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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.*
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(54) Title: MAIZE GLUTATHIONE-S-TRANSFERASE ENZYMES

(57) Abstract: This invention relates to isolated nucleic acid fragments encoding all or a substantial portion of maize glutathione-S-transferase (GST) enzymes involved in the detoxification of xenobiotic compounds in plants and seeds. The invention also relates to the construction of chimeric genes encoding all or a substantial portion of maize GST enzymes, host cells transformed with those genes and methods of the recombinant production of maize GST enzymes. Methods of constructing transgenic plants having altered levels of GST enzymes and screens for identifying maize GST enzyme substrates and maize GST enzyme inhibitor, are also provided.

INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 00/03348

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12N15/54 C12N15/82 C12N9/10 C12Q1/68 C12Q1/48
G01N33/573 A01H5/00

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12N C12Q G01N A01H

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 practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
E	WO 00 18937 A (DU PONT) 6 April 2000 (2000-04-06) the whole document ---	1-22
L,P, X	US 5 962 229 A (MCGONIGLE BRIAN ET AL) 5 October 1999 (1999-10-05) brings into doubt the priority right of the present application the whole document ---	1-22
P,X	WO 99 14337 A (RHONE POULENC AGRICULTURE ;CUMMINS IAN (GB); COLE DAVID J (GB); ED) 25 March 1999 (1999-03-25) see SEQ ID NO:13 ---	1,3, 5-12,18, 21
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Further documents are listed in the continuation of box C.

Patent family members are listed in 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 document but published on or after the international filing date
- *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- *O* document referring to an oral disclosure, use, exhibition or other means
- *P* document published prior to the international filing date but later than the priority date claimed

- *T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- *X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- *Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- *Z* document member of the same patent family

Date of the actual completion of the international search

6 July 2000

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INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 00/03348

