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60/548,891 1 March 2004 (01.03.2004) US
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(54) Title: SORGHUM GENE EXPRESSION PROFILING

(57) Abstract: The present subject matter provides a method for enhancing the drought resistance of a plant. In some embodiments, polynucleotides from *Sorghum bicolor* that hybridize to polynucleotides isolated from rice (*Oryza sativa*) and encode polypeptides for abiotic stress tolerance are also described.



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**INTERNATIONAL SEARCH REPORT**

International application No.

PCT/US05/06729

**A. CLASSIFICATION OF SUBJECT MATTER**  
 IPC(7) : C07K 14/415; C12P 21/02; C12N 15/29, 15/70  
 US CL : 435/71.1; 530/370  
 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. : 435/71.1; 530/370

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)  
 Please See Continuation Sheet

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X,P -- Y,P	Database Uniprot on STIC, European Bioinformatics Institute, (Hinxton, Cambridge, UK), Accession No. Q6AVN6, CHOW et al., Protein Sequence from nucleic acid, 25 October 2004.	1-3 ----- 22, 31-32
X,P -- Y,P	Database GenEmbl on STIC, European Bioinformatics Institute, (Hinxton, Cambridge, UK), Accession No. AC097175, CHOW et al., Gene Sequence, 22 July 2004.	1-3 ----- 22, 31-32
Y	DUBOUZET J.G. et al. OsDREB genes in rice, <i>Oryza sativa</i> L., encode transcription activators that function in drought-, high-salt- and cold-responsive gene expression. The Plant Journal. 2003, Vol. 33, pages 751-763, especially page 761 column one last paragraph.	22, 31-32

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

* Special categories of cited documents:	"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
"A" document defining the general state of the art which is not considered to be of particular relevance	"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
"B" earlier application or patent published on or after the international filing date	"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
"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)	"&"	document member of the same patent family
"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		

Date of the actual completion of the international search: 21 October 2005 (21.10.2005)  
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# INTERNATIONAL SEARCH REPORT

International application No.

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## 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 additional fees, this Authority did not invite payment of any additional fees.
  3.  As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
  
  4.  No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.: 1-3, 22, 31-32 directed to SEQ ID NO:1
- Remark on Protest
- |                          |   |
|--------------------------|---|
| <input type="checkbox"/> | The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.   |
| <input type="checkbox"/> | The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation. |
| <input type="checkbox"/> |   |

## INTERNATIONAL SEARCH REPORT

International application No.

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### 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.

Groups I-LIII, claim(s) 1-3, 22 and 31-32, drawn to an isolated polypeptide, and a method for producing a polypeptide. Group I is directed to an isolated polypeptide encoded by SEQ ID NO:1, Group II is directed to an isolated polypeptide encoded by SEQ ID NO:3, ... Group LIII is directed to an isolated polypeptide encoded by SEQ ID NO:105.

Groups LIV-CVI, claim(s) 4-20, 23-25 and 27-29, drawn to an isolated nucleic acid encoding a polypeptide, an expression cassette, a recombinant vector, a cell, a transgenic plant, and a method for expressing in a plant a nucleic acid encoding a polypeptide. Group LIV is directed to products that comprise and methods that utilize an isolated nucleic acid of SEQ ID NO:1, Group LV is directed to products that comprise and methods that utilize an isolated nucleic acid of SEQ ID NO:3, ... Group CVI is directed to products that comprise and methods that utilize an isolated nucleic acid of SEQ ID NO:105.

Groups CVII-CLIX, claim(s) 21, drawn to a shuffled nucleic acid. Group CVII is directed to a shuffled nucleic acid comprising at least one fragment corresponding to a region of SEQ ID NO:1, CVIII is directed to a shuffled nucleic acid comprising at least one fragment corresponding to a region of SEQ ID NO:3, ... Group CLIX is directed to a shuffled nucleic acid comprising at least one fragment corresponding to a region of SEQ ID NO:105.

Groups CLX-CCXII, claim(s) 23, drawn to a method for decreasing the expression of an isolated nucleic acid molecule of claim 4 in a plant by expressing a ribozyme that specifically cleaves an mRNA transcript encoded by an endogenous gene corresponding to an isolated nucleic acid molecule of claim 4. Group CLX is directed to expressing a ribozyme that specifically cleaves an mRNA transcript encoded by an endogenous gene corresponding to an isolated nucleic acid of SEQ ID NO:1, Group CLXI is directed to expressing a ribozyme that specifically cleaves an mRNA transcript encoded by an endogenous gene corresponding to an isolated nucleic acid of SEQ ID NO:3, ... Group CCXII is directed to expressing a ribozyme that specifically cleaves an mRNA transcript encoded by an endogenous gene corresponding to an isolated nucleic acid of SEQ ID NO:105.

Groups CCXIII-CCLXV, claim(s) 23, drawn to a method for decreasing the expression of an isolated nucleic acid molecule of claim 4 in a plant by expressing an aptamer specifically directed to a polypeptide encoded by an isolated nucleic acid molecule of claim 4. Group CCXIII is directed to expressing an aptamer specifically directed to an isolated polypeptide encoded by SEQ ID NO:1, Group CCXIV is directed to expressing an aptamer specifically directed to an isolated polypeptide encoded by SEQ ID NO:3, ... Group CCLXV is directed to expressing an aptamer specifically directed to an isolated polypeptide encoded by SEQ ID NO:105.

Groups CCLXVI-CCCXVIII, claim(s) 23, drawn to a method for decreasing the expression of an isolated nucleic acid molecule of claim 4 in a plant by expressing a mutated or truncated form of an isolated nucleic acid molecule of claim 4. Group CCLXVI is directed to expressing a mutated or truncated form of an isolated nucleic acid molecule of SEQ ID NO:1, Group CCLXVII is directed to expressing a mutated or truncated form of an isolated nucleic acid molecule of SEQ ID NO:3, ... Group CCCXVIII is directed to expressing a mutated or truncated form of an isolated nucleic acid molecule of SEQ ID NO:105.

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Groups CCCXIX-CCCLXXI, claim(s) 23, drawn to a method for decreasing the expression of an isolated nucleic acid molecule of claim 4 in a plant by homologous recombination with a chromosomal copy of a gene corresponding to an isolated nucleic acid molecule of claim 4. Group CCCXIX is directed to homologous recombination with a chromosomal copy of a gene corresponding to an isolated nucleic acid molecule of SEQ ID NO:1, Group CCCXX is directed to homologous recombination with a chromosomal copy of a gene corresponding to an isolated nucleic acid molecule of SEQ ID NO:3, ... Group CCCLXXI is directed to homologous recombination with a chromosomal copy of a gene corresponding to an isolated nucleic acid molecule of SEQ ID NO:105.

Groups CCCLXXII-CDXXIV, claim(s) 23, drawn to a method for decreasing the expression of an isolated nucleic acid molecule of claim 4 in a plant by homologous recombination with a chromosomal copy of regulatory element of a gene corresponding to an isolated nucleic acid molecule of claim 4. Group CCCLXXII is directed to homologous recombination with a chromosomal copy of regulatory element of a gene corresponding to an isolated nucleic acid molecule of SEQ ID NO:1, Group CCCLXXIII is directed to homologous recombination with a chromosomal copy of regulatory element of a gene corresponding to an isolated nucleic acid molecule of SEQ ID NO:3, ... Group CDXXIV is directed to homologous recombination with a chromosomal copy of regulatory element of a gene corresponding to an isolated nucleic acid molecule of SEQ ID NO:105.

Groups CDXXV -CDLXXVII, claim(s) 26, drawn to an antibody that specifically binds to the polypeptide of claim 1. Group CDXXV is directed to an antibody that specifically binds to an isolated polypeptide encoded by SEQ ID NO:1, Group CDXXVI is directed to an antibody that specifically binds to an isolated polypeptide encoded by SEQ ID NO:3, ... Group CDLXXVII is directed to an antibody that specifically binds to an isolated polypeptide encoded by SEQ ID NO:105.

Groups CDLXXVIII -DXXX, claim(s) 30, drawn to a product isolated from a transformed plant. Group CDLXXVIII is directed to a product isolated from a plant transformed with an isolated nucleic acid of SEQ ID NO:1, Group CDLXXIX is directed to a product isolated from a plant transformed with an isolated nucleic acid of SEQ ID NO:3, ... Group DXXX is directed to a product isolated from a plant transformed with an isolated nucleic acid of SEQ ID NO:105.

