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- (81) Designated States (national): AE, AG, AL, AM, AT, AU,  
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GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC,  
LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW,  
MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG,  
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(BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR,  
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- Published:  
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WO 2003/000941 A3

(54) Title: POLYPEPTIDES HAVING CELLOBIOHYDROLASE I ACTIVITY AND POLYNUCLEOTIDES ENCODING SAME

(57) Abstract: The present invention relates to polypeptides having cellobiohydrolase I activity and polynucleotides having a nucleotide sequence which encodes for the polypeptides. The invention also relates to nucleic acid constructs, vectors, and host cells comprising the nucleic acid constructs as well as methods for producing and using the polypeptides, e.g. for producing ethanol from biomass or for shuffling of DNA.

# INTERNATIONAL SEARCH REPORT

International Application No

PCT/DK 02/00429

**A. CLASSIFICATION OF SUBJECT MATTER**

IPC 7 C12N9/00

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

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)

EPO-Internal, EMBASE

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>DATABASE GENESEQ [Online]                      27 March 2002 (2002-03-27)                      retrieved from GENESEQ                      Database accession no. ABA92722                      XP002902802                      80,6% identity with SEQ ID No1 in 1204nt                      overlap                      -&amp; WO 01 25468 A (EMALFARB MARK AARON)                      12 April 2001 (2001-04-12)                      page 5 sequence listing. SEQ ID No 3                      ---                      -/--</p>	1-29

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.
- "&" document member of the same patent family

Date of the actual completion of the international search

1 December 2002

Date of mailing of the international search report

3 7. 06 03

Name and mailing address of the ISA

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## INTERNATIONAL SEARCH REPORT

International Application No

PCT/DK 02/00429

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT		
Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	MARISTELLA DE OLIVIERA AZEVEDO ET AL: "Sequence of CBH-I gene of humicola grisea var. thermoidea." NUCLEIC ACIDS RESEARCH, vol. 18, no. 3, 1990, page 668 XP002902803 oxford university press the whole document -& DATABASE EMBASE [Online] ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, NL; 13 March 1990 (1990-03-13) XP002902804 78,5% identity with SEQ ID No 1 in 1176 nt overlap ---	1-29
P,X	DATABASE GENESEQ [Online] 13 March 2002 (2002-03-13) retrieved from GENESEQ Database accession no. AAI72045 XP002902805 80.7% identity with SEQ ID No 1 in 1204nt overlap -& WO 01 79507 A (EMALFARB MARK AARON) 25 October 2001 (2001-10-25) page 31 ---	1-29
X	US 5 955 270 A (RADFORD ALAN ET AL) 21 September 1999 (1999-09-21) column 1, line 59 - line 61 column 3, line 24 - line 28 column 6, line 27 - line 31 ---	1-29
X	WO 99 06574 A (VISSER JACOB ;GIELKENS MARCUS MATHEUS CATHAR (NL); GIST BROCADES B) 11 February 1999 (1999-02-11) page 3, line 20 - line 23 page 5, line 25 - line 29 ---	1-29
X	WO 01 04284 A (MID-WEST RESEARCH INSTITUTE) 18 January 2001 (2001-01-18) page 1, line 4 - line 18 ---	1-29
X	F. TALEB ET AL: "The cellulase complex of neurospora crassa: CBH-1 cloning, sequencing and homologies." GENE, vol. 161, 1995, pages 137-138, XP002902806 ELSEVIER SCIENCE BV 1995 ISSN: 0378-1119 the whole document -----	1-29

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/DK 02/00429

## 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.: **1(b), 18, 21, 23, 24**  
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:  
**see FURTHER INFORMATION sheet PCT/ISA/210**
  
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:

see additional 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.:
  
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.:  
**1-29 (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

Continuation of Box I.2

Claims Nos.: 1(b),18, 21, 23, 24

The wording of claim 1 (b), "the polypeptide encoded by the cellobiohydrolase I encoding part of the nucleotide sequence", is vague and undefined. Support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT is lacking. A specified amino acid sequence or polynucleotide sequence is required in order for the polypeptide to be sufficiently defined. The above reasoning is also relevant to the wording of claim 18 and 21.

Claim 23 and 24 relate to an extremely large number of possible compounds. Support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT is to be found, however, for only a very small proportion of the compounds claimed. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible.

The search has been based on the polynucleotide and polypeptide actually provided (SEQ ID No 1-2) and applications thereof.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:2 and the corresponding polynucleotide of SEQ ID N0:1. Methods of production and uses of said polypeptide and polynucleotide.

2. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:4 and the corresponding polynucleotide of SEQ ID N0:3. Methods of production and uses of said polypeptide and polynucleotide.

3. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:6. and the corresponding polynucleotide of SEQ ID N0:5. Methods of production and uses of said polypeptide and polynucleotide

4. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:8. and the corresponding polynucleotide of SEQ ID N0:7. Methods of production and uses of said polypeptide and polynucleotide

5. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:10. and the corresponding polynucleotide of SEQ ID N0:9. Methods of production and uses of said polypeptide and polynucleotide

6. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:12. and the corresponding polynucleotide of SEQ ID N0:11. Methods of production and uses of said polypeptide and polynucleotide

7. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:14. and the corresponding polynucleotide of SEQ ID N0:13. Methods of production and uses of said polypeptide and polynucleotide

8. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:16. and the corresponding

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

polynucleotide of SEQ ID N0:15. Methods of production and uses of said polypeptide and polynucleotide

9. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:37,38. and the corresponding polynucleotide of SEQ ID N0:20. Methods of production and uses of said polypeptide and polynucleotide

10. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:40 and the corresponding polynucleotide of SEQ ID N0:39. Methods of production and uses of said polypeptide and polynucleotide

11. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:42 and the corresponding polynucleotide of SEQ ID N0:41. Methods of production and uses of said polypeptide and polynucleotide

12. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:43,44 and the corresponding polynucleotide of SEQ ID N0:25. Methods of production and uses of said polypeptide and polynucleotide

13. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:45,46,47,48 and the corresponding polynucleotide of SEQ ID N0:24. Methods of production and uses of said polypeptide and polynucleotide

14. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:50. and the corresponding polynucleotide of SEQ ID N0:49. Methods of production and uses of said polypeptide and polynucleotide

15. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:52. and the corresponding polynucleotide of SEQ ID N0:51. Methods of production and uses of said polypeptide and polynucleotide

16. Claims: 1-29 (partially)

## FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

The polypeptide of SEQ ID No:54. and the corresponding polynucleotide of SEQ ID N0:53. Methods of production and uses of said polypeptide and polynucleotide

## 17. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:56. and the corresponding polynucleotide of SEQ ID N0:55. Methods of production and uses of said polypeptide and polynucleotide

## 18. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:58. and the corresponding polynucleotide of SEQ ID N0:57. Methods of production and uses of said polypeptide and polynucleotide

## 19. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:60. and the corresponding polynucleotide of SEQ ID N0:59. Methods of production and uses of said polypeptide and polynucleotide

## 20. Claims: 1-29 (partially)

The polypeptide of SEQ ID No:66 and the corresponding polynucleotide of SEQ ID N0:65. Methods of production and uses of said polypeptide and polynucleotide

## 21. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:17. Methods of production and uses of said polynucleotide.

## 22. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:18. Methods of production and uses of said polynucleotide.

## 23. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:19. Methods of production and uses of said polynucleotide.

## 24. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:21. Methods of production and uses of said polynucleotide.

**FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210**

25. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:22. Methods of production and uses of said polynucleotide.

26. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:23. Methods of production and uses of said polynucleotide.

27. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:26. Methods of production and uses of said polynucleotide.

28. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:27. Methods of production and uses of said polynucleotide.

29. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:28. Methods of production and uses of said polynucleotide.

30. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:29. Methods of production and uses of said polynucleotide.

31. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:30. Methods of production and uses of said polynucleotide.

32. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:31. Methods of production and uses of said polynucleotide.

33. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:32. Methods of production and uses of said polynucleotide.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

34. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:33. Methods of production and uses of said polynucleotide.

35. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:34. Methods of production and uses of said polynucleotide.

36. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:35. Methods of production and uses of said polynucleotide.

37. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:36. Methods of production and uses of said polynucleotide.

38. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:61. Methods of production and uses of said polynucleotide.

39. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:62. Methods of production and uses of said polynucleotide.

40. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:63. Methods of production and uses of said polynucleotide.

41. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:64. Methods of production and uses of said polynucleotide.

42. Claims: 1-29 (partially)

The polynucleotide of SEQ ID No:67. Methods of production and uses of said polynucleotide.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/DK 02/00429

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 0125468	A	12-04-2001	AU 4352700 A 10-05-2001
			AU 6232699 A 26-04-2000
			BR 9914278 A 19-06-2001
			CA 2345356 A1 13-04-2000
			EP 1117808 A2 25-07-2001
			WO 0125468 A1 12-04-2001
-----			
WO 0179507	A	25-10-2001	AU 5066301 A 30-10-2001
			BR 0110090 A 18-02-2003
			EP 1276876 A2 22-01-2003
			WO 0179507 A2 25-10-2001
			NO 20024945 A 12-12-2002
-----			
US 5955270	A	21-09-1999	AU 1389895 A 01-08-1995
			WO 9519441 A1 20-07-1995
-----			
WO 9906574	A	11-02-1999	AU 739445 B2 11-10-2001
			AU 9259698 A 22-02-1999
			BR 9812113 A 18-07-2000
			WO 9906574 A1 11-02-1999
			EP 1002102 A1 24-05-2000
			JP 2001512024 T 21-08-2001
			NZ 502444 A 30-11-2001
			US 6558937 B1 06-05-2003
-----			
WO 0104284	A	18-01-2001	AU 6210800 A 30-01-2001
			CA 2379326 A1 18-01-2001
			EP 1200574 A1 02-05-2002
			WO 0104284 A1 18-01-2001
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