Supporting Information for

Identification of Jumonji AT-Rich Interactive Domain 1A Inhibitor and Their Effect on Cancer Cells

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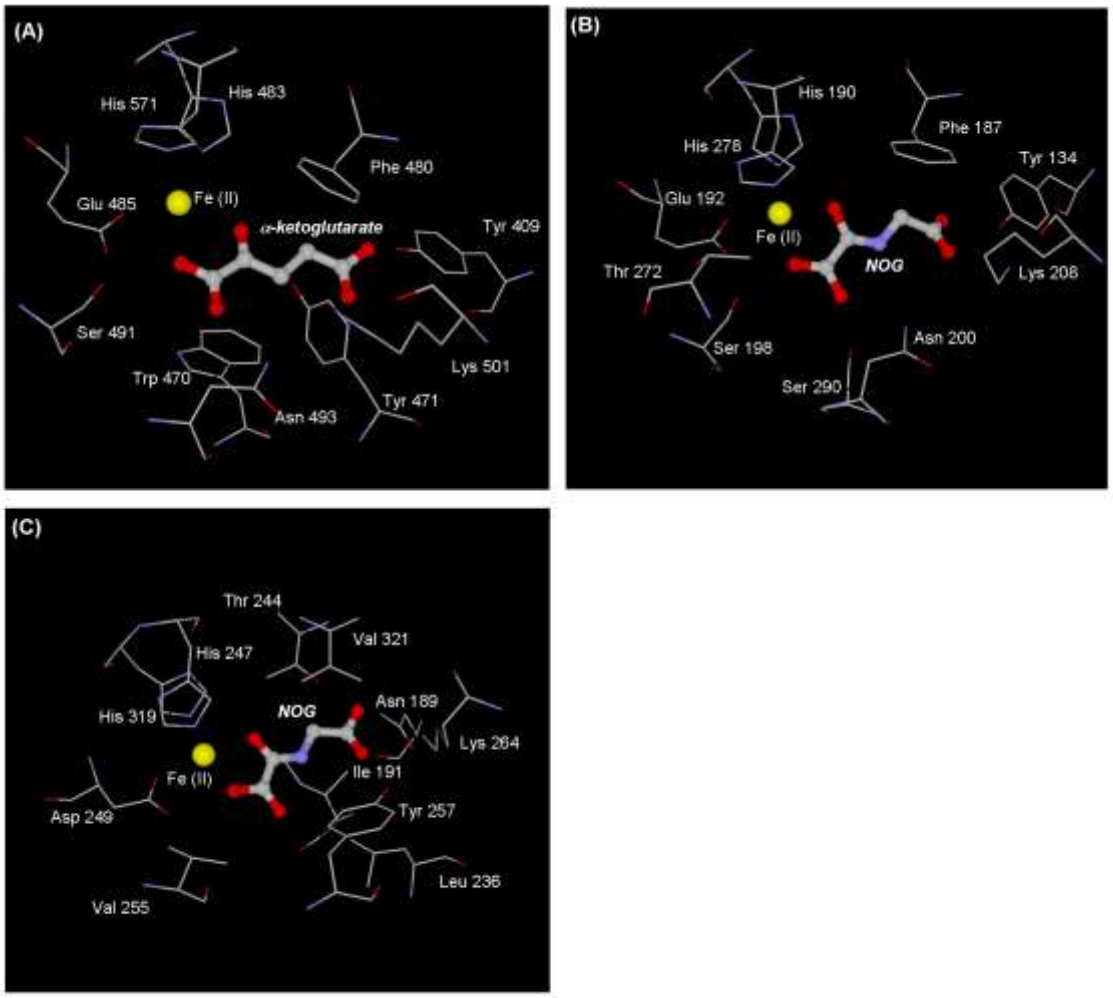


Figure S1. View of the catalytic site of (A) JARID1A (homology model), (B) JMJD2C (PDB code 2XML), and (C) JHDM1F (PDB code 3KV4) with α-ketoglutarate or NOG (**1**) (ball and stick). Amino acid residues are displayed in the wire graphic.

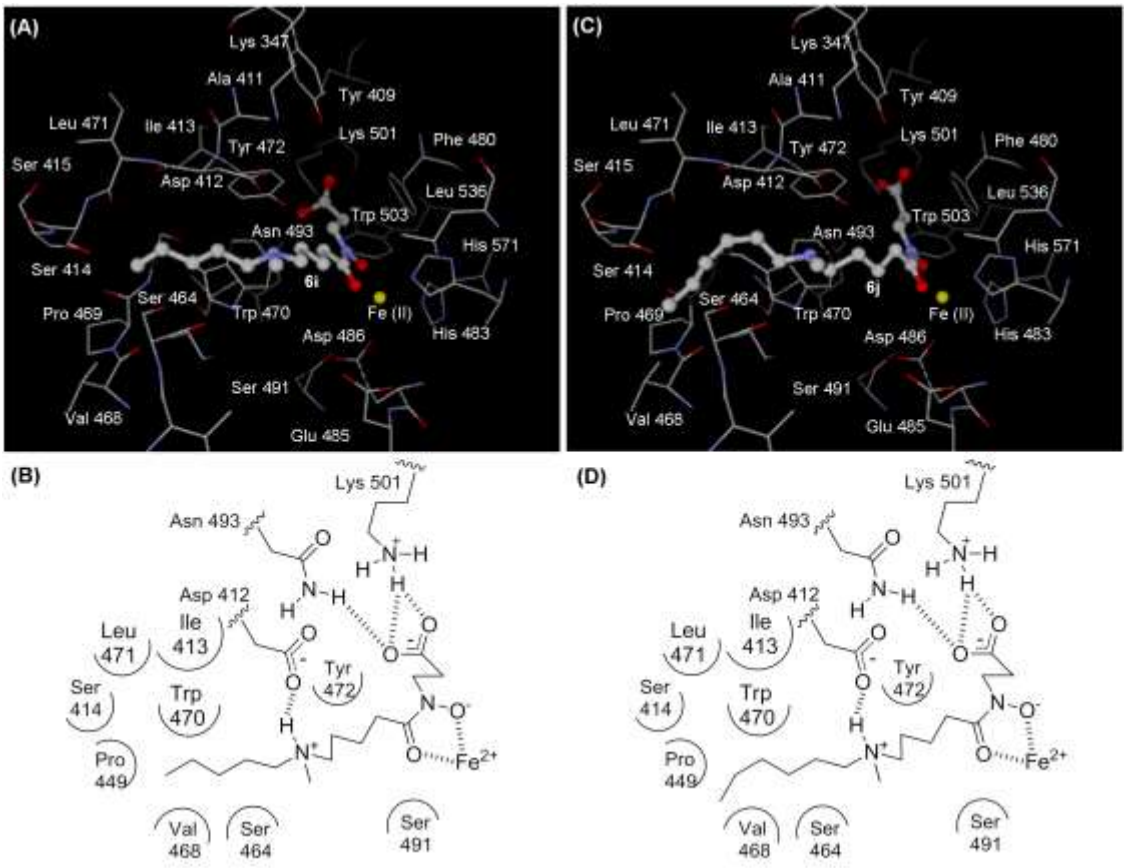


Figure S2. (A) View of the conformation of compound **6i** (ball and stick) docked into the JARID1A active site. (B) Schematic diagram of binding of compound **6i** to JARID1A. (C) View of the conformation of compound **6j** (ball and stick) docked into the JARID1A active site. (D) Schematic diagram of binding of compound **6j** to JARID1A.

The binding modes of compounds **6i** and **6j** were similar to that of compound **6a** (Fig. 2). The *n*-pentyl or *n*-hexyl group was positioned in the small pocket formed by Pro 449, Val 468, and Trp 470. These calculation results suggest that an increase of hydrophobic interaction between the alkyl group and hydrophobic amino acid residues contributes to the strong JARID1A inhibition by compounds **6i** and **6j**.

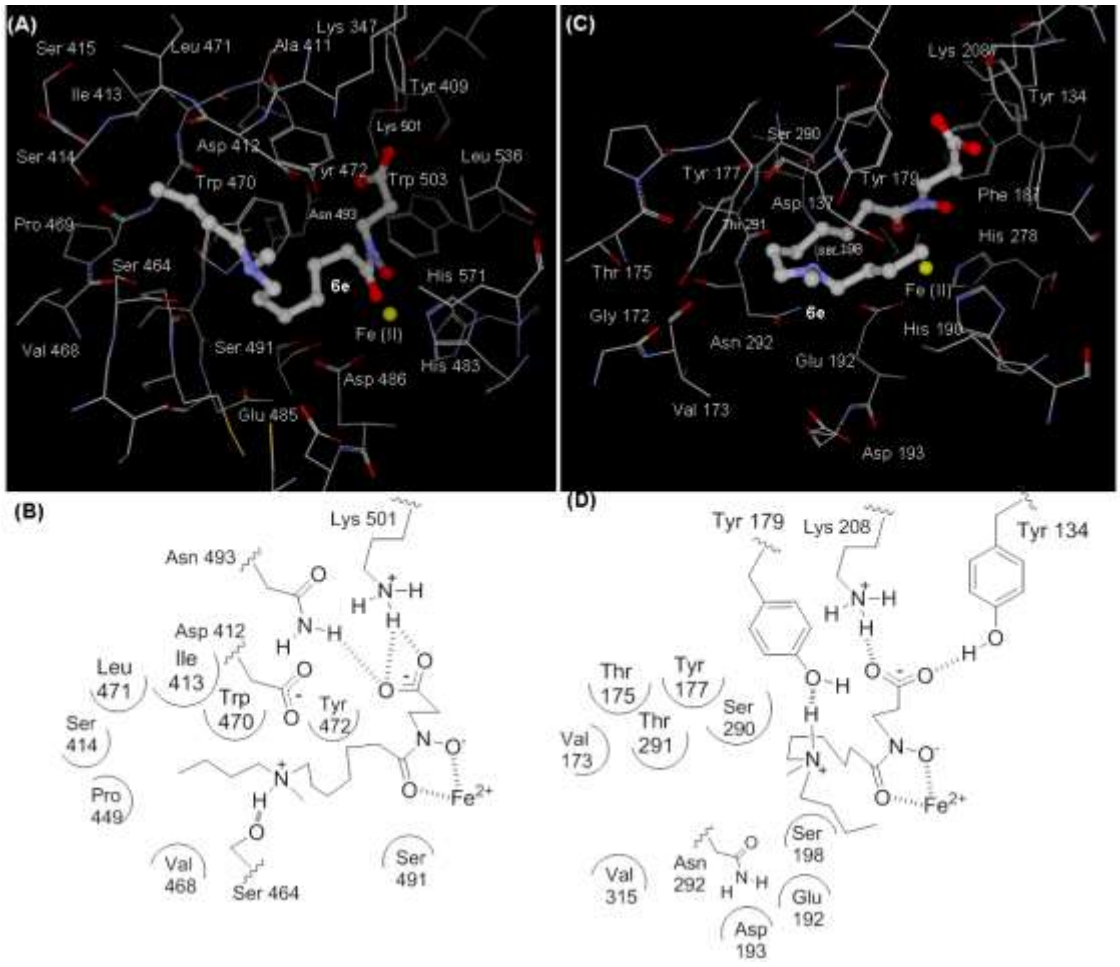


Figure S3. (A) View of the conformation of compound **6e** (ball and stick) docked into the JARID1A active site. (B) Schematic diagram of binding of compound **6e** to JARID1A. (C) View of the conformation of compound **6e** (ball and stick) docked into the JMJD2C active site. (D) Schematic diagram of binding of compound **6e** to JMJD2C.

