



- (51) International Patent Classification:
C12Q 1/6827 (2018.01) C12Q 1/6855 (2018.01)
- (21) International Application Number:
PCT/EP2024/064792
- (22) International Filing Date:
29 May 2024 (29.05.2024)
- (25) Filing Language: English
- (26) Publication Language: English
- (30) Priority Data:
2308068.2 30 May 2023 (30.05.2023) GB
- (71) Applicant: **ALTOS LABS, INC.** [US/US]; 1300 Island Drive, Redwood City, California 94095 (US).
- (71) Applicant (for MN only): **SUTCLIFFE, Nicholas** [GB/G-B]; Aurora Building, Counterslip, Bristol BS1 6BX (GB).

- (72) Inventors: **REIK, Wolf Ulrich**; c/o Altos Labs, The Portway Building, Granta Park, Cambridge Cambridgeshire CB21 6GP (GB). **ABAKIR, Abdulkadir Hassan**; c/o Altos Labs, The Portway Building, Granta Park, Cambridge Cambridgeshire CB21 6GP (GB). **CLARK, Stephen James**; c/o Altos Labs, The Portway Building, Granta Park, Cambridge Cambridgeshire CB21 6GP (GB).
- (74) Agent: **MEWBURN ELLIS LLP**; Aurora Building, Counterslip, Bristol BS1 6BX (GB).
- (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, CV, CZ, DE, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IQ, IR, IS, IT, JM, JO, JP, KE, KG, KH, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, MG, MK, MN, MU, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO,

(54) Title: METHODS FOR DETECTION OF SINGLE STRAND BREAKS IN NUCLEIC ACID

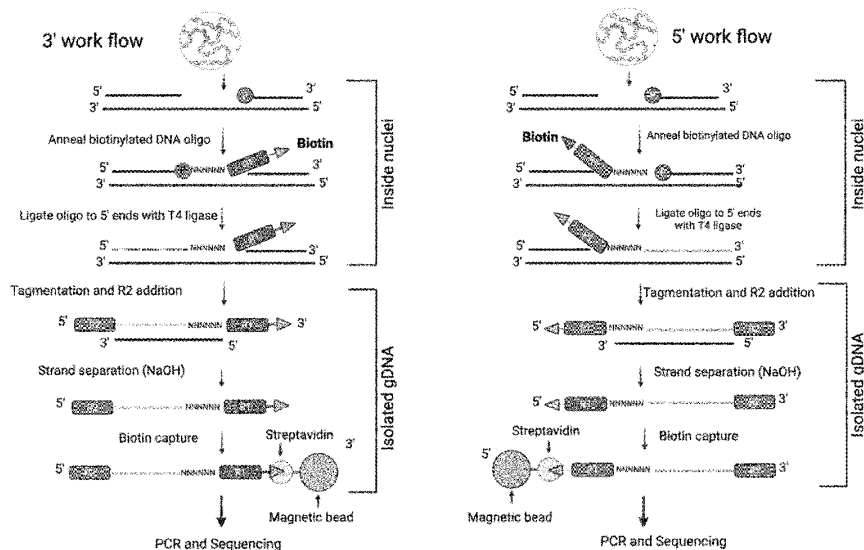


FIGURE 1

(57) Abstract: This invention relates to methods for detecting sites of single strand breaks (SSBs) in double stranded (ds) DNA. A sample of dsDNA is contacted with an oligonucleotide probe comprising a first sequencing adaptor and a tag and then contacted with DNA ligase. The DNA ligase covalently links the oligonucleotide probe to a strand of the dsDNA at the site of a single strand break in the dsDNA. The sample of dsDNA is then contacted with a transposase loaded with a second sequencing adaptor, such that the transposase cleaves the dsDNA to generate a population of DNA fragments comprising the second sequencing adaptor. DNA strands that comprise the first and second sequencing adaptors are isolated from said population and the sequences of the isolated DNA strands determined. Methods and kits for performing the methods are provided.

[Continued on next page]

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, CV, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SC, 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, ME, 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))*
— *in black and white; the international application as filed contained color or greyscale and is available for download from PATENTSCOPE*