Electronic Supplementary Information (ESI)

One-pot approach to functional nucleosides possessing fluorescent group using nucleobase-exchange reaction by thymidine phosphorylase

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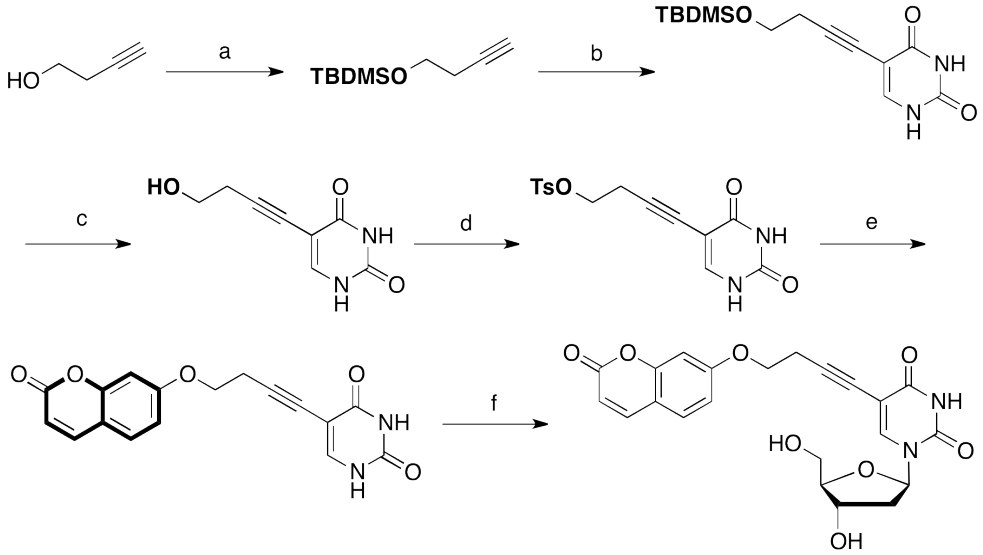
1. Syntheses of 5-modified uracils and unnatural nucleosides.

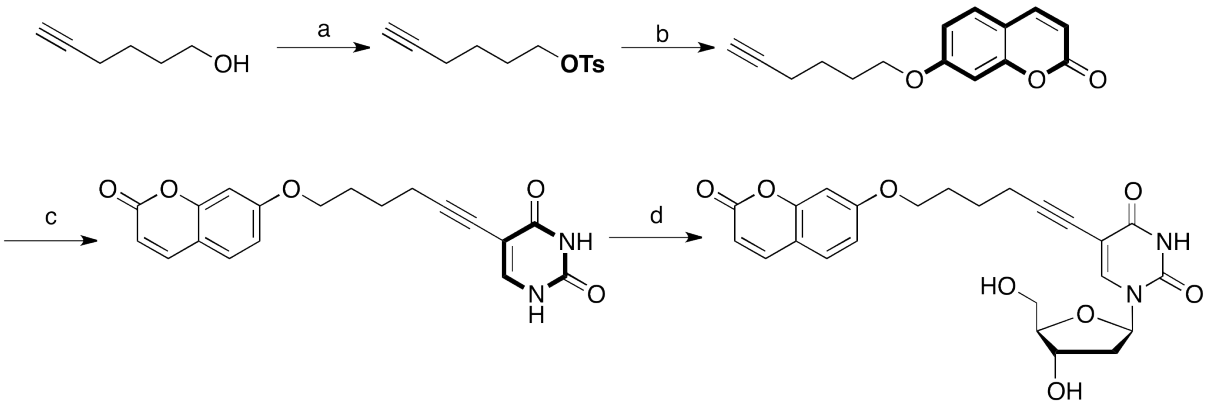
1-1. Syntheses of 2-deoxy-[5-(coumarin-7-oxybut-3-yn)] uridine (dRC2U).



Reagents and conditions. a: TBDMSCl, imidazol in DMF at rt (91 %). b: 5-Iodeuracil, $Pd(PPh_3)_4$, CuI, DIPEA in DMF at rt (91 %). c: TBAF in THF at rt (71 %). d: TsCl in pyridine at rt (88 %). e: Umberiferon, *t*-BuOK in DMF at 70 ˚C (20 %). f: Thymidine, 1 unit/mL of thymidine phosphorylase in 1 mM phosphate buffer at pH 6.8 at 37 ˚C (9.4 %).

5-(Coumarin-7-oxybut-3-yn) uracil. To a solution of 5-(1-tosyloxybut-3-yn)-uracil (88 mg, 0.35 mmol) in 2 mL of DMF was added umbeliferone (57 mg, 0.35 mmol), potassium *t*-butoxide (78 g, 0.70 mmol) at room temperature. The reaction mixture was stirred for 3 h at 70 ˚C. After removal of the DMF in vacuo, the residue was purified by silica gel chromatography eluting with hexane-AcOEt (1:1) to afford 5-(coumarin-7-oxybut-3-yn) uracil as a white powder (17.7 mg, 20 %). This compound was low solubility in many solvents, and then we used this crude compound to next enzymatic reaction without further purification. ^{1}H NMR (DMSO-d_6): δ 1.24 (1H, m), 2.84 (2H, *J* = 6.4 Hz, t), 4.16 (2H, *J* = 6.4 Hz, t), 6.25 (1H, *J* = 9.2 Hz, d), 6.96 (2H, *m*), 7.61 (2H, *J* = 8.8 Hz, d), 7.64 (1H, s), 7.95 (1H, *J* = 9.2 Hz, d), 11.3 (1H, s); ^{13}C NMR (DMSO-d_6): δ 20.3, 67.1, 74.7, 89.8, 97.7, 101.9, 113.1, 113.2, 113.3, 130.1, 144.9, 145.9, 151.0, 155.9, 160.9, 161.9, 163.4.

1-2. Syntheses of 2-deoxy-[5-(coumarin-7-oxyhex-5-yn)] uridine (dRC4U).



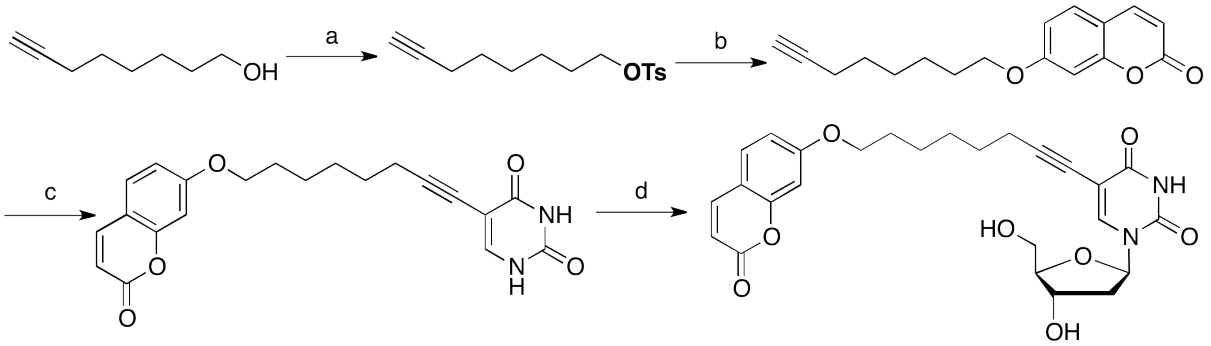
Reagents and conditions. a: TsCl in pyridine at rt (98 %). b: Umberiferon, *t*-BuOK in DMF at rt (99 %). c: 5-Iodeuracil, $Pd(PPh_3)_4$, CuI, DIPEA in DMF at rt (74 %). d: Thymidine, 1 unit/ml thymidine phosphorylase in 1 mM phosphate buffer at pH 6.8 at 37 ˚C (57 %).

Coumarin-7-oxyhex-5-yn. To a solution of *t*-BuOK (62 mg, 055 mmol) in 3 mL of DMF was added umbelliferone (77 mg, 0.48 mmol) and 1-toluensulfonyloxyhex-5-yn (100 mg, 0.40 mmol) at room temperature. The reaction mixture was stirred for 6 h at room temperature. After removal of the DMF in vacuo, the residue was taken up into water and extracted with EtOAc. The organic layer was washed with H_2O twice. The solution was filtered, concentrated, and purified by silica gel chromatography eluting with hexane-diethylether (2:1). The major product was recrystallized from MeOH to afford coumarin-7-oxyhex-5-yn as a pale yellow needles (90 mg, 94 %). ^{1}H NMR (DMSO-d_6): δ 1.74 (2H, *J* = 7.2, 7.4, 7.4, 7.7 Hz, dddd), 1.98 (3H, m), 2.30 (2H, *J* = 2.7, 7.0, 7.0 Hz, td), 4.05 (1H, *J* = 6.4, 6.4 Hz, t), 6.24 (1H, *J* = 9.6 Hz, d), 6.82 (2H, m), 7.36 (1H, *J* = 8.4 Hz, d), 7.63 (1H, *J*= 9.6 Hz, d); ^{13}C NMR (DMSO-d_6): δ 17.9, 25.0, 28.0, 68.3, 71.9, 84.8, 101.6, 112.8, 112.9, 113.3, 130.0, 144.9, 155.9, 161.0, 162.3, 162.4; FABMS *m/e* 243 $[M+H]^+$; Anal. Calcd for $C_{15}H_{14}O_3$: C, 74.36; H, 5.82; N, 0.00. Found: C, 72.87; H, 5.62; N, 0.00.