As a result of the binding simulation of compound **6e** docked to JARID1A, the NH group of compound **6e** forms a hydrogen bond with Ser 464, which is different from the case of compound **6a**, **6i** and **6j**. In addition, the *n*-butyl group is positioned near a small hydrophobic pocket formed by Pro 449, Val 468, and Trp 470. In the case of the binding simulation of compound **6e** docked to JMJD2C, the NH group forms a hydrogen bond with Tyr 179. The methyl group is located at the entrance of active site. It indicates the replacement of the methyl group to a long alkyl group would be allowed in the active site of JMJD2C (See Figure S4).

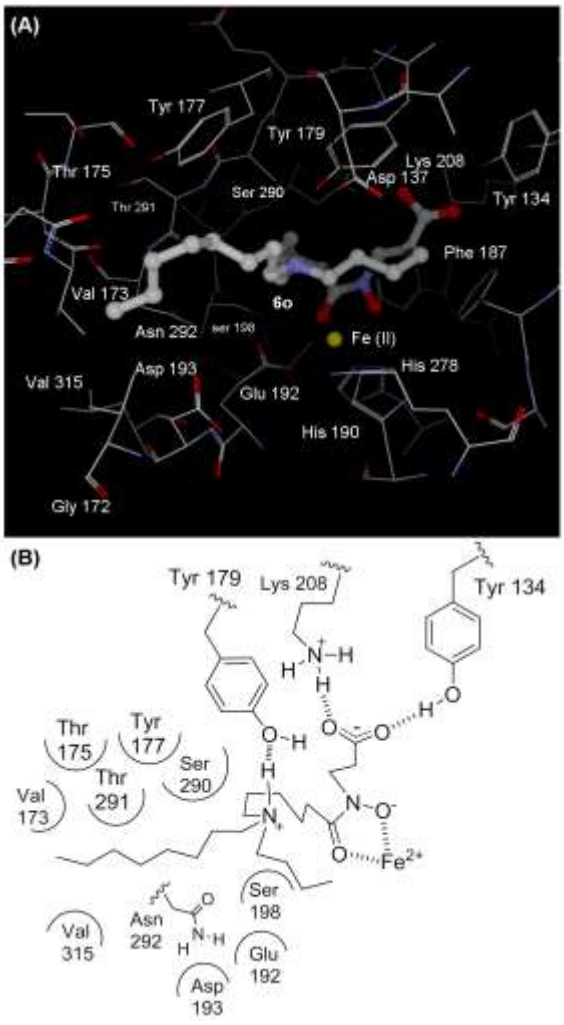


Figure S4. (A) View of the conformation of compound **6o** (ball and stick) docked into the JARID1A active site. (B) Schematic diagram of binding of compound **6o** to JARID1A. (C) Structure of compound **6o**.

The binding mode compound **6o**, designed by the replacement of the methyl group of compound **6e** to *n*-octyl group, was similar to that of compound **6e**. This calculation result suggests that the analogues which have two long alkyl groups might contribute to the strong JMJD2C inhibition. (See also Table S2).

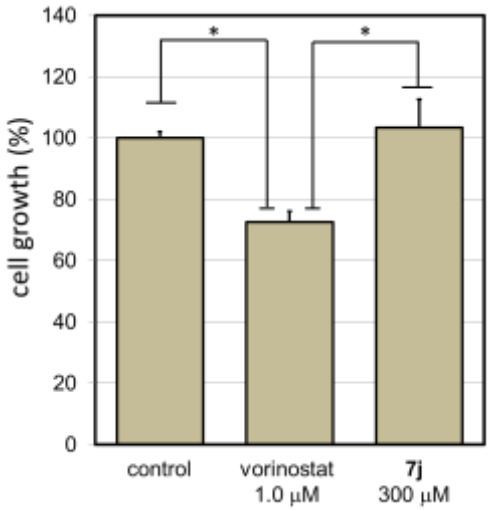


Figure S5. Cell growth inhibition of A549 cells after 72 h incubation with JARID1A inhibitor **7j**. Error bars represent the mean standard deviation (SD) of at least three samples. $^{*}P < 0.05$ (Tukey's test).

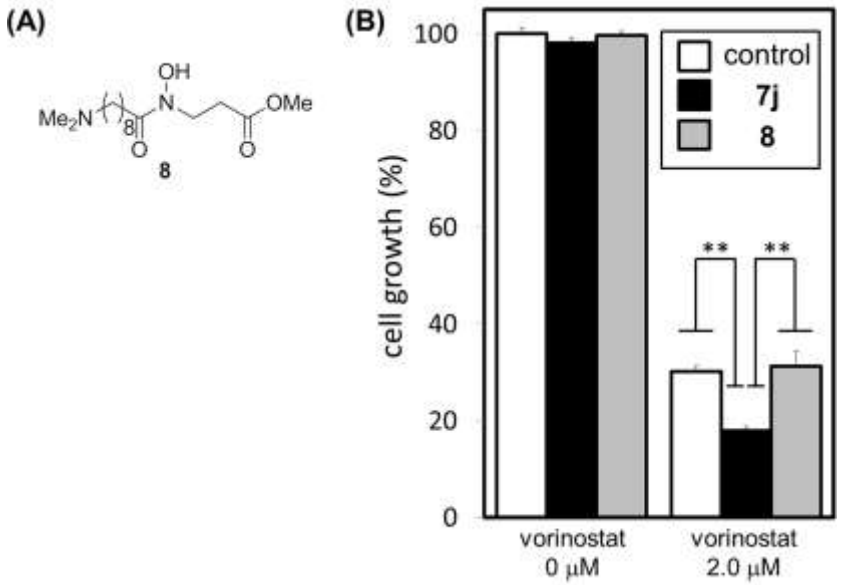


Figure S6. (A) Structure of NCDM-32b (**8**), a prodrug form of NCDM-32a (**3**). (B) Cell growth inhibition of A549 cells after 72 h incubation with combinations of JHDM inhibitors and vorinostat, an HDAC inhibitor. Error bars represent the mean standard deviation (SD) of at least three samples. Combination with 100 μM compound **7j** or NCDM-32b (**8**). $^{**}P < 0.01$ (Tukey's test).

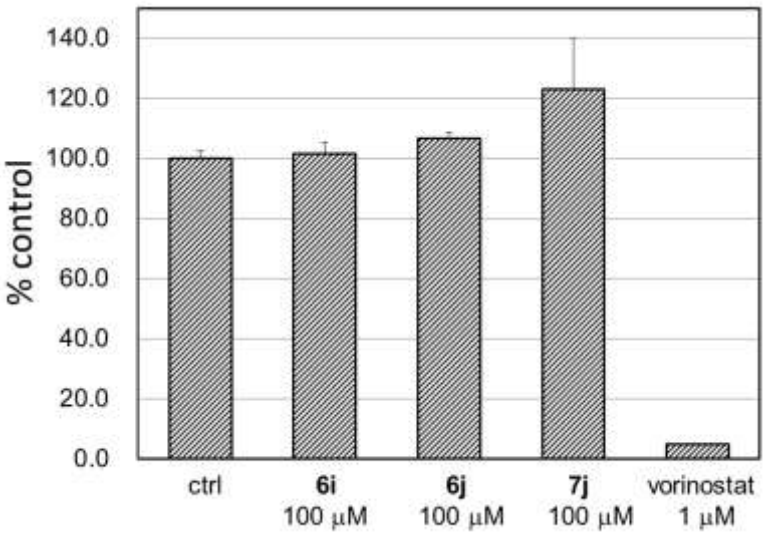
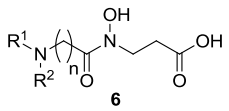


Figure S7. HDAC1-inhibitory activity of compounds **6i, 6j,** and **7j** . Error bars represent the mean standard deviation (SD) of at least three samples.

Table S1. The ratio of IC_{50} value for JARID1A-inhibitory activity of compounds **6** to that for JMJD1A-, JMJD2C-, and JHDM1F-inhibitory activity.



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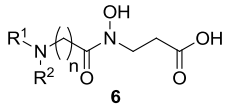
entry	compd	Structure			JMJD1A IC_{50} /JARID1A IC_{50}	JMJD2C IC_{50} /JARID1A IC_{50}	JHDM1F IC_{50} /JARID1A IC_{50}
		R^1	R^2	n			
1	**1**	-	-	-	2.3	1.7	2.6
2	**3**[b]	-	-		-	0.16	1.5
3	**4**[b]	-	-	-	-	1.5	0.022
4	**6a**	*n*-Bu	Me	4	> 23	13	> 23
5	**6b**	*n*-Bu	Me	2	-	> 1.9	-
6	**6c**	*n*-Bu	Me	3	-	10	-
7	**6d**	*n*-Bu	Me	5	-	15	15
8	**6e**	*n*-Bu	Me	6	-	3.6	-
9	**6f**	Me	Me	4	-	2.8	-
10	**6g**	Et	Me	4	-	7.9	-
11	**6h**	*n*-Pr	Me	4	-	8.5	-
12	**6i**	*n*-pentyl	Me	4	> 43	16	> 43
13	**6j**	*n*-hexyl	Me	4	> 30	13	22
14	**6k**	*n*-Bu	Et	4	-	4.6	-
15	**6l**	*n*-Bu	*n*-Bu	4	-	> 5.3	-
16	**6m**	*n*-Bu	*n*-hexyl	4	> 22	> 22	6.4
17	**6n**	*n*-Bu	*n*-octyl	4	> 32	27	7.7

Table S2. MolDock Score of compounds **6** docking into JARID1A and JMJD2C.[a]



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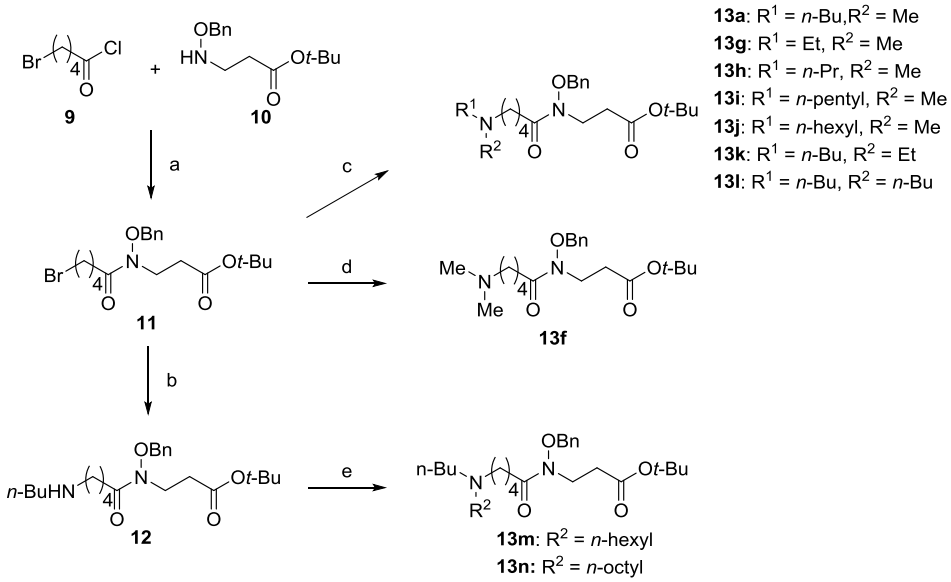
Entry	Compd	structure			JARID1A	JMJD2C
		R^1	R^2	n		
1	**6a**	*n*-Bu	Me	4	-143.70	-118.51
2	**6e**	*n*-Bu	Me	6	-151.91	-136.05
3	**6i**	*n*-pentyl	Me	4	-142.68	NT[b]
4	**6j**	*n*-hexyl	Me	4	-137.16	NT[b]
5	**6o**	*n*-Bu	*n*-octyl	6	ND[c]	-140.55

[a] MolDock Score is derived from the piecewise linear potential scoring functions (ref S1). [b] not tested. [c] ND = no data available. Compound **6o** did not coordinatethe catalytic iron ion in the active site of JARID1A.

Experimental Section

Chemistry. Proton nuclear magnetic resonance spectra (^{1}H NMR) and carbon nuclear magnetic resonance spectra (^{13}C NMR) were recorded on a BRUKER AVANCE 300 spectrometer in the indicated solvent. Chemical shifts (δ) were reported in parts per million relative to the internal standard, tetramethylsilane. High-resolution mass spectra (HRMS) and fast atom bombardment (FAB) mass spectra were recorded on a JEOL JMS-SX102A mass spectrometer. Electrospray ionization (ESI) mass spectra were recorded on a BRUKER HCTplus mass spectrometer. HPLC analysis and Preparative HPLC were performed on an ODS-3 (150 mm x φ4.6 mm, GL Science or Cosmosil) and an Inertsil ODS-3 (250 mm x φ20 mm, GL Science or Cosmosil), respectively. The HPLC system was composed of a pump (HITACHI, L-6050 intelligent pump) and a detector (HITACHI, L-4000 UV detector) Elution of HPLC chromatogram in synthetic procedure was done with a linear gradient (Solvent A: water (0.1%Trifluoroacetic acid (TFA)), Solvent B: MeCN (0.1%TFA); Gradient (I), 0 min (2% B)-2 min (2% B)-20 min (50% B)-30 min (50% B); Gradient (II), 0 min (0% B)-20 min (25% B)-30 min (25% B); Gradient (III), 0 min (0% B)-20 min (15% B)-30 min (15%B); Gradient (IV), 0 min (5% B)-20 min (50% B)-30 min (50% B); Gradient (V), 0 min (10% B)-20 min (40% B)-30 min (40% B); Gradient (VI), 0 min (5% B)-2 min (5% B)-20 min (40% B)-30 min (40%B); Gradient (VII), 0 min (20% B)-20 min (70% B)-30 min (70% B); Gradient (VIII), 0 min (5% B)-20 min (50% B)-30 min (50% B); flow rate = 1.0 mL/min). The detection wavelength was 220 nm. Reagents and solvents were purchased from Aldrich, Merck, Nacalai Tesque, Tokyo Kasei Kogyo, Wako Pure Chemical Industries, Kishida Kagaku, and Kanto Kagaku and were used without purification. Flash column chromatography was performed using silica gel supplied by TOYOTA SILICA GEL (#AP300D).

Scheme S1. Synthesis of Compounds 13a, 13f–n.[a]



[a]Reagents and conditions: (a) *N*,*N*-dimethylaminopyridine (DMAP), Et_3N, CH_2Cl_2, room temperature, 77%; (b) butan-1-amine, MeCN, room temperature, quant; (c) R^1R^2NH, MeCN, reflux, 47–99%; (d) dimethylamine hydrochloride, K_2CO_3, CH_2Cl_2, 35°C, 59%; (e) R^2Br, K_2CO_3, DMF, 60°C, 65–74% (2 steps from **11**).

Preparation of tert-Butyl 3-[N-(benzyloxy)-5-bromopentanamido]propanoate (**11**): A solution of 5-bromovaleryl chloride (1.38 mL, 10.4 mmol) in 5 mL of CH_2Cl_2. was added to a solution of 3-[(benzyloxy)amino]propanoate (**10**)[S1] (2.00 g, 7.96 mmol), triethylamine (1.2 mL, 7.96 mmol), and catalytic amount of DMAP in 5 mL of CH_2Cl_2. After 3.5 hours, the reaction mixture was poured into saturated $NaHCO_3$, and extracted with AcOEt. The organic layer was dried over Na_2SO_4. Filtration, evaporation *in vacuo*, and purification by flash column chromatography (AcOEt/ *n*–hexane = 1/4 to 2/1) gave 2.53 g (77%) of **11** as a yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.92 (2H, t, *J* = 7.1 Hz), 3.37 (1H, t, *J* = 6.6 Hz), 2.53 (2H, t, *J* = 7.1 Hz), 2.36 (2H, t, *J* = 7.2 Hz), 1.90−1.65 (4H, m), 1.42 (9H, s).

*Preparation of tert-butyl 3-[N-(benzyloxy)-5-(butylamino)pentanamido]propanoate (**12**).* A mixture of **11** (950 mg, 2.29 mmol) and *n*-butylamine (1.80 mL, 18.3 mmol) in MeCN (2 mL) was stirred at room temperature for 12 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by silica gel flash column chromatography ($CHCl_3$/MeOH = 97/3 to 81/19) gave 748 mg (quant.) of 1**2** as a yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, *J* = 7.1 Hz), 2.62–2.50 (6H, m), 2.37 (2H, t, *J* = 7.4 Hz), 1.66–1.27 (17H, m), 0.91 (2H, t, *J* = 7.2 Hz): MS (ESI) *m/z* 407 (MH^+).

*Preparation of compounds **13a** and **13g–l***. A solution of bromide (**11**) (1.0 e.q.) and the corresponding secondary amine (4.0–8.0 e.q.) in MeCN was stirred at reflux temperature for 9–12 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by silica gel flash column chromatography gave compounds **13a** and **13g–l** (yield, 47–99%).

*tert-butyl 3-[(benzyloxy)[5-({[butyl(methyl)]amino}valeryl)amino]propanoate (**13a**)*: Yield 99%, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, m), 2.53 (2H, t, *J* = 7.0 Hz), 2.37 (2H, t, *J* = 7.5 Hz), 2.31 (4H, m), 2.20 (3H, s), 1.70 (2H, m), 1.58 (2H, quin, *J* = 7.5 Hz), 1.49–1.43 (2H, m), 1.42 (9H, s), 1.34–1.26 (2H, m), 0.91 (3H, t, *J* = 7.0 Hz).

