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(54) Title: SINGLE PROMOTER SYSTEM FOR MAKING siRNA EXPRESSION CASSETTES AND EXPRESSION LIBRARIES USING A POLYMERASE PRIMER HAIRPIN LINKER

(57) Abstract: The present invention relates to methods and compositions for the elucidation of gene function and the identification of novel genes. Specifically, the present invention relates to methods and compositions for improved functional genomic screening, functional inactivation of specific essential or non-essential genes, and identification of genes that are modulated in response to specific stimuli or encode recognizable phenotypic traits. In particular, the compositions of the present invention include, but are not limited to, expression cassettes comprising a novel polymerase primer hairpin linker that allows rapid construction of a single transcriptional unit encoding both strands of a hairpin siRNA, regardless of sequence. In addition, the present invention includes libraries comprising the expression cassettes of the invention, including vectors for transforming cells, such as replication-deficient retroviral vectors. The invention also includes methods for the production and screening of siRNA libraries, as well as therapeutic uses for the siRNAs expressed in accordance with the invention.

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US03/23239

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : C12N 15/63+
US CL : 435/320.1

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. : 435/320.1; 536/23.1, 24.1, 24.2, 24.5

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.
T	US 2004/0152172 A1 (GEPPERT) 5 August 2004 (05.08.2004) (see entire document).	1-22
Y	MIYAGISHI et al. U6 promoter-driven siRNAs with four uridine 3' overhangs efficiently suppress targeted gene expression in mammalian cells. Nature Biotechnology May 2002, Vol. 19. pp. 497-500 (see entire document, especially page 499).	1-22
Y	BRUMMELKAMP et al. A system for stable expression of short interfering RNAs in mammalian cells. Science April 2002, Vol. 296. pp. 550-553 (see entire document, especially figures 1, 2 and page 552)	1-22
E, Y	US 2004/0005593 A1 (LORENS) 8 January 2004 (08.01.2004) (see entire document, especially figures 1-3 and claims).	1-22
Y	BEGER et al. Identification of Id4 as a regulator of BRCA1 expressoin by using a ribozyme library based inverse genomics approach Proc. Natl. Acad. Sci. January 2001, Vol. 98, pp. 130-135 (see all)	1-22



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
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"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	"&"	document member of the same patent family

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INTERNATIONAL SEARCH REPORT

C. (Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	KAWASAKI et al. Identification of genes by hybrid ribozymes that couple cleavage activity with the unwinding activity of an endogenous RNA helicase. EMBO Reports May 2002, Vol. 3, pp. 443-450 (see entire document)	1-22
E, Y	US 2003/0144239 A1 (AGAMI et al.) 31 July 2003 (31.07.2003) (see entire document, in particular figure 11 and pages 5-11).	1-22
E, Y	US 2004/0002077 A1 (TAIRA et al.) 1 January 2004 (01.01.2004) (see entire document, in particular figures 12, 14--25 and pages 5-11).	1-22

INTERNATIONAL SEARCH REPORT

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Continuation of B. FIELDS SEARCHED Item 3:

EAST, STN (medline, biosis, embase, caplus)

search terms: siRNA, ribozyme, hairpin, expression cassette, random/randomized, sequence, library