

	Espacio reservado para el adhesivo de radicación
---	--

DIRECCIÓN DE NUEVAS CREACIONES
Procedimiento acelerado de exámenes de patente (PPH)

1	NÚMERO DE SOLICITUD ANTE LA SIC: NC2016/0005548		
2	GLOBAL - PPH ☐ PAÍS	3	PCT – PPH ☐ PAÍS
4	PROSUR ☐ PAÍS	5	Alianza del Pacifico (AdP) ☐ PAÍS
6	OFICINA DE PRIMERA PRESENTACIÓN (OPP) ☒	7	OFICINA DE EXAMEN ANTERIOR (OEA) ☐
8	NÚMERO DE SOLICITUD O PUBLICACIÓN DE LA SOLICITUD (OPP) ☒ NÚMERO DE SOLICITUD EN OFICINA DE EXAMEN ANTERIOR (OEA): ☐ NÚMERO DE SOLICITUD PCT: ☐		1. 15/135,106 (Patente No. US 9,988,641 B2)
9	☐ REPRESENTANTE LEGAL ☒ APODERADO		
APELLIDOS DAZA MONTALVO		NOMBRES CAROLINA MERCEDES	IDENTIFICACIÓN C.C 66783790 T.P. 141563 CSJ
DIRECCIÓN Carrera 11 No. 86-53 Piso 3		No. TELÉFONO 6181088	
CIUDAD Bogotá D.C.		CORREO ELECTRÓNICO notificaciones@clark emodet.com.co	
PAÍS Colombia		No. RADICACIÓN O PROTOCOLO DE PODER GENERAL	
10	CONDICIONES PARA ACCEDER A ESTE PROCESO:		
10.1. DATOS DE PRIORIDAD ☒ SI ☐ NO			
(33) PAÍS DE ORIGEN		CÓDIGO PAÍS	(31) NÚMERO
1. ESTADOS UNIDOS DE AMÉRICA		US	15/090926
2. ESTADOS UNIDOS DE AMÉRICA		US	15/135106
			(32) FECHA (AAAA/MM/DD)
			2016/04/05
			2016/04/21
10.2. DATOS SOLICITUD INTERNACIONAL PCT ☐ SI ☒ NO			
Solicitud Internacional No. PCT/		Fecha	
Publicación Internacional No. WO/		Fecha	
*El solicitante presenta una copia de la(s) reivindicación(es) patentable(s) u otorgable(s) de la solicitud de la Oficina de Primera Presentación que profirió el examen de patentabilidad, junto con una traducción al castellano. ☒ SI ☐ NO			
*El solicitante presenta los resultados de búsqueda de la Oficina de Examen Anterior, junto con una traducción al castellano ☒ SI ☐ NO			
REQUISITOS PARA EL RECONOCIMIENTO DE LOS RESULTADOS DE EXÁMENES DE PATENTABILIDAD EFECTUADOS A SOLICITUDES EXTRANJERAS			
Para que la Superintendencia de Industria y Comercio pueda reconocer los resultados de exámenes de patentabilidad producidos por otras Oficinas de Propiedad Industrial se debe cumplir con las siguientes condiciones:			
(a) Al momento de solicitar el examen de patentabilidad, si el solicitante pide que se reconozcan exámenes de patentabilidad de otras oficinas extranjeras, deberá aportar junto con el pago de la tasa correspondiente, los siguientes anexos:			
(i) Copia de las resoluciones, fallos o decisiones que han sido relevantes para la determinación de la patentabilidad de la(s) solicitud(es) en la Oficina que haya proferido el examen de patentabilidad que contienen reivindicaciones patentables u otorgables que son la base de la petición, y si es del caso, su traducción al castellano. ☒ SI ☐ NO			
(ii) Copia de todas las reivindicaciones determinadas como patentables u otorgables en la Oficina que profirió el examen de patentabilidad y, si es del caso, su traducción al castellano. ☒ SI ☐ NO			
(iii) Copia de los antecedentes, información o documentos citados por el examinador de la Oficina de Primera Presentación del examen de patentabilidad, en las resoluciones, fallos o decisiones correspondientes, incluyendo la literatura no patente. Los documentos de patente deberán presentarse siempre y cuando no estén disponibles para la Superintendencia de Industria y Comercio mediante el simple acceso a bases de datos gratuitas. ☒ SI ☐ NO			
(iv) Una tabla de correspondencias de las reivindicaciones que han sido determinadas como patentables u otorgables en la solicitud de la Oficina que haya proferido el examen de patentabilidad. ☒ SI ☐ NO			
11	FIRMA DEL SOLICITANTE, DEL APODERADO O DEL REPRESENTANTE LEGAL Junto a cada firma, indicar el nombre del firmante y su calidad (si tal calidad no es obvia al leer el petitorio)		
 CAROLINA DAZA MONTALVO			



UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	ISSUE DATE	PATENT NO.	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/135,106	06/05/2018	9988641	3459.WHL Cont	8300

86379 7590 05/16/2018
Ingredion Incorporated
Suite C
10 Finderne Avenue
Bridgewater, NJ 08807

ISSUE NOTIFICATION

The projected patent number and issue date are specified above.

Determination of Patent Term Adjustment under 35 U.S.C. 154 (b) (application filed on or after May 29, 2000)

The Patent Term Adjustment is 12 day(s). Any patent to issue from the above-identified application will include an indication of the adjustment on the front page.

If a Continued Prosecution Application (CPA) was filed in the above-identified application, the filing date that determines Patent Term Adjustment is the filing date of the most recent CPA.

Applicant will be able to obtain more detailed information by accessing the Patent Application Information Retrieval (PAIR) WEB site (<http://pair.uspto.gov>).

Any questions regarding the Patent Term Extension or Adjustment determination should be directed to the Office of Patent Legal Administration at (571)-272-7702. Questions relating to issue and publication fee payments should be directed to the Application Assistance Unit (AAU) of the Office of Data Management (ODM) at (571)-272-4200.

APPLICANT(s) (Please see PAIR WEB site <http://pair.uspto.gov> for additional applicants):

Hongxin JIANG, Bridgewater, NJ;
Corn Products Development, Inc., Sao Paulo, BRAZIL
Brad Ostrander, Zionsville, IN;
Chris Lane, Bridgewater, NJ;

The United States represents the largest, most dynamic marketplace in the world and is an unparalleled location for business investment, innovation, and commercialization of new technologies. The USA offers tremendous resources and advantages for those who invest and manufacture goods here. Through SelectUSA, our nation works to encourage and facilitate business investment. To learn more about why the USA is the best country in the world to develop technology, manufacture products, and grow your business, visit SelectUSA.gov.



UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

CORRECTED NOTICE OF ALLOWANCE AND FEE(S) DUE

86379 7590 02/09/2018
Ingredion Incorporated
Suite C
10 FINDERNE AVENUE
BRIDGEWATER, NEW JERSEY 08807

EXAMINER

COHEN, STEFANIE J

ART UNIT

PAPER NUMBER

1732

DATE MAILED: 02/09/2018

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/135,106	04/21/2016	Hongxin JIANG	3459.WHL Cont	8300

TITLE OF INVENTION: COMPOSITIONS AND METHODS FOR PRODUCING STARCH WITH NOVEL FUNCTIONALITY

APPLN. TYPE	ENTITY STATUS	ISSUE FEE DUE	PUBLICATION FEE DUE	PREV. PAID ISSUE FEE	TOTAL FEE(S) DUE	DATE DUE
REGULAR	UNDISCOUNTED	\$1000	\$0.00	\$0.00	\$1000	05/09/2018

THE APPLICATION IDENTIFIED ABOVE HAS BEEN EXAMINED AND IS ALLOWED FOR ISSUANCE AS A PATENT. PROSECUTION ON THE MERITS IS CLOSED. THIS NOTICE OF ALLOWANCE IS NOT A GRANT OF PATENT RIGHTS. THIS APPLICATION IS SUBJECT TO WITHDRAWAL FROM ISSUE AT THE INITIATIVE OF THE OFFICE OR UPON PETITION BY THE APPLICANT. SEE 37 CFR 1.313 AND MPEP 1308.

THE ISSUE FEE AND PUBLICATION FEE (IF REQUIRED) MUST BE PAID WITHIN THREE MONTHS FROM THE MAILING DATE OF THIS NOTICE OR THIS APPLICATION SHALL BE REGARDED AS ABANDONED. THIS STATUTORY PERIOD CANNOT BE EXTENDED. SEE 35 U.S.C. 151. THE ISSUE FEE DUE INDICATED ABOVE DOES NOT REFLECT A CREDIT FOR ANY PREVIOUSLY PAID ISSUE FEE IN THIS APPLICATION. IF AN ISSUE FEE HAS PREVIOUSLY BEEN PAID IN THIS APPLICATION (AS SHOWN ABOVE), THE RETURN OF PART B OF THIS FORM WILL BE CONSIDERED A REQUEST TO REAPPLY THE PREVIOUSLY PAID ISSUE FEE TOWARD THE ISSUE FEE NOW DUE.

HOW TO REPLY TO THIS NOTICE:

I. Review the ENTITY STATUS shown above. If the ENTITY STATUS is shown as SMALL or MICRO, verify whether entitlement to that entity status still applies.

If the ENTITY STATUS is the same as shown above, pay the TOTAL FEE(S) DUE shown above.

If the ENTITY STATUS is changed from that shown above, on PART B - FEE(S) TRANSMITTAL, complete section number 5 titled "Change in Entity Status (from status indicated above)".

For purposes of this notice, small entity fees are 1/2 the amount of undiscounted fees, and micro entity fees are 1/2 the amount of small entity fees.

II. PART B - FEE(S) TRANSMITTAL, or its equivalent, must be completed and returned to the United States Patent and Trademark Office (USPTO) with your ISSUE FEE and PUBLICATION FEE (if required). If you are charging the fee(s) to your deposit account, section "4b" of Part B - Fee(s) Transmittal should be completed and an extra copy of the form should be submitted. If an equivalent of Part B is filed, a request to reapply a previously paid issue fee must be clearly made, and delays in processing may occur due to the difficulty in recognizing the paper as an equivalent of Part B.

III. All communications regarding this application must give the application number. Please direct all communications prior to issuance to Mail Stop ISSUE FEE unless advised to the contrary.

IMPORTANT REMINDER: Utility patents issuing on applications filed on or after Dec. 12, 1980 may require payment of maintenance fees. It is patentee's responsibility to ensure timely payment of maintenance fees when due.

PART B - FEE(S) TRANSMITTAL

Complete and send this form, together with applicable fee(s), to: **Mail** **Mail Stop ISSUE FEE**
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450
or Fax (571)-273-2885

INSTRUCTIONS: This form should be used for transmitting the ISSUE FEE and PUBLICATION FEE (if required). Blocks 1 through 5 should be completed where appropriate. All further correspondence including the Patent, advance orders and notification of maintenance fees will be mailed to the current correspondence address as indicated unless corrected below or directed otherwise in Block 1, by (a) specifying a new correspondence address; and/or (b) indicating a separate "FEE ADDRESS" for maintenance fee notifications.

CURRENT CORRESPONDENCE ADDRESS (Note: Use Block 1 for any change of address)

Note: A certificate of mailing can only be used for domestic mailings of the Fee(s) Transmittal. This certificate cannot be used for any other accompanying papers. Each additional paper, such as an assignment or formal drawing, must have its own certificate of mailing or transmission.

86379 7590 02/09/2018
Ingredion Incorporated
Suite C
10 FINDERNE AVENUE
BRIDGEWATER, NEW JERSEY 08807

Certificate of Mailing or Transmission

I hereby certify that this Fee(s) Transmittal is being deposited with the United States Postal Service with sufficient postage for first class mail in an envelope addressed to the Mail Stop ISSUE FEE address above, or being facsimile transmitted to the USPTO (571) 273-2885, on the date indicated below.

(Depositor's name)
(Signature)
(Date)

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/135,106	04/21/2016	Hongxin JIANG	3459.WHL Cont	8300

TITLE OF INVENTION: COMPOSITIONS AND METHODS FOR PRODUCING STARCH WITH NOVEL FUNCTIONALITY

APPLN. TYPE	ENTITY STATUS	ISSUE FEE DUE	PUBLICATION FEE DUE	PREV. PAID ISSUE FEE	TOTAL FEE(S) DUE	DATE DUE
REGULAR	UNDISCOUNTED	\$1000	\$0.00	\$0.00	\$1000	05/09/2018

EXAMINER	ART UNIT	CLASS-SUBCLASS
COHEN, STEFANIE J	1732	127-037000

1. Change of correspondence address or indication of "Fee Address" (37 CFR 1.363). ☐ Change of correspondence address (or Change of Correspondence Address form PTO/SB/122) attached. ☐ "Fee Address" indication (or "Fee Address" Indication form PTO/SB/47; Rev 03-02 or more recent) attached. Use of a Customer Number is required.	2. For printing on the patent front page, list (1) The names of up to 3 registered patent attorneys or agents OR, alternatively, (2) The name of a single firm (having as a member a registered attorney or agent) and the names of up to 2 registered patent attorneys or agents. If no name is listed, no name will be printed. 1 _____ 2 _____ 3 _____
---	---

3. ASSIGNEE NAME AND RESIDENCE DATA TO BE PRINTED ON THE PATENT (print or type)

PLEASE NOTE: Unless an assignee is identified below, no assignee data will appear on the patent. If an assignee is identified below, the document has been filed for recordation as set forth in 37 CFR 3.11. Completion of this form is NOT a substitute for filing an assignment.

(A) NAME OF ASSIGNEE (B) RESIDENCE: (CITY and STATE OR COUNTRY)

Please check the appropriate assignee category or categories (will not be printed on the patent) : ☐ Individual ☐ Corporation or other private group entity ☐ Government

4a. The following fee(s) are submitted: ☐ Issue Fee ☐ Publication Fee (No small entity discount permitted) ☐ Advance Order - # of Copies _____	4b. Payment of Fee(s): (Please first reapply any previously paid issue fee shown above) ☐ A check is enclosed. ☐ Payment by credit card. Form PTO-2038 is attached. ☐ The director is hereby authorized to charge the required fee(s), any deficiency, or credits any overpayment, to Deposit Account Number _____ (enclose an extra copy of this form).
---	---

5. Change in Entity Status (from status indicated above)

- ☐ Applicant certifying micro entity status. See 37 CFR 1.29
- ☐ Applicant asserting small entity status. See 37 CFR 1.27
- ☐ Applicant changing to regular undiscounted fee status.

NOTE: Absent a valid certification of Micro Entity Status (see forms PTO/SB/15A and 15B), issue fee payment in the micro entity amount will not be accepted at the risk of application abandonment.
NOTE: If the application was previously under micro entity status, checking this box will be taken to be a notification of loss of entitlement to micro entity status.
NOTE: Checking this box will be taken to be a notification of loss of entitlement to small or micro entity status, as applicable.

NOTE: This form must be signed in accordance with 37 CFR 1.31 and 1.33. See 37 CFR 1.4 for signature requirements and certifications.

Authorized Signature _____ Date _____

Typed or printed name _____ Registration No. _____



UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/135,106	04/21/2016	Hongxin JIANG	3459.WHL Cont	8300

86379 7590 02/09/2018

Ingredion Incorporated
Suite C
10 Finderne Avenue
Bridgewater, NEW JERSEY 08807

EXAMINER

COHEN, STEFANIE J

ART UNIT	PAPER NUMBER
----------	--------------

1732

DATE MAILED: 02/09/2018

OMB Clearance and PRA Burden Statement for PTOL-85 Part B

The Paperwork Reduction Act (PRA) of 1995 requires Federal agencies to obtain Office of Management and Budget approval before requesting most types of information from the public. When OMB approves an agency request to collect information from the public, OMB (i) provides a valid OMB Control Number and expiration date for the agency to display on the instrument that will be used to collect the information and (ii) requires the agency to inform the public about the OMB Control Number's legal significance in accordance with 5 CFR 1320.5(b).

The information collected by PTOL-85 Part B is required by 37 CFR 1.311. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, Virginia 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313-1450. Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.



UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

NOTICE OF ALLOWANCE AND FEE(S) DUE

86379 7590 01/16/2018
Ingredion Incorporated
Suite C
10 Finderne Avenue
Bridgewater, NJ 08807

EXAMINER

COHEN, STEFANIE J

ART UNIT

PAPER NUMBER

1732

DATE MAILED: 01/16/2018

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
-----------------	-------------	----------------------	---------------------	------------------

15/135,106

04/21/2016

Hongxin JIANG

3459.WHL Cont

8300

TITLE OF INVENTION: COMPOSITIONS AND METHODS FOR PRODUCING STARCH WITH NOVEL FUNCTIONALITY

APPLN. TYPE	ENTITY STATUS	ISSUE FEE DUE	PUBLICATION FEE DUE	PREV. PAID ISSUE FEE	TOTAL FEE(S) DUE	DATE DUE
nonprovisional	UNDISCOUNTED	\$960	\$0	\$0	\$960	04/16/2018

THE APPLICATION IDENTIFIED ABOVE HAS BEEN EXAMINED AND IS ALLOWED FOR ISSUANCE AS A PATENT. PROSECUTION ON THE MERITS IS CLOSED. THIS NOTICE OF ALLOWANCE IS NOT A GRANT OF PATENT RIGHTS. THIS APPLICATION IS SUBJECT TO WITHDRAWAL FROM ISSUE AT THE INITIATIVE OF THE OFFICE OR UPON PETITION BY THE APPLICANT. SEE 37 CFR 1.313 AND MPEP 1308.

THE ISSUE FEE AND PUBLICATION FEE (IF REQUIRED) MUST BE PAID WITHIN THREE MONTHS FROM THE MAILING DATE OF THIS NOTICE OR THIS APPLICATION SHALL BE REGARDED AS ABANDONED. THIS STATUTORY PERIOD CANNOT BE EXTENDED. SEE 35 U.S.C. 151. THE ISSUE FEE DUE INDICATED ABOVE DOES NOT REFLECT A CREDIT FOR ANY PREVIOUSLY PAID ISSUE FEE IN THIS APPLICATION. IF AN ISSUE FEE HAS PREVIOUSLY BEEN PAID IN THIS APPLICATION (AS SHOWN ABOVE), THE RETURN OF PART B OF THIS FORM WILL BE CONSIDERED A REQUEST TO REAPPLY THE PREVIOUSLY PAID ISSUE FEE TOWARD THE ISSUE FEE NOW DUE.

HOW TO REPLY TO THIS NOTICE:

I. Review the ENTITY STATUS shown above. If the ENTITY STATUS is shown as SMALL or MICRO, verify whether entitlement to that entity status still applies.

If the ENTITY STATUS is the same as shown above, pay the TOTAL FEE(S) DUE shown above.

If the ENTITY STATUS is changed from that shown above, on PART B - FEE(S) TRANSMITTAL, complete section number 5 titled "Change in Entity Status (from status indicated above)".

For purposes of this notice, small entity fees are 1/2 the amount of undiscounted fees, and micro entity fees are 1/2 the amount of small entity fees.

II. PART B - FEE(S) TRANSMITTAL, or its equivalent, must be completed and returned to the United States Patent and Trademark Office (USPTO) with your ISSUE FEE and PUBLICATION FEE (if required). If you are charging the fee(s) to your deposit account, section "4b" of Part B - Fee(s) Transmittal should be completed and an extra copy of the form should be submitted. If an equivalent of Part B is filed, a request to reapply a previously paid issue fee must be clearly made, and delays in processing may occur due to the difficulty in recognizing the paper as an equivalent of Part B.

III. All communications regarding this application must give the application number. Please direct all communications prior to issuance to Mail Stop ISSUE FEE unless advised to the contrary.

IMPORTANT REMINDER: Maintenance fees are due in utility patents issuing on applications filed on or after Dec. 12, 1980. It is patentee's responsibility to ensure timely payment of maintenance fees when due. More information is available at www.uspto.gov/PatentMaintenanceFees.

PART B - FEE(S) TRANSMITTAL

**Complete and send this form, together with applicable fee(s), to: Mail Mail Stop ISSUE FEE
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450
or Fax (571)-273-2885**

INSTRUCTIONS: This form should be used for transmitting the ISSUE FEE and PUBLICATION FEE (if required). Blocks 1 through 5 should be completed where appropriate. All further correspondence including the Patent, advance orders and notification of maintenance fees will be mailed to the current correspondence address as indicated unless corrected below or directed otherwise in Block 1, by (a) specifying a new correspondence address; and/or (b) indicating a separate "FEE ADDRESS" for maintenance fee notifications.

CURRENT CORRESPONDENCE ADDRESS (Note: Use Block 1 for any change of address)

Note: A certificate of mailing can only be used for domestic mailings of the Fee(s) Transmittal. This certificate cannot be used for any other accompanying papers. Each additional paper, such as an assignment or formal drawing, must have its own certificate of mailing or transmission.

86379 7590 01/16/2018
Ingredion Incorporated
Suite C
10 Finderne Avenue
Bridgewater, NJ 08807

Certificate of Mailing or Transmission

I hereby certify that this Fee(s) Transmittal is being deposited with the United States Postal Service with sufficient postage for first class mail in an envelope addressed to the Mail Stop ISSUE FEE address above, or being facsimile transmitted to the USPTO (571) 273-2885, on the date indicated below.

(Depositor's name)
(Signature)
(Date)

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/135,106	04/21/2016	Hongxin JIANG	3459.WHL Cont	8300

TITLE OF INVENTION: COMPOSITIONS AND METHODS FOR PRODUCING STARCH WITH NOVEL FUNCTIONALITY

APPLN. TYPE	ENTITY STATUS	ISSUE FEE DUE	PUBLICATION FEE DUE	PREV. PAID ISSUE FEE	TOTAL FEE(S) DUE	DATE DUE
nonprovisional	UNDISCOUNTED	\$960	\$0	\$0	\$960	04/16/2018

EXAMINER	ART UNIT	CLASS-SUBCLASS
COHEN, STEFANIE J	1732	127-037000

1. Change of correspondence address or indication of "Fee Address" (37 CFR 1.363). ☐ Change of correspondence address (or Change of Correspondence Address form PTO/SB/122) attached. ☐ "Fee Address" indication (or "Fee Address" Indication form PTO/SB/47; Rev 03-02 or more recent) attached. Use of a Customer Number is required.	2. For printing on the patent front page, list (1) The names of up to 3 registered patent attorneys or agents OR, alternatively, 1 _____ (2) The name of a single firm (having as a member a registered attorney or agent) and the names of up to 2 registered patent attorneys or agents. If no name is listed, no name will be printed. 2 _____ 3 _____
---	---

3. ASSIGNEE NAME AND RESIDENCE DATA TO BE PRINTED ON THE PATENT (print or type)

PLEASE NOTE: Unless an assignee is identified below, no assignee data will appear on the patent. If an assignee is identified below, the document has been filed for recordation as set forth in 37 CFR 3.11. Completion of this form is NOT a substitute for filing an assignment.

(A) NAME OF ASSIGNEE (B) RESIDENCE: (CITY and STATE OR COUNTRY)

Please check the appropriate assignee category or categories (will not be printed on the patent) : ☐ Individual ☐ Corporation or other private group entity ☐ Government

4a. The following fee(s) are submitted: ☐ Issue Fee ☐ Publication Fee (No small entity discount permitted) ☐ Advance Order - # of Copies _____	4b. Payment of Fee(s): (Please first reapply any previously paid issue fee shown above) ☐ A check is enclosed. ☐ Payment by credit card. Form PTO-2038 is attached. ☐ The director is hereby authorized to charge the required fee(s), any deficiency, or credits any overpayment, to Deposit Account Number _____ (enclose an extra copy of this form).
---	---

5. Change in Entity Status (from status indicated above)

☐ Applicant certifying micro entity status. See 37 CFR 1.29

☐ Applicant asserting small entity status. See 37 CFR 1.27

☐ Applicant changing to regular undiscounted fee status.

NOTE: Absent a valid certification of Micro Entity Status (see forms PTO/SB/15A and 15B), issue fee payment in the micro entity amount will not be accepted at the risk of application abandonment.

NOTE: If the application was previously under micro entity status, checking this box will be taken to be a notification of loss of entitlement to micro entity status.

NOTE: Checking this box will be taken to be a notification of loss of entitlement to small or micro entity status, as applicable.

NOTE: This form must be signed in accordance with 37 CFR 1.31 and 1.33. See 37 CFR 1.4 for signature requirements and certifications.

Authorized Signature _____ Date _____

Typed or printed name _____ Registration No. _____



UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/135,106	04/21/2016	Hongxin JIANG	3459.WHL Cont	8300
86379	7590	01/16/2018	EXAMINER	
Ingredion Incorporated Suite C 10 FINDERNE AVENUE BRIDGEWATER, NJ 08807			COHEN, STEFANIE J	
			ART UNIT	PAPER NUMBER
			1732	

DATE MAILED: 01/16/2018

Determination of Patent Term Adjustment under 35 U.S.C. 154 (b) (Applications filed on or after May 29, 2000)

The Office has discontinued providing a Patent Term Adjustment (PTA) calculation with the Notice of Allowance.

Section 1(h)(2) of the AIA Technical Corrections Act amended 35 U.S.C. 154(b)(3)(B)(i) to eliminate the requirement that the Office provide a patent term adjustment determination with the notice of allowance. See Revisions to Patent Term Adjustment, 78 Fed. Reg. 19416, 19417 (Apr. 1, 2013). Therefore, the Office is no longer providing an initial patent term adjustment determination with the notice of allowance. The Office will continue to provide a patent term adjustment determination with the Issue Notification Letter that is mailed to applicant approximately three weeks prior to the issue date of the patent, and will include the patent term adjustment on the patent. Any request for reconsideration of the patent term adjustment determination (or reinstatement of patent term adjustment) should follow the process outlined in 37 CFR 1.705.

Any questions regarding the Patent Term Extension or Adjustment determination should be directed to the Office of Patent Legal Administration at (571)-272-7702. Questions relating to issue and publication fee payments should be directed to the Customer Service Center of the Office of Patent Publication at 1-(888)-786-0101 or (571)-272-4200.

OMB Clearance and PRA Burden Statement for PTOL-85 Part B

The Paperwork Reduction Act (PRA) of 1995 requires Federal agencies to obtain Office of Management and Budget approval before requesting most types of information from the public. When OMB approves an agency request to collect information from the public, OMB (i) provides a valid OMB Control Number and expiration date for the agency to display on the instrument that will be used to collect the information and (ii) requires the agency to inform the public about the OMB Control Number's legal significance in accordance with 5 CFR 1320.5(b).

The information collected by PTOL-85 Part B is required by 37 CFR 1.311. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 12 minutes to complete, including gathering, preparing, and submitting the completed application form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, Virginia 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, Virginia 22313-1450. Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it displays a valid OMB control number.

Privacy Act Statement

The Privacy Act of 1974 (P.L. 93-579) requires that you be given certain information in connection with your submission of the attached form related to a patent application or patent. Accordingly, pursuant to the requirements of the Act, please be advised that: (1) the general authority for the collection of this information is 35 U.S.C. 2(b)(2); (2) furnishing of the information solicited is voluntary; and (3) the principal purpose for which the information is used by the U.S. Patent and Trademark Office is to process and/or examine your submission related to a patent application or patent. If you do not furnish the requested information, the U.S. Patent and Trademark Office may not be able to process and/or examine your submission, which may result in termination of proceedings or abandonment of the application or expiration of the patent.

