

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

(19) World Intellectual Property
Organization
International Bureau

(43) International Publication Date
02 February 2023 (02.02.2023)



(10) International Publication Number
WO 2023/009842 A1

- (51) **International Patent Classification:**
C12Q 1/6853 (2018.01) *C12Q 1/6841* (2018.01)
- (21) **International Application Number:**
PCT/US2022/038915
- (22) **International Filing Date:**
29 July 2022 (29.07.2022)
- (25) **Filing Language:** English
- (26) **Publication Language:** English
- (30) **Priority Data:**
63/227,830 30 July 2021 (30.07.2021) US
- (71) **Applicant:** 10X GENOMICS, INC. [US/US]; 6230 Stoneridge Mall Road, Pleasanton, California 94588-3260 (US).
- (72) **Inventors:** HERNÁNDEZ NEUTA, Jorge Iván; c/o 10x Genomics, Inc., 6230 Stoneridge Mall Road, Pleasanton, California 94588-3260 (US). KÜHNEMUND, Malte; c/o

10x Genomics, Inc., 6230 Stoneridge Mall Road, Pleasanton, California 94588-3260 (US). MARKS, Patrick J.; 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, 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, 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,

(54) **Title:** METHODS AND COMPOSITIONS FOR SYNCHRONIZING REACTIONS *IN SITU*

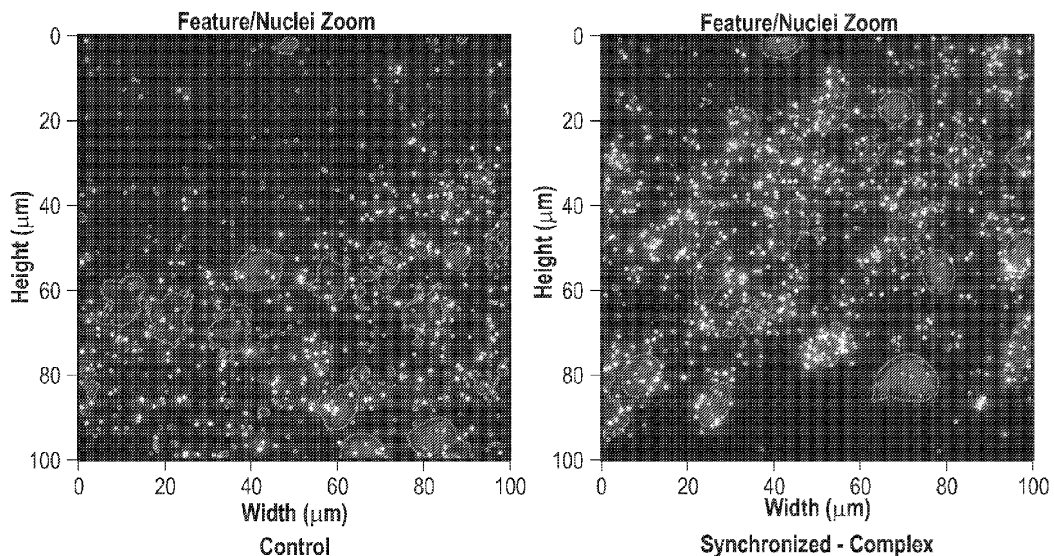


FIG. 4B

(57) **Abstract:** The present disclosure in some aspects relates to methods and compositions for accurately detecting and quantifying multiple analytes present in a biological sample. In some aspects, the methods and compositions provided herein address issues associated with the heterogeneity of analyte abundance (e.g., gene expression levels) and variations among reactions at different locations of a sample (e.g., amplification reaction starting earlier at one location than another location). In some aspects, a method disclosed herein provides a tighter distribution of signal spot size and intensity in a sample, as compared to methods that result in a wide and heterogeneous size and intensity distribution of signal spots.

[Continued on next page]

WO 2023/009842 A1

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).

Published:

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