*tert-Butyl 3-{N-(benzyloxy)-5-[ethyl(methyl)amino]pentanamido}propanoate (**13g**):* Yield 47 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, *J* = 7.1 Hz), 2.59–2.35 (8H,

m), 2.31 (3H, s), 1.66–1.51 (4H, m), 1.42 (9H, m), 1.13 (3H, t, J = 7.2 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 174.95, 170.85, 134.42, 129.34, 128.98, 128.70, 80.83, 56.62, 51.25, 41.69, 41.06, 33.17, 32.15, 28.06, 26.20, 22.28, 11.51: MS (ESI) *m/z* 393 (MH^+).

tert-Butyl 3-{N-(benzyloxy)-5-[methyl(propyl)amino]pentanamido}propanoate ***(13h)****:* Yield 78 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.52 (2H, t, J = 7.1 Hz), 2.44–2.32 (6H, m), 2.27 (3H, s), 1.64–1.45 (6H, m), 1.42 (9H, s), 0.90 (3H, t, J = 7.4 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.11, 170.86, 134.45, 129.33, 128.96, 128.69, 80.82, 59.51, 57.24, 41.88, 33.18, 32.23, 28.06, 26.44, 22.35, 19.96, 11.83: MS (ESI) *m/z* 407 (MH^+).

tert-Butyl 3-{N-(benzyloxy)-5-[methyl(pentyl)amino]pentanamido}propanoate ***(13i)****:* Yield 92 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.2 Hz), 2.52 (2H, t, J = 7.1 Hz), 2.40–2.27 (6H, m), 2.21 (3H, s), 1.65–1.40 (15H, m), 1.37–1.20 (4H, m), 0.89 (3H, t, J = 6.9 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.13, 170.88, 134.48, 129.31, 128.94, 128.68, 80.80, 57.87, 57.50, 42.18, 33.19, 32.36, 29.78, 28.06, 26.90, 26.88, 22.64, 22.51, 14.07; MS (ESI) *m/z* 435 (MH^+).

tert-Butyl 3-{N-(benzyloxy)-5-[hexyl(methyl)amino]pentanamido}propanoate ***(13j)****:* Yield 99 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.52 (2H, t, J = 7.1 Hz), 2.41–2.25 (6H, m), 2.20 (3H, s), 1.64–1.39 (15H, m), 1.36–1.21 (6H, m), 0.88 (3H, t, J = 6.8 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.17, 170.89, 134.48, 129.31, 128.94, 128.68, 80.80, 57.94, 57.51, 42.21, 41.68, 33.19, 32.37, 31.84, 28.06, 27.28, 27.21, 26.94, 22.65, 22.52, 14.07: MS (ESI) *m/z* 449 (MH^+).

tert-Butyl 3-{N-(benzyloxy)-5-[butyl(ethyl)amino]pentanamido}propanoate ***(13k)****:* Yield 86 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.53 (4H, m), 2.46–2.31 (6H, m), 1.63–1.36 (15H, m), 1.36 (9H, s), 1.36–1.20 (2H, m), 1.02 (3H, t, J = 7.2 Hz), 0.91 (3H, t, J = 7.4 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.26, 170.88, 134.49, 129.30, 128.94, 128.68, 80.81, 53.23, 47.46, 41.74, 33.19, 32.39, 28.99, 28.06, 26.62, 22.58, 20.78, 14.07, 11.53: MS (ESI) *m/z* 435 (MH^+).

tert-Butyl 3-[N-(benzyloxy)-5-(dibutylamino)pentanamido]propanoate ***(13l)****:* Yield 93 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.53 (2H, t, J = 7.1 Hz), 2.45–2.22 (8H, m), 1.64–1.50 (2H, m), 1.49–1.34 (15H, m), 1.34–1.21 (4H, m), 0.90 (6H, t, J = 7.2 Hz): ^{13}C

NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.19, 170.89, 134.51, 129.29, 128.92, 128.67, 80.79, 53.91, 41.74, 33.19, 32.45, 29.21, 28.06, 26.82, 22.61, 20.78, 14.11: MS (ESI) *m/z* 463 (MH^+).

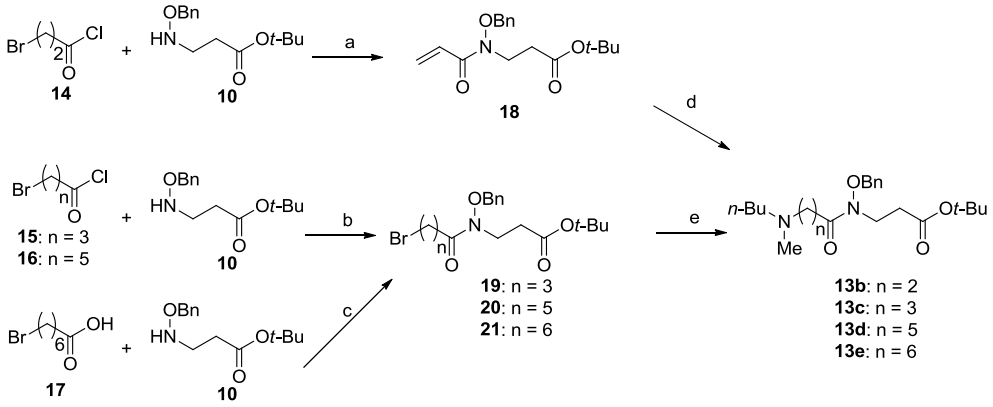
*Preparation of tert-butyl 3-(N-(benzyloxy)-5-bromopentanamido)propanoate (**13f**).* To a solution of **11** (400 mg, 0.97 mmol, 1 eq), dimethylamine hydrochloride (787 mg, 9.65 mmol) and K_2CO_3 (13 g, 9.65 mmol) in CH_2Cl_2 (4 mL) was stirred at 35 °C for 15 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, evaporation *in vacuo*, and purification by silica gel flash chromatography ($CHCl_3$/MeOH = 100 /0 to 90/10) gave 216 mg (59 %) of **13f** as a yellow oil: ^1H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, *J* = 7.1 Hz), 2.52 (2H, t, *J* = 7.1 Hz), 2.37 (2H, t, *J* = 7.4 Hz), 2.32–2.22 (10H, m), 1.64–1.45 (4H, m), 1.41 (9H, s): MS (ESI) *m/z* 379 (MH^+).

*Preparation of compounds **13m** and **13n**.* A mixture of 1**2** (1.0 e.q.), the corresponding bromoalkane (6.0 e.q.), and K_2CO_3 (4.0 e.q.) in DMF was stirred at 60°C for 20 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by silica gel flash chromatography gave compounds **13m** and **13n** (yield, 65–74%).

*tert-Butyl 3-[N-(benzyloxy)-5-[butyl(hexyl)amino]pentanamido]propanoate (**13m**)*: Yield 74 %, brown oil: ^1H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, *J* = 7.1 Hz), 2.52 (8H, t, *J* = 7.2 Hz), 2.38 (2H, t, *J* = 7.1 Hz), 1.64–1.38 (17H, m), 1.37–1.22 (8H, m), 0.95–0.84 (6H, m): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.04, 170.84, 134.44, 129.33, 128.97, 128.70, 80.82, 53.84, 53.59, 41.69, 33.17, 32.21, 31.70, 28.06, 27.14, 26.09, 22.63, 22.36, 20.64, 14.03, 13.97: MS (ESI) *m/z* 491 (MH^+).

*tert-Butyl 3-{N-(benzyloxy)-5-[butyl(octyl)amino]pentanamido}propanoate (**13n**)*: Yield 65 %, brown oil: ^1H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, *J* = 7.1 Hz), 2.71–2.44 (8H, m), 2.38 (2H, m), 1.63–1.46 (8H, m), 1.42 (9H, s), 1.39–1.20 (12H, m), 0.97–0.84 (6H, m): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 174.84, 170.82, 134.41, 129.35, 128.99, 128.71, 80.83, 53.47, 36.46, 33.16, 32.12, 31.81, 29.41, 29.25, 28.06, 27.41, 22.64, 22.26, 20.59, 14.09, 13.92: MS (ESI) *m/z* 519 (MH^+).

Scheme S2. Synthesis of Compounds 13b–e.[a]



[a]Reagents and conditions: (a) *N*,*N*-dimethylaminopyridine (DMAP), Et_3N, CH_2Cl_2, room temperature, 61–83%; (b) *N*-dimethylaminopyridine (DMAP), Et_3N, CH_2Cl_2, room temperature, 61–83%; (c) benzotriazolyloxy-tris[pyrrolidino]phosphonium hexafluorophosphate (PyBOP), *i*-Pr_2NEt, CH_2Cl_2, reflux, 75%; (d) *N*-methylbutan-1-amine, 1,4-dioxane, reflux, 89%; (e) R^1R^2NH, NaI, acetone, reflux, 39–72%.

*Preparation of tert-Butyl 3-[N-(benzyloxy)acrylamido]propanoate (**18**).* A solution of 3-bromopropanoyl chloride (100 μL, 1.13 mmol) in 5 mL of CH_2Cl_2 was added a solution a solution of **10** (190 mg, 0.76 mmol), triethylamine (0.3 mL), and catalytic amount of DMAP in 5 mL of CH_2Cl_2. After 6.5 hours, the reaction mixture was poured into water and extracted with $CHCl_3$. The organic layer was dried over Na_2SO_4. Filtration, evaporation in vacuo, and purification by flash column chromatography (AcOEt/*n*-hexane = 1/9 to 3/7) gave 147 mg (64%) of **18** as yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 6.69 (1H, dd, *J* = 10.3, 17.6 Hz), 6.39 (1H, dd, *J* = 1.8, 17.1 Hz), 5.70 (1H, dd, *J* = 2.0, 10.4 Hz), 4.85 (2H, s), 3.98 (2H, t, *J* = 7.1 Hz), 2.58 (2H, t, *J* = 7.1 Hz), 1.42 (9H, s): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 170.77, 166.87, 134.16, 129.35, 129.15, 129.02, 128.70, 126.32, 80.89, 41.92, 33.24, 28.05, 27.75: MS (ESI) m/z 306 (MH^+).

*Preparation of compounds **19** and **20**.* Bromoalkanoyl chloride (***15*** or **16**) (1.5 e.q.) was added to a solution of compound **10** (1.0 e.q.), triethylamine (1.1–3.0 e.q.), and a catalytic amount of DMAP in CH_2Cl_2 in a

dropwise fashion with cooling in an ice-bath. After stirred at room temperature for 4.0–6.5 h, the reaction mixture was diluted with AcOEt. The organic solution was washed with saturated $NaHCO_3$ aqueous solution. Then, the organic layer was separated and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by silica gel flash column chromatography gave bromides **19** or **20**.

*tert-Butyl 3-[N-(benzyloxy)-4-bromobutanamido]propanoate (**19**)*: Yield 73%, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.39 (5H, m), 4.85 (2H, s), 3.92 (2H, t, J = 7.1 Hz), 3.45 (2H, t, J = 6.3 Hz), 2.54 (2H, q, J = 6.8 Hz), 2.13 (2H, quin, J = 6.6 Hz), 1.42 (9H, s): MS (ESI) *m/z* 400, 402 (MH^+).

*tert-Butyl 3-[N-(benzyloxy)-6-bromohexanamido]propanoate (**20**)*: Yield 83%, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.92 (2H, m), 3.38 (2H, m), 2.53 (2H, m), 2.35 (2H, m), 1.84 (2H, m), 1.75–1.33 (3H, m), 1.41 (9H, s): MS (ESI) m/z 428, 430 (MH^+).

