



(51) International Patent Classification:

C12N 15/63 (2006.01) *C12N 15/113* (2010.01)
A61K 38/47 (2006.01) *C12N 15/86* (2006.01)
C12N 9/34 (2006.01) *C12N 15/90* (2006.01)

(21) International Application Number:

PCT/US2024/039837

(22) International Filing Date:

26 July 2024 (26.07.2024)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

63/516,395 28 July 2023 (28.07.2023) US

(71) Applicant: **REGENERON PHARMACEUTICALS, INC.** [US/US]; 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US).

(72) Inventors: **BAIK, Andrew**; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). **LIN, Allen**; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). **PRAGGASTIS, Maria**; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). **CYGNAR, Katherine**; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). **PEFANIS, Evangelos**; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US). **SABIN, Leah**; c/o Regeneron Pharmaceuticals, Inc., 777 Old Saw Mill River Road, Tarrytown, New York 10591 (US).

(74) Agent: **SCHILLING, Stephen H.** et al.; Alston & Bird LLP, Vantage South End, 1120 South Tryon Street, Suite 300, Charlotte, North Carolina 28203-6818 (US).

(54) Title: ANTI-TFR:GAA AND ANTI-CD63:GAA INSERTION FOR TREATMENT OF POMPE DISEASE

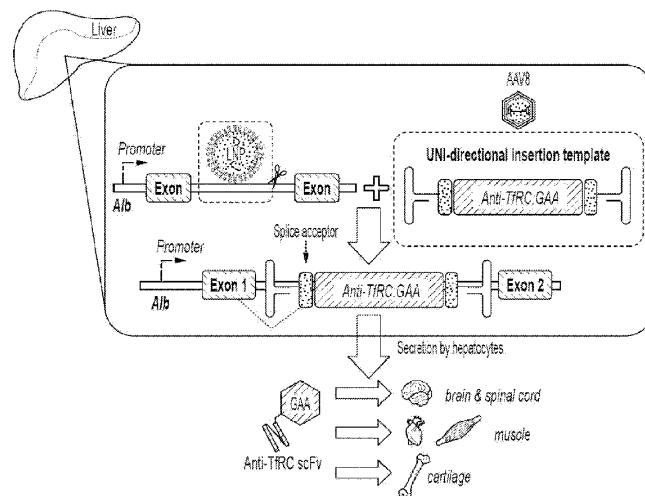


FIG. 36

(57) Abstract: Nucleic acid constructs and compositions that allow insertion of a multidomain therapeutic protein (e.g., GAA fusion protein) coding sequence into a target genomic locus such as an endogenous *ALB* locus and/or expression of the multidomain therapeutic protein (e.g., GAA fusion protein) coding sequence are also provided. The nucleic acid constructs and compositions can be used in methods of integration of a multidomain therapeutic protein (e.g., GAA fusion protein) nucleic acid into a target genomic locus, methods of expression of a multidomain therapeutic protein (e.g., GAA fusion protein) in a cell, methods of reducing glycogen accumulation, methods of treating Pompe disease or GAA deficiency in a subject, and method of preventing or reducing the onset of a sign or symptom of Pompe disease in a subject, including neonatal cells and subjects.

[Continued on next page]

(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, 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:

- *as to the identity of the inventor (Rule 4.17(i))*
- *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*
- *as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii))*

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))*
- *with sequence listing part of description (Rule 5.2(a))*

(88) Date of publication of the international search report:

25 September 2025 (25.09.2025)