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

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
17 September 2020 (17.09.2020)

WIPO | PCT

(10) International Publication Number
WO 2020/186059 A2

(51) International Patent Classification:
A61K 38/00 (2006.01) *C12N 15/63* (2006.01)
C12N 9/22 (2006.01) *C12N 15/90* (2006.01)
C12N 15/10 (2006.01)

(21) International Application Number:
PCT/US2020/022394

(22) International Filing Date:
12 March 2020 (12.03.2020)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
62/817,514 12 March 2019 (12.03.2019) US

(71) Applicants: **CRISPR THERAPEUTICS AG** [CH/CH]; Baarerstrasse 14, 6300 Zug (CH). **BAYER HEALTHCARE, LLC** [US/US]; 100 Bayer Boulevard, Whippany, NJ 07981 (US).

(72) Inventors: **SCHMIDT, Moritz**; c/o Bayer AG, Division: Global Biologics Research, Nattermannallee, Building S19, 50829 Cologne (DE). **KNYPHAUSEN, Philipp**; c/o Bayer AG, Division: Global Biologics Research, Nattermannallee, Building S19, 50829 Cologne (DE). **GALONSKA, Christina**; c/o Bayer AG, Division: Global Biologics Research, Nattermannallee, Building S19, 50829 Cologne (DE). **COCO, Wayne M.**; c/o Bayer AG, Division: Global Biologics Research, Nattermannallee, Building S19, 50829 Cologne (DE). **COHEN, Andre**; c/o Bayer AG, Division: Global Biologics Research, Nattermannallee, Building S19, 50829 Cologne (DE).

(74) Agent: **MANDRAGOURAS, Amy E.** et al.; Nelson Mullins Riley & Scarborough LLP, One Post Office Square, 30th Floor, Boston, MA 02109-2127 (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, IR, IS, 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).

Published:
— *without international search report and to be republished upon receipt of that report (Rule 48.2(g))*

WO 2020/186059 A2

(54) Title: NOVEL HIGH FIDELITY RNA-PROGRAMMABLE ENDONUCLEASE SYSTEMS AND USES THEREOF

(57) Abstract: Described herein are novel systems for targeting, editing or manipulating DNA in a cell, using novel M- SmallCas9 nucleases and variants thereof. The M-SmallCas9 nucleases are derived from wildtype or parental small type II CRISPR Cas9 endonucleases, and display improved fidelity compared to parental type II CRISPR Cas9 enzymes in combination with a simple PAM sequences and are small endonuclease size.