The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether disclosure of these records is required by the Freedom of Information Act.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
3. A record in this system of records may be disclosed, as a routine use, to a Member of Congress submitting a request involving an individual, to whom the record pertains, when the individual has requested assistance from the Member with respect to the subject matter of the record.
4. A record in this system of records may be disclosed, as a routine use, to a contractor of the Agency having need for the information in order to perform a contract. Recipients of information shall be required to comply with the requirements of the Privacy Act of 1974, as amended, pursuant to 5 U.S.C. 552a(m).
5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspection or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

Notice of Allowability	Application No. 15/135,106	Applicant(s) JIANG ET AL.	
	Examiner STEFANIE COHEN	Art Unit 1732	AIA (First Inventor to File) Status Yes

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address--

All claims being allowable, PROSECUTION ON THE MERITS IS (OR REMAINS) CLOSED in this application. If not included herewith (or previously mailed), a Notice of Allowance (PTOL-85) or other appropriate communication will be mailed in due course. **THIS NOTICE OF ALLOWABILITY IS NOT A GRANT OF PATENT RIGHTS.** This application is subject to withdrawal from issue at the initiative of the Office or upon petition by the applicant. See 37 CFR 1.313 and MPEP 1308.

1. ☒ This communication is responsive to 10/3/17.
☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on _____.
2. ☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on _____; the restriction requirement and election have been incorporated into this action.
3. ☒ The allowed claim(s) is/are 1-15. As a result of the allowed claim(s), you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to PPHfeedback@uspto.gov.
4. ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

- a) ☐ All b) ☐ Some *c) ☐ None of the:
1. ☐ Certified copies of the priority documents have been received.
 2. ☐ Certified copies of the priority documents have been received in Application No. _____.
 3. ☐ Copies of the certified copies of the priority documents have been received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

* Certified copies not received: _____.

Applicant has THREE MONTHS FROM THE "MAILING DATE" of this communication to file a reply complying with the requirements noted below. Failure to timely comply will result in ABANDONMENT of this application.

THIS THREE-MONTH PERIOD IS NOT EXTENDABLE.

5. ☐ CORRECTED DRAWINGS (as "replacement sheets") must be submitted.
☐ including changes required by the attached Examiner's Amendment / Comment or in the Office action of Paper No./Mail Date _____.
Identifying indicia such as the application number (see 37 CFR 1.84(c)) should be written on the drawings in the front (not the back) of each sheet. Replacement sheet(s) should be labeled as such in the header according to 37 CFR 1.121(d).
6. ☐ DEPOSIT OF and/or INFORMATION about the deposit of BIOLOGICAL MATERIAL must be submitted. Note the attached Examiner's comment regarding REQUIREMENT FOR THE DEPOSIT OF BIOLOGICAL MATERIAL.

Attachment(s)

- | | |
|---|--|
| 1. ☒ Notice of References Cited (PTO-892) | 5. ☐ Examiner's Amendment/Comment |
| 2. ☒ Information Disclosure Statements (PTO/SB/08),
Paper No./Mail Date <u>8/8/17, 7/21/16</u> | 6. ☒ Examiner's Statement of Reasons for Allowance |
| 3. ☐ Examiner's Comment Regarding Requirement for Deposit
of Biological Material | 7. ☐ Other _____. |
| 4. ☐ Interview Summary (PTO-413),
Paper No./Mail Date _____. | |

/MELVIN C MAYES/
Supervisory Patent Examiner, Art Unit 1732

Election/Restrictions

This application is in condition for allowance except for the presence of claims 15-33 directed to inventions non-elected without traverse. Accordingly, claims 15-33 been cancelled.

REASONS FOR ALLOWANCE

The following is an examiner's statement of reasons for allowance:

US4767849 teaches a starch of the wxsh1 genotype. Table 1 teaches extraction of the starch from a wxsh1 maize kernel. Sample A has an amylose content of .6% and Sample B has an amylose content of .8% which is outside the claimed amylose range claimed in claim 1.

US4615888 teaches bread containing wxsu2 genotype starch but does not teach an amylose content.

Varagona et al (cited on the EP32289635 search report) teaches an alternative splicing induced by insertion of retrotransposons into the maize waxy gene but does not teach an amylose content.

WO03033540 (cited on the EP32289635 search report, corresponding USPGPUB 20060216402) teaches starch from maize plants and/or other plants which

Art Unit: 1732

produce starch storing organs which contain low amylose starch which has an amylose content between 1.5% and 15%.

US20060216402 teaches the starch of the present invention therefore may be viewed as a reduced amylose starch or a special type of waxy starch with new elastic properties. The starch of the present invention is referred to herein as waxy-E or wx-E starch, to emphasize this elastic property not previously available with known waxy starch.

Paragraph 220 of the PGPUB teaches all of the waxy-E starches and waxy starches had a significantly higher peak viscosity and higher final viscosity than the normal starches, indicating that all of the waxy and waxy-E starches excel in the development of viscosity at relatively low starch concentrations. Additionally, all waxy-E and waxy starches had a pasting temperature lower than that of the normal starches, indicating that all of these starches begin to build viscosity earlier than do the normal starches.

Tables 1 and 2 teaches numerous dominant mutant alleles producing an active GBSS protein (Table 1) and recessive mutant alleles producing waxy starch (Table 2) exist.

Although this reference teaches a wx-E allele, this reference does not teach a wxS allele. Further, this reference does not teach a starch derived from a waxy sugary 2 corn plant comprising at least one wxS allele in the endosperm having an aqueous starch pasting temperature that is at least about 5% greater than the starch pasting temperature on a control starch.

Wessler teaches a molecular basis of mutations at the waxy locus of maize.

Wessler, table 1, teaches a wx-stonor allele.

Although this reference shows studies using the wx-stonor allele, this reference does not teach a starch derived from a waxy sugary 2 corn plant comprising at least one wxS allele in the endosperm having an aqueous starch pasting temperature that is at least about 5% greater than the starch pasting temperature on a control starch and further does not teach an amylose content of the starch.

Any comments considered necessary by applicant must be submitted no later than the payment of the issue fee and, to avoid processing delays, should preferably accompany the issue fee. Such submissions should be clearly labeled "Comments on Statement of Reasons for Allowance."

Any inquiry concerning this communication or earlier communications from the examiner should be directed to STEFANIE COHEN whose telephone number is (571)270-5836. The examiner can normally be reached on Monday through Thursday 9:30am-6:00pm.

Examiner interviews are available via telephone, in-person, and video conferencing using a USPTO supplied web-based collaboration tool. To schedule an interview, applicant is encouraged to use the USPTO Automated Interview Request (AIR) at <http://www.uspto.gov/interviewpractice>.

Art Unit: 1732

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Melvin Curtis Mayes can be reached on 5712721234. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov>. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

1/4/17

/STEFANIE COHEN/
Examiner, Art Unit 1732

/MELVIN C MAYES/
Supervisory Patent Examiner, Art Unit 1732



US00988641B2

(12) United States Patent
Jiang et al.**(10) Patent No.: US 9,988,641 B2**
(45) Date of Patent: Jun. 5, 2018**(54) COMPOSITIONS AND METHODS FOR
PRODUCING STARCH WITH NOVEL
FUNCTIONALITY****(58) Field of Classification Search**CPC C12N 15/8245
See application file for complete search history.**(71) Applicant: Corn Products Development, Inc.,** Sao Paulo (BR)**(56) References Cited**

U.S. PATENT DOCUMENTS

4,428,972 A 1/1984 Wurzburg et al.
4,615,888 A * 10/1986 Zallie A21D 2/186
426/21

(Continued)

FOREIGN PATENT DOCUMENTS

WO 2003033540 A2 4/2003
WO WO 03033540 A3 * 10/2003 C08B 30/00

OTHER PUBLICATIONS

Wessler et al, Molecular basis of mutations at the waxy locus of maize: correlation with the fine structure genetic map, 1985, Proc. natl. acad. sci. usa, vol. 82, pp. 4177-4181.*

(Continued)

Primary Examiner — Melvin C Mayes*Assistant Examiner* — Stefanie Cohen**(74) Attorney, Agent, or Firm** — Jason Grauch; Jacqueline Cohen**(*) Notice:** Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 12 days.**(21) Appl. No.: 15/135,106****(22) Filed: Apr. 21, 2016****(65) Prior Publication Data**

US 2017/0283818 A1 Oct. 5, 2017

Related U.S. Application Data**(63)** Continuation of application No. 15/090,926, filed on Apr. 5, 2016, now abandoned.**(51) Int. Cl.****C12N 15/00** (2006.01)
C12N 15/82 (2006.01)
A01H 5/10 (2018.01)
A01H 1/02 (2006.01)
C12N 9/10 (2006.01)
A01H 1/04 (2006.01)
A01G 1/00 (2006.01)
C08B 30/04 (2006.01)**(52) U.S. Cl.**CPC **C12N 15/8245** (2013.01); **A01G 1/001** (2013.01); **A01H 1/02** (2013.01);

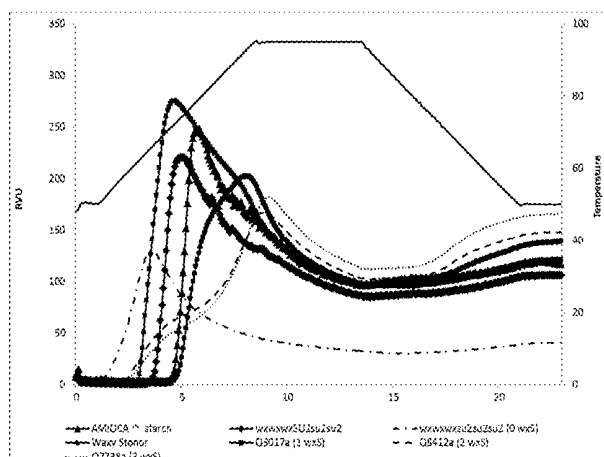
(Continued)

(57) ABSTRACT

This invention disclosure relates to novel maize starch. The starch can be made from the newly developed waxy sugary-2 double-mutant maize that has low activity of Granule Bound Starch Synthase I (GBSSI), which results in low amylose level. The starch from newly developed waxy sugary-2 double-mutant is freeze-thaw stable and has high viscosity. In comparison with the starch of the existing waxy sugary-2 double-mutant maize, the new waxy sugary-2 double-mutant maize starch showed, inter alia, improved pasting profile, starch granule integrity, larger starch granule size, and higher viscosity.

15 Claims, 7 Drawing Sheets

Starch pasting profiles of waxy corn starch samples.



(52) U.S. Cl.

CPC *A01H 1/04* (2013.01); *A01H 5/10*
(2013.01); *C08B 30/044* (2013.01); *C12N*
9/1051 (2013.01); *C12N 15/8241* (2013.01);
C12Y 204/01021 (2013.01); *C12Y 204/01242*
(2013.01)

(56) References Cited

U.S. PATENT DOCUMENTS

4,767,849 A * 8/1988 Friedman A23C 9/1544
127/32
5,648,111 A * 7/1997 Pearlstein A01H 5/10
127/32
5,954,883 A 9/1999 Nagle et al.
6,827,474 B2 12/2004 Clapper
6,960,703 B2 11/2005 Nagle et al.
2006/0216402 A1 * 9/2006 Klucinec C08B 30/00
426/660

OTHER PUBLICATIONS

Varagona, Alternative Splicing Induced by Insertion of
Retrotransposons into the Maize waxy Gene, 1992, The plant cell,
vol. 4, pp. 811-820.*

Marguerite J. Varagona, Altrnative Splicing Induced by Insertion of
Retrotransposons Into The Maize Waxy Gene, The Plant Cell, 1992,
pp. 811-820, vol. 4.

* cited by examiner

FIG. 1
Starch pasting profiles of waxy corn starch samples.

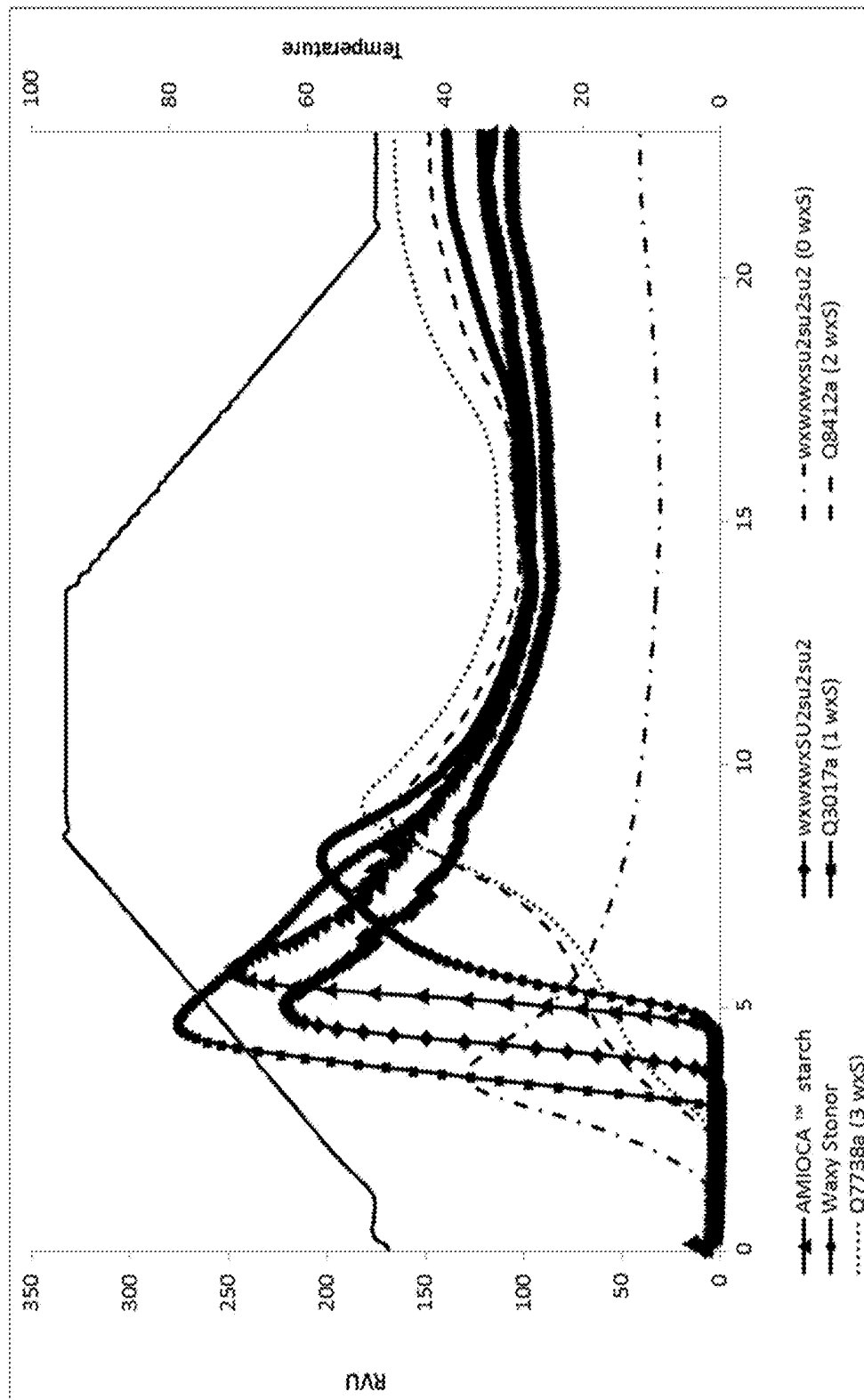


FIG. 2
Starch pasting properties of waxy corn starch samples.

Sample	Pasting Temp. (°C)	Peak (RVU)	Hold (RVU)	Final (RVU)	Break Down (RVU)	Setback (RVU)
Q3017a (1 wxS)	62.0	275.7	95.3	121.3	180.4	26.0
Q8412a (2 wxS)	59.1	167.8	102.1	148.0	65.7	45.9
Q7738a (3 wxS)	59.5	182.2	111.9	165.6	70.2	53.7
<i>wxwxwxsu2su2su2</i> (0 wxS)	54.4	129.3	30.3	40.9	99.0	10.6
K5994a (waxy Stonor)	73.8	202.6	96.3	139.0	106.3	42.7
AMIOCA™ starch	72.2	248.2	96.3	116.8	151.9	20.5
<i>wxwxwxSU2su2su2</i> starch	67.5	220.9	84.9	106.1	136.0	21.2

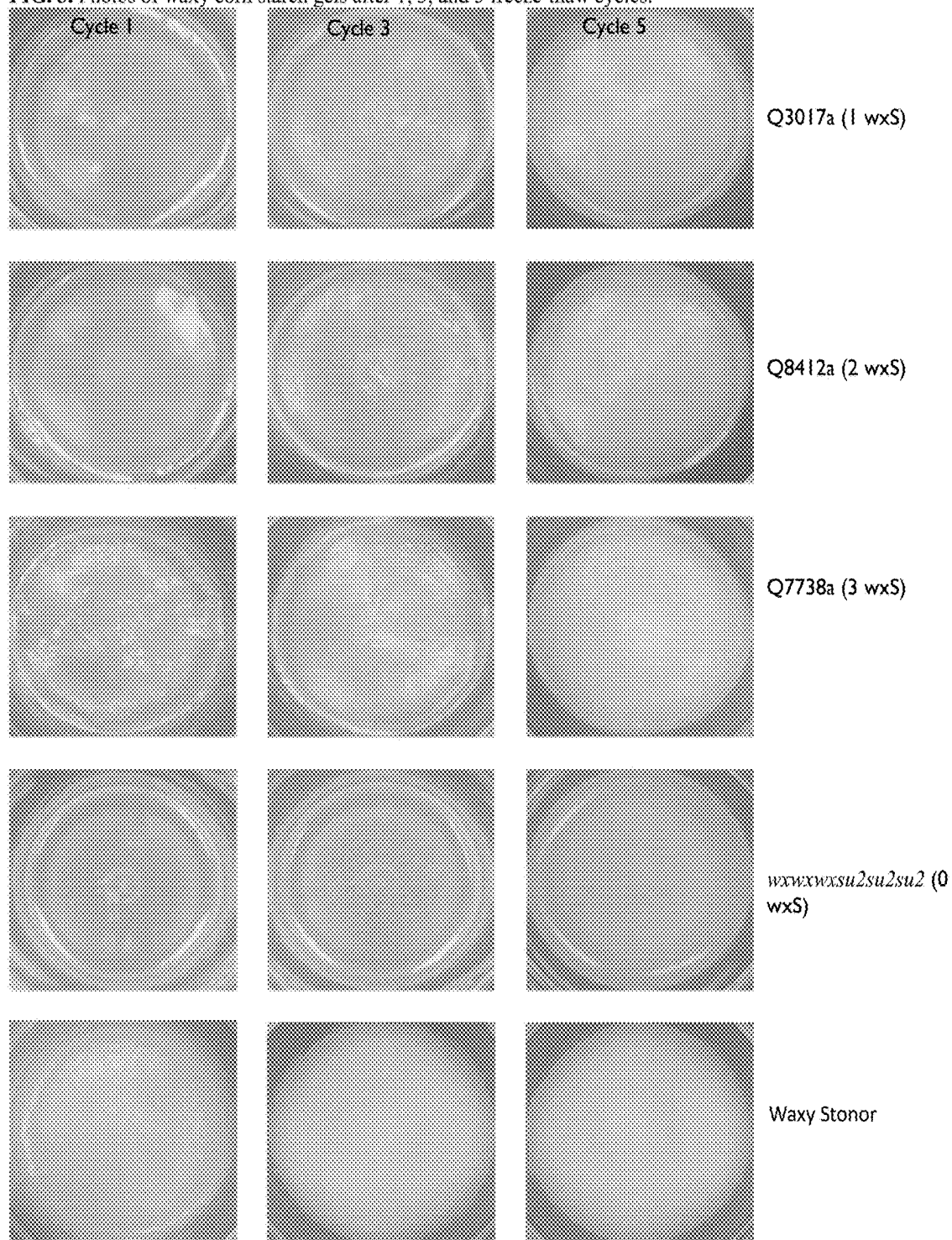
FIG. 3. Photos of waxy corn starch gels after 1, 3, and 5 freeze-thaw cycles.

FIG. 3 Continued

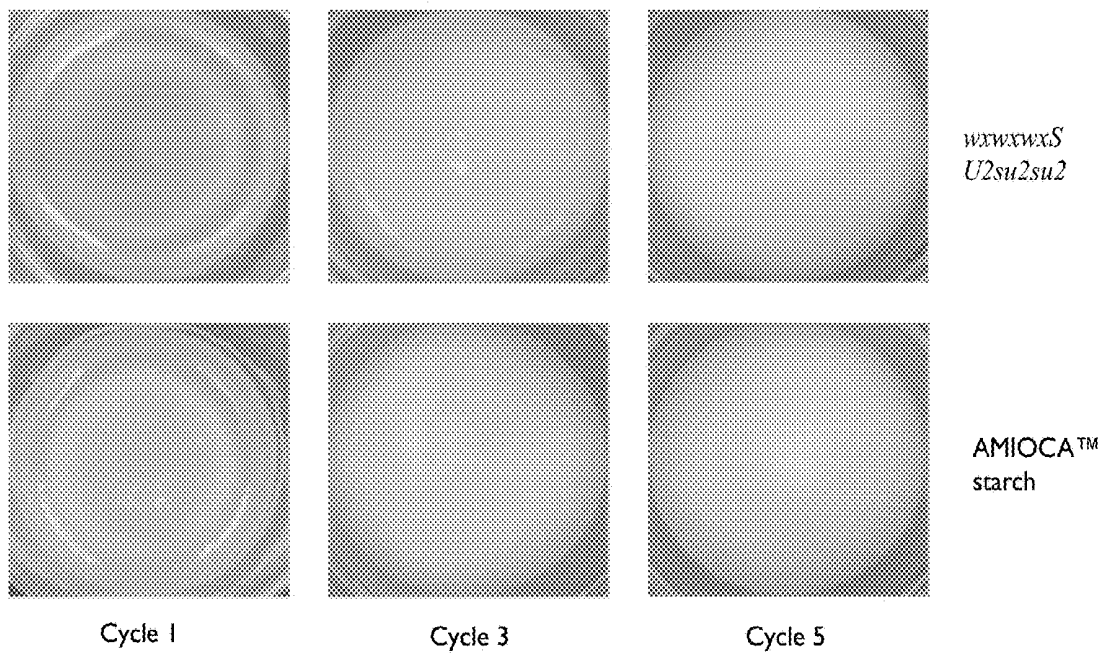


FIG. 4

Thermal Properties of waxy corn starch samples.

Sample	T _o (°C)	T _p (°C)	T _c (°C)	ΔH (J/g)
Q3017a (1 wxS)	57.2±0.0	62.5±0.3	69.3±0.2	12.9±0.0
Q8412a (2 wxS)	51.2±0.1	58.0±0.1	65.7±0.4	6.0±0.1
Q7738a (3 wxS)	51.7±0.2	58.2±0.4	66.8±0.3	8.0±0.1
wxwxwxsu2su2su2	52.2±0.2	58.1±0.2	65.6±0.0	4.5±0.0
K5994a (Waxy Stonor)	68.1	74.6	81.3	14.9
AMIOCA™ starch	66.7±0.5	73.5±0.3	80.6±0.6	16.6±0.2
wxwxwxSU2su2su2 starch	60.8±0.1	67.8±0.1	77.6±0.2	15.4±0.0

FIG. 5
Granule size distributions of waxy corn starch samples.

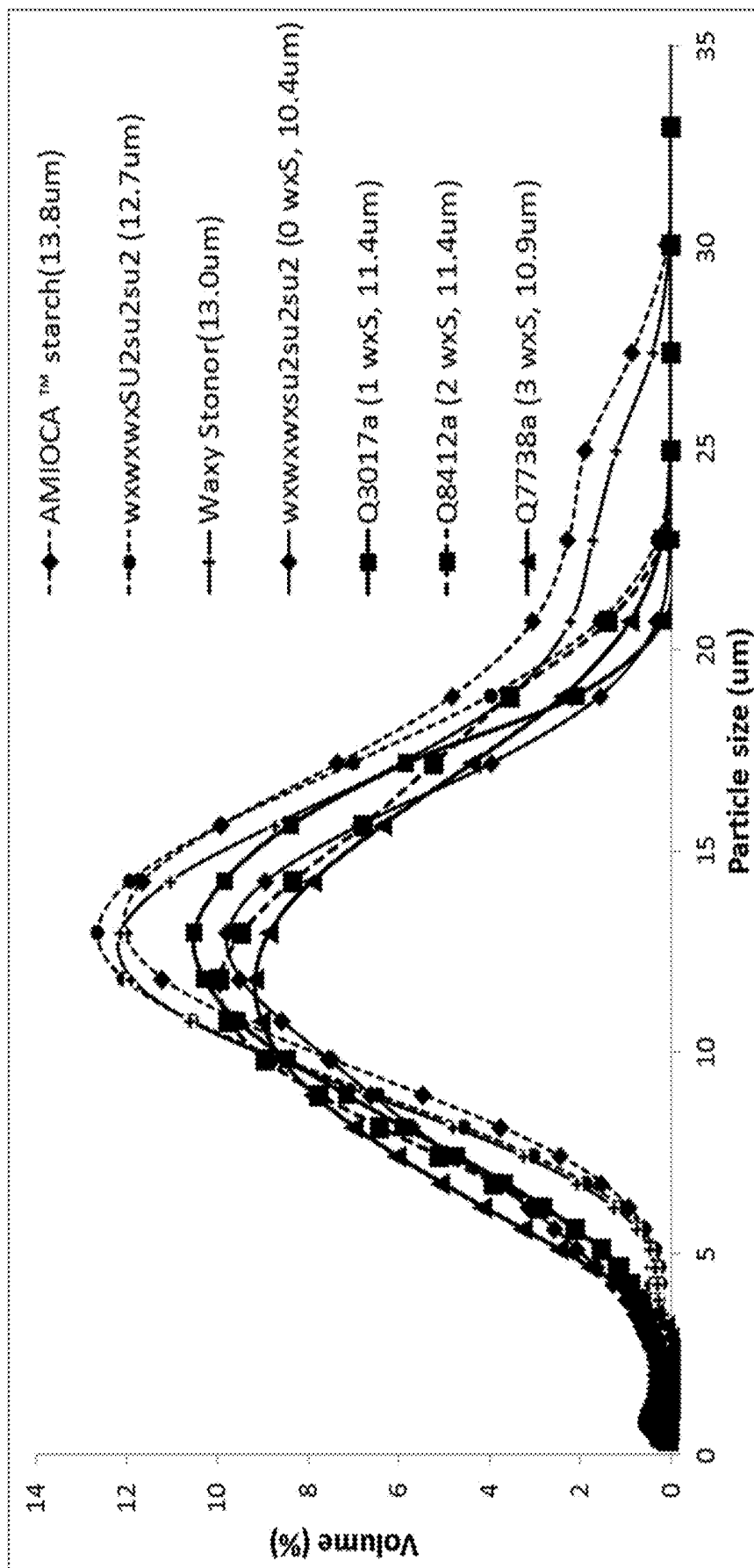
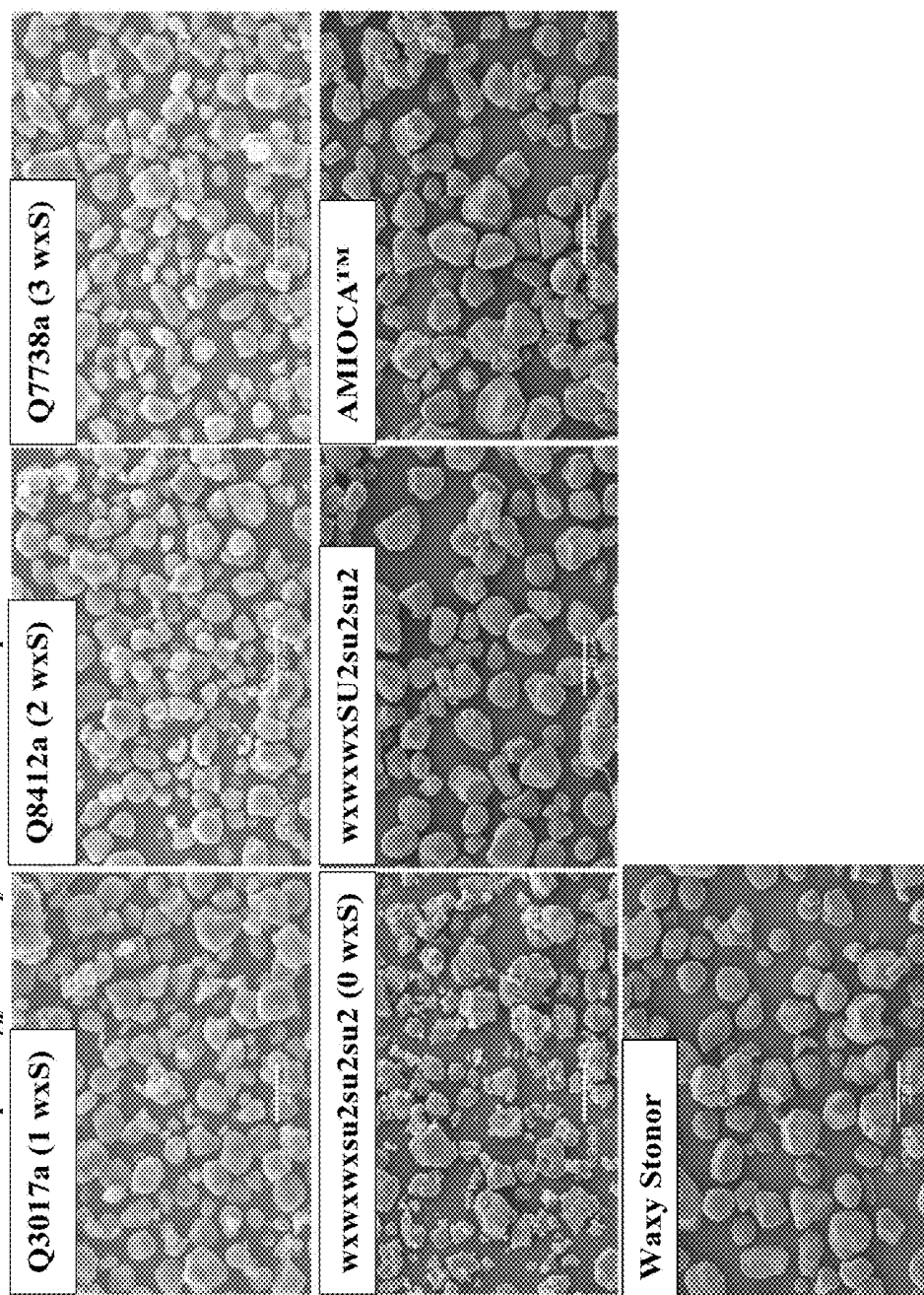


FIG. 6
Granule morphology of waxy corn starch samples.



COMPOSITIONS AND METHODS FOR PRODUCING STARCH WITH NOVEL FUNCTIONALITY

CROSS-REFERENCE TO RELATED APPLICATIONS

The present application is a Continuation of U.S. application Ser. No. 15/090,926, filed Apr. 5, 2016, the disclosure of which is herein incorporated by reference in its entirety for all purposes.

FIELD OF THE INVENTION

The present invention relates to maize starch derived from plants and grains, compositions and methods for producing such starch.

BACKGROUND OF THE INVENTION

The existing waxy sugary-2 double-mutant maize starch shows very good freeze/thaw stability. However, the viscosity of the existing waxy sugary-2 maize starch was substantially lower than commercial waxy maize starch, such as AMIOCA™ starch. In addition, the pasting temperature of the existing waxy sugary-2 double-mutant maize starch is substantially lower than AMIOCA™ starch. Therefore, there remains a need in the art for producing maize starch with good freeze/thaw stability, increased viscosity, and/or higher pasting temperature.

In this invention, we developed new maize starch with higher viscosity and/or higher pasting temperature than commercial waxy maize starch, such as AMIOCA™ starch, by introduction of low activity of GBSSI to the existing waxy sugary-2 double-mutant maize.

SUMMARY OF THE INVENTION

The present invention provides starch with novel functionality which has not been found in nature. In some embodiments, the starch comprises an amylose content of from about 2 wt % to about 20 wt % using an amylose/amylopectin assay kit, e.g., Megazyme®. In some embodiments, the starch comprises an aqueous starch pasting temperature that is at least about 5% greater than the starch pasting temperature of a control starch as measured using a Rapid Visco Analyzer (RVA, Newport Scientific, Sydney, Australia). In some embodiments, the starch is derived from a waxy-sugary 2 double mutant corn plant comprising at least one wx-Stonor (wxS) allele in the endosperm. In some embodiments, the waxy-sugary 2 double mutant corn plant comprises a recessive su2 mutant allele.

In some embodiments, the starch of the present invention is produced from plants which are not transgenic (i.e., non-GMO) for the genotype that produces the disclosed and claimed starch.

In some embodiments, the amylose content of the starch is from about 8 wt % to about 15 wt %.

In some embodiments, the aqueous pasting temperature is from about 5% to about 15% greater than that of a control starch.

In some embodiments, the starch further comprises an enthalpy change of crystallinity. In some embodiments, the enthalpy change of crystallinity is at least about 30% greater than an enthalpy change of crystallinity of a control starch as measured using a differential scanning calorimeter (DSC). In some embodiments, the enthalpy change of crys-

tallinity is from about 30% to about 200% greater than an enthalpy change of crystallinity of a control starch as measured using a DSC.

In some embodiments, the starch further comprises a distribution of starch granule sizes, wherein the distribution of starch granule sizes comprises at least about 40% fewer granules less than 5 microns compared to a proportion of granules less than 5 microns from a control starch as observed using a scanning electron microscope.

In some embodiments, the starch has a granule which exhibits increased integrity compared to a starch derived from the control corn plant as observed using a scanning electron microscope.

In some embodiments, the starch is capable of withstanding at least three freeze-thaw cycles. In some embodiments, the starch is capable of withstanding at least three to at least five freeze-thaw cycles.

In some embodiments, a plant producing the starch of the present invention is generated by cross pollination of a first corn plant having the genotype of wxwxsu2su2, and a second corn plant having the genotype of wxSwxSsu2su2, or the genotype of wxSwxsu2su2. In some embodiments, the endosperm of the waxy-sugary 2 corn plant has one dose, two doses, or three doses of the wxS allele, and the endosperm has the genotype of wxSwwxwsu2su2su2, wxSwxSwxsu2su2su2, or wxSwxSwxSsu2su2su2.

The present invention also provides methods for producing the starch with an improved pasting profile. In some embodiments, the methods comprise introducing at least one wxS allele into the endosperm of a waxy corn plant.

The present invention also provides novel double-mutant plants. In some embodiments, the plants are corn plants. In some embodiments, the plants are homozygous recessive for the starch synthase IIa (su2) gene. In some embodiments, the plants are either homozygous or heterozygous for a mutated Granule-bound starch synthase I (GBSSI) gene. In some embodiments, the mutated GBSSI gene has less GBSSI activity as the wild type GBSSI gene (Wx), but has higher GBSSI activity than the recessive, loss-of-function GBSSI gene (wx).

In some embodiments, the mutated GBSSI gene has about 8% to about 10% of GBSSI activity of the wild type GBSSI gene. In some embodiments, the mutated GBSSI gene is the wx-Stonor (wxS) allele. In some embodiments, the mutated GBSSI gene is the wx-G allele, the wx-B5 allele, or the wx-M allele.

In some embodiments, the double-mutant plant is generated by cross pollination of two parent plants. In some embodiments, the first plant has the genotype of wxwxsu2su2. In some embodiments, the second plant has the genotype of wxSwxSsu2su2, or the genotype of wxSwxsu2su2.

In some embodiments, the endosperm of the plant has one dose, two doses, or three doses of the wxS allele. In some embodiments, the endosperm of the plant has the genotype of wxSwwxwsu2su2su2, wxSwxSwxsu2su2su2, or wxSwxSwxSsu2su2su2.

In some embodiments, starch produced in the endosperm of the plant has a higher viscosity compared to the starch produced in the endosperm of a double, waxy sugary-2 recessive mutant plant having the genotype of wxwxwsu2su2su2.

In some embodiments, starch produced in the endosperm of the plant has a higher starch pasting temperature compared to the starch produced in the endosperm of a double, waxy sugary-2 recessive mutant plant having the genotype of wxwxwsu2su2su2.

The present invention also provides plant parts, plant cells, and tissue cultures of the plants. In some embodiments, the plant part is an endosperm, a leaf, a flower, an ovule, a pollen, a rootstock, or a scion. In some embodiments, the plant part is a seed of the plants.

The present invention also provides starch extracted from plants described above.

The present invention also provides methods for producing starch. In some embodiments, the methods comprise obtaining starch from the endosperm of the plants of the present invention.

In some embodiments, the methods for producing starch comprise growing the plants of the present invention, and harvesting seeds from the plants.

The present invention also provides methods for producing a seed. In some embodiments, the methods comprise crossing a first parent plant with a second parent plant and harvesting the resultant seed. In some embodiments, said first parent plant and/or second parent plant is a plant of the present invention. In some embodiments, the methods comprise self-pollinating a plant of the present invention, and harvesting the resultant seed.

The present invention also provides methods of vegetatively propagating the plant. In some embodiments, the methods comprise collecting part of the plant and regenerating a plant from said part.

The present invention also provides methods of producing a plant derived from the plants of the present invention. In some embodiments, the methods comprise self-pollinating a plant of the present invention at least once to produce a progeny plant derived from the plant. In some embodiments, the methods comprise crossing a plant of the present invention with a second plant to produce a plant progeny.

The present invention also provides methods of producing a transgenic plant. In some embodiments, the methods comprise introducing a transgene of interest into a plant of the present invention. The transgene of interest confers one or more desired phenotypes in the plant.

The present invention also provides methods of producing a plant with desired phenotypes, comprising gene-editing to produce a genetic sequence in a plant of the present invention. The genetic sequence confers one or more desired phenotypes in the plant.

The present invention also provides methods of producing a plant with desired phenotypes, comprising inducing one or more mutations into a plant of the present invention to produce a genetic sequence. The genetic sequence confers one or more desired phenotypes in the plant.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 depicts the pasting profiles of AMIOCA™ starch, wxwxwxSU2su2su2 starch, and starch of waxy-sugary 2 double-mutant plants with zero, one, two, or three doses of wxS gene, as measured by a Rapid Visco Analyzer (RVA) over time during the heating-cooling cycle.

FIG. 2 depicts the pasting properties of AMIOCA™ starch, wxwxwxSU2su2su2 starch, and starch of waxy-sugary 2 double-mutant plants with zero, one, two, or three doses of wxS gene, including the pasting temperature, peak RVU, hold RVU, final RVU, breakdown RVU, and setback RVU.

FIG. 3 depicts the freeze-thaw stability test of starch of waxy-sugary 2 double-mutant plants with one, two, three, or zero doses of wxS gene, starch from Waxy Stonor seed, wxwxwxSU2su2su2 starch, and AMIOCA™ starch. The above-stated starches are presented in rows, and illustrative images of starch gels are provided after the first freeze-thaw (Cycle 1), after the third freeze-thaw (Cycle 3), and after the fifth freeze-thaw (Cycle 5).

FIG. 4 depicts the thermal properties of AMIOCA™ starch, wxwxwxSU2su2su2 starch, and starch of waxy-sugary 2 double-mutant plants with zero, one, two, or three doses of wxS gene.

FIG. 5 depicts the granule size distribution of starch of AMIOCA™ starch, wxwxwxSU2su2su2 starch, and starch of waxy-sugary 2 double-mutant plants with zero, one, two, or three doses of wxS gene.

FIG. 6 depicts the granule morphology of starch of waxy-sugary 2 double-mutant plants with one, two, three, or zero doses of wxS gene, wxwxwxSU2su2su2 starch, AMIOCA™ starch, and starch from Waxy Stonor seed. Starting on the top left of the first row, scanning electron microscopy images are provided for waxy-sugary 2 double-mutant plants with one wxS gene (Q3017a, 1 wxS), two wxS genes (Q8412a, 2 wxS), three wxS genes (Q7738a, 3 wxS), zero doses of wxS gene (wxwxwxSU2su2su2, 0 wxS), wxwxwxSU2su2su2 starch, AMIOCA® starch, and starch from Waxy Stonor seed.

DETAILED DESCRIPTION OF THE INVENTION

Definitions

As used herein, the verb “comprise” as is used in this description and in the claims and its conjugations are used in its non-limiting sense to mean that items following the word are included, but items not specifically mentioned are not excluded.

The invention provides compositions and methods to produce plants. As used herein, the term “plant” refers to any living organism Plantae (i.e., any genus/species in the Plant Kingdom). In some embodiments, the plant is capable of producing starch, including but not limited to maize, potato, wheat, tapioca, rice, potato and sorghum. In some embodiments, the plant is a corn plant.

The invention provides plant parts. As used herein, the term “plant part” refers to any part of a plant including but not limited to the embryo, endosperm, shoot, root, stem, seed, stipule, leaf, petal, flower, ovule, bract, branch, petiole, internode, bark, pubescence, tiller, rhizome, frond, blade, ovule, pollen, stamen, and the like.

The invention provides genes comprising the isolated (e.g., wild type, endogenous to the organism), chimeric, recombinant or synthetic nucleic acid sequence. As used herein, the term “gene” refers to any segment of DNA associated with a biological function. Thus, genes include, but are not limited to, coding sequences and/or the regulatory sequences required for their expression. Genes can also include nonexpressed DNA segments that, for example, form recognition sequences for other proteins. Genes can be obtained from a variety of sources, including cloning from a source of interest or synthesizing from known or predicted sequence information, and may include sequences designed to have desired parameters.

The invention provides polynucleotides with nucleotide change when compared to a wild-type reference sequence. As used herein, the term “nucleotide change” refers to, e.g., nucleotide substitution, deletion, insertion, or transposon, as is well understood in the art. For example, mutations contain alterations that produce silent substitutions, additions, deletions, or alternative splicing, which either alter or do not alter the properties or activities of the encoded protein or how the proteins are made. A nucleotide change can also be insertion and/or exclusion of a retrotransposon element in the gene.

The invention provides polypeptides with protein modification when compared to a wild-type reference sequence. As used herein, the term “protein modification” refers to,

e.g., amino acid substitution, amino acid modification, deletion, insertion, or change of protein activity, as is well understood in the art.

The invention provides polynucleotides and polypeptides derived from wild-type reference sequences. As used herein, the term “derived from” refers to the origin or source, and may include naturally occurring, recombinant, unpurified, or purified molecules. A nucleic acid or an amino acid derived from an origin or source may have all kinds of nucleotide changes or protein modification as defined elsewhere herein.

The invention provides biologically active variants or functional variants of the nucleic acid sequences and polypeptide sequences of the present invention. As used herein, the phrase “a biologically active variant” or “functional variant” with respect to a protein refers to an amino acid sequence that is altered by one or more amino acids with respect to a reference sequence, while still maintains substantial biological activity of the reference sequence. The variant can have “conservative” changes, wherein a substituted amino acid has similar structural or chemical properties, e.g., replacement of leucine with isoleucine. The following table shows exemplary conservative amino acid substitutions. In some embodiments, the variant has one or more amino acid substitutions, wherein one or more or all substitutions are acidic amino acid, such as Aspartic acid, Asparagine, Glutamic acid, or Glutamine.

Original Residue	Very Highly-Conserved Substitutions	Highly Conserved Substitutions (from the Blossum90 Matrix)	Conserved Substitutions (from the Blossum65 Matrix)
Ala	Ser	Gly, Ser, Thr	Cys, Gly, Ser, Thr, Val
Arg	Lys	Gln, His, Lys	Asn, Gln, Glu, His, Lys
Asn	Gln; His	Asp, Gln, His, Lys, Ser, Thr	Arg, Asp, Gln, Glu, His, Lys, Ser, Thr
Asp	Glu	Asn, Glu	Asn, Gln, Glu, Ser
Cys	Ser	None	Ala
Gln	Asn	Arg, Asn, Glu, His, Lys, Met	Arg, Asn, Asp, Glu, His, Lys, Met, Ser
Glu	Asp	Asp, Gln, Lys	Arg, Asn, Asp, Gln, His, Lys, Ser
Gly	Pro	Ala	Ala, Ser
His	Asn; Gln	Arg, Asn, Gln, Tyr	Arg, Asn, Gln, Glu, Tyr
Ile	Leu; Val	Leu, Met, Val	Leu, Met, Phe, Val
Leu	Ile; Val	Ile, Met, Phe, Val	Ile, Met, Phe, Val
Lys	Arg; Gln; Glu	Arg, Asn, Gln, Glu	Arg, Asn, Gln, Glu, Ser, Gln, Ile, Leu, Phe, Val
Met	Leu; Ile	Gln, Ile, Leu, Val	Gln, Ile, Leu, Phe, Val
Phe	Met; Leu; Tyr	Leu, Trp, Tyr	Ile, Leu, Met, Trp, Tyr
Ser	Thr	Ala, Asn, Thr	Ala, Asn, Asp, Gln, Glu, Gly, Lys, Thr
Thr	Ser	Ala, Asn, Ser	Ala, Asn, Ser, Val
Trp	Tyr	Phe, Tyr	Phe, Tyr
Tyr	Trp; Phe	His, Phe, Trp	His, Phe, Trp
Val	Ile; Leu	Ile, Leu, Met	Ala, Ile, Leu, Met, Thr

Alternatively, a variant can have “nonconservative” changes, e.g., replacement of a glycine with a tryptophan. Analogous minor variations can also include amino acid deletion or insertion, or both. Guidance in determining which amino acid residues can be substituted, inserted, or deleted without eliminating biological or immunological activity can be found using computer programs well known in the art, for example, DNASTAR software. For polynucleotides, a variant comprises a polynucleotide having deletions (i.e., truncations) at the 5' and/or 3' end; deletion and/or addition of one or more nucleotides at one or more internal sites in the reference polynucleotide; and/or substitution of one or more nucleotides at one or more sites in the reference polynucleotide. As used herein, a “reference” polynucleotide comprises a nucleotide sequence produced by the methods disclosed herein. Variant polynucleotides also include synthetically derived polynucleotides, such as those

generated, for example, by using site directed mutagenesis but which still comprise genetic regulatory element activity. Generally, variants of a particular polynucleotide or nucleic acid molecule, or polypeptide of the invention will have at least about 60%, 65%, 70%, 75%, 80%, 85%, 90%, 91%, 91.5%, 92%, 92.5%, 93%, 93.5%, 94%, 94.5%, 95%, 95.5%, 96%, 96.5%, 97%, 97.5%, 98%, 98.5%, 99%, 99.1%, 99.2%, 99.3%, 99.4%, 99.5%, 99.6%, 99.7%, 99.8%, 99.9% or more sequence identity to that particular polynucleotide/polypeptides as determined by sequence alignment programs and parameters as described elsewhere herein.

Variant polynucleotides also encompass sequences derived from a mutagenic and recombinogenic procedure such as DNA shuffling. Strategies for such DNA shuffling are known in the art. See, for example, Stemmer (1994) *PNAS* 91:10747-10751; Stemmer (1994) *Nature* 370:389-391; Cramer et al. (1997) *Nature Biotech.* 15:436-438; Moore et al. (1997) *J. Mol. Biol.* 272:336-347; Zhang et al. (1997) *PNAS* 94:4504-4509; Cramer et al. (1998) *Nature* 391:288-291; and U.S. Pat. Nos. 5,605,793 and 5,837,458. For PCR amplifications of the polynucleotides disclosed herein, oligonucleotide primers can be designed for use in PCR reactions to amplify corresponding DNA sequences from cDNA or genomic DNA extracted from any plant of interest. Methods for designing PCR primers and PCR cloning are generally known in the art and are disclosed in Sambrook et al. (1989) *Molecular Cloning: A Laboratory Manual* (2nd ed., Cold Spring Harbor Laboratory Press, Plainview, N.Y.). See also Innis et al., eds. (1990) *PCR Protocols: A Guide to Methods and Applications* (Academic Press, New York); Innis and Gelfand, eds. (1995) *PCR Strategies* (Academic Press, New York); and Innis and Gelfand, eds. (1999) *PCR Methods Manual* (Academic Press, New York). Known methods of PCR include, but are not limited to, methods using paired primers, nested primers, single specific primers, degenerate primers, gene-specific primers, vector-specific primers, partially-mismatched primers, and the like.

The invention provides transgenic plants. As used herein, the term “transgenic” refers to cells, cell cultures, organisms (e.g., plants), and progeny which have received a foreign or modified gene by one of the various methods of transformation, wherein the foreign or modified gene is from the same or different species than the species of the organism receiving the foreign or modified gene.

Waxy Gene

Waxy gene is the granule-bound starch synthase I (GBSSI) gene. The gene encodes a starch granule-bound ADP-glucose-glucosyl transferase (EC 2.4.1.242) responsible for amylose biosynthesis in the triploid endosperm of the developing kernel and in haploid pollen and embryo sac. “Wx” refers to the wild type allele, and “wx” refers to the null, reference mutant-allele. The terms wxS (a.k.a. wx-Stonor), wx-G, wx-B5, and wx-M refer to the intermediate alleles having retrotransposon insertion in the waxy gene, as described in Wessler and Varagona (*PNAS*, 82:4177-4181, 1985) and Varagona et al. (*The Plant Cell*, 4:811-820, 1992), each of which is herein incorporated by reference in its entirety. These intermediate alleles result in less waxy protein activity in plants having the alleles than the waxy protein activity in wild type plants. The maize waxy gene encodes the protein having the sequence of SEQ ID NO: 1.

wx-Stonor

The wx-Stonor allele in maize plant is caused by a retrotransposon insertion at the splice donor site of intron 5 of the waxy gene. Although the insertion does not affect the splicing machinery, it leads to a lower level of enzymatic activity compared to the wild type allele (e.g., only about 9.5% GBSSI activity as of the wild type). As a result, plants

having wx-Stonor allele has higher amylose level than the same plant which does not have the wx-Stonor allele.

Sugary-2 Gene

Sugary-2 gene encodes starch synthase isoform IIa (SSIa, EC 2.4.1.21). "SU2" refers to the wild type allele, and "su2" refers to the recessive, mutant allele. The function of Sugary-2 protein in maize is described in Zhang et al. (Plant Molecular Biology, 54(6):865-879, 2004), which is herein incorporated by reference in its entirety. The maize wild type SU2 gene encodes the wild type protein having the sequence of SEQ ID NO: 2.

More detail of the waxy gene and the sugary-2 gene in maize are described in U.S. Pat. Nos. 4,428,972, 5,954,883, 6,828,474, 6,960,703, and 7,678,898, each of which is herein incorporated by reference in its entirety.

AMIOCA™ Starch

The term refers to the AMIOCA™ corn starch produced by National Starch and Chemical Company, now Ingredion Incorporated.

wxwxwxSU2su2su2 Starch

The term refers to starch isolated from maize endosperm having the genotype of wxwxwxSU2su2su2.

wxwxwxsu2su2su2 Starch

The term refers to starch isolated from maize endosperm having the genotype of wxwxwxsu2su2su2, which is the endosperm of the waxy sugary-2 double recessive mutant. The mutated waxy gene contributes to its good freeze-thaw stability, and the mutated sugary-2 gene results in short branch-chain length. However, the starch has low integrity of starch granules. The granules break down easily, and the starch has low viscosity.

MELOJEL® Starch

The term refers to the MELOJEL® corn starch produced by National Starch and Chemical Company, now Ingredion Incorporated.

Control Corn Plant

The term refers to a proper check plant for comparison. In some embodiments, a check plant can be a wild type plant. In some embodiments, the check plant is a plant that does not have an intermediate waxy gene as described herein. In some embodiments, the check plant is a corn plant that produces a wxwxwxSU2su2su2 starch or a wxwxwxsu2su2su2 starch. Accordingly, in some embodiments, the control corn plant is a corn plant which does not possess a wx-S allele in the endosperm.

Control Starch

The terms refers to a control starch.

wxSwxwxsu2su2su2

The term refers to a maize endosperm with one dose of wxS.

wxSwxwxsu2su2su2 Starch

The term refers to a starch isolated from maize endosperm with one dose of wxS, the endosperm having the genotype wxSwxwxsu2su2su2.

wxSwxSwxsu2su2su2

The term refers to a maize endosperm with two doses of wxS.

wxSwxSwxsu2su2su2 Starch

The term refers to a starch isolated from maize endosperm with two doses of wxS, the endosperm having the genotype wxSwxSwxsu2su2su2.

wxSwxSwxSsu2su2su2

The term refers to a maize endosperm with three doses of wxS.

wxSwxSwxSsu2su2su2 Starch

The term refers to a starch isolated from maize endosperm with three doses of wxS, the endosperm having the genotype wxSwxSwxSsu2su2su2.

The following examples illustrate various aspects of the invention. The examples should, of course, be understood to

be merely illustrative of only certain embodiments of the invention and not to constitute limitations upon the scope of the invention.

Starch with Novel Functionalities

In one aspect, the disclosure provides for a starch derived from a double-mutant corn plant comprising an intermediate GBSSI allele which has less GBSSI activity than a wild type GBSSI gene (Wx), but has higher GBSSI activity than a recessive, loss-of-function GBSSI gene (wx). In some embodiments, the double-mutant corn plant with an intermediate GBSSI allele is a waxy-sugary 2 corn plant comprising at least one wx-Stonor (wxS) allele in the endosperm. In some embodiments, the waxy-sugary 2 corn plant is generated by cross pollination of a first corn plant having the genotype of wxwxsu2su2, and a second corn plant having the genotype of wxSwxSsu2su2 or the genotype of wxSwxsu2su2. In further embodiments, the endosperm of the waxy-sugary 2 corn plant has one dose, two doses, or three doses of the wxS allele, and the endosperm has the genotype of wxSwxwxsu2su2su2, wxSwxSwxsu2su2su2, or wxSwxSwxSsu2su2su2.

In some embodiments, the starch comprises an amylose content of from about 2 wt % to about 20 wt % as measured using a Megazyme® amylose/amylopectin assay kit, and an aqueous starch pasting temperature that is at least about 5% greater than the pasting temperature of a control starch as measured using RVA analysis.

