

(19) World Intellectual Property  
Organization  
International Bureau



(43) International Publication Date  
24 December 2003 (24.12.2003)

PCT

(10) International Publication Number  
**WO 2003/106672 A3**

(51) International Patent Classification<sup>7</sup>: **C12N 15/00**

DNAFORM, 3-35, Mita 1-chome, Minato-ku, Tokyo 108-0073 (JP).

(21) International Application Number:  
PCT/JP2003/007514

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(22) International Filing Date: 12 June 2003 (12.06.2003)

(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, NI, 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

(30) Priority Data:  
2002-171851 12 June 2002 (12.06.2002) JP  
2002-235294 12 August 2002 (12.08.2002) JP

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(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, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

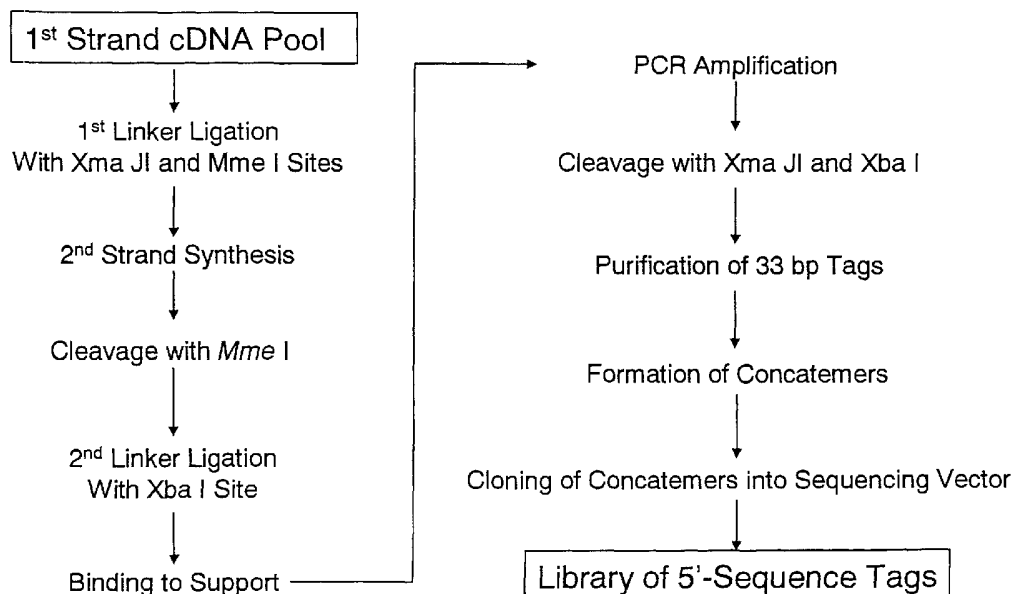
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**Published:**  
— with international search report

[Continued on next page]

(54) Title: METHOD OF UTILIZING THE 5'END OF TRANSCRIBED NUCLEIC ACID REGIONS FOR CLONING AND ANALYSIS



(57) Abstract: A method is disclosed for obtaining the 5'ends of transcribed regions from a plurality of nucleic acid fragments obtained from biological materials or synthetic pools. DNA fragments encoding the 5'ends are enriched for their individual analysis or for the analysis of concatemers thereof. The sequence information derived from 5' ends can be used for characterization and cloning of the transcriptome.



**(88) Date of publication of the international search report:**

1 July 2004

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# INTERNATIONAL SEARCH REPORT

PCT/JP 03/07514

## A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12N15/10

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, WPI Data, PAJ, BIOSIS, 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>WO 02 10438 A (JOHNS HOPKINS UNIVERSITY)<br/> 7 February 2002 (2002-02-07)<br/> cited in the application<br/> the whole document<br/> see especially:<br/> page 4, paragraphs 12,13<br/> page 10, paragraph 31<br/> page 12, paragraph 34<br/> page 14, paragraph 39 -page 15, paragraph 41<br/> page 16, paragraph 43<br/> page 19, paragraph 54 -page 20, paragraph 56<br/> page 25 -page 42; examples 1-5<br/> page 43 -page 60; claims 1-64,75-113<br/> page 61 -page 62; figure 1</p> <p style="text-align: center;">---<br/>-/-</p> | <p>1-6,<br/> 8-16,<br/> 26-55</p> |

☒ Further documents are listed in the continuation of box C.

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Date of the actual completion of the international search

25 February 2004

Date of mailing of the international search report

03/03/2004

Name and mailing address of the ISA

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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                                                    | MARUYAMA, K. & SUGANO, S.:<br>"Oligo-capping: a simple method to replace the cap structure of eukaryotic mRNAs with oligoribonucleotides"<br>GENE,<br>vol. 138, no. 1-2,<br>28 January 1994 (1994-01-28), pages<br>171-174, XP001008855<br>cited in the application<br>the whole document<br>--- | 1-55                  |
| A                                                    | SHIBATA, Y. ET AL.: "Cloning Full-Length, Cap-Trapper-Selected cDNAs by Using the Single-Strand Linker Ligation Method"<br>BIOTECHNIQUES,<br>vol. 30, no. 6, June 2001 (2001-06), pages<br>1250-1254, XP002197302<br>cited in the application<br>the whole document<br>---                       | 1-55                  |
| A                                                    | SUZUKI, Y. ET AL.: "Diverse transcriptional initiation revealed by fine, large-scale mapping of mRNA start sites"<br>EMBO REPORTS,<br>vol. 2, no. 5, May 2001 (2001-05), pages<br>388-393, XP001176665<br>cited in the application<br>the whole document<br>---                                  | 1-55                  |
| A                                                    | CARNINCI, P. & HAYASHIZAKI, Y.:<br>"High-Efficiency Full-Length cDNA Cloning"<br>METHODS IN ENZYMOLOGY,<br>vol. 303, 1999, pages 19-44, XP001022979<br>cited in the application<br>the whole document<br>---                                                                                     | 1-55                  |
| A                                                    | EDERY, I. ET AL.: "An Efficient Strategy To Isolate Full-Length cDNAs Based on an mRNA Cap Retention Procedure (CAPture)"<br>MOLECULAR AND CELLULAR BIOLOGY,<br>vol. 15, no. 6, June 1995 (1995-06), pages<br>3363-3371, XP002915963<br>cited in the application<br>the whole document<br>---    | 1-55                  |
|                                                      | ---                                                                                                                                                                                                                                                                                              |                       |
|                                                      | -/--                                                                                                                                                                                                                                                                                             |                       |

# INTERNATIONAL SEARCH REPORT

PCT/JP 03/07514

| 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>THEISSEN, H. ET AL.: "Cloning of the human cDNA for the U1 RNA-associated 70K protein"<br/>EMBO JOURNAL,<br/>vol. 5, no. 12,<br/>1 December 1986 (1986-12-01), pages 3209-3217, XP009002534<br/>cited in the application<br/>page 3211, column 1, line 38 -column 2, line 16<br/>page 3212 -page 3213; figure 3<br/>----</p>                    | 1-55                                                         |
| A                                                    | <p>EP 1 197 552 A (RIKEN; HAYASHIZAKI, Y.)<br/>17 April 2002 (2002-04-17)<br/>cited in the application<br/>the whole document<br/>----</p>                                                                                                                                                                                                         | 1-55                                                         |
| A                                                    | <p>US 6 352 828 B1 (BRENNER, S.)<br/>5 March 2002 (2002-03-05)<br/>cited in the application<br/>the whole document<br/>----</p>                                                                                                                                                                                                                    | 1-55                                                         |
| E                                                    | <p>WO 03 091416 A (LYNX THERAPEUTICS, INC.)<br/>6 November 2003 (2003-11-06)<br/><br/>the whole document<br/>see especially:<br/>page 18, line 25 -page 21, line 18<br/>and<br/>page 41 -page 43; figures 3A-C<br/>----</p>                                                                                                                        | 1-8,11,<br>12,<br>14-16,<br>26-30,<br>32,<br>37-45,<br>48-55 |
| T                                                    | <p>SHIRAKI, T. ET AL.: "Cap analysis gene expression for high-throughput analysis of transcriptional starting point and identification of promoter usage"<br/>PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF USA,<br/>vol. 100, no. 26,<br/>23 December 2003 (2003-12-23), pages 15776-15781, XP001161070<br/>the whole document<br/>-----</p> | 1-55                                                         |