5-(Coumarin-7-oxyhex-5-yn) uracil (C4U). To a solution of 5-iodouracil (178 mg, 0.75 mmol) in 2 mL of DMF was added compound (242 mg, 1.00 mmol), diisopuropylethylamine (194 mg. 259 µL, 1.5 mmol), CuI (28.6 mg, 0.15 mmol) and $Pd(PPh_3)_4$ (81.7 mg, 0.075 mmol) at room temperature. The reaction mixture was stirred for 3 h at room temperature. After removal of the DMF in vacuo, the residue was purified by silica gel chromatography eluting with hexane-AcOEt (1:1) to afford 5-(coumarin-7-oxyhex-5-yn) uracil as a pale yellow powder (197 mg, 74 %). ^{1}H NMR (DMSO-d_6): δ 1.77 (2H, *J* = 7.2, 7.4, 7.4, 8.0 Hz, dddd), 1.98 (2H, *J* = 6.8, 7.2, 7.4, 8.0 Hz, dddd), 2.48 (2H, *J* = 7.0 Hz, t), 4.14 (1H, *J* = 6.4 Hz, t), 6.23 (1H, *J* = 9.2 Hz, d), 6.93 (2H, m), 7.52 (1H, *J* =

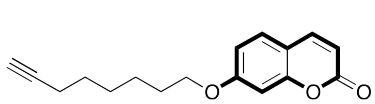
8.0 Hz, d), 7.87 (1H, *J*= 9.6 Hz, d); ^{13}C NMR ($CDCl_3$): δ 18.5, 24.7, 27.5, 40.3, 613, 67.9, 70.7, 85.5, 87.7, 93.3, 100.9, 111.9, 112.9, 129.1, 142.8, 144.4, 156.0, 162.0, 162.7; FABMS *m*/*e* 353 $[M+H]^+$; Anal. Calcd for $C_{19}H_{16}N_2O_5$: C, 64.77; H, 4.58; N, 7.95. Found: C, 63.91; H, 4.68; N, 7.55.

1-3. Syntheses of 2-deoxy-[5-(coumarin-7-oxyoct-7-yn)] uridine (dRC6U).

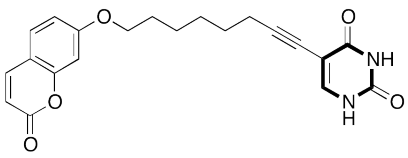


Reagents and conditions. a: TsCl in pyridine at rt (81 %). b: Umberiferon, *t*-BuOK in DMF at rt (38 %). c: 5-Iodeuracil, $Pd(PPh_3)_4$, CuI, DIPEA in DMF at rt (68 %). d: Thymidine, 1 unit/mL thymidine phosphorylase in 1 mM phosphate buffer at pH 6.8 at 37 ˚C (15 %).

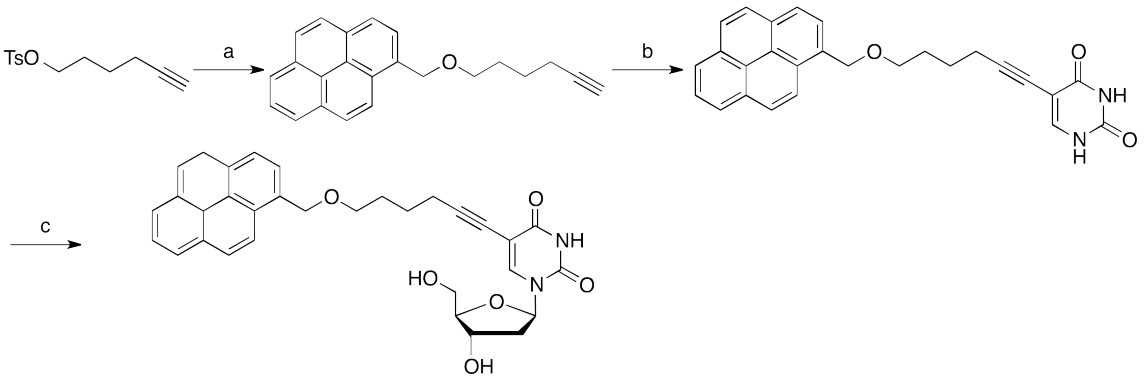
Coumarin-7-oxyoct-7-yn. ^{1}H NMR ($CDCl_3$): δ 1.48 (4H, m), 1.58 (2H, m), 1.82 (2H, *J* = 6.8 Hz, q), 1.94 (1H, *J* = 2.8 Hz, t), 2.20 (2H, *J* = 2.4, 6.8 Hz, td), 4.00 (2H, *J* = 6.8 Hz, t), 6.22 (1H, *J* = 8.8 Hz, d), 6.81 (2H, *m*) , 7.34 (1H, *J* = 8.8 Hz, d) , 7.77 (1H, *J* = 8.4 Hz, d); ^{13}C NMR ($CDCl_3$): δ 18.1, 24.9, 28.0, 67.9, 68.9, 83.8, 101.3, 112.5, 113.0, 113.1, 128.7, 143.5, 155.9, 161.3, 162.2; FABMS *m*/*e* 271 $[M+H]^+$; Anal. Calcd for $C_{17}H_{18}O_3$: C, 75.53; H, 6.71; N, 0.00. Found: C, 75.13; H, 6.68; N, 0.00.



5-(Coumarin-7-oxyoct-7-yn) uracil (C6U). ^{1}H NMR (DMSO-d_6): δ 1.24 (1H, m), 1.50 (6H, m), 1.74 (2H, *J* = 6.4 Hz, q), 2.37 (2H, *J* = 6.4 Hz, t), 4.08 (2H, *J* = 6.4 Hz, td), 4.00 (2H, *J* = 6.8 Hz, t), 6.27 (1H, *J* = 9.6 Hz, d), 6.95 (2H, *m*), 7.62 (2H, *J* = 9.2 Hz, d), 7.98 (1H, *J* = 9.2 Hz, d); ^{13}C NMR (DMSO-d_6): δ19.2, 25.4, 28.4, 28.6, 28.8, 68.8, 73.5, 93.4, 98.2, 101.7, 112.8, 112.9, 113.3, 130.0, 144.9, 145.0, 151.0, 156.0, 160.9, 162.4, 163.4; FABMS *m*/*e* 381 $[M+H]^+$; HRMS calcd for $C_{21}H_{20}N_2O_5$: 380.1372, found: 380.1369.

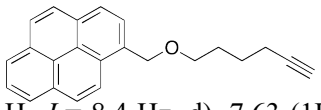


1-4. Syntheses of 2-deoxy-[5-(pyrene-1-methyloxyhex-5-yn)] uridine (dRP4U).

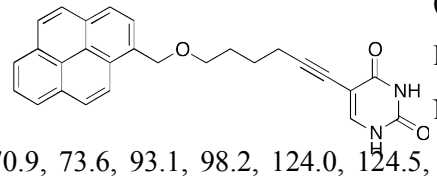


Reagents and conditions. a: 1-Pyrenemthanol, NaH in DMF at rt (80 %). b: 5-Iodeuracil, $Pd(PPh_3)_4$, CuI, DIPEA in DMF at rt (12 %). c: Thymidine, thymidine 1 unit/mL phosphorylase in 1 mM phosphate buffer at pH 6.8 at 37 ˚C (22 %).

Pyrene-1-methyloxyhex-5-yn. ^{1}H NMR ($CDCl_3$): δ 1.74 (2H, *J* = 6.8, 7.2, 7.2, 8.0 Hz, dddd), 1.98 (2H, *J* = 6.8, 7.2, 7.2, 8.0 Hz, dddd), 2.30 (2H, *J* = 2.7, 7.0, 7.0 Hz, td), 4.05 (1H, *J* = 6.4, 6.4 Hz, t), 6.24 (1H, *J* = 9.6 Hz, d), 6.82 (2H, m), 7.36 (1H, *J* = 8.4 Hz, d), 7.63 (1H, *J*= 9.6 Hz, d); ^{13}C NMR ($CDCl_3$): δ 18.3, 25.3, 28.9, 68.5, 69.9, 71.6, 84.4, 123.3, 124.5, 124.8, 125.0, 125.3, 126.0, 127.0, 127.4, 127.5, 127.7, 129.4, 130.9, 131.3, 131.8; Anal. Calcd for $C_{23}H_{22}O$: C, 87.86; H, 7.05; N, 0.00. Found: C, 88.43; H, 6.42; N, 0.00.

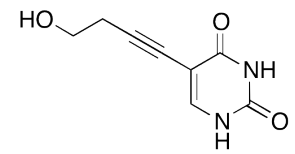


5-(Pyrene-1-methyloxyhex-5-yn) uracil. ^{1}H NMR (DMSO-d_6): δ 1.62 (2H, m), 1.78 (2H, m), 2.41 (2H, *J* = 7.0 Hz, t), 3.68 (2H, *J* = 6.2 Hz, t), d), 7.61 (1H, *J* = 6.0 Hz, d), 8.27 (9H, m), 11.1 (1H, *J*= 4.8 Hz, d), 11.2 (1H, s); ^{13}C NMR (DMSO-d_6): δ 18.6, 19.1, 25.2, 25.7, 29.0, 69.7, 70.0, 70.9, 73.6, 93.1, 98.2, 124.0, 124.5, 124.5, 125.1, 125.7, 125.8, 126.8, 127.5, 127.7, 127.9, 128.0, 129.2, 130.9, 131.1, 131.3, 132.7; FABMS *m*/*e* 422 $[M]^+$; HRMS (FAB) calcd for $C_{27}H_{22}N_2O_3$: 422.1630, found: 422.1628.

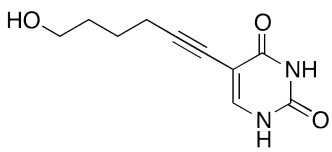


1-5. Syntheses of other compounds.

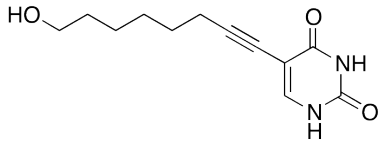
5-(1-Hydroxybut-3-yn) uracil. ^{1}H NMR (CD_3OH-d_3): δ 2.57 (2H, *J* = 6.4 Hz, t), 3.68 (2H, *J* = 6.6 Hz, t), 7.60 (1H, s); ^{13}C NMR (CD_3OH-d_3): δ 24.5, 61.6, 73.5, 92.2, 100.1, 145.9, 152.5, 165.9; Anal. Calcd for $C_8H_8N_2O_3$: C, 53.33; H, 4.48; N, 15.55. Found: C, 53.37; H, 4.53; N, 15.45.