Amylose Content

Starch is comprised of amylose and amylopectin. Amylose is a helical polymer made of α -D-glucose units, connected through $\alpha(1\rightarrow4)$ glycosidic bonds. Amylopectin is a highly branched polymer of glucose comprising $\alpha(1\rightarrow4)$ and $\alpha(1\rightarrow6)$ glycosidic bonds. Branching takes place with the $\alpha(1\rightarrow6)$ bonds, which occurs at approximately every 24 to 30 glucose units. In some embodiments, the amylose content in starch can be measured using an amylose/amylopectin assay kit, such as the amylose/amylopectin assay kit manufactured by Megazyme®, as set forth in the Examples section.

An intermediate mutation of the GBSSI gene in the double-mutant corn plant was observed to increase amylose content in the starch derived therefrom relative to a control corn plant with the null-allele GBSSI gene. For example, amylose content of a starch derived from a waxy-sugary 2 corn plant comprising at least one wx-Stonor (wxS) allele in the endosperm was observed to increase relative to a control corn plant. In some embodiments, the starch derived from a corn plant with at least one wx-Stonor (wxS) allele in the endosperm has an amylose content of from about 2 wt % to about 20 wt %, from about 4 wt % to about 18 wt %, from about 5 wt % to about 15 wt %, or about 8 wt % to about 15 wt %, including all values and subranges in between. In other embodiments, the amylose content is about 5 wt %, about 6 wt %, about 7 wt %, about 8 wt %, about 9 wt %, about 10 wt %, about 11 wt %, about 12 wt %, about 13 wt %, about 14 wt %, about 15 wt %, about 16 wt %, about 17 wt %, or about 18 wt %, including all values between.

Without being limited by theory, an increase in the amylose content would be expected to decrease the amylopectin content in the starch, thereby decreasing the total number glucose units and hydrogen-bonding groups available to interact with an aqueous medium. Such a decrease in hydrogen bonding should correspond to an overall decrease in the strength of the interaction of said starch with the aqueous medium and diminish the pasting properties of the

starch. However, an unexpected improvement in the pasting profile was observed when the amylose content was increased in the starch disclosed herein. Without being limited by theory, this unexpected result can be attributed to an increase in the branch-chain length observed in a waxy-sugary 2 corn plant comprising at least one wx-Stonor (wxS) allele in the endosperm. Longer branch chains on the starch present more glucose units, which increase the number of hydrogen bonding interactions with the aqueous medium compared to shorter branch chain lengths.

The pasting profile of a starch can be measured by known methods, such as using a Rapid Visco Analyzer (RVA) (Newport Scientific, Sydney, Australia). For example, RVA can measure pasting temperature ($^{\circ}$ C.), peak viscosity (RVU), hold viscosity (RVU), final viscosity (RVU), breakdown viscosity (RVU), and set back viscosity (RVU). In some embodiments, the improved pasting profile allows gelatinization to occur, inter alia, at a lower effective amount of the starch and/or at a higher pasting temperature compared to a control starch. Starch gelatinization refers to a process of breaking intermolecular bonds of starch molecules in the presence of water and heat, allowing the hydrogen bonding sites (e.g., hydroxyl groups) to bind more water. To measure a starch pasting profile, an effective amount of a starch can be added to an aqueous solution at neutral pH. For example, starch can be added to an aqueous solution at 8 w/w % solids level.

Pasting Temperature

Pasting temperature, as used herein, is intended to be measured as set forth in the Examples section. The pasting temperature of starch is the temperature at which initial swelling of starch granules takes place when suspended in water. In some embodiments, the starch described herein has an aqueous pasting temperature that is at least about 5% greater than the pasting temperature of a control starch as measured using RVA analysis. In some embodiments, the aqueous pasting temperature is increased by about 1% to about 100%, about 1% to about 50%, about 5% to about 50%, about 5% to 40%, about 5% to about 30%, about 5% to about 25%, about 5% to about 20% or about 5% to about 14%, including all values and subranges in between. In a particular embodiment, the aqueous pasting temperature is from about 5% to about 14% greater than that of a control corn plant, such as a double mutant waxy-sugary 2 corn plant.

In other embodiments, the aqueous pasting temperature is increased by at least about 2%, at least about 3%, at least about 4%, at least about 5%, at least about 6%, at least about 7%, at least about 8%, at least about 9%, at least about 10%, at least about 11%, at least about 12%, at least about 13%, at least about 14%, at least about 15%, at least about 15%, at least about 16%, at least about 17%, at least about 18%, at least about 19%, or at least about 20% greater compared to a pasting temperature of a control starch, including all values in between.

In still other embodiments, the aqueous pasting temperature is increased by 1° C. to about 20° C., or about 5° C. to about 10° C., including all values and subranges in between. In other embodiments, the aqueous pasting temperature is increased at least about 4° C., at least about 5° C., at least about 6° C., at least about 7° C., at least about 8° C., at least about 9° C., at least about 10° C., at least about 11° C., at least about 12° C., at least about 13° C., at least about 14° C., or at or about 15° C. compared to a pasting temperature of a control starch.

In still other embodiments, the starch described herein has an aqueous pasting temperature of at least about 50° C., at

least about 55° C., at least about 60° C., at least about 65° C., at least about 70° C., at least about 75° C., at least about 80° C., at least about 85° C., at least about 90° C., at least about 95° C., or at least about 100° C. In particular embodiments, the starch derived from a waxy-sugary 2 corn plant comprising at least one wx-Stonor (wxS) allele in the endosperm has an aqueous pasting temperature of from about 55° C. to about 68° C., or from about 59° C. to about 65° C., including all values and subranges in between. In yet other embodiments, the aqueous pasting temperature is about 59° C., about 60° C., about 61° C., about 62° C., about 63° C., about 64° C., about 65° C., about 66° C., about 67° C., or about 68° C., including all values in between.

Enthalpy Change of Crystallinity

Enthalpy change of crystallinity, as used herein, is intended to be measured as set forth in the Examples section. Crystallization refers to the process of forming solid crystals from a solution by precipitation. In some embodiments, introduction of at least one wx-Stonor (wxS) allele in the endosperm of a in a waxy-sugary 2 double-mutant corn plant was observed to increase the enthalpy change of crystallinity relative to a starch from a control corn plant. Enthalpy change can be measured using, e.g., differential scanning calorimetry (DSC). In some embodiments, the enthalpy change of crystallinity is increased by about 10% to about 300%, about 10% to about 200%, about 10% to about 150%, about 10% to about 100%, or about 10% to about 50%, including all values and subranges in between. In one such embodiment, the enthalpy change of crystallinity is from about 70% to about 200% greater than an enthalpy change of crystallinity of a control starch as measured using a DSC.

In some embodiments, the enthalpy change of crystallinity is increased by at least about 10%, at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about 60%, at least about 70%, at least about 80%, at least about 90%, at least about 100%, at least about 110%, at least about 120%, at least about 130%, at least about 140%, at least about 150%, at least about 160%, at least about 170%, at least about 180%, at least about 190%, or at least about 200%, including all values in between.

In some embodiments, the enthalpy change of crystallinity is from about 1 J/g to about 20 J/g, about 5 J/g to about 15 J/g, about 6 J/g to about 13 J/g, including all values and subranges in between. In other embodiments, the enthalpy change of crystallinity 8 w/w % is about 4 J/g, about 5 J/g, about 6 J/g, about 7 J/g, about 8 J/g, about 9 J/g, about 10 J/g, about 11 J/g, about 12 J/g, about 13 J/g, including all values in between. In a particular embodiment, the corn plant comprises one wxS allele in the endosperm, and the enthalpy change of crystallinity is at least about 150% greater than that of a starch derived from a control plant with the endosperm genotype of wxwxwxsu2su2su2. In another particular embodiment, the corn plant comprises two wxS alleles in the endosperm, and the enthalpy change of crystallinity is at least about 30% greater than that of a starch derived from a control plant with the endosperm genotype of wxwxwxsu2su2su2. In yet another particular embodiment, the corn plant comprises three wxS alleles in the endosperm, and the enthalpy change of crystallinity is at least about 75% greater than that of a starch derived from a control plant with the endosperm genotype of wxwxwxsu2su2su2.

Freeze Thaw Stability

Freeze thaw stability, as used herein, is intended to be measured as set forth in the Examples section. In some embodiments, the starch described herein exhibits desirable freeze-thaw characteristics, e.g., improved freeze-thaw stability. Freeze-thaw stability can be measured by adding an

effective amount of a starch to water to form a slurry, which is then cooked and syneresis is measured after cycles of freezing/cooling. In some embodiments, introduction of at least one wx-Stonor (wxS) allele into the endosperm of a corn plant did not reduce the freeze-thaw stability of the starch derived therefrom compared to a control starch. In other embodiments, introduction of at least one wx-Stonor (wxS) allele into the endosperm of a corn plant improved the freeze-thaw stability of the starch. In one such embodiment, the starch is capable withstanding a total of at least three to at least five freeze-thaw cycles.

Granule Distribution and Integrity

Granule distribution and integrity, as used herein, is intended to be measured as set forth in the Examples section.

In some embodiments, introduction of at least one wx-Stonor (wxS) allele in the endosperm of a corn plant was observed to alter the distribution of sizes of starch granules relative to a control starch. In some such embodiments, the distribution of starch granule sizes of a starch described herein comprises at least about 40% fewer granules less than 5 microns compared to a proportion of granules less than 5 microns from a control starch as observed using a scanning electron microscope.

Granule integrity, as used herein, refers to the ability of the starch to remain as an intact granule after dissolution in an aqueous medium. In some embodiments, the starch described herein is a granule which exhibits increased integrity compared to a starch derived from the control corn plant as observed using a scanning electron microscope.

Viscosity

Viscosity, as used herein, is intended to be measured as set forth in the Examples section. In some embodiments, the starch described herein has an aqueous peak viscosity that is at least about 10% greater than the aqueous peak viscosity of a control starch as measured using RVA analysis. In some embodiments, the aqueous peak viscosity was increased by about 10% to about 300%, about 10% to about 250%, about 10% to about 200%, about 10% to about 150%, about 10% to about 100%, or about 10% to about 50%, including all values and subranges in between. In some embodiments, the aqueous peak viscosity was increased by at least about 10%, at least about 20%, at least about 30%, at least about 40%, at least about 50%, at least about 60% at least about 70%, at least about 80%, at least about 90%, at least about 100%, at least about 110%, at least about 120%, at least about 130%, at least about 140%, at least about 150%, at least about 160%, at least about 170%, at least about 180%, at least about 190%, or at least about 200%, including all values in between.

In some embodiments, the aqueous peak viscosity of the starch at 8 w/w % solids is about 100 RVU to about 500 RVU, about 100 RVU to about 400 RVU, about 100 RVU to about 300 RVU, about 100 RVU to about 200 RVU, or about 100 RVU to about 150 RVU, including all values and subranges in between. In some embodiments, the aqueous peak viscosity is about 150 RVU, about 155 RVU, about 160 RVU, about 165 RVU, about 170 RVU, about 175 RVU, about 180 RVU, about 185 RVU, about 190 RVU, about 195 RVU, about 200 RVU, about 205 RVU, about 210 RVU, about 215 RVU, about 220 RVU, about 225 RVU, about 230 RVU, about 235 RVU, about 240 RVU, about 245 RVU, about 250 RVU, about 255 RVU, about 260 RVU, about 265 RVU, about 270 RVU, about 275 RVU, about 280 RVU, about 285 RVU, about 290 RVU, about 295 RVU, about 300 RVU, about 305 RVU, about 310 RVU, about 315 RVU, about 320 RVU, about 325 RVU, about 330 RVU, about 335 RVU, about 340 RVU, about 345 RVU, about 350 RVU,

about 355 RVU, about 360 RVU, about 365 RVU, about 370 RVU, about 375 RVU, about 380 RVU, about 385 RVU, about 390 RVU, about 395 RVU, or about 400 RVU, including all values in between.

In a particular embodiment, the waxy-sugary 2 corn plant comprises one wx-Stonor (wxS) allele in the endosperm, and the starch has a peak viscosity of about 110% greater than that of a control plant with the endosperm genotype of wxwxwxsu2su2su2. In another particular embodiment, the waxy-sugary 2 corn plant comprises two wx-Stonor (wxS) alleles in the endosperm, and the starch has a peak viscosity of about 30% greater than that of a control plant with the endosperm genotype of wxwxwxsu2su2su2. In yet another particular embodiment, the waxy-sugary 2 corn plant comprises three wx-Stonor (wxS) alleles in the endosperm, and the starch has a peak viscosity of about 40% greater than that of a control plant with the endosperm genotype of wxwxwxsu2su2su2. In even another particular embodiment, the waxy-sugary 2 corn plant comprises two wx-Stonor (wxS) alleles in the endosperm, and the starch has a peak viscosity of about 20% greater than that of a control plant with the endosperm genotype of wxwxwxSU2su2su2. In still another particular embodiment, the waxy-sugary 2 corn plant comprises two wx-Stonor (wxS) alleles in the endosperm, and the starch has a peak viscosity is at least about 30% greater than that of a control plant with the endosperm genotype of wxwxwxSU2su2su2.

Extraction

Extraction of the starch from the kernel of the plants of the present invention may be carried out in a standard manner by the wet-milling or dry-milling process well known in the art, but is not limited to such methods. In one typical wet-milling process, the plant is cleaned by strong currents of air, sifters and electromagnets to remove unwanted material. It is thereafter steeped in warm water containing a small amount of sulfur dioxide. The steep-water is drawn off and the softened kernels are run through attrition mills to tear them apart. The germ is removed and the remaining mixture is ground, washed and sieved as a slurry. The starch is separated from the gluten by centrifugation, and the remaining slurried starch is then filtered, washed, resuspended and refiltered.

Extraction of flour or variants thereof from the plant can be accomplished by a dry-milling process. In a typical procedure which is suitable herein but not exclusive of other procedures, the grain obtained from a plant of the present invention is first thoroughly cleaned and passed through a scourer and is then tempered or conditioned and passed through a degerminator. Stock from the degerminator is dried and then cooled, passed through a hominy separator and aspirator, ground, and finally sifted according to whether whole or separate fractions are desired.

Modification

It can be understood that some modifications in either of the above extraction processes such, for example, as using a steep-water temperature below that which is commonly employed, may be desirable and will be easily recognized by a starch practitioner.

The starch herein may be modified, if desired, by procedures known in the art, such as by derivatization to form ethers or esters such as hydroxypropyl ethers, acetates, phosphates, succinates, e.g., octenyl succinate, tertiary and quaternary amine ethers, etc., or by any other modification techniques which produce a starch having the characteristics defined herein. The preferred substituent groups herein are hydroxypropyl, phosphate or acetate groups.

For commercial purposes, one modification of the starch herein is crosslinking to strengthen the granules against the handling and processing conditions frequently encountered in manufacturing operations and to provide a starch capable of imparting desirable rheological properties to food systems. Any cross-linking agent known in the art may be employed for this purpose, including but not limited to epichlorohydrin, linear dicarboxylic acid anhydrides, citric acid acrolein, phosphorus oxychloride, adipic/acetic mixed acid anhydrides, and trimetaphosphate salts for food systems and to epichlorohydrin, linear dicarboxylic acid anhydrides, citric acid acrolein, phosphorus oxychloride, adipic/acetic mixed acid anhydrides, trimetaphosphate salts, formaldehyde, cyanuric chloride, diisocyanates, and divinyl sulfones in non-food systems. The cross-linking reaction is carried out using techniques known in the art, for example those described in U.S. Pat. Nos. 2,328,537 and 2,801,242. Procedures for modifying starches are described in the Chapter "Starch and Its Modification" by M. W. Rutenberg, pages 22-26 to 22-47, Handbook of Water Soluble Gums and Resins, R. L. Davidson, Editor (McGraw-Hill, Inc., New York, N.Y. 1980).

The amount of crosslinking agent necessary to give a suitable product will vary depending, for example, on the type of crosslinking agent employed, the concentration of the crosslinking agent, the reaction conditions, and the necessity for having a crosslinked starch which falls within the desired viscosity range. The practitioner will recognize which amounts may be employed, as these are well known in the art. Typically, this amount will range from as low as about 0.001%, by weight of the starch, to as high as is considered acceptable for food use.

The present starches may also be physically modified, such as by thermal inhibition described in WO 95/04082 (published Feb. 9, 1995).

Processing

The starches may also be pregelatinized. Exemplary processes for preparing pregelatinized starches are disclosed in U.S. Pat. No. 4,280,851 (Pitchon, et al.), U.S. Pat. No. 4,465,702 (Eastman, et al.), U.S. Pat. No. 5,037,929 (Rajagopalan), U.S. Pat. No. 5,131,953 (Kasica, et al.), and U.S. Pat. No. 5,149,799 (Rubens). Conventional procedures for pregelatinizing starch are well known to those skilled in the art and described in such articles as Chapter XXII—"Production and Use of Pregelatinized Starch", Starch: Chemistry and Technology, Vol. III—Industrial Aspects, R. L. Whistler and E. F. Paschall, Editors, Academic Press, New York 1967.

The present starches may be purified by any method known in the art to remove off-flavors and colors that are native to the starch or created during starch modification processes. Purification processes preferred for treating the present starches are disclosed in U.S. Ser. No. 07/832,838 filed Feb. 7, 1992, by Kasica, et al. Alkali washing techniques, for starches intended for use in either granular or pregelatinized form, are also useful and described in the family of patents represented by U.S. Pat. No. 5,187,272 (Bertalan, et al.).

Conversion products derived from the present starches, including but not limited to fluidity or thin-boiling starches prepared by oxidation, enzyme conversion particularly by α -amylase, acid hydrolysis, heat and or acid dextrinization, thermal and or sheared products are also useful herein.

Plants Producing Starch

The present invention provides plants producing starch described above. The starch producing plants include, but are not limited to maize, potato, wheat, tapioca, rice, cassava, and sorghum.

The starch of the present invention can be extracted from corn plants, such as from the endosperm of corn plants. The corn plants that produce starch of the present invention are double mutant plants of the waxy/sugary-2 genotype. The waxy gene is located at Chromosome 9 of corn while the sugary-2 gene is located at Chromosome 6. (See M. G. Nueffer, L. Jones, and M. Zuber, "The Mutants of Maize" (Crop Science Society of America, Madison, Wis., 1968), pp. 72 and 73.).

The plants of the present invention are homozygous recessive for the starch synthase IIa (su2) gene, and either homozygous or heterozygous for a mutated granule-bound starch synthase I (GBSSI) gene. The mutated GBSSI gene has less GBSSI activity than the wild type GBSSI gene (Wx), but has higher GBSSI activity than the recessive, loss-of-function GBSSI gene (wx). As used herein, such a waxy mutant gene is called an intermediate waxy mutant. In some embodiments, the intermediate waxy mutant has less than 30%, less than 25%, less than 20%, less than 15%, less than 10%, or less than 5% of the wild type GBSSI gene expression. In some embodiments, the intermediate waxy mutant has about 5%, about 6%, about 7%, about 8%, about 9%, about 10%, about 11%, about 12%, about 13%, about 14%, or about 15% of the wild type GBSSI gene expression.

An intermediate GBSSI gene having less GBSSI activity than the wild type GBSSI gene but higher GBSSI activity than the recessive, loss-of-function GBSSI gene can be either those previously known in the field, or be created by any suitable mutagenesis method.

Mutant alleles such as wxS (a.k.a. wx-Stonor), wx-G, wx-B5, and wx-M have intermediate GBSSI activity. These mutant alleles are due to retrotransposon insertion in the GBSSI gene, as described in Wessler and Varagona (PNAS, 82:4177-4181, 1985) and Varagona et al. (The Plant Cell, 4:811-820, 1992). These intermediate alleles result in a waxy protein having less activity than the wild type waxy protein.

Additional mutant alleles having intermediate GBSSI activity can be created by suitable methods. For example, retrotransposon mutagenesis can be used to generate a population for screening corn plants having a mutated GBSSI gene that has intermediate GBSSI activity. In some embodiments, wxS (a.k.a. wx-Stonor), wx-G, wx-B5, and wx-M can be used as starting materials to produce additional mutant alleles.

Other mutagenesis methods that can be used to produce mutant alleles having intermediate GBSSI activity include, but are not limited to, chemical mutagenesis, radiation mutagenesis, transposon mutagenesis, insertional mutagenesis, signature tagged mutagenesis, site-directed mutagenesis, and natural mutagenesis.

Still in some embodiments, antisense RNA, ribozyme, dsRNAi, RNA interference (RNAi) can be used to produce mutant alleles having intermediate GBSSI activity. Antisense RNA technology involves expressing in, or introducing into, a cell an RNA molecule (or RNA derivative) that is complementary to, or antisense to, sequences found in a particular mRNA in a cell. By associating with the mRNA, the antisense RNA can inhibit translation of the encoded gene product. The use of antisense technology to reduce or inhibit the expression of specific plant genes has been described, for example in European Patent Publication No. 271988, Smith et al., Nature, 334:724-726 (1988); Smith et al., Plant Mol. Biol., 14:369-379 (1990)). A ribozyme is an RNA that has both a catalytic domain and a sequence that is complementary to a particular mRNA. The ribozyme functions by associating with the mRNA (through the comple-

mentary domain of the ribozyme) and then cleaving (degrading) the message using the catalytic domain. RNA interference (RNAi) is the process of sequence-specific, post-transcriptional gene silencing or transcriptional gene silencing in animals and plants, initiated by double-stranded RNA (dsRNA) that is homologous in sequence to the silenced gene. The RNAi technique is discussed, for example, in Elbashir, et al., *Methods Enzymol.* 26:199 (2002); McManus & Sharp, *Nature Rev. Genetics* 3:737 (2002); PCT application WO 01/75164; Martinez et al., *Cell* 110:563 (2002); Elbashir et al., *supra*; Lagos-Quintana et al., *Curr. Biol.* 12:735 (2002); Tuschl et al., *Nature Biotechnol.* 20:446 (2002); Tuschl, *ChemBiochem.* 2:239 (2001); Harborth et al., *J. Cell Sci.* 114:4557 (2001); et al., *EMBO J.* 20:6877 (2001); Lagos-Quintana et al., *Science* 294:8538 (2001); Hutvagner et al., *loc cit.* 834; Elbashir et al., *Nature* 411:494 (2001).

Also within the scope of this invention is the starch resulting from mutants wherein the mutant wx and/or su2 alleles have been moved to another portion of the plant genome by translocation, inversion, or any other methods of chromosome engineering. In addition, starch extracted from a plant grown from artificial mutations and variations of the above genetic composition which may be produced by known standard methods of mutation breeding is also applicable herein.

Also within the scope of this invention is the starch produced from plants other than corn plants, as long as the starch producing plants have a mutant ortholog of the maize GBSSI gene that has intermediate GBSSI activity, and a recessive mutant ortholog of the maize SSIIa gene. In some embodiments, the plants can be wheat, tapioca, rice, cassava, sago, potato, or sorghum.

In some embodiments, the ortholog GBSSI gene in plants other than maize encodes a protein having at least 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or more identity to SEQ ID NO: 1, and have the GBSSI activity. In some embodiments, the ortholog has the sequence of UniProt entry number O82627 (*Antirrhinum majus* (Garden snapdragon)), P12615 (*Avena sativa* (Oat)), Q42968 (*Oryza glaberrima* (African rice)), Q43784 (*Manihot esculenta* (Cassava)), P0C585 (*Oryza sativa* (Rice)), Q00775 (*Solanum tuberosum* (Potato)), P27736 (*Triticum aestivum* (Wheat)), Q43134 (*Sorghum bicolor* (Sorghum)), Q43092 (*Pisum sativum* (Garden pea)), P84766 (*Aegilops tauschii* subsp. *stragulata* (Goatgrass)), Q0DEV5 (*Oryza sativa* subsp. *japonica* (Rice)), Q42857 (*Ipomoea batatas* (Sweet potato)), P84633 (*Fagopyrum esculentum* (Common buckwheat)), P84765 (*Secale cereale* (Rye)), Q9MAQ0 (*Arabidopsis thaliana*), P09842 (*Hordeum vulgare* (Barley)), or A2Y8X2 (*Oryza sativa* subsp. *indica* (Rice)), each of which is herein incorporated by reference in its entirety.

In some embodiments, the ortholog SSIIa gene in plants other than maize encodes a protein having at least 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or more identity to SEQ ID NO: 2, and have the SSIIa activity. In some embodiments, the ortholog has the sequence of UniProt entry number V6BPG5 (*Triticum urartu* (Red wild einkorn)), V6BPN2 (*Triticum monococcum* subsp. *Monococcum*), C3W8L4 (*Hordeum vulgare* var. *distichum* (Domesticated barley)), E2GHR4 (*Oryza sativa*

subsp. *indica* (Rice)), or E2GHR6 (*Oryza sativa* subsp. *japonica* (Rice)), each of which is herein incorporated by reference in its entirety.

In some embodiments, to produce double mutant plants having the intermediate wx mutant and the recessive su2 mutant, a cross pollination between a plant having the intermediate wx mutant and a plant having the recessive su2 mutant can be made.

For example, a cross pollination can be made between a homozygous wxS mutant (wxSwxSSU2SU2) or a heterozygous wxS mutant (wxSWxSU2SU2) and a homozygous waxy su2 mutant (wxwxsu2su2) or a heterozygous waxy su2 mutant (wxWxsu2su2 or wxwxSU2su2). After the cross, self pollination of the F1 generation having the genotype of wxSwxSU2su2 can be used to generate the F2 plants. Plants having either zero, one, two, or three doses of the wxS gene in the endosperm, and three doses of the recessive su2 gene can be isolated accordingly (e.g., endosperm having the genotype of wxwxwxsu2su2su2, wxSwxwxsu2su2su2, wxSwxSwxsu2su2su2, or wxSwxSwSsu2su2su2). This is only one example of the breeding method, and other breeding schemes can be used, and are within the scope of the present invention.

In some embodiments, field production of plants with endosperms that have at least one dose of the intermediate waxy allele and the recessive sugary-2 allele is carried out by crossing female maize plants with male maize plants. A typical planting arrangement is one male row to several female rows. The female rows are either detasseled or rendered male sterile through various other means known in the art such as cytoplasmic or genetic means.

The starch utilized in this invention may be obtained from inbred lines, or the starch can be obtained from hybrids derived from inbreds containing the intermediate waxy/recessive su2 double mutant. While maize is the exemplary plant herein for the source of the waxy starch, the invention is also applicable to other plant species such as, for example, waxy rice, waxy barley and waxy sorghum, provided that they have an intermediate waxy gene and a recessive su2 gene.

Breeding Methods

To make the plants of the present invention, suitable breeding or selection methods can be used. Popular selection methods commonly include pedigree selection, modified pedigree selection, mass selection, recurrent selection, and backcross breeding.

Open-Pollinated Populations

The improvement of open-pollinated populations of such crops as rye, many maizes and sugar beets, herbage grasses, legumes such as alfalfa and clover, and tropical tree crops such as cacao, coconuts, oil palm and some rubber, depends essentially upon changing gene-frequencies towards fixation of favorable alleles while maintaining a high (but far from maximal) degree of heterozygosity. Uniformity in such populations is impossible and trueness-to-type in an open-pollinated variety is a statistical feature of the population as a whole, not a characteristic of individual plants. Thus, the heterogeneity of open-pollinated populations contrasts with the homogeneity (or virtually so) of inbred lines, clones and hybrids.

Population improvement methods fall naturally into two groups, those based on purely phenotypic selection, normally called mass selection, and those based on selection with progeny testing. Interpopulation improvement utilizes the concept of open breeding populations; allowing genes to

flow from one population to another. Plants in one population (cultivar, strain, ecotype, or any germplasm source) are crossed either naturally (e.g., by wind) or by hand or by bees (commonly *Apis mellifera* L. or *Megachile rotundata* F.) with plants from other populations. Selection is applied to improve one (or sometimes both) population(s) by isolating plants with desirable traits from both sources.

There are basically two primary methods of open-pollinated population improvement. First, there is the situation in which a population is changed en masse by a chosen selection procedure. The outcome is an improved population that is indefinitely propagable by random-mating within itself in isolation. Second, the synthetic variety attains the same end result as population improvement but is not itself propagable as such; it has to be reconstructed from parental lines or clones. These plant breeding procedures for improving open-pollinated populations are well known to those skilled in the art and comprehensive reviews of breeding procedures routinely used for improving cross-pollinated plants are provided in numerous texts and articles, including: Allard, *Principles of Plant Breeding*, John Wiley & Sons, Inc. (1960); Simmonds, *Principles of Crop Improvement*, Longman Group Limited (1979); Hallauer and Miranda, *Quantitative Genetics in Maize Breeding*, Iowa State University Press (1981); and, Jensen, *Plant Breeding Methodology*, John Wiley & Sons, Inc. (1988).

Mass Selection

In mass selection, desirable individual plants are chosen, harvested, and the seed composited without progeny testing to produce the following generation. Since selection is based on the maternal parent only, and there is no control over pollination, mass selection amounts to a form of random mating with selection. As stated herein, the purpose of mass selection is to increase the proportion of superior genotypes in the population.

Synthetics

A synthetic variety is produced by crossing inter se a number of genotypes selected for good combining ability in all possible hybrid combinations, with subsequent maintenance of the variety by open pollination. Whether parents are (more or less inbred) seed-propagated lines, as in some sugar beet and beans (*Vicia*) or clones, as in herbage grasses, clovers and alfalfa, makes no difference in principle. Parents are selected on general combining ability, sometimes by test crosses or topcrosses, more generally by polycrosses. Parental seed lines may be deliberately inbred (e.g. by selfing or sib crossing). However, even if the parents are not deliberately inbred, selection within lines during line maintenance will ensure that some inbreeding occurs. Clonal parents will, of course, remain unchanged and highly heterozygous.

Whether a synthetic can go straight from the parental seed production plot to the farmer or must first undergo one or two cycles of multiplication depends on seed production and the scale of demand for seed. In practice, grasses and clovers are generally multiplied once or twice and are thus considerably removed from the original synthetic.

While mass selection is sometimes used, progeny testing is generally preferred for polycrosses, because of their operational simplicity and obvious relevance to the objective, namely exploitation of general combining ability in a synthetic.

The number of parental lines or clones that enter a synthetic vary widely. In practice, numbers of parental lines range from 10 to several hundred, with 100-200 being the average. Broad based synthetics formed from 100 or more clones would be expected to be more stable during seed multiplication than narrow based synthetics.

Pedigreed Varieties

A pedigreed variety is a superior genotype developed from selection of individual plants out of a segregating population followed by propagation and seed increase of self pollinated offspring and careful testing of the genotype over several generations. This is an open pollinated method that works well with naturally self pollinating species. This method can be used in combination with mass selection in variety development. Variations in pedigree and mass selection in combination are the most common methods for generating varieties in self pollinated crops.

Hybrids

A hybrid is an individual plant resulting from a cross between parents of differing genotypes. Commercial hybrids are now used extensively in many crops, including corn (maize), sorghum, sugarbeet, sunflower and broccoli. Hybrids can be formed in a number of different ways, including by crossing two parents directly (single cross hybrids), by crossing a single cross hybrid with another parent (three-way or triple cross hybrids), or by crossing two different hybrids (four-way or double cross hybrids).

Pedigree Selection

Pedigree breeding is used commonly for the improvement of self-pollinating crops or inbred lines of cross-pollinating crops. Two parents possessing favorable, complementary traits are crossed to produce an F_1 . An F_2 population is produced by selfing one or several F_1 s or by intercrossing two F_1 s (sib mating). The dihaploid breeding method or other ploidy reduction techniques could also be used. Selection of the best individuals is usually begun in the F_2 population; then, beginning in the F_3 , the best individuals in the best families are selected. Replicated testing of families, or hybrid combinations involving individuals of these families, often follows in the F_4 generation to improve the effectiveness of selection for traits with low heritability. At an advanced stage of inbreeding (i.e., F_6 and F_7), the best lines or mixtures of phenotypically similar lines are tested for potential release of new cultivars. Similarly, the development of new cultivars through the dihaploid system requires the selection of the cultivars followed by two to five years of testing in replicated plots.

Backcross Breeding

Backcross breeding has been used to transfer genes for a simply inherited, highly heritable trait into a desirable homozygous cultivar or inbred line which is the recurrent parent. The source of the trait to be transferred is called the donor parent. The resulting plant is expected to have the attributes of the recurrent parent (e.g., cultivar) and the desirable trait transferred from the donor parent. After the initial cross, individuals possessing the phenotype of the donor parent are selected and repeatedly crossed (backcrossed) to the recurrent parent. The resulting plant is expected to have the attributes of the recurrent parent (e.g., cultivar) and the desirable trait transferred from the donor parent.

Bulk Segregation Analysis (BSA)

BSA, a.k.a. bulked segregation analysis, or bulk segregant analysis, is a method described by Michelmore et al. (Michelmore et al., 1991, Identification of markers linked to disease-resistance genes by bulked segregant analysis: a rapid method to detect markers in specific genomic regions by using segregating populations. *Proceedings of the National Academy of Sciences, USA*, 99:9828-9832) and Quarrie et al. (Quarrie et al., 1999, *Journal of Experimental Botany*, 50(337):1299-1306).

For BSA of a trait of interest, parental lines with certain different phenotypes are chosen and crossed to generate F_2 ,

doubled haploid or recombinant inbred populations with QTL analysis. The population is then phenotyped to identify individual plants or lines having high or low expression of the trait. Two DNA bulks are prepared, one from the individuals having one phenotype (e.g., resistant to virus), and the other from the individuals having reversed phenotype (e.g., susceptible to virus), and analyzed for allele frequency with molecular markers. Only a few individuals are required in each bulk (e.g., 10 plants each) if the markers are dominant (e.g., RAPDs). More individuals are needed when markers are co-dominant (e.g., RFLPs). Markers linked to the phenotype can be identified and used for breeding or QTL mapping.

Targeting Induced Local Lesions in Genomes (TILLING®)

Breeding schemes of the present application can include crosses with TILLING® plant cultivars. TILLING® is a method in molecular biology that allows directed identification of mutations in a specific gene. TILLING® was introduced in 2000, using the model plant *Arabidopsis thaliana*. TILLING® has since been used as a reverse genetics method in other organisms such as zebrafish, corn, wheat, rice, soybean, tomato and *Brassica rapa* var. *nipposinica*.

The method combines a standard and efficient technique of mutagenesis with a chemical mutagen (e.g., Ethyl methanesulfonate (EMS)) with a sensitive DNA screening-technique that identifies single base mutations (also called point mutations) in a target gene. EcoTILLING is a method that uses TILLING® techniques to look for natural mutations in individuals, usually for population genetics analysis (see Comai, et al., 2003 The Plant Journal 37, 778-786; Gilchrist et al. 2006 Mol. Ecol. 15, 1367-1378; Mejlhede et al. 2006 Plant Breeding 125, 461-467; Nieto et al. 2007 BMC Plant Biology 7, 34-42, each of which is incorporated by reference hereby for all purposes). DEcoTILLING is a modification of TILLING® and EcoTILLING which uses an inexpensive method to identify fragments (Garvin et al., 2007, DEcoTILLING: An inexpensive method for SNP discovery that reduces ascertainment bias. Molecular Ecology Notes 7, 735-746).

The TILLING® method relies on the formation of heteroduplexes that are formed when multiple alleles (which could be from a heterozygote or a pool of multiple homozygotes and heterozygotes) are amplified in a PCR, heated, and then slowly cooled. A "bubble" forms at the mismatch of the two DNA strands (the induced mutation in TILLING® or the natural mutation or SNP in EcoTILLING), which is then cleaved by single stranded nucleases. The products are then separated by size on several different platforms.

More detailed description on methods and compositions on TILLING® can be found in U.S. Pat. No. 5,994,075, US 2004/0053236 A1, WO 2005/055704, and WO 2005/048692, each of which is hereby incorporated by reference for all purposes.

Thus in some embodiments, the breeding methods of the present disclosure include breeding with one or more TILLING® plant lines with one or more identified mutations.

Mutation Breeding

Mutation breeding is another method of introducing new traits into plants. Mutations that occur spontaneously or are artificially induced can be useful sources of variability for a plant breeder. The goal of artificial mutagenesis is to increase the rate of mutation for a desired characteristic. Mutation rates can be increased by many different means or mutating agents including temperature, long-term seed storage, tissue culture conditions, radiation (such as X-rays,

Gamma rays, neutrons, Beta radiation, or ultraviolet radiation), chemical mutagens (such as base analogs like 5-bromo-uracil), antibiotics, alkylating agents (such as sulfur mustards, nitrogen mustards, epoxides, ethyleneamines, sulfates, sulfonates, sulfones, or lactones), azide, hydroxylamine, nitrous acid or acridines. Once a desired trait is observed through mutagenesis the trait may then be incorporated into existing germplasm by traditional breeding techniques. Details of mutation breeding can be found in W. R. Fehr, 1993, Principles of Cultivar Development, Macmillan Publishing Co. New breeding techniques such as the ones involving the uses of Zinc Finger Nucleases or oligonucleotide directed mutagenesis shall also be used to generate genetic variability and introduce new traits into varieties.

In some embodiments, the mutation breeding comprises a single locus conversion. A single locus may contain several genes and/or transgenes, such as a transgene for disease resistance that, in the same expression vector, also contains a transgene for herbicide resistance. The gene for herbicide resistance may be used as a selectable marker and/or as a phenotypic trait. A single locus conversion of site specific integration system allows for the integration of multiple genes at the converted locus. A single locus conversion also allows for making one or more site specific changes to the plant genome. In some embodiments, the single locus conversion is performed by genome editing, a.k.a. genome editing with engineered nucleases (GEEN). In some embodiments, the genome editing comprises using one or more engineered nucleases. In some embodiments, the engineered nucleases include, but are not limited to Zinc finger nucleases (ZFNs), Transcription Activator-Like Effector Nucleases (TALENs), the CRISPR/Cas system, and engineered meganuclease re-engineered homing endonucleases. In some embodiments, the single locus conversion changes one or several nucleic acids of the plant genome.

Double Haploids and Chromosome Doubling

One way to obtain homozygous plants without the need to cross two parental lines followed by a long selection of the segregating progeny, and/or multiple back-crossings is to produce haploids and then double the chromosomes to form doubled haploids. Haploid plants can occur spontaneously, or may be artificially induced via chemical treatments or by crossing plants with inducer lines (Seymour et al. 2012, PNAS vol 109, pg 4227-4232; Zhang et al., 2008 Plant Cell Rep. December 27(12) 1851-60). The production of haploid progeny can occur via a variety of mechanisms which can affect the distribution of chromosomes during gamete formation. The chromosome complements of haploids sometimes double spontaneously to produce homozygous doubled haploids (DHs). Mixoploids, which are plants which contain cells having different ploidies, can sometimes arise and may represent plants that are undergoing chromosome doubling so as to spontaneously produce doubled haploid tissues, organs, shoots, floral parts or plants. Another common technique is to induce the formation of double haploid plants with a chromosome doubling treatment such as colchicine (El-Hennawy et al., 2011 Vol 56, issue 2 pg 63-72; Doubled Haploid Production in Crop Plants 2003 edited by Maluszynski ISBN 1-4020-1544-5). The production of doubled haploid plants yields highly uniform cultivars and is especially desirable as an alternative to sexual inbreeding of longer-generation crops. By producing doubled haploid progeny, the number of possible gene combinations for inherited traits is more manageable. Thus,

an efficient doubled haploid technology can significantly reduce the time and the cost of inbred and cultivar development.

Protoplast Fusion

In another method for breeding plants, protoplast fusion can also be used for the transfer of trait-conferring genomic material from a donor plant to a recipient plant. Protoplast fusion is an induced or spontaneous union, such as a somatic hybridization, between two or more protoplasts (cells of which the cell walls are removed by enzymatic treatment) to produce a single bi- or multi-nucleate cell. The fused cell that may even be obtained with plant species that cannot be interbred in nature is tissue cultured into a hybrid plant exhibiting the desirable combination of traits.

Tissue Culture

As used herein, the term "tissue culture" indicates a composition comprising isolated cells of the same or a different type or a collection of such cells organized into parts of a plant. Exemplary types of tissue cultures are protoplasts, calli, plant clumps, and plant cells that can generate tissue culture that are intact in plants or parts of plants, such as embryos, pollen, flowers, seeds, leaves, stems, roots, root tips, anthers, pistils, meristematic cells, axillary buds, ovaries, seed coat, endosperm, hypocotyls, cotyledons and the like. Means for preparing and maintaining plant tissue culture are well known in the art. By way of example, a tissue culture comprising organs has been used to produce regenerated plants. U.S. Pat. Nos. 5,959,185, 5,973,234, and 5,977,445 describe certain techniques, the disclosures of which are incorporated herein by reference.

This invention is further illustrated by the following examples which should not be construed as limiting. The contents of all references, patents and published patent applications cited throughout this application, as well as the Figures and the Sequence Listing, are incorporated herein by reference.

Example 1

Generating Grain With 1, 2, or 3 Doses of the wxS Allele

Because of the triploid nature of the corn endosperm, wherein two polar nuclei from the maternal parent are fertilized by one sperm cell from the paternal parent, three genotypic combinations are possible. Using a wxSwxSsu2su2 plant as a female, pollinated by a wxwxsu2su2 male results in 'dose-2' or wxSwxSwxsu2su2su2 grain. Using the wxSwxSsu2su2 as a male, pollinating a wxwxsu2su2 female results in 'dose-1' or wxSwwxwsu2su2su2 grain. Self pollination of wxSwxSsu2su2 results in 'dose-3' or wxSwxSwxSsu2su2su2 grain.

wxS dosage	Endosperm Genotype
(1 wxS)	wxSwwxwsu2su2su2
(2 wxS)	wxSwxSwxsu2su2su2
(3 wxS)	wxSwxSwxSsu2su2su2
starch from wxwxwsu2su2su2 seed (0 wxS)	wxwxwsu2su2su2
AMIOCA™ starch (0 wxS)	wxwxwsu2su2su2
starch from wxwxwsu2su2su2 (0 wxS)	wxwxwsu2su2su2

Example 2

Viscosity of Starch Using a Rapid Visco Amylograph (RVA)

The viscosity of maize starch derived from plants containing 0, 1, 2, and 3 doses of the wxS allele in the recessive sugary-2 mutant plants was determined using a Rapid Visco Amylograph (Newport Scientific, Eden Prairie, Minn.). The AMIOCA™ starch and wxwxwsu2su2su2 starch are included as checks. The genotype of endosperms from which the starch was isolated is summarized in the table below. The amylose content was measured using Megazyme® amylose kit. The amylose content of the starch increases with the increase of GBSSI activity. The amylose contents of wxwxwsu2su2su2 starch (7.4%), AMIOCA™ starch (7.1%), and wxwxwsu2su2su2 starch (6.1%) were lower than Waxy Stonor (12.8%) and wxwxwsu2su2su2 starch with 1 to 3 doses of wxS gene in the endosperm (8.2-14.6%). The amylose content of wxwxwsu2su2su2 starch (7.4%, 8.2%, 13.8%, and 14.6%) increased with the increase of wxS dosage (0, 1, 2, and 3 doses of wxS, respectively). The results suggested that the introduction of wxS gene to wxwxwsu2su2su2 slightly increased amylose content of the starch.

Sample	Amylose (%)
Q3017a (1 wxS)	8.2 ± 0.1
Q8412a (2 wxS)	13.8 ± 0.0
Q7738a (3 wxS)	14.6 ± 0.1
wxwxwsu2su2su2 starch (0 wxS)	7.4 ± 0.1
K5594a (Waxy Stonor)	12.8 ± 0.1
AMIOCA™ starch	7.1 ± 0.0
wxwxwsu2su2su2 starch	6.1 ± 0.1

Pasting property of waxy corn starch was analyzed using a Rapid Visco-Analyzer (RVA, Newport Scientific, Sydney, Australia) using "Standard 2" profile method. A suspension (28.0 g) containing 8% starch (w/w, dry starch basis, dsb) was equilibrated at 50° C. for 1 min, heated to 95° C. at a rate of 6° C./min, held at 95° C. for 5 min, and then cooled to 50° C. at a rate of 6° C./min. The rotating speed for the paddle was 160 rpm except that 960 rpm was used for the first 10 s. The results are shown in FIG. 1 and FIG. 2. The existing wxwxwsu2su2su2 starch had the lowest pasting temperature, peak viscosity, and final viscosity. After introduction of wxS gene to wxwxwsu2su2su2 starch, the starch samples (Q3017a [1 wxS], Q8412a [2 wxS], and Q7738a [3 wxS]) showed increased peak viscosity compared to the existing wxwxwsu2su2su2 starch (0 wxS). However, only the wxSwxwsu2su2su2 starch with one dose of wxS gene showed the peak viscosity much higher than Waxy Stonor, AMIOCA™ starch, and wxwxwsu2su2su2 starch.

The results indicate that improvement in pasting temperature will allow starch extraction via the wet-milling process.

Example 3

Freeze-Thaw Stability of Starch

The maize starch derived from plants containing 0, 1, 2, and 3 doses of the wxS allele in the recessive sugary-2 mutant plants was tested for freeze-thaw stability. AMIOCA™ starch and wxwxwsu2su2su2 starch were included as checks. Starch samples 8% solids were cooked in a water bath at 95° C. for 20 minutes. The starch slurry

23

was mixed with a glass rod for three minutes followed by quiescent heating for 17 minutes during cooking. Freeze-thaw stability of starch gel (8%, w/w) at neutral pH was analyzed using a Caron cycling freezer for five freeze-thaw cycles. In each cycle, the sample was frozen for 6 hours and thawed for 6 hours with 2 cycles/day. The samples were visually inspected for syneresis, both on the surface and when pressed, as well as for opacity and gelling changes. This freeze-thaw cycle was repeated. The results are shown in FIG. 3. After first freeze/thaw cycle, only Waxy Stonor starch gel showed high opacity, indicating high degree of starch retrogradation. The opacity of starch gel increased with the increase of freeze/thaw cycles. At third freeze/thaw cycle, the starch gels of AMIOCA™ starch, wxwxwxSU2su2su2 starch, and Waxy Stonor became very opaque, whereas the starch gels of Q3017a (1 wxS), Q8412a (2 wxS), Q7738a (3 wxS), and existing wxwxwxSU2su2su2 starch were still not opaque. After five freeze/thaw cycles, Q3017a (1 wxS), Q8412a (2 wxS), Q7738a (3 wxS), and existing wxwxwxSU2su2su2 starch become slightly opaque.

The results suggested that the slight increase of amylose in starch samples had little effect in the freeze/thaw stability. The Q3017a (1 wxS), Q8412a (2 wxS), Q7738a (3 wxS) starches had freeze/thaw stability better than wxwxwxSU2su2su2 starch and AMIOCA™ starch.

Example 4

Differential Scanning Calorimetry (DSC) Test

Thermal properties of maize starch derived from plants containing 0, 1, 2, and 3 doses of the wxS allele in the recessive sugary-2 mutant plants were determined using Differential Scanning Calorimetry (DSC). The AMIOCA™ starch and wxwxwxSU2su2su2 starch were included as checks. Differential Scanning Calorimetry (DSC) measures heat absorbed or given off by a sample in a controlled atmosphere at specified temperatures. DSC provides information about a specific heat and latent heat of samples as temperature rises, which indicates changes in the amorphous and crystalline structures. Data is recorded in terms of heat flow, and is presented in joules/gram (J/g) (Cassel, 2002). In the analysis of starch, starch gelatinization parameters such as peak onset, peak temperature, end of peak, and gelatinization enthalpy change information were collected. The results are shown in FIG. 4. wxSwxwxSU2su2su2 starch (Q3017a), wxSwxwxSU2su2su2 starch (Q8412a), wxSwxwxSU2su2su2 starch (Q7738a), and wxwxwxSU2su2su2 starch (0 wxS) showed lower gelatini-

24

zation temperatures than AMIOCA™ starch, Waxy Stonor, and wxwxwxSU2su2su2 starch, which is consistent with the better freeze/thaw stability of the starch isolated from endosperm of maize with one, two, and three doses of wxS. The wxSwxwxSU2su2su2 starch with one dose of wxS (Q3017a) had slightly higher onset gelatinization temperature than other hybrids (Q8412a [2 wxS], Q7738a [3 wxS], and wxwxwxSU2su2su2 starch), indicating that the Q3017a had better wet milling characteristics than other hybrids.

Example 5

Starch Granule

The average particle size of waxy maize starch derived from plants containing 1, 2, and 3 doses of the wxS allele in the recessive sugary-2 mutant plants was measured by laser light scattering. The AMIOCA™ starch and wxwxwxSU2su2su2 starch were included as checks. The results are shown in FIG. 5. In addition, the Scanning electron micrograph of each sample was provided in FIG. 6 to show granule morphology. The average starch granule sizes of Q3017a (11.4 μm), Q8412a (11.4 μm), and Q7738a (10.9 μm) were larger than the existing wxwxwxSU2su2su2 starch (10.4 μm), suggesting that the introduction of wxS gene to Waxy VO increased the starch granule size. The result was consistent with the SEM images (FIG. 5) showing that the introduction of wxS gene to wxwxwxSU2su2su2 starch increased the starch granule integrity.

Unless defined otherwise, all technical and scientific terms herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Although any methods and materials, similar or equivalent to those described herein, can be used in the practice or testing of the present disclosure, the preferred methods and materials are described herein. All publications, patents, and patent publications cited are incorporated by reference herein in their entirety for all purposes.

The publications discussed herein are provided solely for their disclosure prior to the filing date of the present application. Nothing herein is to be construed as an admission that the present disclosure is not entitled to antedate such publication by virtue of prior invention.

While the invention has been described in connection with specific embodiments thereof, it will be understood that it is capable of further modifications and this application is intended to cover any variations, uses, or adaptations of the invention following, in general, the principles of the invention and including such departures from the present disclosure as come within known or customary practice within the art to which the invention pertains and as may be applied to the essential features hereinbefore set forth and as follows in the scope of the appended claims.

Sequence Listing

```
SEQ ID NO: 1 Waxy Protein in maize
10      20      30      40      50
MAALATSQLV ATRAGLGVPD ASTFRRGAAQ GLRGARASAA ADTLSMR TSA
60      70      80      90      100
RAAPRHQQQA RRGGRFPSPV VCASAGMNVV FVGAEMAPWS KTGGGLDVLG
110     120     130     140     150
GLPPAMAANG HRVMVSPRY DQYKDAWDT S VVSEIKMGDG YETVRFFHCY
160     170     180     190     200
KRGVDRVFVD HPLFLERVWG KTEEKIYGPV AGTDYRDNQL RFSLLCQAAL
```

Sequence Listing

210	220	230	240	250
EAPRILSLNN	NPYFSGPYGE	DVVVFCNDWH	TGPLSCYLKS	NYQSHGIYRD
260	270	280	290	300
AKTAFCIHNI	SYQGRFAFSD	YPELNLPERF	KSSFDFIDGY	EKPVEGRKIN
310	320	330	340	350
WMKAGILEAD	RVLTVSPYYA	EELISGIARG	CELDNIMRLT	GITGIVNGMD
360	370	380	390	400
VSEWDPSRDK	YIAVKYDVST	AVEAKALNKE	ALQAEVGLPV	DRNIPLVAFI
410	420	430	440	450
GRLEEQKQPD	VMAAAIPQLM	EMVEDVQIVL	LGTGKKKFER	MLMSAEEKFP
460	470	480	490	500
GKVRVAVKFN	AALAHHIMAG	ADVLAVTSRF	EPCGLIQLQG	MRYGTPCACA
510	520	530	540	550
STGGLVDTHI	EGKTGFHMGR	LSVDCNVVEP	ADVKKVATTL	QRAIKVVGTP
560	570	580	590	600
AYEEMVRNCM	IQDLSWKGPA	KNWENVLLSL	GVAGGEPGVE	GEEIAPLAKE
NVAAP				
SEQ ID NO: 2 Sugary 2 Protein in maize				
10	20	30	40	50
MSSAAVSSSS	STFFLALASA	SPGGRRRARV	GSSPFHTGAS	LSFAFWAPPS
60	70	80	90	100
PPRAPRDAAL	VRAEAEAGGK	DAPPERSGDA	ARLPRARRNA	VSKRRDPLQP
110	120	130	140	150
VGRYSATGN	TARTGAASCQ	NAALADVEIK	SIVAAPPTSI	VKFPAPGYRM
160	170	180	190	200
ILPSGDIAP	TVLPAPKPLH	ESPAVDGDSN	GIAPPTVEPL	VQEATWDFKK
210	220	230	240	250
YIGFDEPDEA	KDDSRVGADD	AGSFEHYGDN	DSGPLAGENV	MNVIVVAAEC
260	270	280	290	300
SPWCKTGGLG	DVVGALPKAL	ARRGHRVMVV	VPRYGDYVEA	FDMGIRKYYK
310	320	330	340	350
AAGPVNYFHA	FIDGVDFVFI	DAPLFRHRQD	DIYGGSRQEI	MKRMILFCKV
360	370	380	390	400
AVEVPWHVPC	GGVCYGDGNL	VFIANDWHTA	LLPVYLKAYY	RDHGLMQYTR
410	420	430	440	450
SVLVIHNTAH	QGRGPVDEFP	YMDLPEHYLQ	HFELYDPVGG	EHANIFAAGL
460	470	480	490	500
KMADRVTTVS	RGYLWELKTV	EGGWGLHDII	RSNDWKINGI	VNGIDHQEWN
510	520	530	540	550
PKVDVHLRSD	GYTNYSLETL	DAGKRQCKAA	LQRELGLEVR	DDVPLLFIG
560	570	580	590	600
RLDGQKGVDI	IGDAMPWIAG	QDVQLVMLGT	GRADLERMLQ	HLEREHPNKV
610	620	630	640	650
RGWVGFSVPM	AHRITAGADV	LVMPSTRFPC	GLNQLYAMAY	GTVPVVHAVG
660	670	680	690	700
GLRDTVAFPD	PFS DAGLGWT	FDRAEANKLI	EALRHCLDTY	RNYEESWKS
710	720			
QARGMSQDLS	WDHAAELYED	VLVKAKYQW		

