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

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
4 June 2015 (04.06.2015)

WIPO | PCT

(10) International Publication Number
WO 2015/079376 A1

(51) International Patent Classification:
A61K 47/48 (2006.01)

(21) International Application Number:
PCT/IB2014/066300

(22) International Filing Date:
24 November 2014 (24.11.2014)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
61/909,236 26 November 2013 (26.11.2013) US
62/002,614 23 May 2014 (23.05.2014) US

(71) Applicant *(for all designated States except US)*: **NOVARTIS AG** [CH/CH]; Lichtstrasse 35, CH-4056 Basel (CH).

(72) Inventors; and
(71) Applicants *(for US only)*: **HU, Qi-Ying** [US/US]; c/o Novartis Institutes for BioMedical Research, Inc., 250 Massachusetts Avenue, Cambridge, Massachusetts 02139 (US). **IMASE, Hidetomo** [JP/US]; c/o Novartis Institutes for BioMedical Research, Inc., 250 Massachusetts Avenue, Cambridge, Massachusetts 02139 (US).

(74) Common Representative: **NOVARTIS AG**; Lichtstrasse 35, CH-4056 Basel (CH).

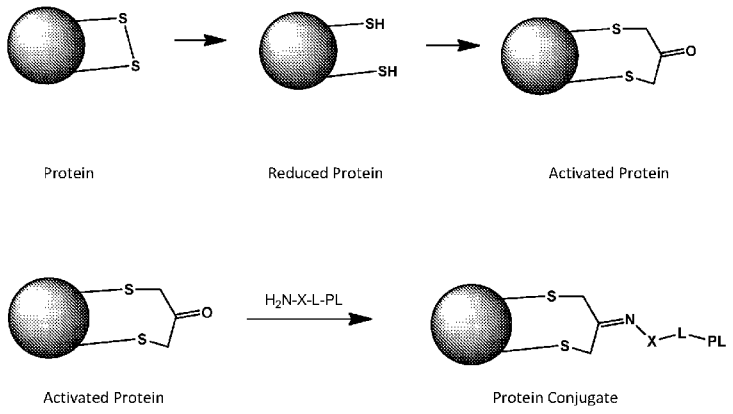
(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, 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, 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, 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))*

(54) **Title**: METHODS FOR OXIME CONJUGATION TO KETONE-MODIFIED POLYPEPTIDES

Figure 1



(57) **Abstract**: The invention provides improved methods to prepare protein conjugates from a ketone-modified protein. In one embodiment, the protein is prepared by linking two free cysteines by reaction with a 1,3-dihaloacetone or similar ketone-containing reactant, linking the sulfur atoms of the two cysteines together. The ketone inserted between the sulfur atoms is then used to form an oxime, thus conjugating the protein to a payload. In another embodiment, two cysteine residues are tied together by reaction with a 1,3-dihaloacetone or similar reactant, and the new ketone is used to form an oxime with a suitable payload molecule, thus conjugating the protein to a payload. The method provides improved reaction conditions for the oxime formation, whereby higher yields and Improved product homogeneity are achieved.

WO 2015/079376 A1