

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

(19) World Intellectual Property
Organization
International Bureau

(43) International Publication Date
30 June 2022 (30.06.2022)



(10) International Publication Number
WO 2022/140389 A1

(51) International Patent Classification:

C12M 1/34 (2006.01) *A61K 39/00* (2006.01)
C12P 21/02 (2006.01) *C07K 1/14* (2006.01)
A61K 38/17 (2006.01) *C07K 14/715* (2006.01)

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

(21) International Application Number:

PCT/US2021/064616

(22) International Filing Date:

21 December 2021 (21.12.2021)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

63/129,349 22 December 2020 (22.12.2020) US

(71) Applicant: **AMGEN INC.**, [US/US]; One Amgen Center Drive, Thousand Oaks, CA 91320-1799 (US).

(72) Inventors: **KRAUS, Eliyahu**; c/o Amgen Inc., One Amgen Center Drive, Mail Stop 28-5-A, Thousand Oaks, CA 91320-1799 (US). **NATARAJAN, Venkatesh**; c/o Amgen Inc., One Amgen Center Drive, Mail Stop 28-5-A, Thousand Oaks, CA 91320-1799 (US). **RODRIGUEZ, Ritsdeliz, Perez**; c/o Amgen Inc., One Amgen Center Drive, Mail Stop 28-5-A, Thousand Oaks, CA 91320-1799 (US). **NIEVES, Omar, Cruz**; c/o Amgen Inc., One Amgen Center Drive, Mail Stop 28-5-A, Thousand Oaks, CA 91320-1799 (US).

(74) Agent: **KISSLING, Heather, R.** et al.; Marshall Gerstein & Borun LLP, 233 S. Wacker Drive, 6300 Willis Tower, Chicago, IL 60606-6357 (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, IT, 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,

(54) Title: CELL CULTURE METHOD

(57) Abstract: The disclosure provides a method of producing etanercept from Chinese hamster ovary cells (CHO), the method comprising running an N-1 bioreactor system using a recirculating tangential flow filtration (RTF) or alternating tangential flow filtration (ATF) cell retention device under conditions that maintain a cell aggregate size of at least 20 μ m, before running an N production bioreactor.



WO 2022/140389 A1