5-(1-Hydroxyhex-5-yn) uracil. ^{1}H NMR (DMSO-d_6): δ 1.56 (2H, *J* = 3.4 Hz, quin), 2.40 (2H, *J* = 6.6 Hz, t), 3.46 (2H, m), 4.47 (1H, *J* = 5.2 Hz, t), 7.69 (1H, s), 11.2 (1H, br), 11.3 (1H, s); ^{13}C NMR (DMSO-d): δ 19.2, 25.4, 32.2, 60.7, 73.5, 93.3, 98.1, 145.1, 151.0, 163.4; FABMS *m*/*e* 209 $[M+H]^+$; Anal. Calcd for $C_{10}H_{12}N_2O_3$: C, 57.68; H, 5.81; N, 13.45. Found: C, 57.24; H, 5.89; N, 13.04.



5-(1-Hydroxyoct-7-yn) uracil. ^{1}H NMR (MeOH-d_3): δ 1.36-1.56 (8H, m), 2.34 (2H, *J* = 7.0 Hz, t), 3.52 (2H, *J* = 6.4 Hz, t), 7.52 (1H, s); ^{13}C NMR (MeOH-d_3): δ 18.7, 25.1, 28.4, 28.4, 32.2, 61.6, 71.1, 93.4, 99.1, 143.9, 151.2, 164.4; FABMS *m*/*e* 237 $[M+H]^+$; Anal. Calcd for $C_{12}H_{16}N_2O_3$: C, 61.00; H, 6.83; N, 11.86. Found: C, 60.68; H, 6.78; N, 11.67.



2. Analysis and isolation of unnatural nucleoside obtained by enzymatic reaction.

Condition of analysis

HPLC system was a GULLIVER model by JASCO. Each conversion was assayed with a C-18 column (150 - 4.6 mm, Imtakt UK-C18) HPLC at a flow rate of 0.5 mL/min. The mobile phase was 8 - 60 % acetonitrile in 10 mM phosphate buffer (pH 6.8). The UV detector was set at 275 to 320 nm and the column was operated at 40 ˚C.

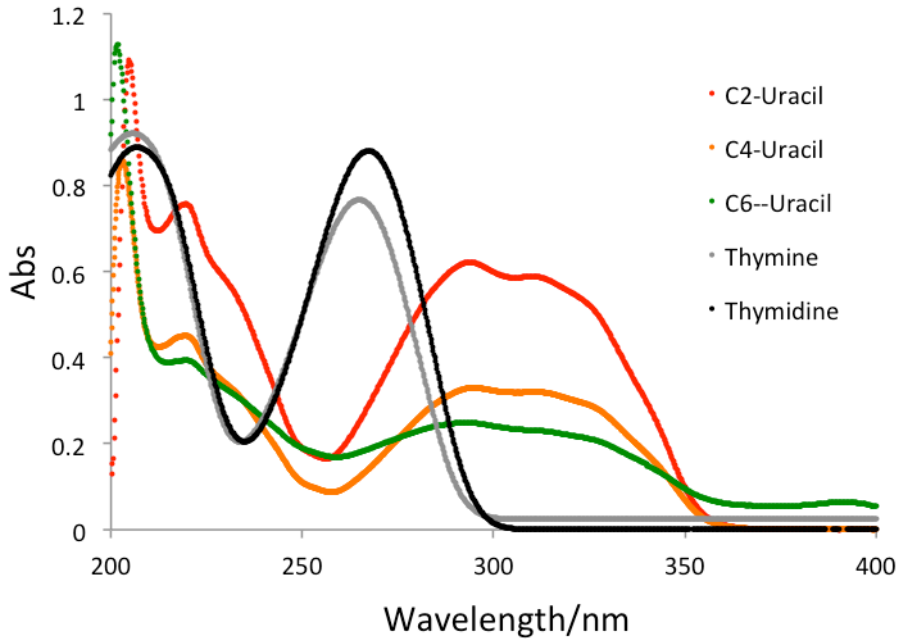
Condition of isolation

The unnatural nucleosides linking a fluorescent to 5 position of uracil were purified by a reversed phase HPLC. Isolations by HPLC were performed with a C-18 column (250 - 25 mm, Shiseido UG-120) at a flow rate of 3.0 mL/min. The mobile phase was 8 - 60 % acetonitrile in 10 mM phosphate buffer (pH 6.8). The UV detector was set at 275 to 320 nm and the column was operated at 40 ˚C.

ST 1 The HPLC conditions (the mobile phase and a UV) to detect each unnatural nucleoside.

Compound	Ratio of mobile phase (%, v/v)		UV (nm)
	MeCN	H_2O	
dRC2U	30	70	294
dRC4U	35	65	320
dRC6U	40	60	288
dRU2-OH	8	92	294
dRU4-OH	10	90	288
dRU6-OH	20	80	275
dRP4U	60	40	294

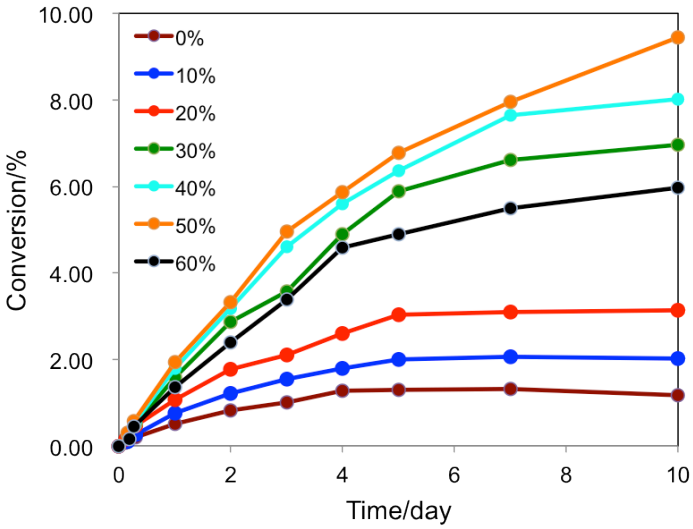
3. UV spectra for the modified uracils containing a fluorescent group.

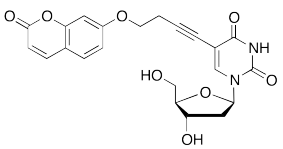


SF 1 UV spectra of modified uracil at 5 position

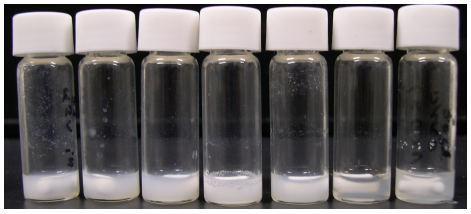
Compound	Concentration (μM)	λ□□□(nm)
5-(coumarin-7-oxybut-3-yn) uracil (C2U)	25	293.8
5-(coumarin-7-oxyhex-5-yn) uracil (C4U)	25	294.8
5-(Coumarin-7-oxyoct-7-yn) uracil, (C6U)	10	294.4
Thymine	1.25	264.8
Thymidine	1.25	267.4

