

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

(19) World Intellectual Property
Organization
International Bureau

(43) International Publication Date
12 November 2020 (12.11.2020)



(10) International Publication Number
WO 2020/227148 A1

(51) International Patent Classification:

C07K 14/415 (2006.01) C12N 15/81 (2006.01)
C12N 9/06 (2006.01) C12P 19/44 (2006.01)
C12N 15/53 (2006.01)

(21) International Application Number:

PCT/US2020/031171

(22) International Filing Date:

01 May 2020 (01.05.2020)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:

62/842,810 03 May 2019 (03.05.2019) US

(71) Applicant: AMYRIS, INC. [US/US]; 5885 Hollis Street, Suite 100, Emeryville, CA 94608 (US).

(72) Inventors: KLEIN, Andrew, P.; 5885 Hollis Street, Suite 100, Emeryville, CA 94608 (US). LUND, Sean; 5885 Hollis Street, Suite 100, Emeryville, CA 94608 (US). WICHMANN, Gale; 5885 Hollis Street, Suite 100, Emeryville, CA 94608 (US). KORYAKINA, Irina; 5885 Hollis Street, Suite 100, Emeryville, CA 94608 (US). HOGAN, Kyle; 5885 Hollis Street, Suite 100, Emeryville, CA 94608 (US). BORISOVA, Svetlana; 5885 Hollis Street, Suite 100, Emeryville, CA 94608 (US).

(74) Agent: ELBING, Karen, L.; Clark & Elbing LLP, 101 Federal Street, 15th Floor, Boston, MA 02110 (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:

- 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))

(54) Title: KAURENOIC ACID 13-HYDROXYLASE (KAH) VARIANTS AND USES THEREOF

FIG. 2



(57) Abstract: Provided herein are genetically modified host cells, compositions, and methods for improved production of steviol glycosides. The host cells are genetically modified to contain a heterologous nucleic acid that expresses novel and optimized variants of Ro.KAH. The host cell further contains one or more heterologous nucleotide sequence encoding further enzymes of a pathway capable of producing one or more steviol glycosides in the host cell. The host cells, compositions, and methods described herein provide an efficient route for the heterologous production of steviol glycosides including rebaudioside A, rebaudioside D, and rebaudioside M.



WO 2020/227148 A1