

(19) World Intellectual Property Organization
International Bureau(43) International Publication Date
19 November 2009 (19.11.2009)

PCT

(10) International Publication Number
WO 2009/140427 A3

(51) International Patent Classification:

C08F 299/00 (2006.01) C08F 293/00 (2006.01)
C08F 299/02 (2006.01) C12N 15/87 (2006.01)
C08F 299/04 (2006.01) A61K 48/00 (2006.01)

(21) International Application Number:

PCT/US2009/043847

(22) International Filing Date:

13 May 2009 (13.05.2009)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

61/052,908 13 May 2008 (13.05.2008) US
61/052,914 13 May 2008 (13.05.2008) US
61/091,294 22 August 2008 (22.08.2008) US
61/112,054 6 November 2008 (06.11.2008) US
61/112,048 6 November 2008 (06.11.2008) US
61/120,769 8 December 2008 (08.12.2008) US
61/140,779 24 December 2008 (24.12.2008) US
61/140,774 24 December 2008 (24.12.2008) US
61/171,377 21 April 2009 (21.04.2009) US

(71) Applicants (for all designated States except US): **UNIVERSITY OF WASHINGTON** [US/US]; 4311 - 11th Avenue Ne, Suite 500, Seattle, WA 98105-4608 (US). **PHASERX** [US/US]; 454 N 34th Street, Seattle, WA 98103 (US).

(72) Inventors; and

(75) Inventors/Applicants (for US only): **STAYTON, Patrick, S.** [US/US]; 3701 Nw 68th Street, Seattle, WA 98117 (US). **HOFFMAN, Alan, S.** [US/US]; 10616 Riviera Place N.e., Seattle, WA 98125 (US). **CONVERTINE, Anthony, J.** [US/US]; 522 North 85th Street, Apt. B113, Seattle, WA 98103 (US). **BENOIT, Danielle** [US/US]; 12348 38th Avenue Ne, Seattle, WA 98125 (US). **DUVALL, Craig, L.** [US/GB]; 4809 Evanston Avenue N., Seattle, WA 98103 (US). **JOHNSON, Paul** [US/US]; 12020 211th Place SE, Snohomish, WA 98296 (US). **GALL, Anna** [US/US]; 20051 170th Avenue NE, Woodinville, WA 98072 (US).

(74) Agent: **RENZONI, George, E.**; Christensen O'connor Johnson Kindness PLLC, 1420 Fifth Avenue, Suite 2800, Seattle, WA 98101 (US).

[Continued on next page]

(54) Title: DIBLOCK COPOLYMERS AND POLYNUCLEOTIDE COMPLEXES THEREOF FOR DELIVERY INTO CELLS

FIG. 1A

Polymer	Structure [D] _{MW1} -[B _x -P _y -D _z] _{MW2}	Mn Kda	Block Ratio MW ₂ /MW ₁
P7v1	[D] _{9.1K} -[B ₄₈ -P ₂₉ -D ₂₃] _{11.37K}	19	1.2
P7v2	[D] _{10K} -[B ₄₆ -P ₁₈ -D ₃₇] _{8.9K}	19	0.9
P7v3	[D] _{6.5K} -[B ₄₁ -P ₃₉ -D ₂₀] _{9.5K}	16	1.5
P7v6	[D] _{9.1K} -[B ₅₂ -P ₂₆ -D ₂₂] _{21.9K}	31	2.4

x, y, z ARE MOLE %. MOLECULAR WEIGHTS WERE DETERMINED BY GEL PERMEATION CHROMATOGRAPHY USING PMMA STANDARDS. COMPOSITIONS WERE DETERMINED BY NMR SPECTROSCOPY.

FIG. 1B

Polymer	Structure [D] _{MW1} -[B-P-D] _{inole% MW2}	Block Ratio MW ₂ /MW ₁	Particle Size (nm)
PRX-1	[D] _{11.3K} -[B ₅₀ -P ₃₀ -D ₂₀] _{20.7K}	1.83	41
PRX-2	[D] _{14.5K} -[B ₅₇ -P ₂₃ -D ₂₀] _{26.4K}	1.82	49
PRX-3	[D] _{11.5K} -[B ₃₅ -P ₂₇ -D ₃₈] _{33.4K}	2.92	60
PRX-4	[D] _{10.7K} -[B ₅₀ -P ₂₇ -D ₂₃] _{33.8K}	3.16	50
PRX-5	[D] _{10.7K} -[B ₄₀ -P ₃₁ -D ₂₉] _{32.2K}	3.00	59
PRX-6	[D] _{14.5K} -[B ₅₃ -P ₃₁ -D ₁₆] _{67.0K}	4.62	115

(57) Abstract: Described herein are copolymers, and methods of making and utilizing such copolymers. Such copolymers have at least two blocks: a first block that has at least one unit that is hydrophilic at physiologic pH, and a second block that has hydrophobic groups. This second block further has at least one unit with a group that is anionic at about physiologic pH. The described copolymers are disruptive of a cellular membrane, including an extracellular membrane, an intracellular membrane, a vesicle, an organelle, an endosome, a liposome, or a red blood cell. Preferably, in certain instances, the copolymer disrupts the membrane and enters the intracellular environment. In specific examples, the copolymer is endosomolytic.



(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, BR, BW, BY, BZ, CA, CH, 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, PG, PH, PL, PT, RO, RS, RU, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, 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, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, MD, RU, TJ,

TM), European (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, SE, SI, SK, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, 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))

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

4 March 2010