4. Plot of the conversion of modified uracil such as C2U, C4U, C6U, P4U vs. time (day).

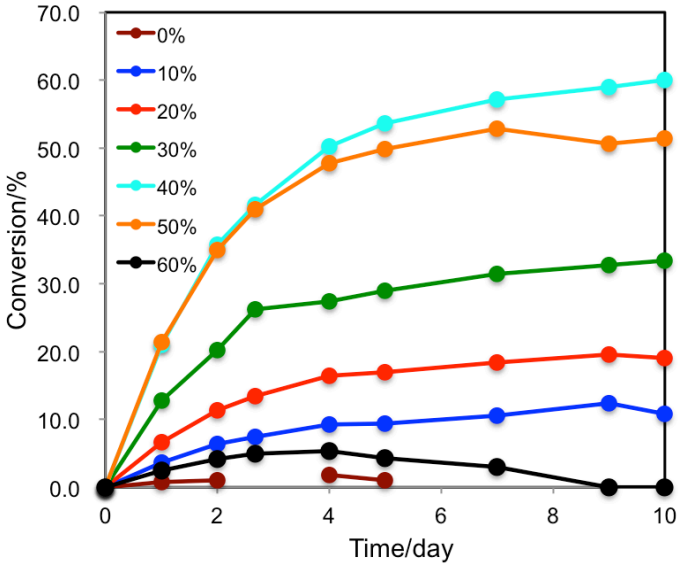


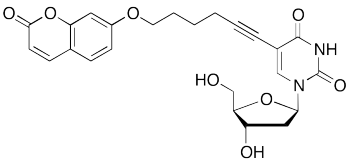


dRC2U

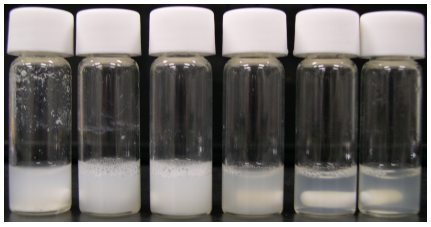


0 DMSO (%) 60

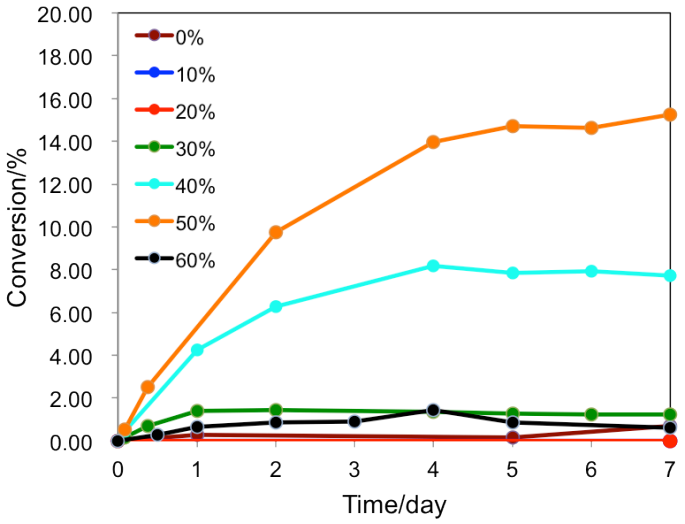


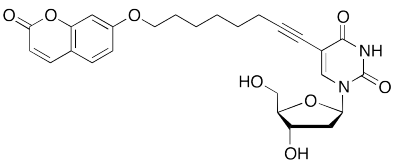


dRC4U

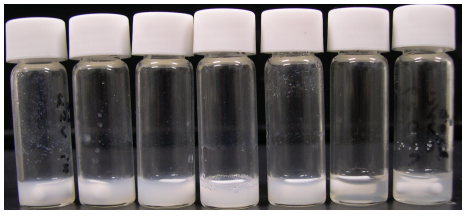


0 DMSO (%) 50

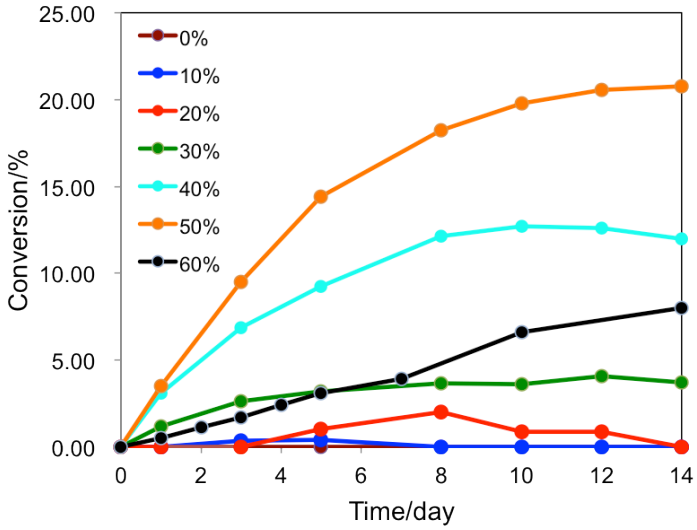


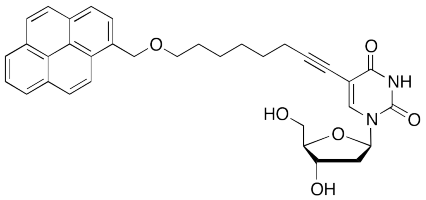


dRC6U

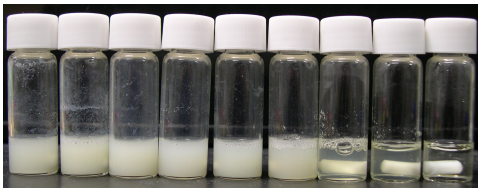


0 DMSO (%) 60





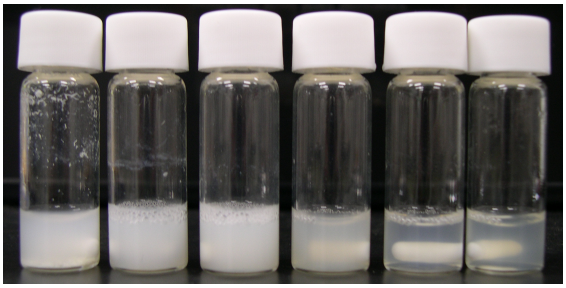
dRP4U



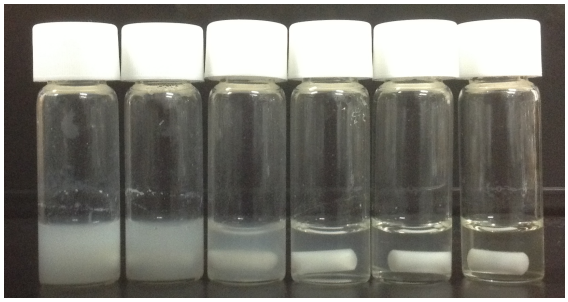
0 **DMSO (%)** **80**

SF 2 Plot of the conversion of modified uracil C2U, C4U, C6U, P4U vs. time (day). Thymidine phosphorylase could convert the unnatural nucleoside possessing fluorescent at 5 position from thymidine and modified uracil. Photo showed the solubility of modified uracil in buffer with additive of various concentration of DMSO. Reaction condition: 5 mM modified uracil, 50 mM thymidine, 1 units/mL thymidine phosphorylase in 1.0 mM phosphate buffer and various concentration of DMSO at 37 ˚C.

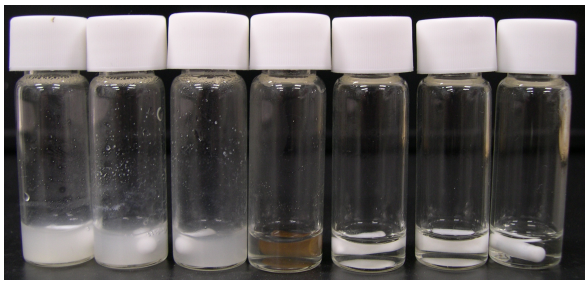
5. Solubility of C4U with mixture of buffer and each organic solvent (DMSO, DMF, THF).



DMSO



DMF



THF

SF 3 Solubility of C4U with mixture of buffer and each organic solvent. Each sample contained 5 mM modified uracil, 50 mM thymidine in 1.0 mM phosphate buffer and each organic solvent. The concentration of organic solvent showed 0, 10, 20, 30, 40, 50 % in phosphate buffer from left side.

6. Fluorescent of P4U and C4U.

P4U **C4U**

SF 4 The fluorescent of P4U and C4U were observed by illumination at 365 nm at room temperature (1.0 mM concentration each compound in MeOH).

7. Log file of docking simulation MF myPresto for Fig. 2.

7-1 Log file for docking simulation of TP and C4U.

```
**********************************************
* *
* sievgene(11/08/01) *
* *
* Biomedicinal Information Research Center *
* 2-3-26 Aomi, Koto-ku, Tokyo 135-0064 *
* JAPAN *
* *
* v4.204 : Aug 1, 2011. *
* *
**********************************************
RANDOM NUMBER METHOD : FORTRAN RANDOM LIBRARY
++++++++++++++++++++++++++++++++++++++++
+ +
+ INFORMATION> sievgene(11/08/01) +
+ READ INPUT SPECIFICATION +
+ +
++++++++++++++++++++++++++++++++++++++++
INFORMATION> INPUT
1) PROTEIN DATA
1-1) TOPOLOGY FILE
FORMAT :ascii
NAME :Pro.tpl
1-2) COORDINATE FILE
FORMAT :ascii
NAME :Pro_md.pdb
1-3) POCKET POINT FILE
FORMAT :ascii
NAME :point.pdb
2) LIGAND DATA
2-1) INPUT TYPE :FILE
FORMAT :MOL2
NAME :0_Q4U.mol2
```

2-2) REFERENCE COORDINATE FILE

FORMAT :no read

NAME :

3) ATOM DATA

3-1) ATOM DATA FILE

NAME :

4) ASA DATA

METHOD :pairwise

NAME :

5) LOG FORMAT :detail

++

+ +

+ INFORMATION> INPUT PROTEIN DATA +

+ +

++

INFORMATION> INPUT POCKET CORRDINATE FILE

FILE NAME :point.pdb

POINT NUMBER : 165

INFORMATION> INPUT PROTEIN DATA FILE

FILE NAME :Pro_md.pdb

ATOM NUMBER : 6640

INFORMATION> INPUT FORMATTED TOPOLOGY FILE

1) TOTAL NUMBER

NUMBER OF MOLECULES : 1

NUMBER OF CHAINS : 1

NUMBER OF RESIDUES : 440

NUMBER OF ATOMS : 6640

NUMBER OF BONDS : 6689

NUMBER OF ANGLES : 12118

NUMBER OF TORSIONS : 21217

NUMBER OF IMPRO. : 1266

MAXIMUM NUMBER OF FUNC. : 1

MAXIMUM NUMBER OF ATOM-TYPE : 40

2) EACH DATA

A) NUMBER OF CHAINS IN EACH MOLECULE

MOLECULE 1 : 1

B) NUMBER OF ATOMS IN EACH MOLECULE

MOLECULE 1 : 6640

C) NUMBER OF BONDS IN EACH MOLECULE

MOLECULE 1 : 6689

D) NUMBER OF ANGLES IN EACH MOLECULE

MOLECULE 1 : 12118

E) NUMBER OF TORSIONS IN EACH MOLECULE

MOLECULE 1 : 21217

F) NUMBER OF IMPROPER IN EACH MOLECULE

MOLECULE 1 : 1266

INFORMATION> PROTEIN SIDE CHAIN

SIDE CHAIN NUMBER : 26

++

+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ GRID SPECIFICATION +

+ +

++

INFORMATION> GRID

1) GRID-POTENTIAL FILE

INPUT FORM :binary

INPUT NAME :grid_file

OUTPUT FORM :no read

OUTPUT NAME :grid_file

2) RECEPTOR DISTANCE (A) : 5.000000

3) OFFSET OF VDW (A) : 0.6000000

4) OFFSET OF COULOMB (A) : 0.6000000

5) RADIUS OF MESH (A) : 1.400000

6) GRID SIZE MERGIN (A) : 5.000000

7) SMOOTHING ITERATION : 3

8) INTERPOLATION : lagrange

++

+ +

+ INFORMATION> GENERATE GRID POTENTIAL +

```
+ +
++++++++++++++++++++++++++++++++++++++++
INFORMATION> GRID SIZE
NUMBER OF GRID : 60
SAMPLING POINT NUMBER : 165
GRID BOUND :
X-AXIS: -12.51100 - 14.48500
Y-AXIS: -19.31200 - 9.298000
Z-AXIS: -26.25100 - -2.826000
1 PARTCLE SIZE:
0.4575593 0.4849153 0.3970339
INFORMATION> READ GRID POTENTIAL:
FILE NAME :grid_file
FORMAT :single
READ MARGIN : 5.000000
READ SMOOTHING : 3
READ VDW RADIUS : 0.6000000
READ ELE RADIUS : 0.6000000
READ MESH RADIUS : 1.400000
READ PROBE RADIUS : 5.000000
++++++++++++++++++++++++++++++++++++++++
+ +
+ INFORMATION> sievgene(11/08/01) +
+ READ CONF SPECIFICATION +
+ +
++++++++++++++++++++++++++++++++++++++++
INFORMATION> CONF
1) GENERATE CONFORMER
TRIAL LIMIT : 100000
CREATE NUMBER : 100
2) SORT ATOM SEQUENCE : T
3) DAMPING FACTOR : 1.000000
4) PHASE NUMBER : 6
5) ROTATE S-H, O-H TERM. : T
++++++++++++++++++++++++++++++++++++++++
```

