

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property
Organization

International Bureau

(43) International Publication Date
16 December 2021 (16.12.2021)



(10) International Publication Number
WO 2021/251905 A1

(51) International Patent Classification:
C12Q 1/68 (2018.01)

(21) International Application Number:
PCT/SG2021/050338

(22) International Filing Date:
11 June 2021 (11.06.2021)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
10202005599R 12 June 2020 (12.06.2020) SG

(71) Applicant: AGENCY FOR SCIENCE, TECHNOLOGY AND RESEARCH [SG/SG]; 1 Fusionopolis Way, #20-10 Connexis North Tower, Singapore 138632 (SG).

(72) Inventors: CHEW, Wei Leong; c/o Genome Institute of Singapore, 60 Biopolis St, Genome #02-01, Singapore 138672 (SG). KENG, Choong Tat; c/o Genome Institute of Singapore, 60 Biopolis St, Genome #02-01, Singapore 138672 (SG). GUO, Ke; c/o Genome Institute of Singapore, 60 Biopolis St, Genome #02-01, Singapore 138672 (SG). LIM, Daryl; c/o Genome Institute of Singapore, 60 Biopolis St, Genome #02-01, Singapore 138672 (SG).

(74) Agent: SPRUSON & FERGUSON (ASIA) PTE LTD; P.O. Box 1531, Robinson Road Post Office, Singapore 903031 (SG).

(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, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, IT, JO, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, 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, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, WS, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every kind of regional protection available): ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK,

EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

— of inventorship (Rule 4.17(iv))

Published:

— with international search report (Art. 21(3))
— with sequence listing part of description (Rule 5.2(a))
— in black and white; the international application as filed contained color or greyscale and is available for download from PATENTSCOPE



WO 2021/251905 A1

(54) Title: A METHOD FOR ASSESSING TRANSDUCTION EFFICIENCY AND/OR SPECIFICITY OF VECTORS AT SINGLE CELL LEVEL

(57) Abstract: Disclosed is a method for assessing the transduction efficiency and/or specificity of vectors at single cell level, comprising: providing a plurality of different vectors; transducing a heterogeneous population of cells with the plurality of different vectors; partitioning the heterogeneous population of cells into a plurality of compartments, wherein each compartment comprises a single cell from the heterogeneous population of cells; subjecting each partitioned cell to nucleotide sequencing; and detecting the presence of the any one or more of the different vectors in each partitioned cell. In particular, each of the plurality of different vectors comprises an oligonucleotide barcode sequence or a marker polynucleotide, wherein the barcode sequence or the marker polynucleotide is different between any two different vectors. In one specific embodiment, the vectors are adeno-associated virus (AAV) vectors.