SEQUENCE LISTING

<160> NUMBER OF SEQ ID NOS: 2

<210> SEQ ID NO 1

<211> LENGTH: 605

<212> TYPE: PRT

<213> ORGANISM: Zea mays

<400> SEQUENCE: 1

```

Met Ala Ala Leu Ala Thr Ser Gln Leu Val Ala Thr Arg Ala Gly Leu
 1             5             10             15

Gly Val Pro Asp Ala Ser Thr Phe Arg Arg Gly Ala Ala Gln Gly Leu
          20             25             30

Arg Gly Ala Arg Ala Ser Ala Ala Ala Asp Thr Leu Ser Met Arg Thr
          35             40             45

Ser Ala Arg Ala Ala Pro Arg His Gln Gln Gln Ala Arg Arg Gly Gly
          50             55             60

Arg Phe Pro Ser Leu Val Val Cys Ala Ser Ala Gly Met Asn Val Val
          65             70             75             80

Phe Val Gly Ala Glu Met Ala Pro Trp Ser Lys Thr Gly Gly Leu Gly
          85             90             95

Asp Val Leu Gly Gly Leu Pro Pro Ala Met Ala Ala Asn Gly His Arg
          100            105            110

Val Met Val Val Ser Pro Arg Tyr Asp Gln Tyr Lys Asp Ala Trp Asp
          115            120            125

Thr Ser Val Val Ser Glu Ile Lys Met Gly Asp Gly Tyr Glu Thr Val
          130            135            140

Arg Phe Phe His Cys Tyr Lys Arg Gly Val Asp Arg Val Phe Val Asp
          145            150            155            160

His Pro Leu Phe Leu Glu Arg Val Trp Gly Lys Thr Glu Glu Lys Ile
          165            170            175

Tyr Gly Pro Val Ala Gly Thr Asp Tyr Arg Asp Asn Gln Leu Arg Phe
          180            185            190

Ser Leu Leu Cys Gln Ala Ala Leu Glu Ala Pro Arg Ile Leu Ser Leu
          195            200            205

Asn Asn Asn Pro Tyr Phe Ser Gly Pro Tyr Gly Glu Asp Val Val Phe
          210            215            220

Val Cys Asn Asp Trp His Thr Gly Pro Leu Ser Cys Tyr Leu Lys Ser
          225            230            235            240

Asn Tyr Gln Ser His Gly Ile Tyr Arg Asp Ala Lys Thr Ala Phe Cys
          245            250            255

Ile His Asn Ile Ser Tyr Gln Gly Arg Phe Ala Phe Ser Asp Tyr Pro
          260            265            270

Glu Leu Asn Leu Pro Glu Arg Phe Lys Ser Ser Phe Asp Phe Ile Asp
          275            280            285

Gly Tyr Glu Lys Pro Val Glu Gly Arg Lys Ile Asn Trp Met Lys Ala
          290            295            300

Gly Ile Leu Glu Ala Asp Arg Val Leu Thr Val Ser Pro Tyr Tyr Ala
          305            310            315            320

Glu Glu Leu Ile Ser Gly Ile Ala Arg Gly Cys Glu Leu Asp Asn Ile
          325            330            335

Met Arg Leu Thr Gly Ile Thr Gly Ile Val Asn Gly Met Asp Val Ser
          340            345            350

Glu Trp Asp Pro Ser Arg Asp Lys Tyr Ile Ala Val Lys Tyr Asp Val
          355            360            365

```

-continued

Ser Thr Ala Val Glu Ala Lys Ala Leu Asn Lys Glu Ala Leu Gln Ala
 370 375 380
 Glu Val Gly Leu Pro Val Asp Arg Asn Ile Pro Leu Val Ala Phe Ile
 385 390 395 400
 Gly Arg Leu Glu Glu Gln Lys Gly Pro Asp Val Met Ala Ala Ala Ile
 405 410 415
 Pro Gln Leu Met Glu Met Val Glu Asp Val Gln Ile Val Leu Leu Gly
 420 425 430
 Thr Gly Lys Lys Lys Phe Glu Arg Met Leu Met Ser Ala Glu Glu Lys
 435 440 445
 Phe Pro Gly Lys Val Arg Ala Val Val Lys Phe Asn Ala Ala Leu Ala
 450 455 460
 His His Ile Met Ala Gly Ala Asp Val Leu Ala Val Thr Ser Arg Phe
 465 470 475 480
 Glu Pro Cys Gly Leu Ile Gln Leu Gln Gly Met Arg Tyr Gly Thr Pro
 485 490 495
 Cys Ala Cys Ala Ser Thr Gly Gly Leu Val Asp Thr Ile Ile Glu Gly
 500 505 510
 Lys Thr Gly Phe His Met Gly Arg Leu Ser Val Asp Cys Asn Val Val
 515 520 525
 Glu Pro Ala Asp Val Lys Lys Val Ala Thr Thr Leu Gln Arg Ala Ile
 530 535 540
 Lys Val Val Gly Thr Pro Ala Tyr Glu Glu Met Val Arg Asn Cys Met
 545 550 555 560
 Ile Gln Asp Leu Ser Trp Lys Gly Pro Ala Lys Asn Trp Glu Asn Val
 565 570 575
 Leu Leu Ser Leu Gly Val Ala Gly Gly Glu Pro Gly Val Glu Gly Glu
 580 585 590
 Glu Ile Ala Pro Leu Ala Lys Glu Asn Val Ala Ala Pro
 595 600 605

<210> SEQ ID NO 2
 <211> LENGTH: 729
 <212> TYPE: PRT
 <213> ORGANISM: Zea mays

<400> SEQUENCE: 2

Met Ser Ser Ala Ala Val Ser Ser Ser Ser Thr Phe Phe Leu Ala
 1 5 10 15
 Leu Ala Ser Ala Ser Pro Gly Gly Arg Arg Arg Ala Arg Val Gly Ser
 20 25 30
 Ser Pro Phe His Thr Gly Ala Ser Leu Ser Phe Ala Phe Trp Ala Pro
 35 40 45
 Pro Ser Pro Pro Arg Ala Pro Arg Asp Ala Ala Leu Val Arg Ala Glu
 50 55 60
 Ala Glu Ala Gly Gly Lys Asp Ala Pro Pro Glu Arg Ser Gly Asp Ala
 65 70 75 80
 Ala Arg Leu Pro Arg Ala Arg Arg Asn Ala Val Ser Lys Arg Arg Asp
 85 90 95
 Pro Leu Gln Pro Val Gly Arg Tyr Gly Ser Ala Thr Gly Asn Thr Ala
 100 105 110
 Arg Thr Gly Ala Ala Ser Cys Gln Asn Ala Ala Leu Ala Asp Val Glu
 115 120 125
 Ile Lys Ser Ile Val Ala Ala Pro Pro Thr Ser Ile Val Lys Phe Pro
 130 135 140

-continued

Ala	Pro	Gly	Tyr	Arg	Met	Ile	Leu	Pro	Ser	Gly	Asp	Ile	Ala	Pro	Glu
145					150					155					160
Thr	Val	Leu	Pro	Ala	Pro	Lys	Pro	Leu	His	Glu	Ser	Pro	Ala	Val	Asp
				165					170					175	
Gly	Asp	Ser	Asn	Gly	Ile	Ala	Pro	Pro	Thr	Val	Glu	Pro	Leu	Val	Gln
			180					185					190		
Glu	Ala	Thr	Trp	Asp	Phe	Lys	Lys	Tyr	Ile	Gly	Phe	Asp	Glu	Pro	Asp
		195					200					205			
Glu	Ala	Lys	Asp	Asp	Ser	Arg	Val	Gly	Ala	Asp	Asp	Ala	Gly	Ser	Phe
		210				215					220				
Glu	His	Tyr	Gly	Asp	Asn	Asp	Ser	Gly	Pro	Leu	Ala	Gly	Glu	Asn	Val
225					230					235					240
Met	Asn	Val	Ile	Val	Val	Ala	Ala	Glu	Cys	Ser	Pro	Trp	Cys	Lys	Thr
			245						250					255	
Gly	Gly	Leu	Gly	Asp	Val	Val	Gly	Ala	Leu	Pro	Lys	Ala	Leu	Ala	Arg
		260						265					270		
Arg	Gly	His	Arg	Val	Met	Val	Val	Pro	Arg	Tyr	Gly	Asp	Tyr	Val	
		275					280					285			
Glu	Ala	Phe	Asp	Met	Gly	Ile	Arg	Lys	Tyr	Tyr	Lys	Ala	Ala	Gly	Pro
		290				295					300				
Val	Asn	Tyr	Phe	His	Ala	Phe	Ile	Asp	Gly	Val	Asp	Phe	Val	Phe	Ile
305				310					315						320
Asp	Ala	Pro	Leu	Phe	Arg	His	Arg	Gln	Asp	Asp	Ile	Tyr	Gly	Gly	Ser
			325						330					335	
Arg	Gln	Glu	Ile	Met	Lys	Arg	Met	Ile	Leu	Phe	Cys	Lys	Val	Ala	Val
		340						345					350		
Glu	Val	Pro	Trp	His	Val	Pro	Cys	Gly	Gly	Val	Cys	Tyr	Gly	Asp	Gly
		355					360					365			
Asn	Leu	Val	Phe	Ile	Ala	Asn	Asp	Trp	His	Thr	Ala	Leu	Leu	Pro	Val
		370				375					380				
Tyr	Leu	Lys	Ala	Tyr	Tyr	Arg	Asp	His	Gly	Leu	Met	Gln	Tyr	Thr	Arg
385				390						395					400
Ser	Val	Leu	Val	Ile	His	Asn	Ile	Ala	His	Gln	Gly	Arg	Gly	Pro	Val
			405					410						415	
Asp	Glu	Phe	Pro	Tyr	Met	Asp	Leu	Pro	Glu	His	Tyr	Leu	Gln	His	Phe
		420						425					430		
Glu	Leu	Tyr	Asp	Pro	Val	Gly	Gly	Glu	His	Ala	Asn	Ile	Phe	Ala	Ala
		435					440					445			
Gly	Leu	Lys	Met	Ala	Asp	Arg	Val	Val	Thr	Val	Ser	Arg	Gly	Tyr	Leu
		450				455					460				
Trp	Glu	Leu	Lys	Thr	Val	Glu	Gly	Gly	Trp	Gly	Leu	His	Asp	Ile	Ile
465				470						475					480
Arg	Ser	Asn	Asp	Trp	Lys	Ile	Asn	Gly	Ile	Val	Asn	Gly	Ile	Asp	His
			485					490						495	
Gln	Glu	Trp	Asn	Pro	Lys	Val	Asp	Val	His	Leu	Arg	Ser	Asp	Gly	Tyr
		500						505						510	
Thr	Asn	Tyr	Ser	Leu	Glu	Thr	Leu	Asp	Ala	Gly	Lys	Arg	Gln	Cys	Lys
		515					520					525			
Ala	Ala	Leu	Gln	Arg	Glu	Leu	Gly	Leu	Glu	Val	Arg	Asp	Asp	Val	Pro
		530				535						540			
Leu	Leu	Gly	Phe	Ile	Gly	Arg	Leu	Asp	Gly	Gln	Lys	Gly	Val	Asp	Ile
545				550						555					560

-continued

Ile	Gly	Asp	Ala	Met	Pro	Trp	Ile	Ala	Gly	Gln	Asp	Val	Gln	Leu	Val
				565					570					575	
Met	Leu	Gly	Thr	Gly	Arg	Ala	Asp	Leu	Glu	Arg	Met	Leu	Gln	His	Leu
			580					585					590		
Glu	Arg	Glu	His	Pro	Asn	Lys	Val	Arg	Gly	Trp	Val	Gly	Phe	Ser	Val
			595				600					605			
Pro	Met	Ala	His	Arg	Ile	Thr	Ala	Gly	Ala	Asp	Val	Leu	Val	Met	Pro
			610			615					620				
Ser	Arg	Phe	Glu	Pro	Cys	Gly	Leu	Asn	Gln	Leu	Tyr	Ala	Met	Ala	Tyr
			625		630					635				640	
Gly	Thr	Val	Pro	Val	Val	His	Ala	Val	Gly	Gly	Leu	Arg	Asp	Thr	Val
			645					650						655	
Ala	Pro	Phe	Asp	Pro	Phe	Ser	Asp	Ala	Gly	Leu	Gly	Trp	Thr	Phe	Asp
			660					665					670		
Arg	Ala	Glu	Ala	Asn	Lys	Leu	Ile	Glu	Ala	Leu	Arg	His	Cys	Leu	Asp
			675			680						685			
Thr	Tyr	Arg	Asn	Tyr	Glu	Glu	Ser	Trp	Lys	Ser	Leu	Gln	Ala	Arg	Gly
			690			695					700				
Met	Ser	Gln	Asp	Leu	Ser	Trp	Asp	His	Ala	Ala	Glu	Leu	Tyr	Glu	Asp
			705		710					715				720	
Val	Leu	Val	Lys	Ala	Lys	Tyr	Gln	Trp							
					725										

What is claimed is:

1. A starch comprising:
an amylose content of from about 2 wt % to about 20 wt % as measured using an amylose/amylopectin assay kit; and
an aqueous starch pasting temperature that is at least about 5% greater than the starch pasting temperature of a control starch as measured using RVA analysis, wherein, the starch is derived from a waxy-sugary 2 corn plant comprising at least one wxS allele in the endosperm.
2. The starch of claim 1, wherein the amylose content is from about 8 wt % to about 15 wt %.
3. The starch of claim 1, wherein the aqueous pasting temperature is from about 5% to about 14% greater than that of the control starch.
4. The starch of claim 1, further comprising an enthalpy change of crystallinity of at least about 30% greater than an enthalpy change of crystallinity of the control starch as measured using RVA analysis.
5. The starch of claim 4, wherein the enthalpy change of crystallinity is from about 70% to about 200% greater than an enthalpy change of crystallinity of the control starch as measured using a differential scanning calorimeter (DSC).
6. The starch of claim 1, further comprising a distribution of starch granule sizes, wherein the distribution of starch granule sizes comprises at least about 40% fewer granules less than 5 microns compared to a proportion of granules less than 5 microns of the control starch as observed using a scanning electron microscope.
7. The starch of claim 1, wherein the starch is a granule which exhibits increased integrity compared to the control starch as observed using a scanning electron microscope.
8. The starch of claim 1, wherein the starch is capable of withstanding at least three freeze-thaw cycles.
9. The starch of claim 1, wherein the starch is capable of withstanding at least three to at least five freeze-thaw cycles.

30

10. The starch of claim 1, wherein the waxy-sugary 2 corn plant is generated by cross pollination of a first corn plant having the genotype of wxwxsu2su2, and a second corn plant having the genotype of wxSwxSsu2su2, or the genotype of wxSwxsu2su2.

35

11. The starch of claim 1, wherein the endosperm of the waxy-sugary 2 corn plant has one dose, two doses, or three doses of the wxS allele, and the endosperm has the genotype of wxSwxwxsu2su2su2, wxSwxSwxsu2su2su2, or wxSwxSwxSsu2su2su2.

40

12. A method for improving at least one characteristics in a pasting profile of a starch comprising introducing at least one wxS allele into the endosperm of a waxy corn plant, wherein the resultant starch derived from the waxy corn plant comprising at least one wxS allele in the endosperm comprises an aqueous starch pasting temperature which is at least about 5% greater than a starch pasting temperature of a control starch as measured using RVA analysis, thereby improving at least one characteristic of the pasting profile of the starch compared a control starch.

50

13. A double-mutant corn plant, wherein the corn plant is homozygous recessive for the starch synthase IIa (su2) gene, and either homozygous or heterozygous for a mutated Granule-bound starch synthase I (GBSSI) gene, wherein the mutated GBSSI gene has less GBSSI activity than the wild type GBSSI gene (Wx), but has higher GBSSI activity than the recessive, loss-of-function GBSSI gene (wx).

55

14. The double-mutant corn plant of claim 13, wherein the mutated GBSSI gene is the wx-Stonor (wxS) allele.

60

15. The double-mutant corn plant of claim 14, wherein the plant is generated by cross pollination of a first corn plant having the genotype of wxwxsu2su2, and a second corn plant having the genotype of wxSwxSsu2su2, or the genotype of wxSwxsu2su2.

65

* * * * *

Notificación de Concesión	Solicitud No. 15/135,106	Solicitante(s) JIANG <i>et al.</i>	
	Examinador COHEN, STEFANIE J.	Unidad de Arte 1732	Estado AIA (Primer Inventor en Presentar) Sí

-- La FECHA DE ENVIO de esta comunicación aparece en la hoja de cubierta con la dirección de correspondencia--

Todas las reivindicaciones son concedidas, TRÁMITE SOBRE EL FONDO ESTÁ (O PERMANECE) CERRADO en esta solicitud. Si no se incluye acá (o enviada previamente), una Notificación de Concesión (PTOL-85) u otra comunicación adecuada será enviada en su debido momento. **ESTA NOTIFICACION DE CONCESION NO ES UNA CONCESION DE DERECHOS DE PATENTE.** Esta solicitud está sujeta a retiro de emisión a la iniciativa de la Oficina o a petición del solicitante. Ver 37 CFR 1.313 y MPEP 1308.

1. Esta comunicación responde a 10/3/17.

Una declaración bajo **37 CFR 1.130(b)** se presentó el_____.

2. Se hizo una elección por el solicitante en respuesta a un requerimiento de restricción definido durante la entrevista el ____; el requerimiento de restricción y la elección se han incorporado en esta acción.

3. Las reivindicaciones concedidas son 1-15. Como resultado de las reivindicaciones aceptadas, usted puede ser elegible del beneficio del programa de **Autopista de Trámite de Patente** en una oficina de propiedad intelectual participante para la correspondiente solicitud. Para más información, por favor ver http://www.uspto.gov/patents/init_events/pph/index.jsp o enviar una pregunta a PPHfeedback@uspto.gov.

4. Se reconoce una reivindicación de prioridad extranjera bajo 35 USC sección 119(a)-(d) o (f).

Copias certificadas:

a) Todas b) Algunas c) Ninguna de las:

- 1. Copias certificadas de los documentos de prioridad se han recibido.
- 2. Copias certificadas de los documentos de prioridad se han recibido en la Solicitud No. _____.
- 3. Copias de las copias certificadas de los documentos de prioridad se han recibido en esta solicitud de fase nacional de la Oficina Internacional (Regla PCT 17.2 (a)).

* Copias certificadas no recibidas: _____.

El Solicitante tiene TRES MESES A PARTIR DE LA “FECHA DE ENVIO” de esta comunicación para presentar una respuesta que cumpla con los requerimientos indicados anteriormente. Falla en cumplir a tiempo resultará en el ABANDONO de esta solicitud.

ESTE PERIODO DE TRES MESES NO ES EXTENSIBLE

5. Se deben presentar FIGURAS CORREGIDAS (como “hojas de reemplazo”).
Incluyendo cambios requeridos por la Enmienda/Comentario del Examinador adjunto o en la Acción Oficial de Papel No./Fecha de Envío ____.

Los indicios de identificación tal como el número de solicitud (ver 37 CFR 1.84(c)) se deben escribir en las figuras en el frente (no atrás) de cada hoja. Las hojas de reemplazo deben estar etiquetadas como tal en el encabezado de acuerdo con 37 CFR 1.121(d).

6. Se debe presentar DEPOSITO DE y/o INFORMACIÓN acerca del depósito de MATERIAL BIOLOGICO. Note el comentario del Examinador adjunto con relación al REQUERIMIENTO PARA EL DEPOSITO DE MATERIAL BIOLOGICO.

Adjunto(s)

- | | |
|--|--|
| 1. Notificación de Referencias Citadas (PTO-892) | 5. Enmienda/Comentario del Examinador |
| 2. Declaraciones de Divulgación de Información (PTO/SB/08), Papel No./Fecha Envío <u>8/8/17</u> , <u>7/21/16</u> | 6. Declaración del Examinador de Razones para Concesión. |
| 3. Comentario del Examinador con Relación a Requerimiento para Depósito de Material Biológico | 7. Otro ____. |
| 4. Resumen de Entrevista (PTO-413), Papel No./Fecha Envío ____. | |

/MELVIN C MAYES/
Examinador Principal, Unidad de Arte 1732

Elección/Restricciones

Esta solicitud está en condiciones de concesión, excepto por la presencia de las reivindicaciones 15-33 dirigidas a invenciones no elegidas sin objeción. En consecuencia, las reivindicaciones 15-33 han sido canceladas.

RAZONES PARA LA CONCESIÓN

La siguiente es una declaración del Examinador de razones para permitir:

El documento US4767849 enseña un almidón del genotipo wxsh1. La Tabla 1 divulga la extracción del almidón de un núcleo de maíz wxsh1. La muestra A tiene un contenido de amilosa de 0,6% y la muestra B tiene un contenido de amilosa de ,8% que está fuera del intervalo de amilosa reivindicado según la reivindicación 1.

El documento US4615888 enseña pan que contiene almidón de genotipo wxsu2 pero no enseña un contenido de amilosa.

Varagona et al. (citado en el informe de búsqueda EP32289635) enseña un empalme alternativo inducido por la inserción de retrotransposones en el gen ceroso del maíz, pero no enseña un contenido de amilosa.

WO03033540 (citado en el informe de búsqueda EP32289635, correspondiente a USPGPUB 20060216402) enseña almidón de plantas de maíz y/u otras plantas que producen órganos que almacenan almidón que contienen almidón bajo en amilosa que tiene un contenido de amilosa entre 1,5% y 15%.

El documento US20060216402 enseña el almidón de la presente invención, por lo tanto, puede verse como un almidón de amilosa reducido o un tipo especial de almidón ceroso con nuevas propiedades elásticas. El almidón de la presente invención se denomina aquí como almidón ceroso-E o wx-E, para enfatizar esta propiedad elástica no disponible anteriormente con almidón céreo conocido.

El Párrafo 220 de la PGPUB enseña que todos los almidones cerosos-E y los almidones cerosos tenían una viscosidad pico significativamente más alta y viscosidad final más alta que los almidones normales, lo que indica que todos los

almidones cerosos y cerosos-E sobresalen en el desarrollo de la viscosidad a relativamente bajas concentraciones de almidón. Además, todos los almidones cerosos y cerosos-E tenían una temperatura de adhesión inferior a la de los almidones normales, lo que indica que todos estos almidones comienzan a acumular viscosidad antes que los almidones normales.

Las Tablas 1 y 2 muestran numerosos alelos mutantes dominantes que producen una proteína GBSS activa (Tabla 1) y existen alelos mutantes recesivos que producen almidón ceroso (Tabla 2).

Aunque esta referencia enseña un alelo wx-E, esta referencia no enseña un alelo wxS. Además, esta referencia no enseña un almidón derivado de una planta de maíz waxy-sugary-2 que comprende al menos un alelo wxS en el endospermo, que tiene una temperatura de adhesión de almidón acuosa que es al menos 5% mayor que la temperatura de adhesión de almidón de un almidón de control.

Wessler enseña una base molecular de mutaciones en el locus ceroso del maíz.

Wessler, Tabla 1, enseña un alelo wx-stonor.

Aunque esta referencia enseña el uso del alelo wx-stonor, esta referencia no enseña un almidón derivado de una planta de maíz waxy-sugary-2 que comprende al menos un alelo wxS en el endospermo, que tiene una temperatura de adhesión de almidón acuosa que es al menos 5% mayor que la temperatura de adhesión de almidón de un almidón de control y, además, no enseña un contenido de amilosa en el almidón.

Cualquier comentario considerado necesario por el solicitante debe ser presentado no más allá del pago de la tasa de expedición y, para evitar retardos, debería preferiblemente acompañar la tasa de expedición. Dichas presentaciones deberían ser etiquetadas claramente como "Comentarios sobre la Declaración de las Razones para la Concesión".

Cualquier duda relacionada con esta comunicación o comunicaciones anteriores del examinador se debe dirigir a STEFANIE COHEN cuyo número de teléfono es (571)270-5836. El examinador puede ser encontrado normalmente de Lunes a Jueves 9:30 am – 6:00 p.m.

Las entrevistas con el examinador están disponibles por teléfono, en persona y por videoconferencia usando una herramienta de colaboración basada en la web suministrada por la USPTO. Para programar una entrevista, se recomienda al solicitante utilizar la Solicitud de Entrevista Automatizada (AIR) de la USPTO en <http://www.uspto.gov/interviewpractice>.

Si los intentos por encontrar al examinador por teléfono son fallidos, el supervisor del examinador, Melvin Curtis Mayes se puede encontrar en 5712721234. El número de fax para la organización donde esta solicitud o procedimiento es asignado es 571-273-8300.

La información relacionada con el estado de una solicitud se puede obtener del sistema de Recuperación de Información de Solicitud de Patente (PAIR). La información de estado para solicitudes publicadas se puede obtener del PAIR Privado o el PAIR Público. La información de estado para solicitudes sin publicar está disponible a través del PAIR Privado únicamente. Para mayor información acerca del sistema PAIR, ver <http://pair-direct.uspto.gov>. Si tiene preguntas sobre el acceso al sistema PAIR Privado, contacte el Centro de Negocio Electrónico (EBC) a 866-217-9197 (totalmente gratis). Si desea asistencia de un Representante de Servicio al Cliente de la USPTO o acceso al sistema de información automatizado, llame 800-786-9199 (EN USA O CANADA) o 571-272-1000.

1/4/17

/STEFANIE COHEN/

Examinador, Unidad de Arte 1732

/MELVIN C MAYES/

Supervisor Examinador de Patentes, Unidad de Arte 1732

B2)

1. A starch comprising:
 - an amylose content of from about 2 wt % to about 20 wt % as measured using an amylose/amylopectin assay kit; and
 - an aqueous starch pasting temperature that is at least about 5% greater than the starch pasting temperature of a control starch as measured using RVA analysis,wherein, the starch is derived from a waxy-sugary 2 corn plant comprising at least one wxS allele in the endosperm.
2. The starch of claim 1, wherein the amylose content is from about 8 wt % to about 15 wt %.
3. The starch of claim 1, wherein the aqueous pasting temperature is from about 5 % to about 14 % greater than that of the control starch.
4. The starch of claim 1, further comprising an enthalpy change of crystallinity of at least about 30 % greater than an enthalpy change of crystallinity of the control starch as measured using RVA analysis.
5. The starch of claim 4, wherein the enthalpy change of crystallinity is from about 70 % to about 200 % greater than an enthalpy change of crystallinity of the control starch as measured using a differential scanning calorimeter (DSC).
6. The starch of claim 1, further comprising a distribution of starch granule sizes, wherein the distribution of starch granule sizes comprises at least about 40 % fewer granules less than 5 microns compared to a proportion of granules less than 5 microns of the control starch as observed using a

scanning electron microscope.

7. The starch of claim 1, wherein the starch is a granule which exhibits increased integrity compared to the control starch as observed using a scanning electron microscope.
8. The starch of claim 1, wherein the starch is capable of withstanding at least three freeze-thaw cycles.
9. The starch of claim 1, wherein the starch is capable of withstanding at least three to at least five freeze-thaw cycles.
10. The starch of claim 1, wherein the waxy-sugary 2 corn plant is generated by cross pollination of a first corn plant having the genotype of $wxwx su2 su2$, and a second corn plant having the genotype of $wxS wxS su2 su2$, or the genotype of $wxS wx su2 su2$.
11. The starch of claim 1, wherein the endosperm of the waxy-sugary 2 corn plant has one dose, two doses, or three doses of the wxS allele, and the endosperm has the genotype of $wxS wx wx su2 su2 su2$, $wxS wx wx su2 su2 su2$, or $wxS wx wx S su2 su2 su2$.
12. A method for improving at least one characteristics in a pasting profile of a starch comprising introducing at least one wxS allele into the endosperm of a waxy corn plant, wherein the resultant starch derived from the waxy corn plant comprising at least one wxS allele in the endosperm comprises an aqueous starch pasting temperature which is at least about 5% greater than a starch pasting temperature of a control starch as measured using RVA analysis, thereby improving at least one characteristic of the pasting profile of the starch compared a control starch

13. A double-mutant corn plant, wherein the corn plant is homozygous recessive for the starch synthase IIa (su2) gene, and either homozygous or heterozygous for a mutated Granule-bound starch synthase I (GBSSI) gene, wherein the mutated GBSSI gene has less GBSSI activity than the wild type GBSSI gene (Wx), but has higher GBSSI activity than the recessive, loss-of-function GBSSI gene (wx).
14. The double-mutant corn plant of claim 13, wherein the mutated GBSSI gene is the wx-Stonor (wxS) allele.
15. The double-mutant corn plant of claim 14, wherein the plant is generated by cross pollination of a first corn plant having the genotype of wxwxsu2su2, and a second corn plant having the genotype of wxSwxSsu2su2, or the genotype of wxSwxsu2su2.