*Preparation of tert-butyl 3-[N-(benzyloxy)-7-bromoheptanamido]propanoate (**21**)*. To a solution of compound **10** (400 mg, 1.59 mmol) in CH_2Cl_2 (5 mL) were added 5-bromoheptanoic acid (**17**) (334 mg, 1.60 mmol), diisopropylethylamine (419 μL, 2.41 mmol), and benzotriazol-1-yl-oxytripyrrolidinophosphonium hexafluorophosphate (PyBOP) (1.25 g, 2.40 mmol). The reaction mixture was heated at reflux temperature overnight. It was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, evaporation in vacuo, and purification by silica gel flash column chromatography (AcOEt/*n*-hexane = 1/10 to AcOEt only) gave 415 mg (61%) of **21** as a yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.36 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 3.51 (2H, t, J = 6.7 Hz), 2.53 (2H, t, J = 7.1 Hz), 2.35 (2H, t, J = 7.4 Hz), 1.81–1.70 (2H, m), 1.70–1.50 (4H, m), 1.42 (9H, s), 1.39–1.25 (2H, m).

*Preparation of tert-butyl 3-{N-(benzyloxy)-3-[butyl(methyl)amino]propanamido}propanoate (**13b**).* A solution of **18** (137 mg, 0.449 mmol) and *N*-methyl-*N*-butylamine (270 μL, 2.24 mmol) in 1,4-dioxane was stirred at reflux temperature for 8 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by silica gel flash column chromatography ($CHCl_3$/MeOH = 49/1 to 24/1) gave 158 mg (89%) of

13b as a yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.84 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.66 (2H, m), 2.53 (4H, m), 2.31 (2H, m), 2.19 (3H, s), 1.50–1.35 (11H, m), 1.35–1.20 (2H, m), 0.90 (3H, J = 7.2 Hz): MS (ESI) *m/z* 393 (MH^+).

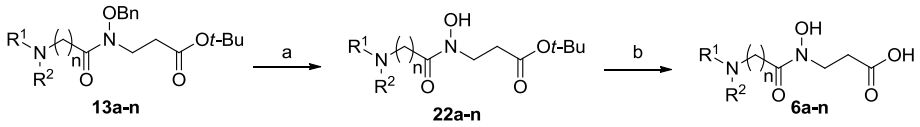
*Prepareation of compounds **13c–e***. A mixture of bromide (**19–21**) (1.0 e.q.), *N*-methyl-*n*-butylamine (5.0–7.0 e.q.), and a catalytic amount of NaI in acetone was heated at reflux temperature for 6–16 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by silica gel flash column chromatography gave compounds **13c–e** (yield, 39–75%).

*tert-Butyl 3-{N-(benzyloxy)-4-[butyl(methyl)amino]butanamido}propanoate (**13c**)*: Yield 61%, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.83 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.53 (2H, t, J = 7.2 Hz), 2.40 (2H, t, J = 7.5 Hz), 2.35–2.27 (4H, m), 2.20 (3H, s), 1.78 (2H, m), 1.50–1.21 (13H, m), 0.90 (3H, t, J = 7.2 Hz): MS (ESI) *m/z* 407 (MH^+).

*tert-Butyl 3-{N-(benzyloxy)-6-[butyl(methyl)amino]hexanamido}propanoate (**13d**)*:Y ield 75%; brown oil; ^{1}H NMR ($CDCl_3$, 300 MHz,δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.52 (2H, t, J = 7.1 Hz), 2.36 (6H, m), 2.24 (3H, s), 1.65–1.43 (6H, m), 1.42 (9H, s), 1.38–1.22 (4H, m), 0.92 (3H, t, J = 7.4 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.25, 170.88, 134.50, 129.30, 128.93, 128.68, 80.80, 57.54, 57.46, 42.11, 41.77, 33.20, 32.44, 29.14, 28.07, 27.32, 26.81, 24.42, 20.72, 14.04: MS (ESI) *m/z* 435 (MH^+).

*tert-Butyl 3-{N-(benzyloxy)-7-[butyl(methyl)amino]heptanamido}propanoate (**13e**)*: Yield 57%, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz,δ; ppm) 7.38 (5H, m), 4.82 (2H, s), 3.91 (2H, t, J = 7.1 Hz), 2.52 (2H, t, J = 7.1 Hz), 2.34 (6H, m), 1.65–1.40 (6H, m), 1.42 (9H, s), 1.38–1.25 (6H, m), 0.92 (3H, t, J = 7.4 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.33, 170.89, 134.52, 129.29, 128.92, 128.67, 80.79, 57.75, 57.51, 42.18, 33.20, 32.44, 29.34, 29.25, 28.06, 27.38, 26.96, 24.47, 20.73, 14.05: MS (ESI) *m/z* 449 (MH^+).

Scheme S3. Synthesis of Compounds 6.[a]



6a, 13a, 22a: R^1 = *n*-Bu,R^2 = Me, n = 4
6b, 13b, 22b: R^1 = *n*-Bu, R^2 = Me, n = 2
6c, 13c, 22c: R^1 = *n*-Bu, R^2 = Me, n = 3
6d, 13d, 22d: R^1 = *n*-Bu, R^2 = Me, n = 5
6e, 13e, 22e: R^1 = *n*-Bu, R^2 = Me, n = 6
6f, 13f, 22f: R^1 = R^2 = Me, n = 4
6g, 13g, 22g: R^1 = Et, R^2 = Me, n = 4
6h, 13h, 22h: R^1 = *n*-Pr, R^2 = Me, n = 4
6i, 13i, 22i: R^1 = *n*-pentyl, R^2 = Me, n = 4
6j, 13j, 22j: R^1 = *n*-hexyl, R^2 = Me, n = 4
6k, 13k, 22k: R^1 = *n*-Bu, R^2 = Et, n = 4
6l, 13l, 22l: R^1 = *n*-Bu, R^2 = *n*-Bu, n = 4
6m, 13m, 22m: R^1 = *n*-Bu, R^2 = *n*-hexyl, n = 4
6n, 13n, 22n: R^1 = *n*-Bu, R^2 = *n*-octyl, n = 4

[a]Reagents and conditions: (a) H_2, Pd/C, AcOEt or MeOH, room temperature; (b) HCl, 1,4-dioxane, CH_2Cl_2, room temperature, 31–100% (2 steps).

*Preparation of compounds **22a–n***. A catalytic amount of 5% of Pd/C was added to a solution of compound **21** in AcOEt. After stirred under H_2 at room temperature for 3–23 h, the reaction mixture was filtered through Celite. The filtrate was concentrated in vacuo to give compounds **22a** and **22g–l**, which was used without further purification.

*tert-butyl 3-({5-[butyl(methyl)]amino}valeryl(hydroxy)amino]propanoate **(22a)***: Yellow oil: ^{1}H NMR ($CDCl_3$, 500 MHz, δ; ppm) 3.91 (2H, t, J = 7.0 Hz), 2.59–2.54 (4H, m), 2.43–2.38 (4H, m), 2.24 (3H, s), 1.74 (2H, m), 1.53–1.48 (4H, m), 1.45 (9H, s), 1.35–1.25 (2H, m), 0.93 (3H, t, J = 7.5 Hz).

*tert-Butyl 3-{3-[butyl(methyl)amino]-N-hydroxypropanamido}propanoate **(22b)***: Yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.89 (2H, t, J = 7.2 Hz), 2.79 (2H, m), 2.50 (4H, m), 2.34 (3H, s), 1.55–1.40 (2H, m), 1.45 (9H, s), 1.41–1.22 (2H, m), 0.93 (3H, t, J = 7.2 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 171.00, 170.93, 80.65, 57.93, 56.66, 43.23, 42.21, 33.48, 29.00, 28.07, 20.42, 13.89: MS (ESI) *m/z* 303 (MH^+).

*tert-Butyl 3-{4-[butyl(methyl)amino]-N-hydroxybutanamido}propanoate **(22c)***: Yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.90 (2H, t, J = 7.2 Hz), 2.61–2.35 (8H, m), 2.24 (3H, s), 1.57–1.40 (11H, m), 1.40–1.23 (2H, m), 0.94 (3H, t, J = 7.4 Hz); MS (ESI) *m/z* 317 (MH^+).

*tert-Butyl 3-{6-[butyl(methyl)amino]-N-hydroxyhexanamido}propanoate **(22d)***: Yellow oil, ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.88 (2H, t, J = 6.6 Hz), 2.61 (2H, m), 2.43 (2H, m), 2.29 (3H, s), 1.75–1.60 (2H, m), 1.60–1.47 (4H, m), 1.45 (9H, s), 1.42–1.23 (2H, m), 0.92 (3H, t, J = 7.2 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.70, 172.83, 81.46, 57.29, 57.10, 44.47, 41.84, 33.62, 31.52, 28.20, 28.05, 26.60, 25.87, 24.41, 20.67, 13.97: MS (ESI) *m/z* 345 (MH^+).

*tert-Butyl 3-{7-[butyl(methyl)amino]-N-hydroxyheptanamido}propanoate **(22e)***: Yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.87 (2H, t, J = 6.6 Hz), 2.60 (2H, m), 2.54–2.29 (6H, m), 2.24 (3H, s), 1.73–1.57 (2H, m), 1.57–1.39 (13H, m), 1.39–1.21 (6H, m), 0.92 (3H, t, J = 7.2 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 174.13, 172.70, 81.39, 57.12, 56.77, 44.53, 41.88, 33.51, 31.92, 29.17, 28.40, 28.06, 26.48, 26.26, 24.84, 20.77, 14.02: MS (ESI) *m/z* 359 (MH^+).

*tert-Butyl 3-[5-(dimethylamino)valeryl(hydroxyl)amino]propanoate **(22f)***: Yellow solid: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.91 (2H, t, J = 6.9 Hz), 2.65–2.40 (6H, m), 2.35 (6H, s), 1.80–1.65 (2H, m), 1.65–1.50 (2H,

m), 1.45 (9H, m): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.02, 171.84, 81.02, 58.69, 44.64, 43.98, 33.61, 30.22, 28.06, 24.94, 23.96.

