

(19) World Intellectual Property  
Organization  
International Bureau



(43) International Publication Date  
12 September 2003 (12.09.2003)

PCT

(10) International Publication Number  
**WO 2003/074657 A3**

- (51) International Patent Classification<sup>7</sup>: **C12N 1/21**, [CN/US]; 900 Davidson Road, 69 Nichols Apt., Piscataway, NJ 08854 (US). **KE, Haiping** [CN/US]; 102-3G Montgomery Street, Highland Park, NJ 08904 (US).  
5/10, 15/63, C12P 21/02, G01V 3/14
- (21) International Application Number: PCT/US2003/005531 (74) Agents: **BROWDY AND NEIMARK, P.L.L.C.** et al.; 624 Ninth Street N.W., Suite 300, Washington, DC 20001-5303 (US).
- (22) International Filing Date: 25 February 2003 (25.02.2003)
- (25) Filing Language: English (81) Designated States (national): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, 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, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.
- (26) Publication Language: English (84) Designated States (regional): ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).
- (30) Priority Data:  
60/361,069 1 March 2002 (01.03.2002) US  
60/402,921 14 August 2002 (14.08.2002) US
- (71) Applicant (for all designated States except US): **TAKARA BIO, INC.** [JP/JP]; 4-1, Seta 3-chome, Otsu-shi, Shiga 520-2193 (JP).
- (71) Applicant (for SD only): **MCINNIS, Patricia** [US/US]; 2325 42nd Street, N.W., Apt. 203, Washington, D.C., DC 20007 (US).
- (72) Inventors; and
- (75) Inventors/Applicants (for US only): **INOUE, Masayori** [JP/US]; The Colony House, Apt. 16K, 1050 George Street, New Brunswick, NJ 08901 (US). **PHADTARE, Sangita** [IN/US]; 12-D Bartle Court, Highland Park, NJ 08904 (US). **XIA, Bing** [CN/US]; Apartment 9, 280 Grove Street, Newton, MA 02466 (US). **QING, Guoliang**
- Published:  
— with international search report
- (88) Date of publication of the international search report:  
16 September 2004
- 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: COLD SHOCK INDUCIBLE EXPRESSION AND PRODUCTION OF HETEROLOGOUS POLYPEPTIDES

(57) Abstract: The present invention relates to a DNA molecule or vector and a host cell containing this DNA molecule or vector which can be used to produce a heterologous polypeptide under conditions that elicit a cold shock response in the host cell. The DNA molecule and vector include a nucleotide sequence encoding a heterologous polypeptide and a promoter and 5'-UTR from a cold shock inducible gene which directs its expression. In addition, an AT-rich sequence that enhances translation under cold shock inducible conditions is either present in the coding sequence of the heterologous polypeptide or in an additional element inserted between the coding sequence and the cold shock inducible promoter and 5'-UTR.

WO 2003/074657 A3

**INTERNATIONAL SEARCH REPORT**

International application No.

PCT/US03/05531

**A. CLASSIFICATION OF SUBJECT MATTER**

IPC(7) : C12N 1/21, 15/63, 5/10; C12P 21/02; G01V 3/14  
 US CL : 324/309; 435/69.1, 252.3, 252.33, 320.1, 325, 419

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. : 324/309; 435/69.1, 252.3, 252.33, 320.1, 325, 419

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.
A	US 6,294,711 B1 (MEULEWAETER et al) 25 September 2001 (25.09.2001), see entire document.	1-36
A, P	US 6,479,260 B1 (TAKAYAMA et al) 12 November 2002 (12.11.2002), see entire document.	1-36
A	US 6,333,191 B1 (INOUE et al) 25 December 2001 (25.12.2001), see entire document.	1-36

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	*"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
*"E" earlier application or patent published on or after the international filing date	*"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
*"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)	*"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
*"O" document referring to an oral disclosure, use, exhibition or other means	*"Z" document member of the same patent family
*"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

22 August 2003 (22.08.2003)

Date of mailing of the international search report

09 JUN 2004

Name and mailing address of the ISA/US

Mail Stop PCT, Attn: ISA/US  
 Commissioner for Patents  
 P.O. Box 1450  
 Alexandria, Virginia 22313-1450

Authorized officer

Terry A. McKelvey

Telephone No. 703-308-0196

*Janice Ford*  
*for*

Facsimile No.

**INTERNATIONAL SEARCH REPORT**

PCT/US03/05531

**Continuation of B. FIELDS SEARCHED Item 3:**

EAST (USPAT, PGPUB, EPO, JPO, Derwent databases), Dialog OneSeach (biotech databases)  
search terms: cold shock, induc?, promoter?, regulat?, AT rich, translation, enhanc?, utr, laci