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C07H 21/04 (2006.01)

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(81) Designated States (unless otherwise indicated, for every kind of national protection available): AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LT, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

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[Continued on next page]

(54) Title: ORTHOGONAL CAS9 PROTEINS FOR RNA-GUIDED GENE REGULATION AND EDITING

(57) Abstract: Methods of modulating expression of a target nucleic acid in a cell are provided including use of multiple orthogonal Cas9 proteins to simultaneously and independently regulate corresponding genes or simultaneously and independently edit corresponding genes.

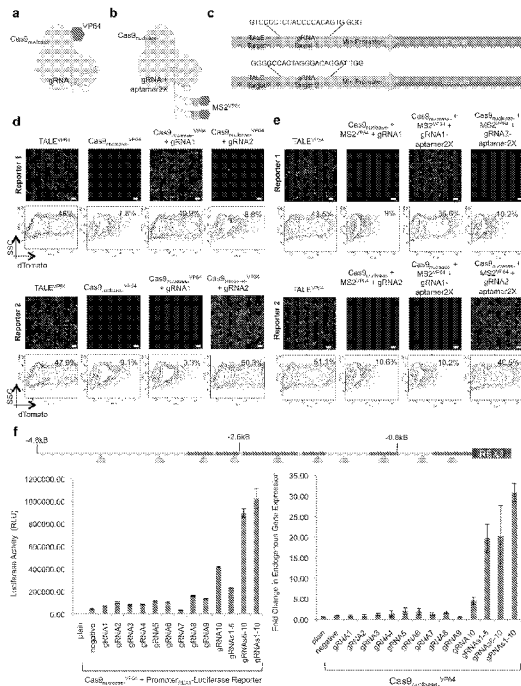


Figure 1

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## INTERNATIONAL SEARCH REPORT

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International application No.

PCT/US14/45700

A. CLASSIFICATION OF SUBJECT MATTER  
 IPC(8) - A61K 48/00; C07H 21/04; C12P 19/34 (2014.01)  
 CPC - A61K 48/00; C07H 21/04

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(8) Classification(s): C07H 21/04; A61K 48/00; C12P 19/34 (2014.01); CPC Classification(s): C12N 15/10, 15/113; C12Q 1/686, 1/6869; A61K 48/00, 38/00; C07H 21/04 USPC Classification(s): 435/91.1, 89, 85, 72, 41; 514/44R, 43, 42, 23, 1; 536/23.1, 22.1, 18.7

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)

MicroPatent (US-G, US-A, EP-A, EP-B, WO, JP-bib, DE-C,B, DE-A, DE-T, DE-U, GB-A, FR-A); Pubmed; Google Scholar; ScienceDirect; Cas9, double strand break, nickase, nuclease null, inactive Cas9, VP64, CRISPR

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	MALI, P et al. RNA-Guided Human Genome Engineering Via Cas9. Science. February 15, 2013; Vol. 339, No. 6121, pp. 823-826; abstract; page 823, middle column, second paragraph to right column, second paragraph; Figures 1, 2. doi: 10.1126/science.123203	1-40
A	CONG, L et al. Multiplex Genome Engineering Using CRISPR/Cas Systems. Science. February 15, 2013; Vol. 339, No. 6121, pp. 819-823; page 820, center column, second paragraph to right column, first paragraph; Figure 2. doi: 10.1126/science.123114	1-40
A	QI, L et al. Repurposing CRISPR as an RNA-guided platform for sequence-specific control of gene expression. Cell. February 28, 2013; Vol. 152, No. 5, pp. 1173-1183; page 1175, right column, fourth paragraph to page 1177, left column, first paragraph; Figures 2, 4. doi: 10.1016/j.cell.2013.02.022	1-40
A	ZHANG, F et al. Nature Biotechnology. February 2011. Vol. 29, No. 2, pp. 149-153; page 150, figure 1C and 1D. doi: 10.1038/nbt.177	1-40

Further documents are listed in the continuation of Box C.

\* 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 application or patent but published on or after the international filing date

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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

"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

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**Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)**

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing filed or furnished:

a. (means)



on paper



in electronic form

b. (time)



in the international application as filed



together with the international application in electronic form



subsequently to this Authority for the purposes of search

2.  In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments: