



- (51) **International Patent Classification:**
C12Q 1/68 (2006.01) *G01N 33/52* (2006.01)
- (21) **International Application Number:**
PCT/US2014/022163
- (22) **International Filing Date:**
7 March 2014 (07.03.2014)
- (25) **Filing Language:** English
- (26) **Publication Language:** English
- (30) **Priority Data:**
61/775,415 8 March 2013 (08.03.2013) US
- (71) **Applicant:** **BIO-RAD LABORATORIES, INC.**
[US/US]; 1000 Alfred Nobel Drive, Hercules, California
94547 (US).
- (72) **Inventors:** **LOWE, Adam**; 4862 Bernal Avenue, Apartment C, Pleasanton, California 94566 (US). **MCDERMOTT, Geoff**; 4862 Bernal Avenue, Apartment B, Pleasanton, California 94566 (US). **HINDSON, Chris**; 4862 Bernal Avenue, Apartment B, Pleasanton, California 94566 (US). **CHIA, Erin**; 1988 MLK Jr. Way, Apartment 317, Berkeley, California 94704 (US). **HIDDESEN, Amy**; 5050 Hacienda Drive, Dublin, California 94568 (US). **HINDSON, Ben**; 1789 Terra Court, Pleasanton, California 94566 (US). **HAN, Chunxiao**; 4750 Tassajara Road, Apartment 5310, Dublin, California 94568 (US). **WANG, Yaqi**; 5605 Maymont Lane, Dublin, California 94568 (US).
- (74) **Agents:** **ALEMOZAFAR, Ali, R.** et al.; Wilson Sonsini Goodrich & Rosati, 650 Page Mill Road, Palo Alto, California 94304-1050 (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, 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.
- (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, 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))*
 - *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments (Rule 48.2(h))*

(54) **Title:** COMPOSITIONS, METHODS AND SYSTEMS FOR POLYMERASE CHAIN REACTION ASSAYS

(57) **Abstract:** The present disclosure provides methods, devices, systems and compositions for detecting nucleic acids in polymerase chain reaction assays, such as droplet digital polymerase chain reaction (ddPCR) assays. The present disclosure provides methods, devices, systems and compositions for detecting nucleic acids in ddPCR assays using intercalating dyes. A dual surfactant system with at least one fluorosurfactant and at least one non-ionic non-fluorosurfactant may be employed for droplet generation and nucleic acid detection.

