

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

(19) World Intellectual Property
Organization
International Bureau

(43) International Publication Date
08 December 2022 (08.12.2022)



(10) International Publication Number
WO 2022/256324 A1

- (51) **International Patent Classification:**
C12Q 1/6841 (2018.01) *C12Q 1/6816* (2018.01)
- (21) **International Application Number:**
PCT/US2022/031601
- (22) **International Filing Date:**
31 May 2022 (31.05.2022)
- (25) **Filing Language:** English
- (26) **Publication Language:** English
- (30) **Priority Data:**
63/195,613 01 June 2021 (01.06.2021) US
- (71) **Applicant:** 10X GENOMICS, INC. [US/US]; 6230 Stoneridge Mall Road, Pleasanton, California 94588-3260 (US).
- (72) **Inventors:** KÜHNEMUND, Malte; c/o 10x Genomics, Inc., 6230 Stoneridge Mall Road, Pleasanton, California 94588-3260 (US). QIAN, Xiaoyan; c/o 10x Genomics, Inc., 6230 Stoneridge Mall Road, Pleasanton, California 94588-3260 (US).
- (74) **Agent:** ROBERTS KINGMAN, Kelsey et al.; Morrison & Foerster LLP, 755 Page Mill Road, Palo Alto, California 94304 (US).
- (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, IQ, IR, IS, IT, JM, 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.

(54) **Title:** METHODS AND COMPOSITIONS FOR ANALYTE DETECTION AND PROBE RESOLUTION

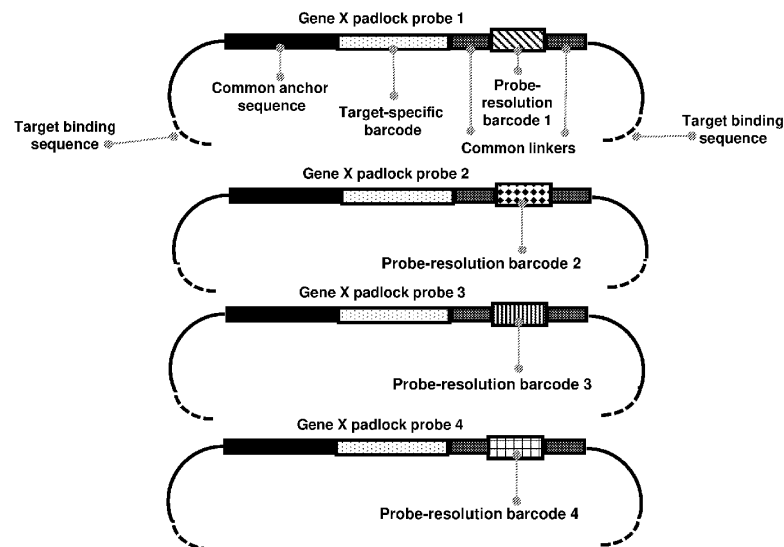


FIG. 1A

(57) **Abstract:** The present disclosure in some aspects relates to methods and compositions for accurately detecting and quantifying analytes present at high levels, such as highly expressed genes in a sample. In some embodiments, a probe-resolution barcode sequence disclosed herein does not specifically correspond to any particular target analyte(s) but can be used to resolve dense optical signals due to spatially overlapping signals associated with different molecules of a target analyte, thereby enabling resolution of signals in a dense "spot" and accurate counting of spots associated with molecules that are in spatial proximity. Also provided are kits comprising probes for use in such methods.

[Continued on next page]

(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).

Published:

— with international search report (Art. 21(3))