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— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

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For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: DNA MOLECULES ENCODING A COTTON CHITINASE AND PROMOTER

(57) Abstract: The present invention relates to isolated nucleic acid molecules encoding endogenous cotton chitinases and corresponding promoters, which are preferentially expressed in secondary walled cells during secondary wall deposition. The polypeptide encoded by the nucleic acid molecule, a DNA construct linking the isolated nucleic acid molecule with a promoter, the DNA construct incorporated in an expression system, a host cell, a plant, or a plant seed are also disclosed. The present invention also relates to a DNA construct linking the isolated promoters with a second DNA as well as expression systems, host cells, plants, or plant seeds containing the DNA construct. Methods of imparting resistance to insects and fungi, regulating the fiber cellulose content, and methods of expressing a gene preferentially in secondary walled cells during secondary wall deposition are also disclosed.

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US04/01816

A. CLASSIFICATION OF SUBJECT MATTER
IPC(7) : C12N 15/29, 15/82, 15/87, 5/04; A01H 5/00
US CL : 800/287, 278, 278, 298; 435/468, 419, 320.1; 536/23.1, 24.1
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 800/287, 278, 278, 298; 435/468, 419, 320.1; 536/23.1, 24.1

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

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
WEST, STN:agricola, biosis, caplus,caba; sequence search of SEQ ID NO:1, 2, 7

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- A	US 6,096,950 (JOHN) 01 AUGUST 2000 (01.08.2000), column 3, lines 15 to column 4, line 55, column 20, number 12; column 25, number 14; column 27, lines 20-32; column 32-36, table 1-9.	42-48, 50-57, 72-84, 91-94 --- 8-12, 24-28, 49, 58, 65-67, 69
X	US 6,259,003 B1 (FUKJISAWA et al) 10 JULY 2001 (10.07.2001), entire document.	42-48, 50-57, 72-84, 91-94
Y --- A	US 5,530,187 (LAMB et al) 25 JUNE 1996 (25.06.1996), column 4, lines 40-60; columns 11 to14; sequence listing, SEQ ID NO:1).	1-7, 16-23, 29-41, 85-90 --- 8-12, 24-28, 49, 58, 65-67, 69

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

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search
15 March 2005 (15.03.2005)

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/US04/01816

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

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, the international search was carried out on the basis of:

   a. type of material

   ☒ a sequence listing

   ☐ table(s) related to the sequence listing

   b. format of material

   ☐ in written format

   ☒ in computer readable form

   c. time of filing/furnishing

   ☐ contained in the international application as filed

   ☐ filed together with the international application in computer readable form

   ☒ furnished subsequently to this Authority for the purposes of search

2. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US04/01816

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

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

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

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

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

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

This International Searching Authority found multiple inventions in this international application, as follows:
Please See Continuation Sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☒ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.: 1-12, 16-67, 69, 72-94 including SEQ ID NO:1, 2, and 7

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

Remark on Protest
☐ The additional search fees were accompanied by the applicant's protest.
☐ No protest accompanied the payment of additional search fees.

BOX III. OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING

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

Group I, claim(s) 1-12, 16-41, 85-90, drawn to an isolated nucleic acid molecule of SEQ ID NO:1, or variants thereof, from cotton, encoding a chitinase of SEQ ID NO:2 or variants thereof, a DNA construct comprising a promoter operably linked to said nucleic acid molecule, an expression system comprising said construct, a host cell, or plant transformed with said construct, and a method of imparting resistance to plants against insects and fungi comprising transforming a plant with said nucleic acid molecule.

Group II, claim(s) 1-12, 16-41, 85-90, drawn to an isolated nucleic acid molecule of SEQ ID NO:3, or variants thereof, from cotton, encoding a chitinase of SEQ ID NO:4 or variants thereof, a DNA construct comprising a promoter operably linked to said nucleic acid molecule, an expression system comprising said construct, a host cell, or plant transformed with said construct, and a method of imparting resistance to plants against insects and fungi comprising transforming a plant with said nucleic acid molecule.

Group III, claim(s) 1-12, 16-41, 85-90, drawn to an isolated nucleic acid molecule of SEQ ID NO:1, or variants thereof, from cotton, encoding a chitinase of SEQ ID NO:5 or variants thereof, a DNA construct comprising a promoter operably linked to said nucleic acid molecule, an expression system comprising said construct, a host cell, or plant transformed with said construct, and a method of imparting resistance to plants against insects and fungi comprising transforming a plant with said nucleic acid molecule.