+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ DOCK SPECIFICATION +

+ +

++

INFORMATION> DOCK

1) DOCKING

1-1) METHOD : flex

1-2) PROTEIN SURFACE : electrostatic

1-3) GENERATION : 4

1-4) PLATE NUMBER : 1000

1-5) MATCHING : 4

1-6) CANDIDATE ATOM DISTANCE (A)

LOWER : 2.500000 - 4.000000

UPPER : 4.500000 - 8.000000

1-7) DOCKING SPEED : NORMAL

1-8) DOCKING OR ENERGY ONLY : DOCKING

1-9) EVALUATE H-BOND : YES

1-10) ROTATE PROTEIN SIDE CHAIN : NO

1-11) ROTATE LIGAND -OH : NO

2) SCORE COEFFICIENT

2-1) VAN DER WAALS WEIGHT : 3.000000

2-2) DIELECTRIC WEIGHT : 3.000000

2-3) A.S.A WEIGHT : 1.500000

2-4) HYDROGEN-BOND WEIGHT : 1.000000

2-4) ANISOTROPIC H-BOND WEIGHT : 5.000000

2-5) INSIDE POCKET DISTANCE (A) : 0.000000

7-2 Log file for docking simulation of TP and C2U.

```
**********************************************
* *
* sievgene(11/08/01) *
* *
* Biomedicinal Information Research Center *
* 2-3-26 Aomi, Koto-ku, Tokyo 135-0064 *
* JAPAN *
* *
* v4.204 : Aug 1, 2011. *
* *
**********************************************
RANDOM NUMBER METHOD : FORTRAN RANDOM LIBRARY
++++++++++++++++++++++++++++++++++++++++++
+ +
+ INFORMATION> sievgene(11/08/01) +
+ READ INPUT SPECIFICATION +
+ +
++++++++++++++++++++++++++++++++++++++++++
INFORMATION> INPUT
1) PROTEIN DATA
1-1) TOPOLOGY FILE
FORMAT :ascii
NAME :Pro.tpl
1-2) COORDINATE FILE
FORMAT :ascii
NAME :Pro_md.pdb
1-3) POCKET POINT FILE
FORMAT :ascii
NAME :point.pdb
2) LIGAND DATA
2-1) INPUT TYPE :FILE
FORMAT :MOL2
NAME :0_Q2U.mol2
2-2) REFERENCE COORDINATE FILE
```

FORMAT :no read

NAME :

3) ATOM DATA

3-1) ATOM DATA FILE

NAME :

4) ASA DATA

METHOD :pairwise

NAME :

5) LOG FORMAT :detail

++

+ +

+ INFORMATION> INPUT PROTEIN DATA +

+ +

++

INFORMATION> INPUT POCKET CORRDINATE FILE

FILE NAME :point.pdb

POINT NUMBER : 61

INFORMATION> INPUT PROTEIN DATA FILE

FILE NAME :Pro_md.pdb

ATOM NUMBER : 6640

INFORMATION> INPUT FORMATTED TOPOLOGY FILE

1) TOTAL NUMBER

NUMBER OF MOLECULES : 1

NUMBER OF CHAINS : 1

NUMBER OF RESIDUES : 440

NUMBER OF ATOMS : 6640

NUMBER OF BONDS : 6689

NUMBER OF ANGLES : 12118

NUMBER OF TORSIONS : 21217

NUMBER OF IMPRO. : 1266

MAXIMUM NUMBER OF FUNC. : 1

MAXIMUM NUMBER OF ATOM-TYPE : 40

2) EACH DATA

A) NUMBER OF CHAINS IN EACH MOLECULE

MOLECULE 1 : 1

B) NUMBER OF ATOMS IN EACH MOLECULE

MOLECULE 1 : 6640

C) NUMBER OF BONDS IN EACH MOLECULE

MOLECULE 1 : 6689

D) NUMBER OF ANGLES IN EACH MOLECULE

MOLECULE 1 : 12118

E) NUMBER OF TORSIONS IN EACH MOLECULE

MOLECULE 1 : 21217

F) NUMBER OF IMPROPER IN EACH MOLECULE

MOLECULE 1 : 1266

INFORMATION> PROTEIN SIDE CHAIN

SIDE CHAIN NUMBER : 15

++

+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ GRID SPECIFICATION +

+ +

++

INFORMATION> GRID

1) GRID-POTENTIAL FILE

INPUT FORM :binary

INPUT NAME :grid_file

OUTPUT FORM :no read

OUTPUT NAME :grid_file

2) RECEPTOR DISTANCE (A) : 5.500000

3) OFFSET OF VDW (A) : 0.6000000

4) OFFSET OF COULOMB (A) : 0.6000000

5) RADIUS OF MESH (A) : 1.400000

6) GRID SIZE MERGIN (A) : 5.000000

7) SMOOTHING ITERATION : 3

8) INTERPOLATION : lagrange

++

+ +

+ INFORMATION> GENERATE GRID POTENTIAL +

+ +

```
++++++++++++++++++++++++++++++++++++++++
INFORMATION> GRID SIZE
NUMBER OF GRID : 60
SAMPLING POINT NUMBER : 61
GRID BOUND :
X-AXIS: -6.519000 - 15.49600
Y-AXIS: -15.71700 - 6.500000
Z-AXIS: -24.17700 - -3.012000
1 PARTCLE SIZE:
0.3731356 0.3765593 0.3587288
INFORMATION> READ GRID POTENTIAL:
FILE NAME :grid_file
FORMAT :single
READ MARGIN : 5.000000
READ SMOOTHING : 3
READ VDW RADIUS : 0.6000000
READ ELE RADIUS : 0.6000000
READ MESH RADIUS : 1.400000
READ PROBE RADIUS : 5.500000
++++++++++++++++++++++++++++++++++++++++
+ +
+ INFORMATION> sievgene(11/08/01) +
+ READ CONF SPECIFICATION +
+ +
++++++++++++++++++++++++++++++++++++++++
INFORMATION> CONF
1) GENERATE CONFORMER
TRIAL LIMIT : 100000
CREATE NUMBER : 100
2) SORT ATOM SEQUENCE : T
3) DAMPING FACTOR : 0.7000000
4) PHASE NUMBER : 3
5) ROTATE S-H, O-H TERM. : T
++++++++++++++++++++++++++++++++++++++++
+ +
```

+ INFORMATION> sievgene(11/08/01) +

+ READ DOCK SPECIFICATION +

+ +

++

INFORMATION> DOCK

1) DOCKING

1-1) METHOD : flex

1-2) PROTEIN SURFACE : electrostatic

1-3) GENERATION : 1

1-4) PLATE NUMBER : 1000

1-5) MATCHING : 0

1-6) CANDIDATE ATOM DISTANCE (A)

LOWER : 1.000000 - 1.200000

UPPER : 8.000000 - 12.00000

1-7) DOCKING SPEED : NORMAL

1-8) DOCKING OR ENERGY ONLY : DOCKING

1-9) EVALUATE H-BOND : NO

2) SCORE COEFFICIENT

2-1) VAN DER WAALS WEIGHT : 1.000000

2-2) DIELECTRIC WEIGHT : 1.000000

2-3) A.S.A WEIGHT : 1.000000

2-4) HYDROGEN-BOND WEIGHT : 1.000000

2-5) INSIDE POCKET DISTANCE (A) : 6.000000

++

+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ MINI SPECIFICATION +

+ +

++

PARAMETERS FOR MINIMIZATION

1) GENERAL PARAMETERS

METHOD : steep

CPU LIMIT (S) : 360000.000000000

LOOP LIMIT : 100

UPDATE : 20

MONITORING LOG : 100

CALC. OF RMSD : true

CONVERGE (RMSF) : 0.100000000000000

INITIAL STEP(A) : 1.000000000000000E-002

UP RATE : 1.00000000000000

DOWN RATE : 0.300000000000000

2) ANALYSIS FILE

LOGICAL UNIT : 30

CLOSE : F

3) PARAMETERS FOR ENERGY CALCULATION

POTENTIAL FUNC. : AMBER

CUT-OFF LOGIC : Residue-surface

CUT-OFF DIST. : 22.0000000000000

DIELECTRIC IS DEPEND DISTANCE: true

DIELECTRIC VAL. : 4.00000000000000

CALCULATE BOND

CALCULATE ANGLE

CALCULATE TORS.

CALCULATE IMPRO.

CALCULATE VDW14

CALCULATE ELE14

CALCULATE VDW15

CALCULATE ELE15

CALCULATE HYD.

ENERGY IS NOT DIVIDED. NCLUSTer= 1

++

+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ OUTPUT SPECIFICATION +

+ +

++

INFORMATION> OUTPUT

1) LIGAND COORDINATE FILE

FORMAT :MOL2

NUMBER : 10

NAME :0C_Q2U.mol2

2) SCORE FILE

NUMBER : 10

NAME :0S_Q2U.mol2

INFORMATION> GENERATE PROTEIN POCKET

H-DONAR/H-ACCEPTOR NUMBER : 139

MESH NUMBER : 108

ATOM ID= 754 NAME= C TYPE= 5

ATOM ID= 754 NAME= C TYPE= 5

ATOM ID= 756 NAME= C TYPE= 5

ATOM ID= 756 NAME= C TYPE= 5

ATOM ID= 756 NAME= C TYPE= 5

ATOM ID= 757 NAME= H TYPE= 3

ATOM ID= 758 NAME= C TYPE= 5

ATOM ID= 758 NAME= C TYPE= 5

ATOM ID= 1379 NAME= C TYPE= 5

ATOM ID= 1389 NAME= C TYPE= 5

ATOM ID= 1390 NAME= O TYPE= 4

ATOM ID= 1390 NAME= O TYPE= 4

ATOM ID= 1390 NAME= O TYPE= 4

ATOM ID= 1393 NAME= C TYPE= 5

ATOM ID= 1395 NAME= H TYPE= 3

ATOM ID= 1778 NAME= C TYPE= 5

ATOM ID= 1779 NAME= H TYPE= 3

ATOM ID= 1780 NAME= H TYPE= 3

ATOM ID= 1781 NAME= C TYPE= 5

ATOM ID= 1783 NAME= C TYPE= 5

ATOM ID= 1783 NAME= C TYPE= 5

ATOM ID= 1783 NAME= C TYPE= 5

ATOM ID= 1785 NAME= H TYPE= 3

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1793 NAME= N TYPE= 4

ATOM ID= 1797 NAME= H TYPE= 3

ATOM ID= 1808 NAME= N TYPE= 4

ATOM ID= 1808 NAME= N TYPE= 4

ATOM ID= 1810 NAME= C TYPE= 5

ATOM ID= 1827 NAME= O TYPE= 4

ATOM ID= 1827 NAME= O TYPE= 4

ATOM ID= 2560 NAME= C TYPE= 5

ATOM ID= 2561 NAME= C TYPE= 5

ATOM ID= 2563 NAME= C TYPE= 5

ATOM ID= 2566 NAME= O TYPE= 4

ATOM ID= 2570 NAME= C TYPE= 5

ATOM ID= 2571 NAME= H TYPE= 3

ATOM ID= 2630 NAME= O TYPE= 4

ATOM ID= 2630 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2633 NAME= O TYPE= 4

