agcaaaaagc ccactggctc acgctaggaa  360
ccaaaaggcc cagcagtgat ccagccccaa aagagatctc ctttgccccg gagattacaa  420
tggacgattt cctctatctt tacgatctag gaaggaagtt cgaaggtgaa ggtgacgaca  480
ctatgttcac cactgataat gagaaggtta gcctcttcaa tttcagaaag aatgctgacc  540
cacagatggt tagagaggcc tacgcagcag gtctcatcaa gacgatctac ccgagtaaca  600
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FEATURE                Location/Qualifiers
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                       mol_type = genomic DNA
                       organism = Cauliflower mosaic virus
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What is claimed:

1. A plant that lacks root nodules and is engineered to overexpress an asparaginyl-tRNA synthetase 1 (NARS1) protein as compared to a control plant.

2. The plant of claim **1**, wherein the plant comprises a construct comprising a heterologous promoter operably linked to a polynucleotide encoding the NARS1 protein.

3. The plant of claim **2**, wherein the heterologous promoter is a constitutive promoter.

4. The plant of claim **3**, wherein the constitutive promoter is a cauliflower mosaic virus (CaMV) 35S promoter or a ubiquitin promoter.

5. The plant of claim **1**, wherein the plant is a non-leguminous plant.

6. The plant of claim **1**, wherein the plant is selected from the group consisting of maize, tobacco, hemp, rice, canola, potato, wheat, cotton, and sugar beet.

7. The plant of claim **1**, wherein the plant is *Arabidopsis thaliana* or maize.

8. The plant of claim **1**, wherein the NARS1 protein is an endogenous NARS1 protein.

9. The plant of claim **8**, wherein expression of an endogenous gene encoding the NARS1 protein is upregulated in the plant.

10. The plant of claim **1**, wherein the NARS1 protein is a heterologous NARS1 protein.

11. The plant of claim **1**, wherein the NARS1 protein is at least 90% identical to an amino acid sequence selected from SEQ ID NOs: 2-19.

12. The plant of claim **1**, wherein the NARS1 protein is overexpressed in at least one tissue selected from roots, leaves, fruit, or seeds of the plant.

13. A seed produced by the plant of claim **1**.

14. A method of generating a plant that overexpresses a NARS1 protein, the method comprising:

a) introducing a construct comprising a heterologous promoter operably linked to a polynucleotide encoding the NARS1 protein into a plant cell from a plant that lacks root nodules; and

b) growing the plant cell into the plant that overexpresses the NARS1 protein.

15. The method of claim **14**, wherein the plant is selected from the group consisting of *Arabidopsis thaliana*, maize, tobacco, hemp, rice, canola, potato, wheat, cotton, and sugar beet.

16. The method of claim **14**, wherein the NARS1 protein is at least 90% identical to an amino acid sequence selected from SEQ ID NOs: 2-19.

17. A plant or seed produced by the method of claim **14**.

18. A method of growing a plant that overexpresses NARS1, the method comprising:

a) planting the seed of claim **13**; and

b) growing the seed into a plant that overexpresses NARS1.

19. The method of claim **18**, wherein the plant has an increased growth rate as compared to a control plant.

20. The method of claim **18**, wherein the plant has an increased yield as compared to a control plant.

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