*tert-Butyl 3-{5-[ethyl(methyl)amino]-N-hydroxypentanamido}propanoate (**22g**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.91 (2H, t, J = 6.9 Hz), 2.70–2.42 (8H, m), 2.33 (3H, s), 1.80–1.65 (2H, m), 1.65–1.55 (2H, m), 1.45 (9H, s), 1.16 (3H, t, J = 7.4 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.02, 171.87, 81.05, 56.06, 51.37, 44.00, 40.72, 33.61, 30.07, 28.13, 28.06, 24.48, 24.16, 20.73: MS (ESI) *m/z* 303 (MH^+).

*tert-Butyl 3-{N-hydroxy-5-[methyl(propyl)amino]pentanamido}propanoate (**22h**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.90 (2H, t, J = 6.9 Hz), 2.65–2.33 (8H, m), 2.27 (3H, s), 1.78–1.65 (2H, m), 1.62–1.42 (13H, m), 0.91 (3H, t, J = 7.4 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.10, 171.94, 81.07, 59.71, 56.80, 43.99, 41.62, 33.63, 30.10, 28.09, 24.75, 24.44, 19.30, 11.82: MS (ESI) *m/z* 317 (MH^+).

*tert-Butyl 3-{N-hydroxy-5-[methyl(pentyl)amino]pentanamido}propanoate (**22i**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.90 (2H, t, J = 7.1 Hz), 2.70–2.49 (4H, m), 2.49–2.31 (4H, m), 2.27 (3H, s), 1.79–1.65 (2H, m), 1.60–1.40 (13H, m), 1.38–1.20 (4H, m), 0.90 (3H, t, J = 7.1 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.07, 171.88, 81.04, 57.86, 56.80, 43.98, 41.65, 33.62, 30.07, 29.66, 28.05, 26.68, 25.68, 24.70, 24.50, 22.51, 14.01: MS (ESI) *m/z* 345 (MH^+).

*tert-Butyl 3-{5-[hexyl(methyl)amino]-N-hydroxypentanamido}propanoate (**22j**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.90 (2H, t, J = 6.9 Hz), 2.70–2.50 (4H, m), 2.43–2.28 (4H, m), 2.22 (3H, s), 1.77–1.63 (2H, m), 1.56–1.40 (13H, m), 1.36–1.21 (6H, m), 0.92 (3H, t, J = 6.8 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.07, 171.90, 81.03, 58.12, 56.96, 43.93, 41.86, 33.66, 31.70, 29.95, 28.05, 27.27, 26.24, 24.87, 24.80, 22.61, 14.03: MS (ESI) *m/z* 359 (MH^+).

*tert-Butyl 3-{5-[butyl(ethyl)amino]-N-hydroxypentanamido}propanoate (**22k**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.90 (2H, t, J = 6.9 Hz), 2.70–2.36 (10H, m), 1.71 (2H, m), 1.57–1.38 (13H, m), 1.30 (2H, sext, J = 7.3 Hz), 1.12–0.87 (6H, m): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.22, 172.05, 81.10, 52.82, 52.63, 47.09, 44.16, 33.60, 30.26, 28.05, 27.47, 24.74, 24.50, 20.78, 13.98, 10.17: MS (ESI) *m/z* 345 (MH^+).

*tert-Butyl 3-[5-(dibutylamino)-N-hydroxypentanamido]propanoate (**22l**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.89 (2H, t, J = 6.8 Hz), 2.70–2.33 (10H, m), 1.69 (2H, m), 1.56–1.36 (15H, m), 1.36–1.22 (2H,

m), 0.92 (6H, m): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.27, 172.12, 81.15, 53.45, 53.28, 44.20, 33.60, 30.40, 28.06, 27.47, 24.89, 24.28, 20.75, 13.98: MS (ESI) *m/z* 373 (MH^+).

*tert-Butyl 3-{5-[butyl(hexyl)amino]-N-hydroxypentanamido}propanoate (**22m**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.90 (2H, t, *J* = 6.9 Hz), 2.75–2.42 (10H, m), 1.80–1.41 (17H, m), 1.41–1.22 (8H, m), 0.99–0.84 (6H, m): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.12, 172.15, 81.18, 53.46, 53.18, 44.22, 33.60, 31.59, 30.38, 28.14, 28.06, 27.12, 27.00, 24.85, 24.52, 23.88, 22.59, 20.65, 14.00, 13.91: MS (ESI) *m/z* 401 (MH^+).

*tert-Butyl 3-{5-[butyl(octyl)amino]-N-hydroxypentanamido}propanoate (**22n**)*: Brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.91 (2H, t, *J* = 6.8 Hz), 2.88–2.53 (10H, m), 1.85–1.53 (8H, m), 1.52–1.22 (21H, m), 1.00–0.82 (6H, m): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 172.94, 172.12, 81.19, 52.83, 44.31, 35.18, 33.54, 31.72, 31.61, 30.61, 29.15, 29.02, 28.14, 28.06, 27.15, 25.98, 23.91, 22.60, 20.40, 14.07, 13.74: MS (ESI) *m/z* 429 (MH^+).

*Preparation of compounds **6a–n***. To a solution of compound **22** (1.0 e.q.) in CH_2Cl_2 was added 4N HCl in 1,4-dioxane (10–15 e.q.) in a dropwise fashion at 0°C. After stirred at room temperature for 5–10 h, the mixture was concentrated in vacuo. The residue was purified by HPLC gave compounds **6a**–**n**.

*3-{5-[butyl(methyl)amino]-N-hydroxypentanamido}propanoic acid (**6a**)*: Yield, 32.0 mg, 62% from **21a**: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.88 (2H, t, *J* = 7.0 Hz), 3.20–3.16 (2H, m), 3.09–3.06 (2H, m), 2.84 (3H, s), 2.61–2.55 (2H, m), 2.39 (2H, t, *J* = 6.7 Hz), 1.75–1.67 (6H, m), 1.44–1.40 (2H, m), 1.00 (3H, t, *J* = 7.6 Hz): ^{13}C NMR ($CDCl_3$, 125 MHz, δ; ppm) 176.67, 175.29, 57.25, 57.02, 45.19, 40.42, 33.86, 32.31, 27.21, 24.77, 22.73, 20.81, 13.87: MS (FAB) m/z 275 (MH^+): HRMS calcd. for $C_{13}H_{27}O_4N_2$, 275.19709, found 275.19592: HPLC t_R = 11.72 min (Gradient (I), purity 95%).

*3-{3-[butyl(methyl)amino]-N-hydroxypropanamido}propanoic acid (**6b**)*: Yield 38.8 mg, 73% from **13b**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.90 (2H, t, *J* = 7.1 Hz), 3.53 (2H, m), 3.25–3.15 (2H, m), 2.97 (2H, t, *J* = 6.3 Hz), 2.86 (3H, s), 2.63 (2H, t, *J* = 6.9 Hz), 1.75 (2H, quin, *J* = 7.8 Hz), 1.44 (2H, sext, *J* = 7.4 Hz), 1.01 (3H, t, *J* = 7.4 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.08, 172.07, 57.83, 53.01, 45.17,

40.82, 32.10, 27.92, 27.17, 20.77, 13.86: MS (ESI) *m/z* 247 (MH^+): HRMS calcd. for $C_{11}H_{23}N_2O_4$ (MH^+), 247.1658, found 247.1662; HPLC t_R = 10.55 min (Gradient (VI), purity 98%).

3-{4-[Butyl(methyl)amino]-N-hydroxybutanamido}propanoic acid ***(6c)***: yield, 30.3 mg, 67% from **13c**: yellow oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.89 (2H, t, *J* = 7.1 Hz), 3.29–3.00 (4H, m), 2.87 (3H, s), 2.70–2.57 (4H, m), 2.10–1.90 (2H, m), 1.81–1.61 (2H, m), 1.43 (2H, sext, *J* = 7.4 Hz), 1.00 (3H, t, *J* = 7.4 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.08, 174.60, 57.32, 57.21, 45.29, 40.60, 32.23, 30.39, 27.27, 20.82, 20.29, 13.88; MS (ESI) *m/z* 261 (MH^+); HRMS calcd. for $C_{12}H_{25}N_2O_4$, 261.1814, found 261.1810; HPLC t_R = 11.34 min (Gradient (IV), purity 98%).

3-{6-[Butyl(methyl)amino]-N-hydroxyhexanamido}propanoic acid ***(6d)***: Yield, 13.4 mg, 31% from **13d**: yellow oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.88 (2H, t, *J* = 7.1 Hz), 3.27–2.98 (4H, m), 2.85 (3H, s), 2.60 (2H, t, *J* = 7.1 Hz), 2.51 (2H, t, *J* = 7.2 Hz), 1.81–1.61 (6H, m), 1.50–1.35 (4H, m), 1.00 (3H, t, *J* = 7.4 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.81, 175.16, 57.19, 40.42, 34.42, 32.67, 32.46, 27.18, 26.98, 25.35, 24.87, 20.81, 13.84: MS (ESI) *m/z* 289 (MH^+): HRMS calcd. for $C_{15}H_{31}N_2O_4$, 289.2127, found 289.2121: HPLC t_R = 14.27 min (Gradient (I), purity 99%).

3-{7-[Butyl(methyl)amino]-N-hydroxyheptanamido}propanoic acid ***(6e)***: Yield, 21.6 mg, 45% from **13e**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.87 (2H, t, *J* = 7.1 Hz), 3.25–2.98 (4H, m), 2.85 (3H, s), 2.60 (2H, m), 2.48 (2H, t, *J* = 7.4 Hz), 1.80–1.57 (6H, m), 1.50–1.35 (6H, m), 1.01 (3H, t, *J* = 7.2 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 176.17, 175.15, 57.36, 57.26, 45.27, 40.46, 32.96, 32.51, 29.58, 27.23, 27.20, 25.39, 24.99, 20.86, 13.89: MS (ESI) *m/z* 303 (MH^+): HRMS calcd. for $C_{15}H_{31}N_2O_4$, 303.2284, found 303.2281: HPLC t_R = 15.47 min (Gradient (VI), purity 97%).