ATOM ID= 2633 NAME= O TYPE= 4

ATOM ID= 2633 NAME= O TYPE= 4

ATOM ID= 2637 NAME= H TYPE= 3

ATOM ID= 2667 NAME= N TYPE= 4

ATOM ID= 2669 NAME= C TYPE= 5

ATOM ID= 2675 NAME= C TYPE= 5

ATOM ID= 2676 NAME= O TYPE= 4

ATOM ID= 2676 NAME= O TYPE= 4

ATOM ID= 2691 NAME= N TYPE= 4

ATOM ID= 2699 NAME= H TYPE= 3

ATOM ID= 2699 NAME= H TYPE= 3

ATOM ID= 2701 NAME= C TYPE= 5

ATOM ID= 2701 NAME= C TYPE= 5

ATOM ID= 2703 NAME= H TYPE= 3

ATOM ID= 2711 NAME= C TYPE= 5

ATOM ID= 2713 NAME= H TYPE= 3

ATOM ID= 2714 NAME= C TYPE= 5

ATOM ID= 2715 NAME= O TYPE= 4

ATOM ID= 2716 NAME= O TYPE= 4

ATOM ID= 2716 NAME= O TYPE= 4

ATOM ID= 2716 NAME= O TYPE= 4

ATOM ID= 2718 NAME= O TYPE= 4

ATOM ID= 2718 NAME= O TYPE= 4

ATOM ID= 2722 NAME= H TYPE= 3

ATOM ID= 2829 NAME= C TYPE= 5

ATOM ID= 2830 NAME= H TYPE= 3

ATOM ID= 2846 NAME= C TYPE= 5

ATOM ID= 2897 NAME= C TYPE= 5

ATOM ID= 2897 NAME= C TYPE= 5

ATOM ID= 2898 NAME= H TYPE= 3

ATOM ID= 2898 NAME= H TYPE= 3

ATOM ID= 2900 NAME= N TYPE= 4

ATOM ID= 2900 NAME= N TYPE= 4

ATOM ID= 2900 NAME= N TYPE= 4

ATOM ID= 3172 NAME= O TYPE= 4

ATOM ID= 3172 NAME= O TYPE= 4

ATOM ID= 3178 NAME= H TYPE= 3

ATOM ID= 3179 NAME= H TYPE= 3

ATOM ID= 3180 NAME= C TYPE= 5

ATOM ID= 3183 NAME= C TYPE= 5

ATOM ID= 3183 NAME= C TYPE= 5

ATOM ID= 3192 NAME= O TYPE= 4

ATOM ID= 3192 NAME= O TYPE= 4

ATOM ID= 3214 NAME= C TYPE= 5

ATOM ID= 3214 NAME= C TYPE= 5

ATOM ID= 3214 NAME= C TYPE= 5

ATOM ID= 3215 NAME= H TYPE= 3

ATOM ID= 3216 NAME= H TYPE= 3

ATOM ID= 3216 NAME= H TYPE= 3

ATOM ID= 3220 NAME= C TYPE= 5

ATOM ID= 3221 NAME= H TYPE= 3

ATOM ID= 3221 NAME= H TYPE= 3

ATOM ID= 3289 NAME= H TYPE= 3

ATOM ID= 3289 NAME= H TYPE= 3

ATOM ID= 5384 NAME= H TYPE= 3

ATOM ID= 5384 NAME= H TYPE= 3

ATOM ID= 5386 NAME= N TYPE= 4

ATOM ID= 5386 NAME= N TYPE= 4

ATOM ID= 5386 NAME= N TYPE= 4

TOTAL POCKET TRIANGLE: 17730

++

+ +

+ INFORMATION> SCREENING START +

+ +

++

INFORMATION> POCKET CENTER COORDINATE

X-AXIS : 4.492983

Y-AXIS : -3.696033

Z-AXIS : -13.93182

INFORMATION> SCREENING NUMBER : 1

INFORMATION> REFERENCE COODINATE NOT READ

(USE LIGAND COORDINATE)