TRADUCCIÓN DE LAS REIVINDICACIONES OTORGADAS EN ESTADOS
UNIDOS SOLICITUD No. 15/135106 (PATENTE US No. 9988641 B2)

1. Un almidón, caracterizado porque comprende:
 - un contenido de amilosa de aproximadamente 2% en peso a aproximadamente 20% en peso como es medido usando un kit de ensayo de amilosa/amilopectina; y
 - una temperatura de adhesión de almidón acuosa que es menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA,en donde, el almidón se deriva de una planta de maíz waxy-sugary-2 que comprende al menos un alelo wxS en el endospermo.
2. El almidón de conformidad con la reivindicación 1, caracterizado porque el contenido de amilosa es de aproximadamente 8% en peso a aproximadamente 15% en peso.
3. El almidón de conformidad con la reivindicación 1, caracterizado porque la temperatura de adhesión acuosa es de aproximadamente 5% a aproximadamente 14% mayor que aquella del almidón de control.
4. El almidón de conformidad con la reivindicación 1, caracterizado porque además comprende un cambio de entalpía de cristalinidad de al menos aproximadamente 30% mayor que un cambio de entalpía de cristalinidad del almidón de control como es medido usando el análisis RVA.
5. El almidón de conformidad con la reivindicación 4, caracterizado porque el cambio de entalpía de la cristalinidad es de aproximadamente 70% a aproximadamente 200% mayor que un cambio de entalpía de

cristalinidad del almidón de control como es medido usando un calorímetro de barrido diferencial (DSC).

6. El almidón de conformidad con la reivindicación 1, caracterizado además porque comprende una distribución de tamaños de gránulos de almidón, en donde la distribución de los tamaños de granulo de almidón comprende al menos aproximadamente 40% menos gránulos menores que 5 micras comparado con una proporción de gránulos menores que 5 micras del almidón de control como es observado usando un microscopio electrónico de barrido.
7. El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es un gránulo que muestra integridad incrementada comparada con el almidón de control como es observado usando un microscopio electrónico de barrido.
8. El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es capaz de soportar al menos tres ciclos de congelación/descongelación.
9. El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es capaz de soportar al menos tres a al menos cinco ciclos de congelación/descongelación.
10. El almidón de conformidad con la reivindicación 1, caracterizado porque la planta de maíz waxy-sugary-2 se genera por polinización cruzada de una primera planta de maíz que tiene el genotipo de *wxwxsu2su2*, y una segunda planta de maíz que tiene el genotipo de *wxSwxSsu2su2*, o el

genotipo de *wxSwxsu2su2*.

11. El almidón de conformidad con la reivindicación 1, caracterizado porque el endospermo de la planta de maíz waxy-sugary-2 tiene una dosis, dos dosis, o tres dosis del alelo *wxS*, y el endospermo tiene el genotipo de *wxSwxwxsu2su2su2*, *wxSwxSwxsu2su2su2*, o *wxSwxSwxSsu2su2su2*.
12. Un método para mejorar al menos una característica en un perfil de adhesión de un almidón, caracterizado porque comprende introducir al menos un alelo *wxS* en el endospermo de una planta de maíz waxy, en donde el almidón resultante derivado de la planta de maíz waxy que comprende al menos un alelo *wxS* en el endospermo comprende una temperatura de adhesión de almidón acuosa que es al menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA, mejorando de esta manera al menos una característica del perfil de adhesión del almidón comparado con el almidón de control.
13. Una planta de maíz mutante doble, caracterizada porque la planta de maíz es homocigota recesiva para el gen de almidón sintasa IIa (*su2*), y es ya sea homocigoto o heterocigoto para un gen de almidón sintasa I ligada a gránulos mutado (*GBSSI*), en donde el gen *GBSSI* mutado tiene menos actividad de *GBSSI* que el gen *GBSSI* de tipo natural (*Wx*), pero tiene mayor actividad de *GBSSI* que el gen *GBSSI* de pérdida de función, recesivo (*wx*).
14. La planta de maíz mutante doble de conformidad con la reivindicación 13, caracterizada porque el gen *GBSSI* mutado es el alelo *wx-Stonor* (*wxS*).

15. La planta de maíz mutante doble de conformidad con la reivindicación 14, caracterizada porque la planta se genera por polinización cruzada de una primera planta de maíz que tiene el genotipo de *wxwxsu2su2*, y una segunda planta de maíz que tiene el genotipo de *wxSwxSsu2su2*, o el genotipo de *wxSwxsu2su2*.



UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER FOR PATENTS
P.O. Box 1450
Alexandria, Virginia 22313-1450
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
15/135,106	04/21/2016	Hongxin JIANG	3459.WHL Cont	8300
86379	7590	07/03/2017		
Ingredion Incorporated Suite C 10 Finderne Avenue Bridgewater, NJ 08807			EXAMINER COHEN, STEFANIE J	
			ART UNIT	PAPER NUMBER
			1732	
			NOTIFICATION DATE	DELIVERY MODE
			07/03/2017	ELECTRONIC

Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Notice of the Office communication was sent electronically on above-indicated "Notification Date" to the following e-mail address(es):

patents@nstarch.com
patents@ingredion.com

Office Action Summary	Application No. 15/135,106	Applicant(s) JIANG ET AL.	
	Examiner STEFANIE COHEN	Art Unit 1732	AIA (First Inventor to File) Status Yes

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 2 MONTHS FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

- 1) ☒ Responsive to communication(s) filed on 12/27/16.
☐ A declaration(s)/affidavit(s) under **37 CFR 1.130(b)** was/were filed on ____.
- 2a) ☐ This action is **FINAL**. 2b) ☐ This action is non-final.
- 3) ☐ An election was made by the applicant in response to a restriction requirement set forth during the interview on ____; the restriction requirement and election have been incorporated into this action.
- 4) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims*

- 5) ☒ Claim(s) 1-33 is/are pending in the application.
5a) Of the above claim(s) ____ is/are withdrawn from consideration.
- 6) ☐ Claim(s) ____ is/are allowed.
- 7) ☐ Claim(s) ____ is/are rejected.
- 8) ☐ Claim(s) ____ is/are objected to.
- 9) ☒ Claim(s) 1-33 are subject to restriction and/or election requirement.

* If any claims have been determined allowable, you may be eligible to benefit from the **Patent Prosecution Highway** program at a participating intellectual property office for the corresponding application. For more information, please see http://www.uspto.gov/patents/init_events/pph/index.jsp or send an inquiry to PPHfeedback@uspto.gov.

Application Papers

- 10) ☐ The specification is objected to by the Examiner.
- 11) ☐ The drawing(s) filed on ____ is/are: a) ☐ accepted or b) ☐ objected to by the Examiner.
Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).
Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

Priority under 35 U.S.C. § 119

- 12) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

Certified copies:

- a) ☐ All b) ☐ Some** c) ☐ None of the:
1. ☐ Certified copies of the priority documents have been received.
 2. ☐ Certified copies of the priority documents have been received in Application No. ____.
 3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

** See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

- | | |
|---|---|
| 1) ☐ Notice of References Cited (PTO-892) | 3) ☐ Interview Summary (PTO-413)
Paper No(s)/Mail Date. ____. |
| 2) ☐ Information Disclosure Statement(s) (PTO/SB/08a and/or PTO/SB/08b)
Paper No(s)/Mail Date ____. | 4) ☐ Other: ____. |

Art Unit: 1732

The present application, filed on or after March 16, 2013, is being examined under the first inventor to file provisions of the AIA.

DETAILED ACTION

Election/Restrictions

Restriction to one of the following inventions is required under 35 U.S.C. 121:

- I. Claims 1-11, drawn to a starch, classified in C08L3/00.
- II. Claim 12, drawn to a method for improving at least one characteristic in a pasting profile, classified in A0H1/00.
- III. Claims 13-33, drawn to a double mutant corn plant, classified in A0H5/00.

The inventions are distinct, each from the other because of the following reasons:

Inventions I and II are related as process of making and product made. The inventions are distinct if either or both of the following can be shown: (1) that the process as claimed can be used to make another and materially different product or (2) that the product as claimed can be made by another and materially different process (MPEP § 806.05(f)). In the instant case, the process as claimed can be used to make a different product, such as a starch comprising an amylose content outside the range of about 2 wt% to about 20 wt% as measured using an amylose/ amylopectin assay kit.

Inventions II and III are related as process of making and product made. The inventions are distinct if either or both of the following can be shown: (1) that the process as claimed can be used to make another and materially different product or (2) that the product as claimed can be made by another and materially different process (MPEP § 806.05(f)). In the instant case, the product claimed in claim 13 can be made by a different process such as a *su2* gene that is not a *wxS* allele.

Inventions I and III are directed to related distinct inventions. The related inventions are distinct if: (1) the inventions as claimed are either not capable of use together or can have a materially different design, mode of operation, function, or effect; (2) the inventions do not overlap in scope, i.e., are mutually exclusive; and (3) the inventions as claimed are not obvious variants. See MPEP § 806.05(j). In the instant case, the inventions as claimed cannot be used together, do not overlap in scope. Furthermore, the inventions as claimed do not encompass overlapping subject matter and there is nothing of record to show them to be obvious variants.

Invention I is to a starch while Invention III is to a double mutant corn plant. These cannot be used together.

Invention I is to a starch derived from a waxy corn plant comprising at least one *wxS* allele in the endosperm while Invention III is to a mutated *GBSSI* gene. These do not overlap in scope and are not obvious variants.

Restriction for examination purposes as indicated is proper because all the inventions listed in this action are independent or distinct for the reasons given above and there would be a serious search and/or examination burden if restriction were not required because one or more of the following reasons apply:

- the inventions have acquired a separate status in the art in view of their different classification

- the inventions have acquired a separate status in the art due to their recognized divergent subject matter

- the inventions require a different field of search as they require searching different classes/subclasses

- prior art applicable to one invention would not likely be applicable to another invention

Applicant is advised that the reply to this requirement to be complete must include (i) an election of a invention to be examined even though the requirement may be traversed (37 CFR 1.143) **and (ii) identification of the claims encompassing the elected invention.**

The election of an invention may be made with or without traverse. To reserve a right to petition, the election must be made with traverse. If the reply does not distinctly and specifically point out supposed errors in the restriction requirement, the election shall be treated as an election without traverse. Traversal must be presented at the time of election in order to be considered timely. Failure to timely traverse the requirement

Art Unit: 1732

will result in the loss of right to petition under 37 CFR 1.144. If claims are added after the election, applicant must indicate which of these claims are readable upon the elected invention.

Should applicant traverse on the ground that the inventions are not patentably distinct, applicant should submit evidence or identify such evidence now of record showing the inventions to be obvious variants or clearly admit on the record that this is the case. In either instance, if the examiner finds one of the inventions unpatentable over the prior art, the evidence or admission may be used in a rejection under 35 U.S.C. 103 or pre-AIA 35 U.S.C. 103(a) of the other invention.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to STEFANIE COHEN whose telephone number is (571)270-5836. The examiner can normally be reached on Monday through Thursday 9:3am-6:00pm.

Examiner interviews are available via telephone, in-person, and video conferencing using a USPTO supplied web-based collaboration tool. To schedule an interview, applicant is encouraged to use the USPTO Automated Interview Request (AIR) at <http://www.uspto.gov/interviewpractice>.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Melvin Curtis Mayes can be reached on 5712721234. The fax phone

Art Unit: 1732

number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov>. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

6/19/17

/STEFANIE COHEN/
Examiner, Art Unit 1732

Application/Control Number: 15/135,106
Art Unit: 1732

Page 7

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:

Hongxin JIANG *et al.*

App. No.: 15/135,106

Filing Date: April 21, 2016

For: **COMPOSITIONS AND METHODS
FOR PRODUCING STARCH WITH
NOVEL FUNCTIONALITY**

Confirmation No.: 8300

Art Unit: 1732

Examiner: Cohen, Stefanie J

Customer No.: 86379

Submitted Electronically October 3, 2017

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

RESPONSE TO RESTRICTION REQUIREMENT DATED JULY 3, 2017

Sir:

Applicant herein responds to the Restriction Requirement dated July 3, 2017, setting a shortened statutory period of two-months to respond (hereinafter "Office Action"). A request for a one-month extension of time along with the requisite fee accompanies this response.

The Director is hereby authorized to charge any fees which may be required, or credit any overpayment, to Deposit Account No. 504636 referencing attorney Docket No. 3459.WHL.Cont.

Listing of Claims begins at **page 2**.

Remarks begin at **page 6**.

Amendments to the Claims

This listing of claims will replace all prior versions and listings of claims in the application:

1. (Original) A starch comprising:
an amylose content of from about 2 wt % to about 20 wt % as measured using an amylose/amylopectin assay kit; and
an aqueous starch pasting temperature that is at least about 5% greater than the starch pasting temperature of a control starch as measured using RVA analysis,
wherein, the starch is derived from a waxy-sugary 2 corn plant comprising at least one wxS allele in the endosperm.
2. (Original) The starch of claim 1, wherein the amylose content is from about 8 wt % to about 15 wt %.
3. (Previously Presented) The starch of claim 1, wherein the aqueous pasting temperature is from about 5 % to about 14 % greater than that of the control starch.
4. (Previously Presented) The starch of claim 1, further comprising an enthalpy change of crystallinity of at least about 30 % greater than an enthalpy change of crystallinity of the control starch as measured using RVA analysis.
5. (Original) The starch of claim 4, wherein the enthalpy change of crystallinity is from about 70 % to about 200 % greater than an enthalpy change of crystallinity of the control starch as measured using a differential scanning calorimeter (DSC).
6. (Previously Presented) The starch of claim 1, further comprising a distribution of starch granule sizes, wherein the distribution of starch granule sizes comprises at least about 40 % fewer granules less than 5 microns compared to a proportion of granules less than 5 microns of the control starch as observed using a scanning electron microscope.
7. (Previously Presented) The starch of claim 1, wherein the starch is a granule which exhibits increased integrity compared to the control starch as observed using a scanning electron microscope.

8. (Previously Presented) The starch of claim 1, wherein the starch is capable of withstanding at least three freeze-thaw cycles.

9. (Currently Amended) The starch of claim 1~~claim 9~~, wherein the starch is capable of withstanding at least three to at least five freeze-thaw cycles.

10. (Previously Presented) The starch of claim 1, wherein the waxy-sugary 2 corn plant is generated by cross pollination of a first corn plant having the genotype of *wxwxsu2su2*, and a second corn plant having the genotype of *wxSwxSsu2su2*, or the genotype of *wxSwxsu2su2*.

11. (Previously Presented) The starch of claim 1, wherein the endosperm of the waxy-sugary 2 corn plant has one dose, two doses, or three doses of the *wxS* allele, and the endosperm has the genotype of *wxSwxwxsu2su2su2*, *wxSwxSwxsu2su2su2*, or *wxSwxSwxSsu2su2su2*.

12. (Withdrawn) A method for improving at least one characteristics in a pasting profile of a starch comprising introducing at least one *wxS* allele into the endosperm of a waxy corn plant, wherein the resultant starch derived from the waxy corn plant comprising at least one *wxS* allele in the endosperm comprises an aqueous starch pasting temperature which is at least about 5% greater than a starch pasting temperature of a control starch as measured using RVA analysis, thereby improving at least one characteristic of the pasting profile of the starch compared a control starch.

13. (Withdrawn) A double-mutant corn plant, wherein the corn plant is homozygous recessive for the starch synthase IIa (*su2*) gene, and either homozygous or heterozygous for a mutated Granule-bound starch synthase I (GBSSI) gene, wherein the mutated GBSSI gene has less GBSSI activity than the wild type GBSSI gene (*Wx*), but has higher GBSSI activity than the recessive, loss-of-function GBSSI gene (*wx*).

14. (Withdrawn) The double-mutant corn plant of claim 13, wherein the mutated GBSSI gene is the *wx-Stonor* (*wxS*) allele.

15. (Withdrawn) The double-mutant corn plant of claim 14, wherein the plant is generated by cross pollination of a first corn plant having the genotype of *wxwx su2 su2*, and a second corn plant having the genotype of *wxS wxS su2 su2*, or the genotype of *wxS wx su2 su2*.
16. (Withdrawn) The double-mutant corn plant of claim 15, wherein the endosperm of the corn plant has one dose, two doses, or three doses of the *wxS* allele, wherein the endosperm has the genotype of *wxS wx wx su2 su2 su2*, *wxS wx S wx su2 su2 su2*, or *wxS wx S wx S su2 su2 su2*.
17. (Withdrawn) The double-mutant corn plant of claim 13, wherein the starch produced in the endosperm of the corn plant has a higher pasting temperature compared to the starch produced in the endosperm of a double, waxy sugary-2 recessive mutant corn plant having the genotype of *wx wx wx su2 su2 su2*.
18. (Withdrawn) A seed of the double-mutant corn plant of claim 13.
19. (Withdrawn) A plant part, plant cell, or tissue culture of the double-mutant corn plant of claim 13.
20. (Withdrawn) The plant part of claim 19, wherein the plant part is selected from the group consisting of an endosperm, a leaf, a flower, an ovule, a pollen, a rootstock, and a scion.
21. (Withdrawn) A method for producing starch comprising obtaining starch from the endosperm of the double-mutant corn plant of claim 13.
22. (Withdrawn) A method for producing starch comprising growing the double-mutant corn plant of claim 13, and harvesting seeds from the double-mutant corn plant.
23. (Withdrawn) A method for producing a corn seed comprising crossing a first parent corn plant with a second parent corn plant and harvesting the resultant corn seed, wherein said first parent corn plant and/or second parent corn plant is the double-mutant corn plant of claim 13.
24. (Withdrawn) A method for producing a corn seed comprising self-pollinating the double-mutant corn plant of claim 13, and harvesting the resultant corn seed.
25. (Withdrawn) A method of vegetatively propagating the corn plant of claim 13, said method comprising collecting part of the plant of claim 13 and regenerating a plant from said part.

26. (Withdrawn) A method of producing a corn plant derived from the double-mutant corn plant of claim 13, the method comprising self-pollinating the double-mutant plant of claim 13 at least once to produce a progeny plant derived from the double-mutant plant of claim 13.
27. (Withdrawn) A method of producing a corn plant derived from the double-mutant plant of claim 13, the method comprising crossing the double-mutant plant of claim 13 with a second corn plant to produce a progeny plant of the double-mutant plant of claim 13.
28. (Withdrawn) A method of producing a corn plant comprising introducing a transgene into a corn plant, wherein the transgene confers the desired phenotype of the double-mutant plant of claim 13.
29. (Withdrawn) A method of producing a corn plant comprising gene-editing to produce a genetic sequence to confer the desired phenotype of the double-mutant plant of claim 13.
30. (Withdrawn) A method of producing a corn plant comprising introducing a mutation to produce a genetic sequence to confer the desired phenotype of the double-mutant plant of claim 13.
31. (Withdrawn) A starch extracted from the double-mutant corn plant of claim 13.
32. (Withdrawn) A starch extracted from the double-mutant corn plant of claim 15.
33. (Withdrawn) A starch extracted from the endosperm of the double-mutant corn plant of claim

REMARKS

Applicant herein amends Claim 9 and withdraws claims 12-33 such that upon entry of the election and amendments provided herein, Claims 1-11 will be restricted to the elected invention and under examination.

RESTRICTION REQUIREMENT

The Office requires restriction under 35 U.S.C 121 to one of the following inventions:

“I. Claims 1-11, drawn to a starch, classified in C08L3/00.

II. Claim 12, drawn to a method for improving at least one characteristic in a pasting profile, classified in AOH1 /00.

III. Claims 13-33, drawn to a double mutant corn plant, classified in AOH5/00.”

Office Action at page 2.

Applicant herein elects Group I without traverse. Claims 1-11 encompass the elected invention.

CONCLUSION

Applicant submits that all of the claims are in condition for allowance, which action is requested. If the Office finds that a telephone conference would expedite prosecution of this application, please telephone the undersigned at the number provided.

Respectfully submitted,

Dated: October 3, 2017

/Jacqueline Marie Cohen/
Jacqueline Marie Cohen
Registration No. 51574

Ingredion Inc.
10 Finderne Avenue
Brisgewater, NJ 08807
Tel.: (908)575-6184

Oficina de Patentes y Marcas de los Estados Unidos

INGREDION INCORPORATED

Suite C

10 FINDERNE AVENUE

BRIDGEWATER, NJ 08807

Solicitud No.: 15/135.106

Fecha de presentación: 04/21/2016

Primer inventor nombrado: Hongxin JIANG

Número de expediente del abogado: 3459.WHL Cont

Número de confirmación: 8300

Examinador: COHEN, STEFANIE J.

Unidad de técnica: 1732

Fecha de notificación: 07/03/2017

Modo de envío: Electrónico

A continuación encontrará una comunicación de la Oficina relativa a esta solicitud o procedimiento.

El período de respuesta, si lo hay, se establece en la comunicación adjunta.

La notificación de la comunicación de la Oficina se envió electrónicamente en la "Fecha de notificación" indicada más arriba a la(s) siguiente(s) dirección de correo electrónico:

patents@nstarch.com

patents@ingredion.com

Resumen de la Acción Oficial

Solicitud No.: 15/135.106

Solicitante: JIANG ET AL.

Examinador: STEFANIE COHEN

Unidad de técnica: 1732

AIA (primer inventor en el archivo): Sí

La Fecha de Envío de esta comunicación aparece en la hoja de cubierta con la dirección de correspondencia-

Periodo de Respuesta

Un periodo estatutario acortado para responder se establece para que venza en 2 meses desde la fecha de envío de esta comunicación.

Nota del traductor: Se hace a continuación traducción únicamente de las casillas señaladas con una X en el Resumen de la Acción Oficial.

Estatus

1) X En respuesta a comunicación presentada el 12/27/16

(...)

Disposición de las Reivindicaciones

5) X Las reivindicaciones 1 a 33 están pendientes en la solicitud

7) X Las reivindicaciones 1 a 33 están sujetas a restricción y/o requerimiento de elección

(...)

Número de solicitud/control: 15/135.106

Unidad de técnica: 1732

La presente solicitud, presentada en o después del 16 de marzo de 2013, está siendo examinada bajo el primer inventor para presentar las disposiciones de la AIA.

ACCIÓN DETALLADA

Elección / Restricciones

La restricción a una de las siguientes invenciones se requiere bajo 35 US.C. 121:

- i. Reivindicaciones 1 A 11, hacen referencia a un almidón, clasificado en C08L3/00.
- ii. Reivindicación 12, hare referencia a un método para mejorar al menos una característica en un perfil de adhesión, clasificado en A0H1/00.
- iii. Reivindicaciones 13 a 33, hacen referencia a una planta de maíz mutante doble, clasificada en A0H5/00.

Las invenciones son distintas cada una de la otra por las siguientes razones:

Las invenciones I y II están relacionadas como proceso de fabricación y producto realizado. Las invenciones son distintas si puede mostrarse una o

ambas de las siguientes opciones: (1) que el proceso según lo reivindicado se puede usar para hacer otro producto materialmente diferente o (2) que el producto reivindicado puede ser fabricado por otro y materialmente diferente proceso (MPEP § 806.05(f)). En el presente caso, el proceso según se reivindica puede usarse para fabricar un producto diferente, tal como un almidón que comprende un contenido de amilosa fuera del intervalo de aproximadamente 2% en peso a aproximadamente 20% en peso medido usando un kit de ensayo de amilosa/amilopectina.

Las invenciones I y III están relacionadas como proceso de fabricación y producto realizado. Las invenciones son distintas si puede mostrarse una o ambas de las siguientes opciones: (1) que el proceso según lo reivindicado se puede usar para hacer otro producto materialmente diferente o (2) que el producto reivindicado puede ser fabricado por otro y materialmente diferente proceso (MPEP § 806.05(f)). En el presente caso, el producto reclamado en la reivindicación 13 puede fabricarse mediante un proceso diferente tal como un gen *su2* que no es un alelo *wxS*.

Las invenciones I y III están dirigidas a invenciones distintas relacionadas. Las invenciones relacionadas son distintas si: (1) las invenciones tal como se reivindican no son capaces de usarse juntas o pueden tener un diseño, modo de operación, función o efecto materialmente diferente; (2) las invenciones no se solapan en su alcance, es decir, son mutuamente excluyentes; y (3) las invenciones tal como se reivindican no son variantes obvias. Ver MPEP § 806.05(j). En el presente caso, las invenciones tal como se reivindican no se pueden usar juntas, no se superponen en su alcance. Además, las invenciones

tal como se reivindican no abarcan el tema superpuesto y no hay nada de registro que las muestre como variantes obvias.

La invención I es a un almidón mientras que la invención III es a una planta de maíz mutante doble. Estas no pueden ser usadas juntas.

La invención I es a un almidón derivado de una planta de maíz cerosa que comprende por lo menos un alelo wxS en el endospermo, mientras que la invención III es a un gen GBSSI mutado. Estas no se solapan en alcance y no son variantes obvias.

La restricción para fines de examen como se indica es adecuada porque todas las invenciones enumeradas en esta acción son independientes o distintas por las razones indicadas anteriormente y habría una búsqueda seria y/o examen si no se requiriera restricción debido a una o más de las siguientes razones que aplican:

- Las invenciones han adquirido un estado separado en la técnica en vista de su diferente clasificación.
- Las invenciones han adquirido un estado separado en la técnica debido a su materia divergente reconocida.
- Las invenciones requieren un campo de búsqueda diferente ya que requieren buscar diferentes clases/subclases
- La técnica anterior aplicable a una invención probablemente no sería aplicable a otra invención.

Se aconseja al solicitante que la respuesta a este requisito sea completa debe incluir (i) una elección de una invención para ser examinada aunque se

pueda cruzarse el requisito (37 CFR 1.143) y (ii) **identificación de las reivindicaciones que abarcan la invención elegida.**

La elección de una invención puede hacerse con o sin objeción. Para reservar un derecho de petición, la elección debe hacerse con objeción. Si la respuesta no señala de manera clara y específica los supuestos errores en el requisito de restricción, la elección se tratará como una elección sin objeción. El recorrido debe presentarse al momento de la elección para que se considere oportuno. Si no se cruza oportunamente el requisito resultará en la pérdida del derecho de petición bajo 37 CFR 1.144. Si se agregan reivindicaciones después de la elección, el solicitante debe indicar cuáles de estas reivindicaciones son legibles a la invención elegida.