3-(5-(dimethylamino)-N-hydroxypentanamido)propanoic acid ***(6f)***. Yield, 35.2 mg, 87% from **13f**: ^{1}H NMR (CD_3OD, 300 MHz,δ; ppm) 3.88 (2H, t, *J* = 6.9 Hz), 3.12 (2H, m), 2.88 (6H, s), 2.65–2.52 (4H, m), 1.81–1.61 (4H, m): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.34, 175.10, 58.67, 45.20, 43.43, 32.32, 32.23, 25.16, 22.11: HRMS calcd. for $C_{10}H_{21}N_2O_4$ (MH^+-HCl), 233.1501, found 233.1504: HPLC t_R = 11.12 min (Gradient (III), purity 99%).

*3-{5-[ethyl(methyl)amino]-N-hydroxypentanamido}propanoic acid (**6g**)*: Yield, 25.6 mg, 98% from **13g**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.88 (2H, t, J = 6.9 Hz), 3.28–3.00 (4H, m), 2.84 (3H, s), 2.66–2.53 (4H, m), 1.83–1.61 (4H, m), 1.33 (3H, t, J = 7.4 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.39, 175.15, 56.51, 52.46, 45.25, 39.82, 32.38, 32.29, 24.85, 22.35, 9.56; MS (ESI) *m/z* 247 (MH^+): HRMS calcd. for $C_{11}H_{23}N_2O_4$, 247.1658, found 247.1662: HPLC t_R = 11.39 min (Gradient (II), purity 98%).

*3-{N-hydroxy-5-[methyl(propyl)amino]pentanamido}propanoic acid (**6h**)*: Yield, 36.3 mg, 84% from **13h**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.88 (2H, t, J = 6.9 Hz), 3.27–2.96 (4H, m), 2.85 (3H, s), 2.67–2.52 (4H, m), 1.85–1.61 (6H, m), 1.02 (3H, t, J = 7.5 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.40, 175.13, 58.92, 57.05, 45.25, 40.44, 32.38, 32.29, 24.76, 22.35, 18.70, 11.10: MS (ESI) *m/z* 261 (MH^+): HRMS calcd. for $C_{12}H_{25}N_2O_4$, 261.1814, found 261.1810; HPLC t_R = 7.32 min (Gradient (III), purity 95%).

*3-{N-hydroxy-5-[methyl(pentyl)amino]pentanamido}propanoic acid (**6i**)*: Yield, 30.4 mg, 90% from **13i**: brown oil; ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.88 (2H, t, J = 6.9 Hz), 3.36–2.94 (4H, m), 2.85 (3H, s), 2.65–2.52 (4H, m), 1.85–1.62 (6H, m), 1.48–1.30 (4H, m), 0.96 (3H, t, J = 6.9 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.31, 175.04, 57.44, 57.00, 45.21, 40.38, 32.33, 32.24, 29.65, 24.92, 24.66, 23.23, 22.28, 14.12: MS (ESI) *m/z* 289 (MH^+): HRMS calcd. for $C_{14}H_{29}N_2O_4$, 289.2127, found 289.2121: HPLC t_R = 12.52 min (Gradient (IV), purity 99%).

*3-{5-[hexyl(methyl)amino]-N-hydroxypentanamido}propanoic acid (**6j**)*: Yield, 38.9 mg, 96% from **13j**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.88 (2H, t, J = 6.9 Hz), 3.28–2.97 (4H, m), 2.85 (3H, s), 2.68–2.50 (4H, m), 1.86–1.60 (6H, m), 1.48–1.29 (6H, m), 0.93 (3H, m): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.40, 175.12, 57.51, 57.04, 45.25, 40.42, 32.43, 32.37, 32.28, 27.27, 25.23, 24.78, 23.51, 22.32, 14.28; HRMS calcd. for $C_{15}H_{31}N_2O_4$ (MH^+), 303.2284, found 303.2280; HPLC t_R = 14.90 min (Gradient (IV), purity 98%).

*3-{5-[butyl(ethyl)amino]-N-hydroxypentanamido}propanoic acid (**6k**)*: yield, 28.6 mg, 92% from **13k**: brown oil; ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.89 (2H, t, J = 6.9 Hz), 3.28–3.08 (6H, m), 2.67–2.52 (4H, m), 1.81–1.61 (6H, m), 1.73 (2H, sext, J = 7.4 Hz), 1.31 (3H, t, J = 7.2 Hz), 1.01 (3H, t, J = 7.2 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.36, 175.05, 53.54, 53.43, 45.27, 33.93, 32.34, 26.96, 24.45, 22.90, 22.49,

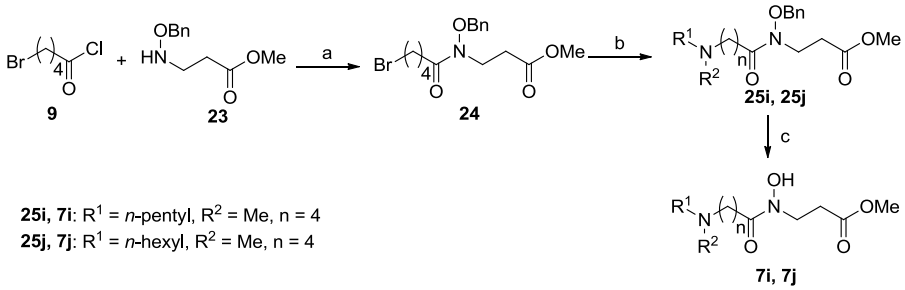
20.94, 13.92, 9.13; MS (ESI) *m/z* 289 (MH^+): HRMS calcd. for $C_{14}H_{29}N_2O_4$, 289.2127, found 289.2131: HPLC t_R = 11.14 min (Gradient (IV), purity 95%).

*3-[5-(dibutylamino)-N-hydroxypentanamido]propanoic acid (**6l**)*: Yield, 38.9 mg, 87% from **13l**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.88 (2H, t, *J* = 7.1 Hz), 3.23–3.05 (6H, m), 2.67–2.50 (4H, m), 1.82–1.60 (8H, m), 1.42 (4H, sext, *J* = 7.4 Hz), 1.01 (3H, t, *J* = 7.4 Hz): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.40, 175.05, 54.07, 53.94, 45.23, 32.36, 32.24, 26.85, 24.39, 22.44, 20.89, 13.88; MS (ESI) *m/z* 317 (MH^+); HRMS calcd. for $C_{16}H_{33}N_2O_4$, 317.2440, found 317.2436; HPLC t_R = 13.46 min (Gradient (V), purity 99%).

*3-{5-[Butyl(hexyl)amino]-N-hydroxypentanamido}propanoic acid (**6m**)*. Yield, 32.6 mg 50% from **21m**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.91 (2H, t, J = 7.1 Hz), 3.22–3.09 (6H, m), 2.68–2.55 (4H, m), 1.83–1.64 (8H, m), 1.51–1.36 (8H, m), 1.08–0.90 (6H, m): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.36, 175.05, 54.30, 54.05, 53.92, 45.23, 32.40, 32.24, 27.30, 26.85, 24.86, 24.38, 23.49, 22.42, 20.89, 14.24, 13.88; MS (ESI) m/z 345 (MH^+): HRMS calcd. for $C_{18}H_{37}N_2O_4$, 345.2753, found 345.2758: HPLC t_R = 11.19 min (Gradient (VII), purity 97%).

*3-{5-[Butyl(octyl)amino]-N-hydroxypentanamido}propanoic acid (**6n**)*. Yield, 58.6 mg, 89% from **21n**: brown oil: ^{1}H NMR (CD_3OD, 300 MHz, δ; ppm) 3.89 (2H, t, *J* = 7.1 Hz), 3.21–3.05 (6H, m), 2.68–2.50 (4H, m), 1.81–1.59 (8H, m), 1.50–1.25 (12H, m), 1.01 (3H, t, *J* = 7.4 Hz), 0.91 (3H, m): ^{13}C NMR (CD_3OD, 75 MHz, δ; ppm) 175.35, 175.03, 54.30, 54.06, 53.93, 45.24, 32.87, 32.35, 32.22, 30.19, 30.18, 27.62, 26.86, 24.90, 24.40, 23.67, 22.41, 20.89, 14.38, 13.89; MS (ESI) *m/z* 373 (MH^+): HRMS calcd. for $C_{20}H_{41}N_2O_4$, 373.3066, found 373.3071: HPLC t_R = 14.38 min (Gradient (VII), purity 97%).

Scheme S4. Synthesis of Compounds 7.[a]



25i, 7i: R^1 = *n*-pentyl, R^2 = Me, n = 4
25j, 7j: R^1 = *n*-hexyl, R^2 = Me, n = 4

[a]Reagents and conditions: (a) DMAP, Et_3N, CH_2Cl_2, room temperature, 78%; (b) R^1R^2NH, MeCN, reflux; (c) H_2, Pd/C, AcOEt, room temperature, 76–87% (2 steps).

*Preparation of methyl 3-(5-bromo-N-hydroxypentanamido)propanoate (**24**).* A solution of 5-bromovaleryl chloride (400 μL, 3.00 mmol) in CH_2Cl_2 was added to a solution of **23**[S3] (209 mg, 1.00 mmol), triethylamine (277 μL, 2.00 mmol), and catalytic amount of DMAP in CH_2Cl_2 (2 mL) in a dropwise fashion at 0 °C and stirred at rt for 4 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with saturated aqueous $NaHCO_3$ aquerous solution three times and dried over Na_2SO_4. Filtration, evaporation in vacuo, purification by flash column chromatography (AcOEt/Hexane = 1/4 to 1/1) gave 289 mg (78 %) of **24** as a yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.81 (2H, s), 3.97 (2H, t, *J* = 6.8 Hz), 3.64 (3H, s), 3.40 (2H, t, *J* = 6.6 Hz), 2.59 (2H, t, *J* = 6.8 Hz), 2.38 (2H, t, *J* = 7.1 Hz), 1.92–1.65 (4H, m): MS (ESI) *m/z* 372, 374 (MH^+).

*Preparation of compounds **25i** and **25j**.* A solution of bromide (**24**) (1.0 e.q.) and the corresponding secondary amine (3.0 e.q.) in MeCN was stirred at reflux temperature for 5 h. The reaction mixture was poured into water and extracted with AcOEt. The organic layer was washed with brine and dried over Na_2SO_4. Filtration, concentration in vacuo, and purification by silica gel flash column chromatography gave compounds **25i** and **25j** (yield, 83–84%).