INFORMATION> INPUT LIGAND FILE

FILE NAME :0_Q2U.mol2

ATOM NUMBER : 36

BOND NUMBER : 38

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATION> GENERATE CONFOMER

PHASE NUMBER : 3.000000

ROTATIVE TORSION NUMBER : 2

TRIAL ROTRATION : 10000000

TRIAL EXECUTION : 9

CONFOMER NUMBER : 100

INFORMATION> LIGAND ACCEPTOR, DONAR

DONAR NUMBER: 2

25 H

26 H

ACCEPTOR NUMBER: 5

7 O

8 O

13 O

23 O

24 O

INFORMATION> GLOBAL SEARCH START

INFORMATION> CONFORMER NUMBER = 10 GENERATION = 1

ID EDGE-LOW EDGE-UPR PLATE NUM TRIAL NUMBER

1 1.199 8.016 184296 475

2 1.199 8.016 300270 651

3 1.199 8.016 296232 593

4 1.199 8.016 198066 787

5 1.199 8.016 299184 1111

6 1.199 8.016 288834 652

7 1.199 8.016 184296 574

8 1.199 8.016 313716 288

9 1.199 8.016 341826 1160

10 1.199 8.016 192504 293

INFORMATINO> CREATE NEXT CONFORMER

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATION> GENERATE CONFOMER

PHASE NUMBER : 3.000000

ROTATIVE TORSION NUMBER : 2

TRIAL ROTRATION : 10000000

TRIAL EXECUTION : 9

CONFOMER NUMBER : 10

INFORMATINO> NEW CONFOMER NUMBER = 8

INFORMATION> BEFORE MINIMIZE RANKING :EXPERIMENT ID=1

INFORMATION> LOCAL SCORE RANKING 10

SCORE(/100) dG HIT MTS rotNum ASA ELE HYD VDW SURFACE RMSD

1 -2.64 -6.78 -71.55 -248.54 2 -249.79 0.00 -13.81 0.00 0.00 17.45

2 -2.63 -6.88 -69.06 -250.79 2 -240.62 0.00 -22.02 0.00 0.00 17.41

3 -2.60 -6.80 -68.22 -247.82 2 -237.70 0.00 -21.81 0.00 0.00 15.68

4 -2.55 -6.66 -67.31 -243.14 2 -234.61 0.00 -20.41 0.00 0.00 15.87

5 -2.53 -6.61 -66.34 -241.34 2 -231.14 0.00 -21.49 0.00 0.00 15.89

6 -2.50 -6.55 -65.80 -239.06 2 -229.31 0.00 -21.04 0.00 0.00 16.08

7 -2.49 -6.42 -66.96 -235.45 2 -233.70 0.00 -15.17 0.00 0.00 17.48

8 -2.44 -6.45 -62.58 -234.98 2 -217.74 0.00 -26.10 0.00 0.00 15.21

9 -2.42 -6.29 -63.85 -230.16 2 -222.60 0.00 -18.97 0.00 0.00 14.36

10 -2.40 -6.28 -62.83 -229.48 2 -218.93 0.00 -21.05 0.00 0.00 15.07

REFERENCE COORDINATE DATA

999.90 999.90 0.00 0.00

INFORMATION> CREATED LIGAND TOPOLOGY

ATOM NUMBER : 36

BOND NUMBER : 38

ANGLE NUMBER : 61

TORSION NUMBER : 84

IMPROPER NUMBER : 15

INFORMATION> LOCAL SEARCH START

10 %

20 %

30 %

40 %

50 %

60 %

70 %

80 %

90 %

100 %

INFORMATION> LOCAL SEARCH FINISHED

INFORMATION> AFTER MINIMIZE RANKING

INFORMATION> AFTER MINIMIZE RANKING :EXPERIMENT ID=1

INFORMATION> LOCAL SCORE RANKING 10

COMPOUND NAME : Q2U.mol2

FILE NAME : 0_Q2U.mol2

SCORE(/100) dG HIT MTS rotNum ASA ELE HYD VDW SURFACE RMSD

@ 1 -2.86 -7.27 -77.87 -267.79 2 -270.04 0.00 -12.82 -3.22 66.52 17.37

@ 2 -2.86 -7.23 -78.23 -266.84 2 -271.40 0.00 -11.13 -3.21 61.22 17.10

@ 3 -2.81 -7.11 -76.90 -262.29 2 -266.78 0.00 -10.92 -3.21 60.98 17.18

@ 4 -2.76 -7.17 -72.29 -262.65 2 -250.05 0.00 -23.11 -3.04 62.78 17.43

@ 5 -2.72 -6.92 -73.62 -255.44 2 -255.11 0.00 -13.90 -3.28 66.90 17.45

@ 6 -2.71 -6.98 -71.48 -256.53 2 -247.30 0.00 -20.32 -3.16 64.12 16.33

@ 7 -2.71 -7.08 -69.64 -258.91 2 -240.54 0.00 -26.97 -3.05 64.34 15.21

@ 8 -2.70 -7.00 -70.74 -256.87 2 -244.68 0.00 -22.49 -3.03 59.70 15.87

@ 9 -2.70 -6.98 -70.92 -256.26 2 -245.38 0.00 -21.53 -3.03 58.81 15.77

@ 10 -2.69 -6.98 -70.49 -256.09 2 -243.81 0.00 -22.51 -3.03 58.95 15.68

REFERENCE COORDINATE DATA

999.90 999.90 0.00 0.00

INFORMATION> ALL EXPERIMENT FINISHED

INFORMATION> COMPOUND RANKING 1

COMPOUND NAME SCORE(/100) dG HIT MTS

1 Q2U.mol2 -2.86 -7.27 -77.87 -267.79

TOTAL EXPERIMENT NUMBER : 1

TOTAL TRIAL NUMBER : 6584

TOTAL CPU TIME(S) : 9.474840

INPUT : 1.543103

GRID : 9.7509623E-03

CONF : 7.7228546E-03

DOCK : 0.2884932

MIN : 7.417803

```
**********************************************
* *
* sievgene(11/08/01) *
* *
* Biomedicinal Information Research Center *
* 2-3-26 Aomi, Koto-ku, Tokyo 135-0064 *
* JAPAN *
* *
* v4.204 : Aug 1, 2011. *
* *
**********************************************
RANDOM NUMBER METHOD : FORTRAN RANDOM LIBRARY
++++++++++++++++++++++++++++++++++++++++++
+ +
+ INFORMATION> sievgene(11/08/01) +
+ READ INPUT SPECIFICATION +
+ +
++++++++++++++++++++++++++++++++++++++++++
INFORMATION> INPUT
1) PROTEIN DATA
1-1) TOPOLOGY FILE
FORMAT :ascii
NAME :Pro.tpl
1-2) COORDINATE FILE
FORMAT :ascii
NAME :Pro_md.pdb
1-3) POCKET POINT FILE
FORMAT :ascii
NAME :point.pdb
2) LIGAND DATA
2-1) INPUT TYPE :FILE
FORMAT :MOL2
NAME :0_Q2U.mol2
2-2) REFERENCE COORDINATE FILE
FORMAT :no read
```

NAME :

3) ATOM DATA

3-1) ATOM DATA FILE

NAME :

4) ASA DATA

METHOD :pairwise

NAME :

5) LOG FORMAT :detail

++

+ +

+ INFORMATION> INPUT PROTEIN DATA +

+ +

++

INFORMATION> INPUT POCKET CORRDINATE FILE

FILE NAME :point.pdb

POINT NUMBER : 61

INFORMATION> INPUT PROTEIN DATA FILE

FILE NAME :Pro_md.pdb

ATOM NUMBER : 6640

INFORMATION> INPUT FORMATTED TOPOLOGY FILE

1) TOTAL NUMBER

NUMBER OF MOLECULES : 1

NUMBER OF CHAINS : 1

NUMBER OF RESIDUES : 440

NUMBER OF ATOMS : 6640

NUMBER OF BONDS : 6689

NUMBER OF ANGLES : 12118

NUMBER OF TORSIONS : 21217

NUMBER OF IMPRO. : 1266

MAXIMUM NUMBER OF FUNC. : 1

MAXIMUM NUMBER OF ATOM-TYPE : 40

2) EACH DATA

A) NUMBER OF CHAINS IN EACH MOLECULE

MOLECULE 1 : 1

B) NUMBER OF ATOMS IN EACH MOLECULE

MOLECULE 1 : 6640

C) NUMBER OF BONDS IN EACH MOLECULE

MOLECULE 1 : 6689

D) NUMBER OF ANGLES IN EACH MOLECULE

MOLECULE 1 : 12118

E) NUMBER OF TORSIONS IN EACH MOLECULE

MOLECULE 1 : 21217

F) NUMBER OF IMPROPER IN EACH MOLECULE

MOLECULE 1 : 1266

INFORMATION> PROTEIN SIDE CHAIN

SIDE CHAIN NUMBER : 15

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+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ GRID SPECIFICATION +

+ +

++

INFORMATION> GRID

1) GRID-POTENTIAL FILE

INPUT FORM :binary

INPUT NAME :grid_file

OUTPUT FORM :no read

OUTPUT NAME :grid_file

2) RECEPTOR DISTANCE (A) : 5.500000

3) OFFSET OF VDW (A) : 0.6000000

4) OFFSET OF COULOMB (A) : 0.6000000

5) RADIUS OF MESH (A) : 1.400000

6) GRID SIZE MERGIN (A) : 5.000000

7) SMOOTHING ITERATION : 3

8) INTERPOLATION : lagrange

++

+ +

+ INFORMATION> GENERATE GRID POTENTIAL +

+ +

++

INFORMATION> GRID SIZE

NUMBER OF GRID : 60

SAMPLING POINT NUMBER : 61

GRID BOUND :

X-AXIS: -6.519000 - 15.49600

Y-AXIS: -15.71700 - 6.500000

Z-AXIS: -24.17700 - -3.012000

1 PARTCLE SIZE:

0.3731356 0.3765593 0.3587288

INFORMATION> READ GRID POTENTIAL:

FILE NAME :grid_file

FORMAT :single

READ MARGIN : 5.000000

READ SMOOTHING : 3

READ VDW RADIUS : 0.6000000

READ ELE RADIUS : 0.6000000

READ MESH RADIUS : 1.400000

READ PROBE RADIUS : 5.500000

++

+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ CONF SPECIFICATION +

+ +

++

INFORMATION> CONF

1) GENERATE CONFORMER

TRIAL LIMIT : 100000

CREATE NUMBER : 100

2) SORT ATOM SEQUENCE : T

3) DAMPING FACTOR : 1.000000

4) PHASE NUMBER : 6

5) ROTATE S-H, O-H TERM. : T

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+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ DOCK SPECIFICATION +

+ +

++

INFORMATION> DOCK

1) DOCKING

1-1) METHOD : rigid

1-2) PROTEIN SURFACE : electrostatic

1-3) GENERATION : 6

1-4) PLATE NUMBER : 1000

1-5) MATCHING : 4

1-6) CANDIDATE ATOM DISTANCE (A)

LOWER : 2.500000 - 4.000000

UPPER : 5.000000 - 10.00000

1-7) DOCKING SPEED : NORMAL

1-8) DOCKING OR ENERGY ONLY : DOCKING

1-9) EVALUATE H-BOND : YES

1-10) ROTATE PROTEIN SIDE CHAIN : NO

1-11) ROTATE LIGAND -OH : NO

2) SCORE COEFFICIENT

2-1) VAN DER WAALS WEIGHT : 3.000000

2-2) DIELECTRIC WEIGHT : 4.000000

2-3) A.S.A WEIGHT : 2.000000

2-4) HYDROGEN-BOND WEIGHT : 3.000000

2-4) ANISOTROPIC H-BOND WEIGHT : 4.000000

2-5) INSIDE POCKET DISTANCE (A) : 0.000000

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+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ MINI SPECIFICATION +

+ +

++

PARAMETERS FOR MINIMIZATION

1) GENERAL PARAMETERS

METHOD : steep

CPU LIMIT (S) : 360000.000000000

LOOP LIMIT : 100

UPDATE : 20

MONITORING LOG : 100

CALC. OF RMSD : true

CONVERGE (RMSF) : 0.100000000000000

INITIAL STEP(A) : 1.000000000000000E-002

UP RATE : 1.00000000000000

DOWN RATE : 0.300000000000000

2) ANALYSIS FILE

LOGICAL UNIT : 30

CLOSE : F

3) PARAMETERS FOR ENERGY CALCULATION

POTENTIAL FUNC. : AMBER

CUT-OFF LOGIC : Residue-surface

CUT-OFF DIST. : 22.0000000000000

DIELECTRIC IS DEPEND DISTANCE: true

DIELECTRIC VAL. : 4.00000000000000

CALCULATE BOND

CALCULATE ANGLE

CALCULATE TORS.

CALCULATE IMPRO.

CALCULATE VDW14

CALCULATE ELE14

CALCULATE VDW15

CALCULATE ELE15

CALCULATE HYD.

ENERGY IS NOT DIVIDED. NCLUSTer= 1

++

+ +

+ INFORMATION> sievgene(11/08/01) +

+ READ OUTPUT SPECIFICATION +

+ +

++

INFORMATION> OUTPUT

1) LIGAND COORDINATE FILE

FORMAT :MOL2

NUMBER : 10

NAME :0C_Q2U.mol2

2) SCORE FILE

NUMBER : 10

NAME :0S_Q2U.mol2

INFORMATION> GENERATE PROTEIN POCKET

H-DONAR/H-ACCEPTOR NUMBER : 139

MESH NUMBER : 108

ATOM ID= 754 NAME= C TYPE= 5

ATOM ID= 754 NAME= C TYPE= 5

ATOM ID= 756 NAME= C TYPE= 5

ATOM ID= 756 NAME= C TYPE= 5

ATOM ID= 756 NAME= C TYPE= 5

ATOM ID= 757 NAME= H TYPE= 3

ATOM ID= 758 NAME= C TYPE= 5

ATOM ID= 758 NAME= C TYPE= 5

ATOM ID= 1379 NAME= C TYPE= 5

ATOM ID= 1389 NAME= C TYPE= 5

ATOM ID= 1390 NAME= O TYPE= 4

ATOM ID= 1390 NAME= O TYPE= 4

ATOM ID= 1390 NAME= O TYPE= 4

ATOM ID= 1393 NAME= C TYPE= 5

ATOM ID= 1395 NAME= H TYPE= 3

ATOM ID= 1778 NAME= C TYPE= 5

ATOM ID= 1779 NAME= H TYPE= 3

ATOM ID= 1780 NAME= H TYPE= 3

ATOM ID= 1781 NAME= C TYPE= 5

ATOM ID= 1783 NAME= C TYPE= 5

ATOM ID= 1783 NAME= C TYPE= 5

ATOM ID= 1783 NAME= C TYPE= 5

ATOM ID= 1785 NAME= H TYPE= 3

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1787 NAME= C TYPE= 5

ATOM ID= 1793 NAME= N TYPE= 4

ATOM ID= 1797 NAME= H TYPE= 3

ATOM ID= 1808 NAME= N TYPE= 4

ATOM ID= 1808 NAME= N TYPE= 4

ATOM ID= 1810 NAME= C TYPE= 5

ATOM ID= 1827 NAME= O TYPE= 4

ATOM ID= 1827 NAME= O TYPE= 4

ATOM ID= 2560 NAME= C TYPE= 5

ATOM ID= 2561 NAME= C TYPE= 5

ATOM ID= 2563 NAME= C TYPE= 5

ATOM ID= 2566 NAME= O TYPE= 4

ATOM ID= 2570 NAME= C TYPE= 5

ATOM ID= 2571 NAME= H TYPE= 3

ATOM ID= 2630 NAME= O TYPE= 4

ATOM ID= 2630 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2631 NAME= O TYPE= 4