The inventions listed as Groups I-DXXX do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

The inventions listed as Groups I-DXXX 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:

A) The invention has no special technical feature that defined the contribution over the prior art, or  
B) Unity of invention between different categories of inventions will only be found to exist if specific combinations of inventions are present. Those combinations include:

- 1) A product and a special process of manufacture of said product.
- 2) A product and a process of use of said product.
- 3) A product, a special process of manufacture of said product, and a process of use of said product.
- 4) A product and an apparatus specially designed to carry out said process.

5) A product, a special process of manufacture of said product, and an apparatus specially designed to carry out said process.

A) The technical feature linking the inventions of Groups I-LIII is a polypeptide for abiotic stress tolerance encoded by a nucleic acid molecule isolated from *Oryza sativa*. However, a polypeptide for abiotic stress tolerance encoded by a nucleic acid molecule isolated from *Oryza sativa* is obvious or anticipated over DUBOUZET et al. (The Plant Journal, February 2003, Vol. 33, pages 751-763), for example, and therefore does not constitute a special technical feature as defined by PCT Rule 13.2, because it does not define a contribution over the prior art. The special technical feature of each of Groups I-LIII is the particular sequence characteristic of each group.

The technical feature linking the inventions of Groups LIV-CLIX is a nucleic acid molecule isolated from *Oryza sativa* that encodes a polypeptide for abiotic stress tolerance. However, a nucleic acid molecule isolated from *Oryza sativa* that encodes a polypeptide for abiotic stress tolerance is obvious or anticipated over DUBOUZET et al. (The Plant Journal, February 2003, Vol. 33, pages 751-763), for example, and therefore does not constitute a special technical feature as defined by PCT Rule 13.2, because it does not define a contribution over the prior art. The special technical feature of each of Groups LIV-CLIX is the particular sequence characteristic of each group.

The technical feature linking the inventions of Groups CLX-CDXXIV is decreasing the expression of nucleic acid molecules isolated from *Oryza sativa* that encode polypeptides for abiotic stress tolerance. However, decreasing the expression of nucleic acid molecules isolated from *Oryza sativa* that encode polypeptides for abiotic stress tolerance is obvious or anticipated over LI et al. (Biol. Pharm. Bull., February 2003, Vol. 26, No. 2, pages 256-261), for example, and therefore does not constitute a special technical feature as defined by PCT Rule 13.2, because it does not define a contribution over the prior art. The special technical feature of each of Groups CLX-CDXXIV is the particular materials required to practice the particular methodologies.

The technical feature linking the inventions of Groups CDXXV-CDLXXVII is an antibody that specifically binds to a polypeptide for abiotic stress tolerance encoded by a nucleic acid molecule isolated from *Oryza sativa*. However, an antibody that specifically binds to a polypeptide for abiotic stress tolerance encoded by a nucleic acid molecule isolated from *Oryza sativa* is obvious or anticipated over LI et al. (Biol. Pharm. Bull., February 2003, Vol. 26, No. 2, pages 256-261), for example, and therefore does not

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constitute a special technical feature as defined by PCT Rule 13.2, because it does not define a contribution over the prior art. The special technical feature of each of CDXXV-CDLXXVII is the particular antibody characteristic of each group.

The technical feature linking the inventions of Groups CDLXXVIII-DXXX is a product isolated from a transformed plant. However, a product isolated from a transformed plant is obvious or anticipated over either of DUBOUZET et al. (The Plant Journal, February 2003, Vol. 33, pages 751-763) or LI et al. (Biol. Pharm. Bull., February 2003, Vol. 26, No. 2, pages 256-261), for example, and therefore does not constitute a special technical feature as defined by PCT Rule 13.2, because it does not define a contribution over the prior art. The special technical feature of each of Groups CDLXXVIII-DXXX is the particular product isolated.

Groups I-LIII, Groups LIV-CLIX, Groups CLX-CDXXIV, Groups CDXXV-CDLXXVII and Groups CDLXXVIII-DXXX are not linked by a technical feature.

B) The allowed combinations do not include multiple products and multiple methods of using said products as claimed in the instant application; See MPEP § 1850. Applicant's claims encompass multiple inventions and do not have a special technical feature which links the inventions one to the other and they thus lack unity of invention.

Continuation of B. FIELDS SEARCHED Item 3:

STN (agricola, biosis, biotecnologia, caba, caplus, medline, uspatfull): inventor names, plant, stress, protein. STIC sequence search for SEQ ID NO:1.