*Methyl 3-[(N-(benzyloxy)-5-[methyl(pentyl)amino]pentanamido] propanoate (**25i**)*: Yield 84 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.81 (2H, s), 3.97 (2H, t, *J* = 6.8 Hz), 3.63 (3H, s), 2.59 (2H, t, *J* = 6.9 Hz), 2.45–2.25 (6H, m), 2.21 (3H, s), 1.65–1.19 (10H, m), 0.89 (3H, t, *J* = 6.9 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 175.30, 172.18, 134.36, 129.32, 128.99, 128.70, 57.89, 57.51, 51.77, 42.20, 41.53, 32.33, 31.94, 29.78, 26.93, 22.65, 22.48, 14.07.

*methyl 3-[(N-(benzyloxy)-5-[hexyl(methyl)amino]pentanamido] propanoate (**25j**)*: Yield 83 %, yellow oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 7.38 (5H, m), 4.81 (2H, s), 3.97 (2H, t, J = 6.8 Hz), 3.63 (3H, s), 2.59 (2H, t, J = 6.9 Hz), 2.44–2.25 (6H, m), 2.20 (3H, s), 1.67–1.21 (12H, m), 0.88 (3H, t, J = 6.6 Hz): ^{13}C NMR (CDCl3, 75 MHz, δ; ppm) 175.36, 172.18, 134.36, 129.32, 129.00, 128.70, 57.95, 57.51, 51.77, 42.22, 41.55, 32.34, 31.94, 31.85, 27.28, 27.23, 26.94, 22.65, 22.48, 14.07.

*Preparation of compounds **7i** and **7j**.* A catalytic amount of 5% of Pd/C was added to a solution of compound **25** in AcOEt. After stirred under H_2 at room temperature for **1**2 h, the reaction mixture was filtered

through Celite. The filtrate was concentrated in vacuo to give compound **7.** Regarding synthesis of compound **7i**, the further purification by HPLC was performed.

*Methyl 3-{N-hydroxy-5-[methyl(pentyl)amino]pentanamido}propanoate (**7i**)*: Yield, 26.2 mg, 76% from **24**: brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.95 (2H, t, J = 6.9 Hz), 3.69 (3H, s), 2.80–2.31 (8H, m), 2.27 (3H, s), 1.74 (2H, m), 1.62–1.42 (4H, m), 1.41–1.19 (4H, m), 0.91 (3H, t, J = 6.9 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.25, 172.75, 57.88, 56.80, 51.82, 43.86, 41.51, 32.10, 29.89, 29.65, 25.56, 24.61, 24.52, 22.51, 14.00: MS (ESI) *m/z* 303 (MH^+): HRMS calcd. for $C_{15}H_{31}N_2O_4$, 303.2284, found 303.2278; HPLC t_R = 14.44 min (Gradient (VIII), purity 96%).

*Methyl 3-{5-[hexyl(methyl)amino]-N-hydroxypentanamido}propanoate (**7j**)*: Yield, 71.7 mg, 70% from **24**: brown oil: ^{1}H NMR ($CDCl_3$, 300 MHz, δ; ppm) 3.95 (2H, t, J = 6.9 Hz), 3.69 (3H, s), 2.80–2.32 (8H, m), 2.27 (3H, s), 1.74 (2H, m) 1.61–1.40 (4H, m), 1.39–1.20 (6H, m), 0.89 (3H, t, J = 6.6 Hz): ^{13}C NMR ($CDCl_3$, 75 MHz, δ; ppm) 173.27, 172.74, 57.89, 56.78, 51.82, 43.87, 41.49, 32.08, 31.63, 29.94, 27.17, 25.82, 24.54, 22.58, 14.00: MS (ESI) m/z 317 (MH^+); HRMS calcd. for $C_{16}H_{33}N_2O_4$ (MH^+), 317.2440, found 317.2437: HPLC t_R = 16.54 min (Gradient (VIII), purity 96%).

Binding simulation. A homology model of JARID1A based on the crystal structure of JMJD2A (PDB code 3PDQ) was built using the homology modeling module of Protein Discovery Full Automatic Modeling System (PDFAMS) (In-Silico Sciences, Inc.).[S4] The amino acids were numbered according to the accession No. P29375 (amino acid residues 318–611). Docking was performed using Molegro Virtual Docker 6.0 software. The simulation was carried out under two conditions: (1) the hydroxamate group of compound **6a** bidentately coordinates to the catalytic Fe(II) ion; (2) the carboxylate group of compound **6a** forms hydrogen bonds with the amino acid residues of the enzyme. The structures of compounds **6a**, **6e, 6i**, **6j** and **6o** bound to JARID1A or JMJD2C (PDB code 2MXL) were constructed by MolDock, which is based on a heuristic search algorithm that combines differential evolution with a cavity prediction algorithm. The docking parameters were as follow: Grid Resolution: 0.30, Max iterations: 1500, Population size: 50, Energy threshold: 100.00, Simplex evolution: 300 (Max steps) and 1.00 (Neighbour distance factor), Search space (JARID1A): (X, Y, Z) = (-26.58, -21.59, -23.91) with radius 15, Search space (JMJD2C): (X, Y, Z) = (49.00, 24.79, 54.61) with radius 15, distance constraints (JARID1A): constraint center (X, Y, Z) = (-29.70, -21.38, -20.83) with hard constraint between minimum 2.2 to maximum 2.7 (two O atom of hydroxamate of compounds **6a**, **6e, 6i**, **6j** and **6o**) and (X, Y, Z) = (-24.08, -21.96, -24.52) with hard constraint between minimum 0.0 to maximum 3.0 (C atom of carboxylate of compounds **6a**, **6e, 6i**, **6j** and **6o**), distance constraints (JMJD2C): constraint center (X, Y, Z) = (53.30, 26.45, 49.72) with hard constraint between minimum 2.2 to maximum 2.7 (two O atoms of hydroxamate of compounds **6a**, **6e** and **6o**) and (X, Y, Z) = (50.72, 22.62, 59.14) with hard constraint between minimum 0.0 to maximum 3.0 (C atom of carboxylate of compounds **6a**, **6e** and **6o**).

Biology. *JARID1A, JMJD2C, and JHDM1F Inhibition Assay.* The JARID1A, JMJD2C, and JHDM1F activity were measured as described in ref S1 and S2.

JMJD1A Inhibition Assay. The JMJD1A activity assay was performed by Alpha screen assay system using JMJD1A (BPS #50130), biotinylated H3K9me1 peptide (AnaSpec, #64358), α-ketoglutarate (2OG, Aldrich K2000), $(NH_4)_2Fe(SO_4)_2 \cdot 6H_2O$ (Aldrich, #215406), (+)-Sodium L-ascorbate (Aldrich, #11140), AlphaLISA acceptor beads (PerkinElmer #AL138), Alpha Streptavidin donor beads (PerkinElmer #AL6760002), AlphaLISA epigenetic buffer (PerkinElmer #AL008). The inhibitors and JMJD1A (10 ng) in 7.5 μL of assay buffer (50 mM HEPES pH 7.5, 0.01% Tween, 0.1% BSA) were added to each well of a 384 white plate and were incubated for 5–10 minutes at room temperature. Then, 2.5 μL of mixture of the peptide (final concentration: 50 nM), 2OG (final concentration: 5 μM), $(NH_4)_2Fe(SO_4)_2 \cdot 6H_2O$ (final concentration: 5 μM), and (+)-Sodium L-ascorbate (final concentration: 100 μM) in assay buffer were added to the each well and incubated for 60 minutes at room temperature. Then, the reaction was treated with AlphaLISA acceptor beads in epigenetic buffer for 60 minutes hour and Alpha Streptavidin donor beads for 30 minutes, according to the supplier's instruction. The Alpha signal was read by Ensight® readers from PerkinElmer Ltd.

Western Blot Analysis. Human lung cancer A549 cells (provided by RIKEN BRC through the National Bio-Resource Project of the MEXT, Japan) were cultured in RPMI 1640 containing 10% heat-inactivated fetal bovine serum (FBS), penicillin and streptomycin mixture at 37 °C in a humidified atmosphere of 5% CO_2 in air. A549 cells (5×10^5 cells/2 mL/dish) were treated for the indicated period with synthetic compounds at the indicated concentrations in 10% FBS-supplemented in appropriate cell culture medium, then the cells were collected and extracted with SDS buffer. Protein concentrations of the lysates were determined using a BCA protein assay. Equivalent amounts of protein from each lysate were resolved in 5–20% SDS-polyacrylamide gels and transferred onto PVDF membranes. After blocking with TBS-T containing 5% skimed milk, the transblotted membranes were probed with rabbit polyclonal H3K4me3 antibody (Abcam, #ab8580) (1:5000 dilution), rabbit polyclonal H3K9me3 antibody (Abcam, #ab8898) (1:1000 dilution), and rabbit polyclonal H3K27me2 antibody (Cell Signal Technology, #9755S) (1:500 dilution) and rabbit polyclonal H3 antibody (Abcam, #ab1791) (1:200000 dilution) in TBS-T containing 5% skimed milk. The probed membranes were

washed three times with TBS-T, incubated with ECL rabbit IgG, HRP-linked whole Anti-body (GE Healthcare Life Sciences, #NA934) (1:2500 dilution), and again washed three times with TBS-T. The immunoblots were visualized by enhanced chemiluminescence with Immobilon™ Western Chemiluminescent HRP Substrate (Millipore, #P90718).

Cell growth inhibition assay. A549 cells were plated in 96-well plates at the initial density of 1 x 10^3 cells/well (50 μL/well) and incubated at 37°C. After 24 h, cells were exposed to test compounds by adding solutions (50 μL/well) of the compounds at various concentrations in medium at 37 °C under 5% CO_2 in air for 72 h. The mixtures were then treated with 10 μL of AlamarBlue® (AbD Serotec, #BUF012A), and incubation was continued at 37 °C for 3 h. The fluorescence in each well was measured with an ARVO™ X3 microplate reader (excitation at 540 nm, emission, at 590 nm). The percentage cell growth was calculated from the fluorescence readings.

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