Group IV, claim(s) 1-12, 16-41, 85-90, drawn to an isolated nucleic acid molecule of SEQ ID NO:1, or variants thereof, from cotton, encoding a chitinase of SEQ ID NO:6 or variants thereof, a DNA construct comprising a promoter operably linked to said nucleic acid molecule, an expression system comprising said construct, a host cell, or plant transformed with said construct, and a method of imparting resistance to plants against insects and fungi comprising transforming a plant with said nucleic acid molecule.

Group V, claim(s) 1-12, 16-41, 85-90, drawn to an isolated nucleic acid molecule of SEQ ID NO:3, or variants thereof, from cotton, encoding a chitinase of SEQ ID NO:5 or variants thereof, a DNA construct comprising a promoter operably linked to said nucleic acid molecule, an expression system comprising said construct, a host cell, or plant transformed with said construct, and a method of imparting resistance to plants against insects and fungi comprising transforming a plant with said nucleic acid molecule.

Group VI, claim(s) 1-12, 16-41, 85-90, drawn to an isolated nucleic acid molecule of SEQ ID NO:3, or variants thereof, from cotton, encoding a chitinase of SEQ ID NO:6 or variants thereof, a DNA construct comprising a promoter operably linked to said nucleic acid molecule, an expression system comprising said construct, a host cell, or plant transformed with said construct, and a method of imparting resistance to plants against insects and fungi comprising transforming a plant with said nucleic acid molecule.

Group VII, claim(s) 13-15, drawn to a polypeptide of SEQ ID NO:2.

Group VIII, claim(s) 13-15, drawn to a polypeptide of SEQ ID NO:4.

Group IX, claim(s) 13-15, drawn to a polypeptide comprising the amino acid sequence of SEQ ID NO:5.

~~Group X, claim(s) 13-15, drawn to a polypeptide comprising the amino acid sequence of SEQ ID NO:6.~~

Group XI, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:7, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:2, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XII, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:7, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XIII, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:7, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XIV, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:7, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:4, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XV, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:7, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XVI, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:7, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XVII, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:8, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:2, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XVIII, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:8, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XIX, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:8, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XX, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:8, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:4, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXI, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:8, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXII, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:8, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXIII, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:9, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:2, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXIV, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:9, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXV, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:9, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXVI, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:9, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:4, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXVII, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:9, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXVIII, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:9, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXIX, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:10, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:2, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXX, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:10, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXXI, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:10, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:1 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXXII, claim(s) 42-67, 69, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:10, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:4, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXXIII, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:10, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:5, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXXIV, claim(s) 42-67, 68, 72-84, 91-94, drawn to an isolated DNA promoter from cotton of SEQ ID NO:10, a DNA construct comprising said promoter operably linked to a DNA encoding a protein or polypeptide or operably linked to SEQ ID NO:3 or variants thereof, encoding SEQ ID NO:6, host cell or plant transformed with said construct and method of expressing a gene preferentially in secondary walled cells during secondary wall deposition in a plant comprising transforming a plant with said construct.

Group XXXV, claim(s) 42, 50, 70-71, drawn to a DNA construct comprising a promoter operably linked to a DNA encoding a protein or polypeptide wherein the DNA encodes any of the proteins in claims 70 or 71.

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The inventions listed as Groups I-XXXV do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons: an isolated nucleic acid molecule from cotton encoding an endogenous cotton chitinase is taught in the prior art. Hudspeth et al (1996, Plant Molecular Biology 31(4):911-916) teach an isolated cDNA clone from cotton that encodes a chitinase.

Applicants' chemical compounds, i.e., different DNA sequences encoding different polypeptides, each have different properties and different core structures that elicit different activities; and as such, the Groups I-XVI are not linked by, or share, a single special technical feature.

In addition, the claims are not linked by or share a single technical feature because they are each drawn to products not required by the other. The isolated nucleic acids and encoded proteins of groups I-VI do not share a technical feature, as the polypeptides of groups VII-X do not share a technical feature, as the constructs comprising a promoter sequence operably linked to a nucleic acid encoding a protein of groups XI to XXXIV do not share a technical feature. Lastly, each of the products in groups I to XXXV do not share a technical feature..