Si el solicitante se atraviesa argumentando que las invenciones no son patentablemente distintas, el solicitante deberá presentar evidencia o identificar tal evidencia ahora de registro que demuestre que las invenciones son variantes obvias o admitir claramente en el expediente que este es el caso. En cualquier caso, si el examinador encuentra que una de las invenciones no es patentable con respecto a la técnica anterior, la evidencia puede usarse en un rechazo bajo 35 U.S.C. 103 o pre-AIA U.S.C. 103 (a) de la otra invención.

Cualquier duda relacionada con esta comunicación o comunicaciones anteriores del examinador se debe dirigir a STEFANIE COHEN cuyo número de teléfono es (571)270-5836. El examinador puede ser encontrado normalmente de Lunes a Jueves 9:30 am – 6:00 p.m.

Las entrevistas con el examinador están disponibles por teléfono, en persona y por videoconferencia usando una herramienta de colaboración basada en la web suministrada por la USPTO. Para programar una entrevista, se recomienda al solicitante utilizar la Solicitud de Entrevista Automatizada (AIR) de la USPTO en <http://www.uspto.gov/interviewpractice>.

Si los intentos por encontrar al examinador por teléfono son fallidos, el supervisor del examinador, Melvin Curtis Mayes se puede encontrar en 5712721234. El número de fax para la organización donde esta solicitud o procedimiento es asignado es 571-273-8300.

La información relacionada con el estado de una solicitud se puede obtener del sistema de Recuperación de Información de Solicitud de Patente (PAIR). La información de estado para solicitudes publicadas se puede obtener del PAIR Privado o el PAIR Público. La información de estado para solicitudes sin publicar está disponible a través del PAIR Privado únicamente. Para mayor información acerca del sistema PAIR, ver <http://pair-direct.uspto.gov>. Si tiene preguntas sobre el acceso al sistema PAIR Privado, contacte el Centro de Negocio Electrónico (EBC) a 866-217-9197 (totalmente gratis). Si desea asistencia de un Representante de Servicio al Cliente de la USPTO o acceso al sistema de información automatizado, llame 800-786-9199 (EN USA O CANADA) o 571-272-1000.

6/19/17

/STEFANIE COHEN/

Examinador, Unidad de Arte 1732

EN LA OFICINA DE PATENTES Y MARCAS DE ESTADOS UNIDOS DE
AMÉRICA

En solicitud de:	Confirmación No. 8300
Hongxin JIANG <i>et al.</i>	Unidad de arte: 1732
Solicitud No. 15/135,106	Examinador: Cohen, Stefanie J.
Fecha de presentación: 21 de abril de 2016	Cliente No. 86379
Para: COMPOSICIONES Y MÉTODOS PARA PRODUCIR ALMIDÓN CON FUNCIONALIDAD NOVEDOSA	

Enviado Electrónicamente el 3 de octubre de 2017

Comisionado de patentes

P.O. Box 1450

Alexandria, VA 22313-1450

**RESPUESTA AL REQUERIMIENTO DE RESTRICCIÓN CON FECHA DEL 3 DE
JULIO DE 2017**

Señor,

El solicitante responde al Requisito de Restricción fechado el 3 de julio de 2017, estableciendo un período estatutario acortado de dos meses para responder (en adelante, "Acción Oficial"). Una solicitud de prórroga de un mes junto con la tarifa requerida acompaña a esta respuesta.

Se autoriza al Director a cobrar cualquier tarifa que se requiera, o acreditar cualquier pago en exceso, a la Cuenta de Depósito N° 504636 haciendo referencia al abogado Expediente No. 3459.WHL.Cont.

Listado de Reivindicaciones comienza en la **página 2**.

Las observaciones comienzan en la **página 6**.

Modificaciones a las Reivindicaciones

La lista de reivindicaciones reemplazará todas las versiones anteriores y las listas de reivindicaciones en la solicitud:

1. (Original) Un almidón, caracterizado porque comprende:
 - un contenido de amilosa de aproximadamente 2% en peso a aproximadamente 20% en peso como es medido usando un kit de ensayo de amilosa/amilopectina; y
 - una temperatura de adhesión de almidón acuosa que es menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA,en donde, el almidón se deriva de una planta de maíz waxy-sugary-2 que comprende al menos un alelo wxS en el endospermo.
2. (Original) El almidón de conformidad con la reivindicación 1, caracterizado porque el contenido de amilosa es de aproximadamente 8% en peso a aproximadamente 15% en peso.
3. (Presentada anteriormente) El almidón de conformidad con la reivindicación 1, caracterizado porque la temperatura de adhesión acuosa es de aproximadamente 5% a aproximadamente 14% mayor que aquella del almidón de control.
4. (Presentada anteriormente) El almidón de conformidad con la reivindicación 1, caracterizado porque además comprende un cambio de entalpía de cristalinidad de al menos aproximadamente 30% mayor que un cambio de entalpía de cristalinidad del almidón de control como es medido usando el análisis RVA.

5. (Original) El almidón de conformidad con la reivindicación 4, caracterizado porque el cambio de entalpía de la cristalinidad es de aproximadamente 70% a aproximadamente 200% mayor que un cambio de entalpía de cristalinidad del almidón de control como es medido usando un calorímetro de barrido diferencial (DSC).
6. (Presentada anteriormente) El almidón de conformidad con la reivindicación 1, caracterizado además porque comprende una distribución de tamaños de gránulos de almidón, en donde la distribución de los tamaños de gránulo de almidón comprende al menos aproximadamente 40% menos gránulos menores que 5 micras comparado con una proporción de gránulos menores que 5 micras del almidón de control como es observado usando un microscopio electrónico de barrido.
7. (Presentada anteriormente) El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es un gránulo que muestra integridad incrementada comparada con el almidón de control como es observado usando un microscopio electrónico de barrido.
8. (Presentada anteriormente) El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es capaz de soportar al menos tres ciclos de congelación/descongelación.
9. (Modificada) El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es capaz de soportar al menos tres a al menos cinco ciclos de congelación/descongelación.
10. (Presentada anteriormente) El almidón de conformidad con la reivindicación 1, caracterizado porque la planta de maíz waxy-sugary-2 se genera por polinización cruzada de una primera planta de maíz que tiene el

genotipo de *wxwxsu2su2*, y una segunda planta de maíz que tiene el genotipo de *wxSwxSsu2su2*, o el genotipo de *wxSwxsu2su2*.

- 11.(Presentada anteriormente) El almidón de conformidad con la reivindicación 1, caracterizado porque el endospermo de la planta de maíz waxy-sugary-2 tiene una dosis, dos dosis, tres dosis del alelo *wxS*, y el endospermo tiene el genotipo de *wxSwxwxsu2su2su2*, *wxSwxSwxsu2su2su2*, o *wxSwxSwxSsu2su2su2*.
- 12.(Retirada) Un método para mejorar al menos una característica en un perfil de adhesión de un almidón, caracterizado porque comprende introducir al menos un alelo *wxS* en el endospermo de una planta de maíz waxy, en donde el almidón resultante derivado de la planta de maíz waxy que comprende al menos un alelo *wxS* en el endospermo comprende una temperatura de adhesión de almidón acuosa que es al menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA, mejorando de esta manera al menos una característica del perfil de adhesión del almidón comparado con el almidón de control.
- 13.(Retirada) Una planta de maíz mutante doble, caracterizada porque la planta de maíz es homocigota recesiva para el gen de almidón sintasa IIa (*su2*), y es ya sea homocigoto o heterocigoto para un gen de almidón sintasa I ligada a gránulos mutado (GBSSI), en donde el gen GBSSI mutado tiene menos actividad de GBSSI que el gen GBSSI de tipo natural (*Wx*), pero tiene mayor actividad de GBSSI que el gen GBSSI de pérdida de función, recesivo (*wx*).
- 14.(Retirada) La planta de maíz mutante doble de conformidad con la

reivindicación 13, caracterizada porque el gen GBSSI mutado es el alelo *wx-Stonor* (*wxS*).

- 15.(Retirada) La planta de maíz mutante doble de conformidad con la reivindicación 14, caracterizada porque la planta se genera por polinización cruzada de una primera planta de maíz que tiene el genotipo de *wxwxsu2su2*, y una segunda planta de maíz que tiene el genotipo de *wxSwxSsu2su2*, o el genotipo de *wxSwxsu2su2*.
- 16.(Retirada) La planta de maíz mutante doble de conformidad con la reivindicación 15, caracterizada porque el endospermo de la planta de maíz tiene una dosis, dos dosis, o tres dosis del alelo *wxS*, en donde el endospermo tiene el genotipo de *wxSwxwxsu2su2su2*, *wxSwxSwxsu2su2su2*, o *wxSwxSwxSsu2su2su2*.
- 17.(Retirada) La planta de maíz mutante doble de conformidad con la reivindicación 13, caracterizada porque el almidón producido en el endospermo de la planta de maíz tiene una temperatura de adhesión más alta comparado con el almidón producido en el endospermo de una planta de maíz mutante recesiva *waxy-sugary-2* doble que tiene el genotipo de *wxwxwxsu2su2su2*.
- 18.(Retirada) Una semilla, caracterizada porque es de la planta de maíz mutante doble de conformidad con la reivindicación 13.
- 19.(Retirada) Una parte de la planta, célula de la planta, o cultivo de tejido de la planta de maíz mutante doble de conformidad con la reivindicación 13.
- 20.(Retirada) La parte de la planta de conformidad con la reivindicación 19, caracterizada porque la parte de la planta se selecciona del grupo que

consiste de un endospermo, una hoja, una flor, un óvulo, un polen, un rizoma y un vástago.

21.(Retirada) Un método para producir almidón, caracterizado porque comprende obtener almidón del endospermo de la planta de maíz mutante doble de conformidad con la reivindicación 13.

22.(Retirada) Un método para producir almidón, caracterizado porque comprende desarrollar la planta de maíz mutante doble de conformidad con la reivindicación 13, y cosechar las semillas de la planta de maíz mutante doble.

23.(Retirada) Un método para producir una semilla de maíz, caracterizado porque comprende cruzar una primera planta de maíz precursora con una segunda planta de maíz precursora y cosechar la semilla de maíz resultante, en donde la primera planta de maíz precursora, y/o la segunda a planta de maíz precursora es la planta de maíz mutante doble de conformidad con la reivindicación 13.

24.(Retirada) Un método para producir una semilla de maíz, caracterizado porque comprende auto-polinizar la planta de maíz mutante doble de conformidad con la reivindicación 13, y cosechar la semilla de maíz resultante.

25.(Retirada) Un método para propagar de manera vegetativa la planta de maíz de conformidad con la reivindicación 13, el método caracterizado porque comprende recolectar parte de la planta de la reivindicación 13 y regenerar una planta de dicha parte.

26.(Retirada) Un método para producir una planta de maíz derivada de la

planta de maíz mutante doble de conformidad con la reivindicación 13, el método caracterizado porque comprende auto-polinizar la planta mutante doble de la reivindicación 13 al menos una vez para producir una planta progenie derivada de la planta mutante doble de la reivindicación 13.

27.(Retirada) Un método para producir una planta de maíz derivada de la planta mutante doble de conformidad con la reivindicación 13, el método caracterizado porque comprende cruzar la planta mutante doble de la reivindicación 13 con una segunda planta de maíz para producir una planta progenie de la planta mutante doble de la reivindicación 13.

28.(Retirada) Un método para producir una planta de maíz, caracterizado porque comprende introducir un transgen en una planta de maíz, en donde el transgen confiere el fenotipo deseado de la planta mutante doble de conformidad con la reivindicación 13.

29.(Retirada) Un método para producir una planta de maíz, caracterizado porque comprende editar el gen para producir una secuencia genética para conferir el fenotipo deseado de la planta mutante doble de conformidad con la reivindicación 13.

30.(Retirada) Un método para producir una planta de maíz, caracterizado porque comprende introducir una mutación para producir una secuencia genética para conferir el fenotipo deseado de la planta mutante doble de conformidad con la reivindicación 13.

31.(Retirada) Un almidón, caracterizado porque se extrae de la planta de maíz mutante doble de conformidad con la reivindicación 13.

32.(Retirada) Un almidón, caracterizado porque se extrae de la planta de maíz

mutante doble de conformidad con la reivindicación 15.

33. (Retirada) Un almidón, caracterizado porque se extrae del endospermo de la planta de maíz mutante doble de conformidad con la reivindicación.

OBSERVACIONES

El solicitante en este documento modifica la Reivindicación 9 y retira las Reivindicaciones 12-33 de tal manera que al ingresar la elección y las modificaciones provistas en este documento. Las Reivindicaciones 1-11 estarán restringidas a la invención elegida y bajo examen.

SOLICITUD DE RESTRICCIÓN

Lo oficina solicita la restricción a una de las siguientes invenciones bajo 35 U.S.C. 121:

- i. Reivindicaciones 1 A 11, hacen referencia a un almidón, clasificado en C08L3/00.
- ii. Reivindicación 12, hare referencia a un método para mejorar al menos una característica en un perfil de adhesión, clasificado en A0H1/00.
- iii. Reivindicaciones 13 a 33, hacen referencia a una planta de maíz mutante doble, clasificada en A0H5/00” . Acción oficial en página 2.

El solicitante en este documento elige el Grupo I sin objeción. Las reivindicaciones 1-11 abarcan la invención elegida.

CONCLUSIÓN

El solicitante sostiene que todas las reivindicaciones están en condiciones para la concesión, acción que se solicita. Si la Oficina determina que una

conferencia telefónica agilizaría el enjuiciamiento de esta solicitud, llame al abajo firmante al número provisto.

Respetuosamente,

/Jacqueline Marie Cohen/

Jacqueline Marie Cohen

Registration No. 51574

Fecha: 3 de octubre de 2017

Ingredion Inc.

10 Finderne Avenue

Brisgewater, NJ 08807

Tel.: (908) 575-6184

Doc code: IDS

PTO/SB/08a (01-10)

Doc description: Information Disclosure Statement (IDS) Filed

Approved for use through 07/31/2012. OMB 0651-0031

U.S. Patent and Trademark Office; U.S. DEPARTMENT OF COMMERCE

Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

INFORMATION DISCLOSURE STATEMENT BY APPLICANT (Not for submission under 37 CFR 1.99)	Application Number	15135106
	Filing Date	2016-04-21
	First Named Inventor	Hongxin JIANG
	Art Unit	1732
	Examiner Name	N/A
	Attorney Docket Number	INGR-036/01US(3459)

U.S.PATENTS						Remove
Examiner Initial*	Cite No	Patent Number	Kind Code ¹	Issue Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear
Change(s) applied to document, /P.K./ 1/19/2018		4428972		1984-01-31	~~National Starch and Chemical Corporation~~ Wurzburg et al.	
	2	5954883		1999-09-21	~~National Starch and Chemical Corporation~~ Nagle et al.	
	3	6827474		2004-12-07	~~National Starch & Chemical Investment Holding Corp~~ Clapper	
	4	6960703		2005-11-01	~~National Starch and Chemical Investment Holdings~~ Nagle et al.	
If you wish to add additional U.S. Patent citation information please click the Add button.						Add

U.S.PATENT APPLICATION PUBLICATIONS						Remove
Examiner Initial*	Cite No	Publication Number	Kind Code ¹	Publication Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear
	1					
If you wish to add additional U.S. Published Application citation information please click the Add button.						Add

FOREIGN PATENT DOCUMENTS								Remove
Examiner Initial*	Cite No	Foreign Document Number ³	Country Code ²ⁱ	Kind Code ⁴	Publication Date	Name of Patentee or Applicant of cited Document	Pages,Columns,Lines where Relevant Passages or Relevant Figures Appear	T ⁵

Notice of References Cited	Application/Control No. 15/135,106		Applicant(s)/Patent Under Reexamination JIANG ET AL.	
	Examiner STEFANIE COHEN		Art Unit 1732	Page 1 of 1

U.S. PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Name	CPC Classification	US Classification
*	A	US-4,615,888 A	10-1986	Zallie; James	A21D2/186	426/21
*	B	US-4,767,849 A	08-1988	Friedman; Robert B.	A23C9/1544	127/32
*	C	US-2006/0216402 A1	09-2006	Klucinec; Jeffrey D.	C08B30/00	426/660
*	D	US-5,648,111 A	07-1997	Pearlstein; Richard Warren	A01H5/10	127/32
	E	US-				
	F	US-				
	G	US-				
	H	US-				
	I	US-				
	J	US-				
	K	US-				
	L	US-				
	M	US-				

FOREIGN PATENT DOCUMENTS

*		Document Number Country Code-Number-Kind Code	Date MM-YYYY	Country ~~WO~~	Name	CPC Classification
	N	WO 03033540 A3	10-2003	~~UNITED STATES~~	CHANG MING-TANG	C08B30/00
	O					
	P					
	Q					
	R					
	S					
	T					

NON-PATENT DOCUMENTS

*		Include as applicable: Author, Title Date, Publisher, Edition or Volume, Pertinent Pages)
	U	Wessler et al, molecular basis of mutations at the waxy locus of maize: correlation with the fine structure genetic map, 1985, proc. natl. acad. sci. usa, vol. 82, pp. 4177-4181
	V	Varagona, Alternative Splicing Induced by Insertion of Retrotransposons into the Maize waxy Gene, 1992, the plant cell, vol. 4, pgs. 811-820
	W	
	X	

*A copy of this reference is not being furnished with this Office action. (See MPEP § 707.05(a).)
Dates in MM-YYYY format are publication dates. Classifications may be US or foreign.

6	TABLA DE CORRESPONDENCIA DE REIVINDICACIONES		
	Reivindicaciones en la solicitud en Colombia (NOTA. LAS REIVINDICACIONES ACÁ INDICADAS CORRESPONDEN A AQUELLAS MODIFICADAS EN MODIFICACIÓN VOLUNTARIA EL 07/09/2018)	Reivindicaciones otorgables en la solicitud internacional	Explicación sobre la suficiente correspondencia o Comentarios que explican la correspondencia
1.	Un almidón derivado de una planta de maíz waxy-sugary-2 que comprende un alelo wxS o alelos wxS en el endospermo, en donde el almidón comprende: - un contenido de amilosa de 2% en peso a 20% en peso como es medido usando un kit de ensayo de amilosa/amilopectina; y - una temperatura de adhesión de almidón acuosa entre 55 °C y 68 °C.	1. Un almidón, caracterizado porque comprende: – un contenido de amilosa de aproximadamente 2% en peso a aproximadamente 20% en peso como es medido usando un kit de ensayo de amilosa/amilopectina; y – una temperatura de adhesión de almidón acuosa que es menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA, en donde, el almidón se deriva de una planta de maíz waxy-sugary-2 que comprende al menos un alelo wxS en el endospermo.	Esta reivindicación fue modificada con el fin de: - Relocalizar la expresión “el almidón se deriva de una planta de maíz waxy-sugary-2 que comprende al menos un alelo wxS en el endospermo” para especificar en el preámbulo que el almidón se deriva de una planta de maíz waxy-sugary-2 que comprende un alelo wxS o alelos wxS en el endospermo. - Eliminar los términos imprecisos “aproximadamente” y “al menos un”. - Reemplazar la expresión “que es menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA” por un rango de temperatura de adhesión de almidón acuosa.
2.	El almidón de conformidad con la reivindicación 1, caracterizado porque el contenido de amilosa es de 8% en peso a 15% en peso.	2. El almidón de conformidad con la reivindicación 1, caracterizado porque el contenido de amilosa es de aproximadamente 8% en peso a aproximadamente 15% en peso.	Esta reivindicación fue modificada con el fin de eliminar el término impreciso “aproximadamente”.
3.	El almidón de conformidad con la reivindicación 1, caracterizado porque la temperatura de adhesión acuosa es entre 59 °C y 68 °C.	3. El almidón de conformidad con la reivindicación 1, caracterizado porque la temperatura de adhesión acuosa es de aproximadamente 5% a aproximadamente 14% mayor que aquella del almidón de control.	Esta reivindicación fue modificada con el fin de reemplazar la expresión “de aproximadamente 5% a aproximadamente 14% mayor que aquella del almidón de control” por un rango de temperatura de adhesión acuosa.
4.	El almidón de conformidad con la reivindicación 1, caracterizado porque además comprende un cambio de entalpía de cristalinidad de 8% p/p de entre 4 J/g y 13 J/g.	4. El almidón de conformidad con la reivindicación 1, caracterizado porque además comprende un cambio de entalpía de cristalinidad de al menos aproximadamente 30% mayor que un cambio de entalpía de cristalinidad del almidón de control como es medido usando el análisis RVA.	Esta reivindicación fue modificada con el fin de reemplazar la expresión “al menos aproximadamente 30% mayor que un cambio de entalpía de cristalinidad del almidón de control como es medido usando el análisis RVA” por un rango de cambio de entalpía de cristalinidad
5.	El almidón de conformidad con la reivindicación 4, caracterizado porque el cambio de entalpía de la cristalinidad es de 1 J/g a 20 J/g.	5. El almidón de conformidad con la reivindicación 4, caracterizado porque el cambio de entalpía de la cristalinidad es de aproximadamente 70% a aproximadamente 200% mayor que un cambio de entalpía de cristalinidad del almidón de control como es medido usando un calorímetro de barrido diferencial (DSC).	Esta reivindicación fue modificada con el fin de reemplazar la expresión “de aproximadamente 70% a aproximadamente 200% mayor que un cambio de entalpía de cristalinidad del almidón de control como es medido usando un calorímetro de barrido diferencial (DSC)” por un rango de cambio de entalpía de cristalinidad
		6. El almidón de conformidad con la reivindicación 1, caracterizado además porque comprende una distribución de tamaños de gránulos de almidón, en donde la distribución de los tamaños de granulo de almidón comprende al menos aproximadamente 40% menos gránulos menores que 5 micras comparado con una	Esta reivindicación fue eliminada mediante modificación voluntaria presentada el 7 de septiembre de 2018

	proporción de gránulos menores que 5 micras del almidón de control como es observado usando un microscopio electrónico de barrido.	
	7. El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es un gránulo que muestra integridad incrementada comparada con el almidón de control como es observado usando un microscopio electrónico de barrido.	<i>Esta reivindicación fue eliminada mediante modificación voluntaria presentada el 7 de septiembre de 2018</i>
	8. El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es capaz de soportar al menos tres ciclos de congelación/descongelación.	<i>Esta reivindicación fue eliminada mediante modificación voluntaria presentada el 7 de septiembre de 2018</i>
	9. El almidón de conformidad con la reivindicación 1, caracterizado porque el almidón es capaz de soportar al menos tres a al menos cinco ciclos de congelación/descongelación.	<i>Esta reivindicación fue eliminada mediante modificación voluntaria presentada el 7 de septiembre de 2018</i>
6. El almidón de conformidad con la reivindicación 1, caracterizado porque la planta de maíz waxy-sugary-2 se genera por polinización cruzada de una primera planta de maíz que tiene el genotipo de wxwxsu2su2, y una segunda planta de maíz que tiene el genotipo de wxSwxSsu2su2, o el genotipo de wxSwxsu2su2.	10. El almidón de conformidad con la reivindicación 1, caracterizado porque la planta de maíz waxy-sugary-2 se genera por polinización cruzada de una primera planta de maíz que tiene el genotipo de wxwxsu2su2, y una segunda planta de maíz que tiene el genotipo de wxSwxSsu2su2, o el genotipo de wxSwxsu2su2.	<i>Es la misma reivindicación</i>
7. El almidón de conformidad con la reivindicación 1, caracterizado porque el endospermo de la planta de maíz waxy-sugary-2 tiene una dosis, dos dosis, o tres dosis del alelo wxS, y el endospermo tiene el genotipo de wxSwxwxsu2su2su2, wxSwxSwxsu2su2su2, o wxSwxSwxSsu2su2su2.	11. El almidón de conformidad con la reivindicación 1, caracterizado porque el endospermo de la planta de maíz waxy-sugary-2 tiene una dosis, dos dosis, o tres dosis del alelo wxS, y el endospermo tiene el genotipo de wxSwxwxsu2su2su2, wxSwxSwxsu2su2su2, o wxSwxSwxSsu2su2su2.	<i>Es la misma reivindicación</i>
8. Un método para mejorar una característica o características en un perfil de adhesión de un almidón, caracterizado porque comprende introducir un alelo wxS o alelos wxS en el endospermo de una planta de maíz waxy, en donde el almidón resultante derivado de la planta de maíz waxy que comprende un alelo wxS o alelos wxS en el endospermo comprende una temperatura de adhesión de almidón acuosa entre 55 °C y 68 °C, mejorando de esta manera una característica o características del perfil de adhesión del almidón comparado con el almidón de control.	12. Un método para mejorar al menos una característica en un perfil de adhesión de un almidón, caracterizado porque comprende introducir al menos un alelo wxS en el endospermo de una planta de maíz waxy, en donde el almidón resultante derivado de la planta de maíz waxy que comprende al menos un alelo wxS en el endospermo comprende una temperatura de adhesión de almidón acuosa que es al menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA, mejorando de esta manera al menos una característica del perfil de adhesión del almidón comparado con el almidón de control.	<i>Esta reivindicación fue modificada con el fin de:</i> - <i>Eliminar los términos imprecisos “al menos una” y “al menos un”.</i> - <i>Reemplazar la expresión “que es al menos aproximadamente 5% mayor que la temperatura de adhesión de almidón de un almidón de control como es medido usando el análisis RVA” por un rango de temperatura de adhesión de almidón acuosa.</i>
9. Una planta de maíz mutante doble, caracterizada porque la planta de maíz es homocigota recesiva para el gen de almidón sintasa IIa (su2), y es ya sea homocigoto o	13. Una planta de maíz mutante doble, caracterizada porque la planta de maíz es homocigota recesiva para el gen de almidón sintasa IIa (su2), y es ya sea homocigoto o	<i>Es la misma reivindicación</i>

heterocigoto para un gen de almidón sintasa I ligada a gránulos mutado (GBSSI), en donde el gen GBSSI mutado tiene menos actividad de GBSSI que el gen GBSSI de tipo natural (Wx), pero tiene mayor actividad de GBSSI que el gen GBSSI de pérdida de función, recesivo (wx).	heterocigoto para un gen de almidón sintasa I ligada a gránulos mutado (GBSSI), en donde el gen GBSSI mutado tiene menos actividad de GBSSI que el gen GBSSI de tipo natural (Wx), pero tiene mayor actividad de GBSSI que el gen GBSSI de pérdida de función, recesivo (wx).	
10. La planta de maíz mutante doble de conformidad con la reivindicación 9, caracterizada porque el gen GBSSI mutado es el alelo wx-Stonor (wxS).	14. La planta de maíz mutante doble de conformidad con la reivindicación 13, caracterizada porque el gen GBSSI mutado es el alelo wx-Stonor (wxS).	<i>Es la misma reivindicación</i>
11. La planta de maíz mutante doble de conformidad con la reivindicación 10, caracterizada porque la planta se genera por polinización cruzada de una primera planta de maíz que tiene el genotipo de wxwxsu2su2, y una segunda planta de maíz que tiene el genotipo de wxSwxSsu2su2, o el genotipo de wxSwxsu2su2.	15. La planta de maíz mutante doble de conformidad con la reivindicación 14, caracterizada porque la planta se genera por polinización cruzada de una primera planta de maíz que tiene el genotipo de wxwxsu2su2, y una segunda planta de maíz que tiene el genotipo de wxSwxSsu2su2, o el genotipo de wxSwxsu2su2.	<i>Es la misma reivindicación</i>