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	GROVE G ET AL: "CHARACTERIZATION AND HETEROSPECIFIC EXPRESSION OF CDNA CLONES OF GENES IN THE MAIZE GSH S-TRANSFERASE MULTIGENE FAMILY" NUCLEIC ACIDS RESEARCH,GB,OXFORD UNIVERSITY PRESS, SURREY, vol. 16, no. 2, 1988, pages 425-438, XP002000776 ISSN: 0305-1048 the whole document & EMBL ACCESSION NO:X06754, 16 July 1998 (1998-07-16), ---	1,3,5,6, 8,9,12, 18,19,21
X	WOSNICK M A ET AL: "TOTAL CHEMICAL SYNTHESIS AND EXPRESSION IN ESCHERICHIA-COLI OF A MAIZE GLUTATHIONE TRANSFERASE GST GENE" GENE (AMSTERDAM), vol. 76, no. 1, 1989, pages 153-160, XP002141993 ISSN: 0378-1119 the whole document & EMBL ACCESSION NO:M24889, 23 November 1989 (1989-11-23), ---	1,3,5,6, 8,9,12, 18,19,21
X	JEPSON I ET AL: "CLONING AND CHARACTERIZATION OF MAIZE HERBICIDE SAFENER-INDUCED CDNAS ENCODING SUBUNITS OF GLUTATHIONE S-TRANSFERASE ISOFORMS I, II AND IV" PLANT MOLECULAR BIOLOGY,NL,NIJHOFF PUBLISHERS, DORDRECHT, vol. 26, no. 6, 1 December 1994 (1994-12-01), pages 1855-1866, XP000616303 ISSN: 0167-4412 the whole document & EMBL ACCESSION NO:X79515, 28 February 1995 (1995-02-28), XP002101643 ---	1,3,12
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X	WO 96 23072 A (BAYER AG ;BIESELER BARBARA (DE); REINEMER PETER (DE); HAIN RUEDIGE) 1 August 1996 (1996-08-01) the whole document ---	1,3,5-9, 12
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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>DATABASE BIOSIS [Online] BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US October 1998 (1998-10) MCGONIGLE BRIAN ET AL: "Homoglutathione selectivity by soybean glutathione S-transferases." Database accession no. PREV199800516177 XP002142056 abstract & PESTICIDE BIOCHEMISTRY AND PHYSIOLOGY, vol. 62, no. 1, October 1998 (1998-10), pages 15-25, ISSN: 0048-3575</p>	1-22
A	<p>--- MOORE R E ET AL: "CLONING AND EXPRESSION OF A CDNA ENCODING A MAIZE GLUTATHIONE-S-TRANSFERASE IN E. COLI" NUCLEIC ACIDS RESEARCH, GB, OXFORD UNIVERSITY PRESS, SURREY, vol. 14, 1986, pages 7227-7235, XP002000777 ISSN: 0305-1048 the whole document</p>	1-22
A	<p>--- DIXON ET AL: "purification, regulation and cloning of a glutathione transferase (GST) from maize resembling the auxin-inducible type-III GSTs" PLANT MOLECULAR BIOLOGY, NL, NIJHOFF PUBLISHERS, DORDRECHT, vol. 36, no. 36, 1998, pages 75-87-87, XP002112985 ISSN: 0167-4412 the whole document</p>	1-22
A	<p>--- DIXON ET AL: "glutathione-mediated detoxification systems in plants" CURRENT OPINION IN PLANT BIOLOGY, GB, QUADRANT SUBSCRIPTION SERVICES, vol. 1, no. 3, June 1998 (1998-06), pages 258-266-266, XP002112989 ISSN: 1369-5266 the whole document</p>	1-22
A	<p>--- TIMMERMAN K P: "MOLECULAR CHARACTERIZATION OF CORN GLUTATHIONE S-TRANSFERASE ISOZYMES INVOLVED IN HERBICIDE DETOXICATION" PHYSIOLOGIA PLANTARUM, DK, MUNKSGAARD INTERNATIONAL PUBLISHERS, COPENHAGEN, vol. 77, 1 January 1989 (1989-01-01), pages 465-471, XP002000778 ISSN: 0031-9317 the whole document</p>	1-22
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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	NEUEFEIND ET AL: "plant glutathione S-transferases and herbicide detoxification" BIOLOGICAL CHEMISTRY,XX,XX, vol. 378, no. 3/04, March 1997 (1997-03), pages 199-205-205, XP002112990 the whole document	1-22
A	--- IRZYK GERARD ET AL: "A cDNA clone encoding the 27-kilodalton subunits of glutathione S-transferase IV from Zea mays." PLANT PHYSIOLOGY (ROCKVILLE), vol. 107, no. 1, 1995, pages 311-312, XP002142007 ISSN: 0032-0889 the whole document & EMBL ACCESSION NO:U12679, 23 August 1994 (1994-08-23),	10,11
A	--- DATABASE BIOSIS [Online] BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US1979 GUDEWAR M B ET AL: "PURIFICATION AND PROPERTIES OF A GLUTATHIONE S TRANSFERASE FROM CORN WHICH CONJUGATES S TRIAZINE HERBICIDES" Database accession no. PREV197968050399 XP002141995 abstract & PHYTOCHEMISTRY (OXFORD), vol. 18, no. 5, 1979, pages 735-740, EN ISSN: 0031-9422	13,16
A	--- DROOG FRANS N J ET AL: "2,4-Dichlorophenoxyacetic acid and related chlorinated compounds inhibit two auxin-regulated type-III tobacco glutathione S-transferases." PLANT PHYSIOLOGY (ROCKVILLE), vol. 107, no. 4, 1995, pages 1139-1146, XP002141994 ISSN: 0032-0889 the whole document -----	13,16

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 00/03348

Box I Observations where certain claims were found unsearchable (Continuation of Item 1 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 II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

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

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.:

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.:

1-22 all partially

Remark on Protest

- The additional search fees were accompanied by the applicant's protest.
- No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Invention 1: Claims 1-22 all partially

Nucleic acid fragments encoding a maize GST enzyme as represented by SEQ ID NO: 1 and the corresponding amino acid SEQ ID NO:2, chimeric genes, transformed hosts, methods for altering GST levels, obtaining the fragment and identifying substrates and inhibitors of the GST, using said fragment and polypeptide as defined above.

Inventions 2-8. Claims 1-22 all partially.

As first group but for fragments represented by SEQ ID NO:3 through 15(odd numbers) independently and corresponding amino acid SEQ ID NO:4 through 16(even numbers).

Inventions 9. Claims 1-22 all partially.

As first group but for type II maize GST fragments represented by SEQ ID NOS:17 and 19 and corresponding amino acid SEQ ID NOS:18 and 20.

Invention 10. Claims 1-22 all partially.

As first group but for fragments represented by SEQ ID NO:21 and corresponding amino acid SEQ ID NO:22.

Invention 11. Claims 1-22 all partially.

As first group but for fragments represented by SEQ ID NO:23 and corresponding amino acid SEQ ID NO:24.

Inventions 12-36. Claims 23-44 all partially

As first group but for fragments represented by SEQ ID NO:25 through 73(odd numbers) independently and corresponding amino acid SEQ ID NO:26 through 74(even numbers)

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/US 00/03348

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