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

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
5 May 2011 (05.05.2011)

PCT

(10) International Publication Number
WO 2011/053612 A1

(51) International Patent Classification:
C12N 1/19 (2006.01) *C12N 15/09* (2006.01)

(21) International Application Number:
PCT/US2010/054183

(22) International Filing Date:
27 October 2010 (27.10.2010)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
61/256,369 30 October 2009 (30.10.2009) US

(71) Applicant *(for all designated States except US)*: **MERCK SHARP & DOHME CORP.** [US/US]; 126 East Lincoln Avenue, Rahway, New Jersey 07065-0907 (US).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **HAMILTON, Stephen, R.** [GB/US]; 98 Mascoma Heights Drive, Enfield, New Hampshire 03748 (US). **STADHEIM, Terrance, A.** [US/US]; 22 Market Street, Lyme, New Hampshire 03768 (US).

(74) Common Representative: **MERCK SHARP & DOHME CORP.**; 126 East Lincoln Avenue, Rahway, New Jersey 07065-0907 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM,

[Continued on next page]

(54) Title: METHOD FOR PRODUCING THERAPEUTIC PROTEINS IN PICHIA PASTORIS LACKING DIPEPTIDYL AMINOPEPTIDASE ACTIVITY

(57) Abstract: The present invention related to methods and compositions for producing therapeutic proteins in yeast cell lines, and in particular Pichia pastoris, lacking dipeptidyl aminopeptidase (DAP) activity. DAP activity has been eliminated by genetically modifying a Pichia pastoris cell line such that STE13 and DAP2 have been deleted.

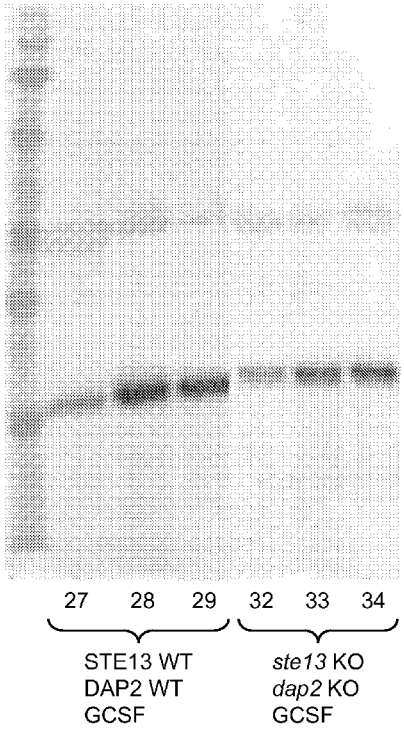


FIG.6

WO 2011/053612 A1

AO, AT, AU, AZ, BA, BB, BG, BH, 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, IS, JP, KE, KG, KM, 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, PE, PG, PH, PL, PT, RO, RS, RU, 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, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, 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, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

— *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))*

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