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(54) Title: METHODS AND MEANS FOR REGULATION OF GENE EXPRESSION

(57) Abstract: A transcriptional activator which comprises a fusion protein, the fusion protein comprising at least two polypeptide components, a first polypeptide component which binds in a sequence specific manner to an operator sequence in DNA, and a second polypeptide component, comprising a Lux-R transcription factor regulatory domain which binds cognate AcylHSL or an analogue thereof, whereby upon binding of the AcylHSL or analogue the DNA binding function of the first polypeptide component is activated; the transcriptional activator additionally comprising a third polypeptide component which activates transcription in eukaryotic cells.



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# INTERNATIONAL SEARCH REPORT

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## A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12N15/62 C07K19/00 C12N15/12 C07K14/47 C12N15/31  
C07K14/195 C07K14/28 C12N15/38 C07K14/035 C12N15/67  
C12N15/85 C07D307/33

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 C07K C07D

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.                                |
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| P, X       | WO 00 09704 A (ZENECA LIMITED (GB); MARTINEZ ALBERTO; JEPSON IAN; FRAY RUPERT GEORGE) 24 February 2000 (2000-02-24)<br><br>page 17 -page 23; figures 9,10; examples 1-4<br>page 29 -page 35; claims<br>--- | 1-7, 12,<br>15-17,<br>19, 20,<br>22-33,<br>36-38, 42 |
| E          | WO 01 02593 A (CALGENE LLC (US); MCBRIDE OULMASSOV; MILLER; ANDERSON; CROSSLAND ADAMS) 11 January 2001 (2001-01-11)<br><br>page 36 -page 40; figures 10,13,15,16,18<br>---<br>-/--                         | 1-7, 12,<br>15-17,<br>22-33,<br>36-38, 42            |



Further documents are listed in the continuation of box C.



Patent family members are listed in annex.

\* Special categories of cited documents:

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- \*O\* document referring to an oral disclosure, use, exhibition or other means
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- \*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

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## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

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| X        | US 5 196 318 A (TEXAS A&M UNIVERSITY SYSTEM (US); BALDWIN T.O; DEVINE J.H. SHADEL G.S.) 23 March 1993 (1993-03-23)                                                                                                                                                                                                                                                            | 1,3-6,<br>20,<br>22-33,<br>36,37,42 |
| Y        | column 4, line 47 -column 8, line 28<br>---                                                                                                                                                                                                                                                                                                                                   | 7,12                                |
| Y        | FUQUA C. ET AL.: "Census and consensus in bacterial ecosystems: the LuxR-LuxI family of quorum-sensing transcriptional regulators"<br>ANNUAL REVIEW OF MICROBIOLOGY,<br>vol. 50, 1996, pages 727-751, XP000857492<br>ISSN: 0066-4227<br>cited in the application<br>page 734; figure 1<br>---                                                                                 | 7,12                                |
| X        | HWANG I. ET AL.: "Modulating quorum sensing by antiactivation: TraM interacts with TraR to inhibit activation of Ti plasmid conjugal transfer genes"<br>MOLECULAR MICROBIOLOGY,<br>vol. 34, no. 2, October 1999 (1999-10),<br>pages 282-294, XP001002985<br>page 283, right-hand column<br>---                                                                                | 1-4,7,<br>12,19,<br>20,22-26        |
| X        | ZHU J. AND WINANS S.C.: "Autoinducer binding by the quorum-sensing regulator TraR increases affinity for target promoters in vitro and decreases TraR turnover rates in whole cells"<br>PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE USA,<br>vol. 96, April 1999 (1999-04), pages 4832-4837, XP002173293<br>cited in the application<br>page 4832 -page 4835<br>--- | 1,3,4,7,<br>12,20,<br>22-27         |
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| A        | GYURIS J. ET AL.: "Cdi1, a human G1 and S phase protein phosphatase that associates with Cdk2"<br>CELL,<br>vol. 75, 19 November 1993 (1993-11-19),<br>pages 791-803, XP000673597<br>ISSN: 0092-8674<br>page 792; figure 1<br>---<br>-/--                                                                                                                                      | 2                                   |

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International Application No

PC1/IB 00/01828

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

| Category | Citation of document, with indication, where appropriate, of the relevant passages                                                                                                                                                                                                                                                       | Relevant to claim No. |
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| A        | DEVINE J.H. ET AL.: "Nucleotide sequence of the luxR and luxI genes and structure of the primary regulatory region of the lux regulon of <i>Vibrio fischeri</i> ATCC 7744" BIOCHEMISTRY, vol. 27, 1988, pages 837-842, XP002152246 ISSN: 0006-2960 cited in the application page 839 -page 840; figures 3,4<br>---                       | 6                     |
| A        | SLOCK J. ET AL.: "Critical regions of the <i>Vibrio fischeri</i> LuxR protein defined by mutational analysis" JOURNAL OF BACTERIOLOGY, vol. 172, no. 7, July 1990 (1990-07), pages 3974-3979, XP000884181 ISSN: 0021-9193 page 3977; figure 3<br>---                                                                                     | 6                     |
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| A        | CHOI S.H. AND GREENBERG E.P.: "Genetic dissection of DNA binding and luminescence gene activation by the <i>Vibrio fischeri</i> LuxR protein" JOURNAL OF BACTERIOLOGY, vol. 174, no. 12, June 1992 (1992-06), pages 4064-4069, XP001002993 page 4066; figure 2<br>---                                                                    | 6                     |
| A        | FUQUA W.C. AND WINANS S.C.: "A LuxR-LuxI type regulatory system activates <i>Agrobacterium</i> Ti plasmid conjugal transfer in the presence of a plant tumor metabolite" JOURNAL OF BACTERIOLOGY, vol. 176, no. 10, May 1994 (1994-05), pages 2796-2806, XP001010680 cited in the application page 2798, left-hand column<br>---<br>-/-- | 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. |
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| A          | <p>EBERHARD A. ET AL.: "Analog of the autoinducer of bioluminescence in <i>Vibrio fischeri</i>"</p> <p>ARCHIVES OF MICROBIOLOGY, vol. 146, no. 1, 1986, pages 35-40, XP000884126</p> <p>ISSN: 0302-8933</p> <p>page 37; table 1</p> <p style="text-align: center;">---</p>                                                                                                                                | 30                    |
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**FURTHER INFORMATION CONTINUED FROM PCT/SA/ 210**

Continuation of Box I.1

Although claims 27, 28 and 30-35 (insofar as in vivo methods of treatment of the human/animal body by therapy are concerned, see pages 27-32) are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/IB 00/01828

| Patent document<br>cited in search report | Publication<br>date | Patent family<br>member(s)   | Publication<br>date      |
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