Supplementary Material

Chemical synthesis of palmitoylated histone H4

Hironobu Hojo*\textsuperscript{a} and Isao Suetake*\textsuperscript{b}

\textsuperscript{a} Institute for Protein Research, Osaka University, Suita, Osaka 5650871, Japan
\textsuperscript{b} Nakamura Gakuen Graduate School University, Fukuoka 8140198, Japan

Email: hojo@protein.osaka-u.ac.jp, suetake@nakamura-u.ac.jp

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Trial synthetic routes of segment 2 .................................................................................................................................................. S2
Boc-[Arg(Ts)2]-NH-resin

1) TFA, 2) DIEA/DMF, 3) Boc-Gly-SCH2CH2COOH, DIC, Oxyma

Synthesis by the Boc method
[1] TFA, 2) Boc-AA-OH, HBTU, DIEA]

Boc-Gly-Leu-Ile-Tyr(BrZ)-Glu(Obn)-Glu(Obn)-Thr(Bn)-Arg(Ts)-Gly-
SCH2CH2CO-[Arg(Ts)]2-NH-resin

1) TFA, 2) Boc-Ser(Pal)-OH, DIC, Oxyma

Synthesis by the Boc method
[1] TFA, 2) Boc-AA-OH, HBTU, DIEA]

Boc-Ile-Thr(Bn)-Lys(Noc)-Pro-Ala-Ile-Arg(Ts)-Arg(Ts)-Leu-Ala-Arg(Ts)-
Arg(Ts)-Gly-Gly-Val-Lys(Noc)-Arg(Ts)-Ile-Ser(Pal)-Gly-Leu-Ile-Tyr(Bn) -
Glu(Obn)-Glu(Obn)-Thr(Bn)-Arg(Ts)-Gly-SCH2CH2CO-[Arg(Ts)]2-NH-resin

10% anisole/ HF

H-Ile-Thr-Lys(Noc)-Pro-Ala-Ile-Arg-Arg-Ala-Arg-Gly-Gly-Val-Lys(Noc) -
Arg-Ile-Ser-Gly-Leu-Ile-Tyr-Glu-Thr-Arg-Gly-SCH2CH2CO-Arg2-NH2

Fig. S1. Trial synthesis of segment 2 by the Boc method. Elution conditions: column, Cosmosil 5C18 AR-II (4.6 x 150 mm, Nacalai Tesque, Kyoto) at the flow rate of 1 mL min⁻¹; eluent, A, 0.1% TFA, B acetonitrile containing 0.1% TFA; temperature, 50 °C; gradient, 20% B at 0 min to 40% B at 20 min linearly.

Fmoc = Gly

Et

O

[Lys(Boc)]2-Rink amide MBHA resin (NH2-resin)

Microwave-assisted synthesis by Fmoc method
1) 10% piperazine, 0.1 M HOBr, 10% Eth/NMP, 2) Fmoc-AA, DIC, HOBr

Gly-Leu-Ile-Tyr(Bu')-Glu(Obu')-Glu(Obu')-Thr(Bu')-Arg(Pbf)-Gly-
[Lys(Boc)]2-NH-resin

TrtS

Fmoc-Ser(Pal)-OH, DIC, HOBr in DCE

Manual synthesis by Fmoc method

Lys(Noc)-Arg(Pbf)-Ile-Ser(Pal)-Gly-Leu-Ile-Tyr(Bu')-Glu(Obu')-Glu(Obu')-
Thr(Bu')-Arg(Pbf)-Gly

[Lys(Boc)]2-NH-resin

Microwave-assisted synthesis by Fmoc method
1) 10% piperazine, 0.1 M HOBr, 10% Eth/NMP (With or without microwave irradiation)
2) Fmoc-AA, DIC, HOBr

TFA cocktail (TFA-TIS-DODT-H2O, 93 : 2.5 : 2 : 2.5), r.t., 2 h

Pro-Ala-Ile-Arg-Leu-Ala-Arg-Arg-Gly-Gly-Val-Lys(Noc) -
Arg-Ile-Ser(Pal)-Gly-Leu-Ile-Tyr-Glu-Thr-Arg-Gly-
[Lys]2-NH2

Fig. S2. Trial synthesis of the partial sequence (32-56) of segment 2 by the Fmoc-method. The introduction of Ile⁵⁹ to Lys⁶¹ was abandoned. The HPLC profile a) shows the analysis of crude peptide obtained with microwave irradiation during Fmoc group removal for Pro⁶⁰ to Lys⁶⁴. Profile b) shows the analysis of crude peptide obtained without microwave irradiation during Fmoc group removal for Pro⁶² to Lys⁶⁴. Elution conditions: column, Cosmosil 5C18 AR-II (4.6 x 150 mm, Nacalai Tesque, Kyoto) at the flow rate of 1 mL min⁻¹; eluent, A, 0.1% TFA, B acetonitrile containing 0.1% TFA; temperature, 50 °C; gradient, 20% B at 0 min to 60% B at 20 min linearly.