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(81) **Designated States** *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, 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, IR, IS, JP, KE, KG, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

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Published:

— *with international search report (Art. 21(3))*

— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments (Rule 48.2(h))*

(88) **Date of publication of the international search report**:
20 August 2015

(54) **Title**: PLANTS CONTAINING BENEFICIAL ENDOPHYTES

(57) **Abstract**: This application relates to methods and materials for providing a benefit to a seed, seedling or plant by producing seeds that are internally colonized with endophytes. Beneficial endophytes with particular characteristics are provided. These may be used in the methods described to provide a benefit to a seed, seedling or plant.

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US14/72399

A. CLASSIFICATION OF SUBJECT MATTER
IPC(8) - A01N 63/00, 63/02 (2015.01)
CPC - A01N 63/00, 63/02
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC(8): A01N 63/00, 63/02, 63/04 (2015.01)
CPC: A01N 63/00, 63/02; C05G 3/02; C12N 1/20; C12R1/01, 1/07

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

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
PatSeer (US, EP, WO, JP, DE, GB, CN, FR, KR, ES, AU, IN, CA, INPADOC Data); Dialog ProQuest; IP.com; Google; Google Scholar; preparation, composition, agricultural, plant, seed, bacterial, endophyte

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X — Y	WO 2013/090628 A1 (SYNTHETIC GENOMICS, INC.) June 20, 2013; abstract; paragraphs [0008], [0009], [0014], [0015], [0020], [0025], [0026], [0028], [0049], [0052], [0099], [00111], [00116], [00127]	1-3, 53, 89-91, 104 — 77-85, 92-98
Y	ROSENBLUETH, M et al. Bacterial Endophytes And Their Interactions With Hosts. MPMI. 2006, Vol. 19, No. 8; pages 827-837; abstract; page 828, column 1, paragraph 1; page 832, column 1, paragraph 7; page 833, column 1, paragraph 4. DOI: 10.1094/MPMI-19-0827.	77-85
Y	US 2012/0131696 A1 (AAYAL, S et al.) May 24, 2012; abstract; paragraph [0347]	92-98
Y	US 2012/0178624 A1 (KAMINSKYJ, SGW et al.) July 12, 2012; abstract; paragraphs [0011], [0013], [0026], [0037], [0045], [0083]	81-85
Y	US 2009/0155214 A1 (ISAWA, T et al.) June 18, 2009; figures 1-1, 2-2; paragraphs [0003], [0008], [0012], [0051]	93, 98

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

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

Date of the actual completion of the international search	Date of mailing of the international search report
29 May 2015 (29.05.2015)	26 JUN 2015
Name and mailing address of the ISA/ Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-3201	Authorized officer Shane Thomas PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

Form PCT/ISA/210 (second sheet) (January 2015)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US14/72399

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☒ Claims Nos.: 4-52, 86-88, 99-103
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

-***-Please See Supplemental Page-***-

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Group I: Claims 1-3, 53, 77-85, 89-98, 104

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.

☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

☐ No protest accompanied the payment of additional search fees.

International application No.
PCT/US14/72399

-***-Continued from Box No. III: Observations Where Unity of Invention Is Lacking:

This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees must be paid.

Group I: Claims 1-3, 53, 77-85, 89-98 and 104 are directed toward a method for generating an agricultural seed preparation; and a synthetic combination of a purified bacterial population in association with a seed of an agricultural plant.

Group II: Claims 54-65 are directed toward methods of screening and identifying a modulator of a plant trait.

Group III: Claims 66-76 are directed toward a method for preparing an agricultural seed preparation comprising a microbe localized in the seed, comprising a bacterial endophyte exhibiting the ability to induce an alteration in the population of microbes present in the seed or plant, as compared to a reference seed, seedling or plant.

The inventions listed as Groups I-III do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons: the special technical features of Group I include an activity selected from catalase activity, oxidase activity and casein hydrolysis activity; which are not present in any other Group, the special technical features of Group II including measuring a trait in plants selected from the group consisting of root biomass, root length and height; which are not present in any other Group, the special technical features of Group III including a bacterial population comprising a bacterial endophyte exhibiting the ability to induce an alteration in the population of microbes present in the seed or plant.

Groups I-III share the technical features including contacting at least one flower of a plurality of agricultural plants with at least one bacterial population; the bacterial population comprising a bacterial endophyte; a seed made by the flower, a seedling grown from the seed or an agricultural plant grown from the seed; and a reference seedling or plant. Groups I-II share the technical features including applying a purified bacterial population to flowers of an agricultural plant, the bacterial population comprising bacteria of one or more species of bacterial endophytes; and altered protein expression in seedlings or plants derived from the seeds produced by the flowers; Groups I and III share the technical features including a method for preparing an agricultural seed preparation comprising a microbe localized in the seed, comprising: a) contacting at least one flower of a plurality of agricultural plants with a formulation comprising a purified bacterial population, the bacterial population comprising a bacterial endophyte.

However, these shared technical features are previously disclosed by WO 2013/090628 A1 (SYNTHETIC GENOMICS, INC.) (hereinafter 'SGI') in view of the publication entitled 'Bacterial Endophytes And Their Interactions With Hosts' by Rosenblueth, et al. (hereinafter 'Rosenblueth').

SGI discloses contacting at least one flower (applying to a flower; paragraph [0014]) of a plurality (paragraph [0020]) of agricultural plants (paragraph [00116]) with at least one bacterial population (paragraphs [0009], [0014]); the bacterial population comprising a bacterial endophyte (paragraph [0008]); a seed made by the flower (seed of plants from flowers artificially infected with and endophytic microbial strain; paragraphs [0014], [0015]), a seedling grown from the seed or an agricultural plant grown from the seed (progeny, including plants or plant parts (a seedling or plant grown from the seed); paragraph [0015]); and a reference seedling or plant (a control plant; paragraph [0025]); applying a purified bacterial population (applying an isolated (purified) bacterial population; paragraphs [0009], [0014]) to flowers (paragraph [0014]) of an agricultural plant (paragraphs [0014], [00116]), the bacterial population comprising bacteria of one or more species of bacterial endophytes (the bacterial population comprising bacteria of one or more species of bacterial endophytes; paragraphs [0008], [0009]); a method for preparing (a method for producing (preparing); paragraph [0014]) an agricultural seed preparation (seed; paragraphs [0015], [0037]) comprising a microbe localized in the seed (comprising endophytic microbes; paragraphs [0015], [0053]), comprising: a) contacting at least one flower (applying to a flower; paragraph [0014]) of a plurality (paragraph [0020]) of agricultural plants (paragraphs [0014], [00116]) with a formulation comprising a purified bacterial population (paragraphs [0010], [0014]), the bacterial population comprising a bacterial endophyte (paragraph [0008]); and wherein the plants with endophytic microbes demonstrate enhanced growth and yield (paragraph [0008]). SGI does not disclose altered protein expression in seedlings or plants derived from the seeds produced by the flowers. Rosenblueth discloses altered protein expression in seedlings or plants comprising or contacted with endophytes (altered plant gene (protein) expression in seedlings or plants comprising or contacted with endophytes; abstract; page 832, column 1, paragraph 7). It would have been obvious to a person of ordinary skill in the art, at the time of the invention, to have modified the disclosure of SGI, for integrating the modification of protein expression disclosed by Rosenblueth as a natural means by which the endophyte microbes induce alterations in gene expression in the host plants, which result in the increased growth and yield disclosed by SGI.

Since none of the special technical features of the Groups I-III inventions is found in more than one of the inventions, and since all of the shared technical features are previously disclosed by a combination of the references, unity of invention is lacking.