## INTERNATIONAL SEARCH REPORT

International Application No

PCT/JP 03/07514

| Patent document<br>cited in search report |    | Publication<br>date | Patent family<br>member(s) | Publication<br>date |
|-------------------------------------------|----|---------------------|----------------------------|---------------------|
| WO 0210438                                | A  | 07-02-2002          | AU 8087501 A               | 13-02-2002          |
|                                           |    |                     | WO 0210438 A2              | 07-02-2002          |
|                                           |    |                     | US 2003008290 A1           | 09-01-2003          |
| EP 1197552                                | A  | 17-04-2002          | CA 2356123 A1              | 25-02-2002          |
|                                           |    |                     | EP 1197552 A2              | 17-04-2002          |
|                                           |    |                     | JP 2002253237 A            | 10-09-2002          |
|                                           |    |                     | US 2002106666 A1           | 08-08-2002          |
| US 6352828                                | B1 | 05-03-2002          | US 5846719 A               | 08-12-1998          |
|                                           |    |                     | US 5604097 A               | 18-02-1997          |
|                                           |    |                     | AU 733782 B2               | 24-05-2001          |
|                                           |    |                     | AU 3374097 A               | 05-01-1998          |
|                                           |    |                     | CA 2256700 A1              | 11-12-1997          |
|                                           |    |                     | CN 1230226 A               | 29-09-1999          |
|                                           |    |                     | CZ 9803979 A3              | 14-07-1999          |
|                                           |    |                     | EP 0923650 A1              | 23-06-1999          |
|                                           |    |                     | HU 0003944 A2              | 28-03-2001          |
|                                           |    |                     | JP 2000515006 T            | 14-11-2000          |
|                                           |    |                     | NO 985698 A                | 08-02-1999          |
|                                           |    |                     | PL 331513 A1               | 19-07-1999          |
|                                           |    |                     | US 6138077 A               | 24-10-2000          |
|                                           |    |                     | US 6172218 B1              | 09-01-2001          |
|                                           |    |                     | WO 9746704 A1              | 11-12-1997          |
|                                           |    |                     | US 6235475 B1              | 22-05-2001          |
|                                           |    |                     | US 6172214 B1              | 09-01-2001          |
|                                           |    |                     | US 6150516 A               | 21-11-2000          |
|                                           |    |                     | US 6013445 A               | 11-01-2000          |
|                                           |    |                     | AU 712929 B2               | 18-11-1999          |
|                                           |    |                     | AU 4277896 A               | 06-05-1996          |
|                                           |    |                     | AU 5266399 A               | 09-12-1999          |
|                                           |    |                     | CA 2202167 A1              | 25-04-1996          |
|                                           |    |                     | CA 2441634 A1              | 25-04-1996          |
|                                           |    |                     | CZ 9700866 A3              | 17-09-1997          |
|                                           |    |                     | EP 0793718 A1              | 10-09-1997          |
|                                           |    |                     | FI 971473 A                | 04-06-1997          |
|                                           |    |                     | HU 77916 A2                | 28-10-1998          |
|                                           |    |                     | JP 10507357 T              | 21-07-1998          |
|                                           |    |                     | NO 971644 A                | 02-06-1997          |
|                                           |    |                     | US 6280935 B1              | 28-08-2001          |
|                                           |    |                     | WO 9612014 A1              | 25-04-1996          |
|                                           |    |                     | US 5635400 A               | 03-06-1997          |
|                                           |    |                     | US 5654413 A               | 05-08-1997          |
|                                           |    |                     | AU 3946195 A               | 06-05-1996          |
|                                           |    |                     | DE 69513997 D1             | 20-01-2000          |
|                                           |    |                     | DE 69513997 T2             | 27-07-2000          |
|                                           |    |                     | EP 0786014 A1              | 30-07-1997          |
|                                           |    |                     | EP 0952216 A2              | 27-10-1999          |
|                                           |    |                     | WO 9612039 A1              | 25-04-1996          |
|                                           |    |                     | US 6140489 A               | 31-10-2000          |
|                                           |    |                     | US 5695934 A               | 09-12-1997          |
|                                           |    |                     | US 5863722 A               | 26-01-1999          |
| WO 03091416                               | A  | 06-11-2003          | WO 03091416 A2             | 06-11-2003          |
|                                           |    |                     | US 2004002104 A1           | 01-01-2004          |