ATOM ID= 2633 NAME= O TYPE= 4

ATOM ID= 2633 NAME= O TYPE= 4

ATOM ID= 2633 NAME= O TYPE= 4

ATOM ID= 2637 NAME= H TYPE= 3

ATOM ID= 2667 NAME= N TYPE= 4

ATOM ID= 2669 NAME= C TYPE= 5

ATOM ID= 2675 NAME= C TYPE= 5

ATOM ID= 2676 NAME= O TYPE= 4

ATOM ID= 2676 NAME= O TYPE= 4

ATOM ID= 2691 NAME= N TYPE= 4

ATOM ID= 2699 NAME= H TYPE= 3

ATOM ID= 2699 NAME= H TYPE= 3

ATOM ID= 2701 NAME= C TYPE= 5

ATOM ID= 2701 NAME= C TYPE= 5

ATOM ID= 2703 NAME= H TYPE= 3

ATOM ID= 2711 NAME= C TYPE= 5

ATOM ID= 2713 NAME= H TYPE= 3

ATOM ID= 2714 NAME= C TYPE= 5

ATOM ID= 2715 NAME= O TYPE= 4

ATOM ID= 2716 NAME= O TYPE= 4

ATOM ID= 2716 NAME= O TYPE= 4

ATOM ID= 2716 NAME= O TYPE= 4

ATOM ID= 2718 NAME= O TYPE= 4

ATOM ID= 2718 NAME= O TYPE= 4

ATOM ID= 2722 NAME= H TYPE= 3

ATOM ID= 2829 NAME= C TYPE= 5

ATOM ID= 2830 NAME= H TYPE= 3

ATOM ID= 2846 NAME= C TYPE= 5

ATOM ID= 2897 NAME= C TYPE= 5

ATOM ID= 2897 NAME= C TYPE= 5

ATOM ID= 2898 NAME= H TYPE= 3

ATOM ID= 2898 NAME= H TYPE= 3

ATOM ID= 2900 NAME= N TYPE= 4

ATOM ID= 2900 NAME= N TYPE= 4

ATOM ID= 2900 NAME= N TYPE= 4

ATOM ID= 3172 NAME= O TYPE= 4

ATOM ID= 3172 NAME= O TYPE= 4

ATOM ID= 3178 NAME= H TYPE= 3

ATOM ID= 3179 NAME= H TYPE= 3

ATOM ID= 3180 NAME= C TYPE= 5

ATOM ID= 3183 NAME= C TYPE= 5

ATOM ID= 3183 NAME= C TYPE= 5

ATOM ID= 3192 NAME= O TYPE= 4

ATOM ID= 3192 NAME= O TYPE= 4

ATOM ID= 3214 NAME= C TYPE= 5

ATOM ID= 3214 NAME= C TYPE= 5

ATOM ID= 3214 NAME= C TYPE= 5

ATOM ID= 3215 NAME= H TYPE= 3

ATOM ID= 3216 NAME= H TYPE= 3

ATOM ID= 3216 NAME= H TYPE= 3

ATOM ID= 3220 NAME= C TYPE= 5

ATOM ID= 3221 NAME= H TYPE= 3

ATOM ID= 3221 NAME= H TYPE= 3

ATOM ID= 3289 NAME= H TYPE= 3

ATOM ID= 3289 NAME= H TYPE= 3

ATOM ID= 5384 NAME= H TYPE= 3

ATOM ID= 5384 NAME= H TYPE= 3

ATOM ID= 5386 NAME= N TYPE= 4

ATOM ID= 5386 NAME= N TYPE= 4

ATOM ID= 5386 NAME= N TYPE= 4

TOTAL POCKET TRIANGLE: 17730

++

+ +

+ INFORMATION> SCREENING START +

+ +

++

INFORMATION> POCKET CENTER COORDINATE

X-AXIS : 4.492983

Y-AXIS : -3.696033

Z-AXIS : -13.93182

INFORMATION> SCREENING NUMBER : 1

INFORMATION> REFERENCE COODINATE NOT READ

(USE LIGAND COORDINATE)

INFORMATION> INPUT LIGAND FILE

FILE NAME :0_Q2U.mol2

ATOM NUMBER : 36

BOND NUMBER : 38

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATION> LIGAND ACCEPTOR, DONAR

DONAR NUMBER: 2

25 H

26 H

ACCEPTOR NUMBER: 5

7 O

8 O

13 O

23 O

24 O

INFORMATION> GLOBAL SEARCH START

INFORMATION> CONFORMER NUMBER = 1 GENERATION = 1

ID EDGE-LOW EDGE-UPR PLATE NUM TRIAL NUMBER

1 3.643 6.191 786 125

INFORMATINO> CREATE NEXT CONFORMER

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATINO> NEW CONFOMER NUMBER = 1

INFORMATION> CONFORMER NUMBER = 1 GENERATION = 2

ID EDGE-LOW EDGE-UPR PLATE NUM TRIAL NUMBER

1 3.643 6.191 786 125

INFORMATINO> CREATE NEXT CONFORMER

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATINO> NEW CONFOMER NUMBER = 1

INFORMATION> CONFORMER NUMBER = 1 GENERATION = 3

ID EDGE-LOW EDGE-UPR PLATE NUM TRIAL NUMBER

1 3.643 6.191 786 125

INFORMATINO> CREATE NEXT CONFORMER

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATINO> NEW CONFOMER NUMBER = 1

INFORMATION> CONFORMER NUMBER = 1 GENERATION = 4

ID EDGE-LOW EDGE-UPR PLATE NUM TRIAL NUMBER

1 3.643 6.191 786 125

INFORMATINO> CREATE NEXT CONFORMER

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATINO> NEW CONFOMER NUMBER = 1

INFORMATION> CONFORMER NUMBER = 1 GENERATION = 5

ID EDGE-LOW EDGE-UPR PLATE NUM TRIAL NUMBER

1 3.643 6.191 786 125

INFORMATINO> CREATE NEXT CONFORMER

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATINO> NEW CONFOMER NUMBER = 1

INFORMATION> CONFORMER NUMBER = 1 GENERATION = 6

ID EDGE-LOW EDGE-UPR PLATE NUM TRIAL NUMBER

1 3.643 6.191 786 125

INFORMATINO> CREATE NEXT CONFORMER

INFORMATION> LIGAND ANALYZE

ATOM BELONGS RING STRUCTURE:

1 3 4 5 6 8 10 11 12 13 21 22 24 25 26 27 28 30 32 33 35

ROTATIVE TORSION NUMBER : 2

(O-H, S-H TERMAINAL) : 0

INFORMATINO> NEW CONFOMER NUMBER = 1

INFORMATION> BEFORE MINIMIZE RANKING :EXPERIMENT ID=1

INFORMATION> LOCAL SCORE RANKING 10

SCORE(/100) dG HIT MTS rotNum ASA ELE HYD VDW SURFACE RMSD

1 -4.13 -4.98 -48.58 -183.39 2 -169.20 0.00 -20.93 0.00 0.00 22.06

2 -3.87 -4.64 -45.09 -171.48 2 -157.06 0.00 -20.40 0.00 0.00 21.72

3 -3.69 -4.40 -42.17 -162.69 2 -146.82 0.00 -20.92 0.00 0.00 21.19

4 -3.66 -4.55 -30.27 -163.34 2 -103.39 0.00 -52.15 0.00 0.00 16.40

5 -3.61 -4.28 -40.54 -158.52 2 -141.09 0.00 -21.78 0.00 0.00 21.51

6 -3.58 -4.24 -40.15 -157.04 2 -139.73 0.00 -21.61 0.00 0.00 21.44

7 -3.44 -4.25 -27.89 -153.10 2 -95.21 0.00 -50.10 0.00 0.00 16.25

8 -3.34 -4.04 -41.52 -150.99 2 -145.03 0.00 -13.22 0.00 0.00 18.03

9 -3.25 -3.93 -40.45 -147.23 2 -141.31 0.00 -12.98 0.00 0.00 20.00

10 -3.12 -3.64 -32.83 -135.45 2 -114.08 0.00 -23.22 0.00 0.00 21.68

REFERENCE COORDINATE DATA

999.90 999.90 0.00 0.00

INFORMATION> CREATED LIGAND TOPOLOGY

ATOM NUMBER : 36

BOND NUMBER : 38

ANGLE NUMBER : 61

TORSION NUMBER : 84

IMPROPER NUMBER : 15

INFORMATION> LOCAL SEARCH START

10 %

20 %

30 %

40 %

50 %

60 %

70 %

80 %

90 %

100 %

INFORMATION> LOCAL SEARCH FINISHED

INFORMATION> AFTER MINIMIZE RANKING

INFORMATION> AFTER MINIMIZE RANKING :EXPERIMENT ID=1

INFORMATION> LOCAL SCORE RANKING 10

COMPOUND NAME : Q2U.mol2

FILE NAME : 0_Q2U.mol2

SCORE(/100) dG HIT MTS rotNum ASA ELE HYD VDW SURFACE RMSD

@ 1 -5.06 -6.33 -50.30 -228.37 2 -171.67 0.00 -52.44 -2.76 72.11 16.67

@ 2 -4.63 -5.78 -43.16 -208.37 2 -146.61 0.00 -54.87 -2.73 72.63 16.76

@ 3 -4.15 -5.03 -49.17 -185.27 2 -171.25 0.00 -20.93 0.00 57.61 22.06

@ 4 -3.89 -4.70 -45.71 -173.47 2 -159.22 0.00 -20.40 0.00 60.72 21.72

@ 5 -3.87 -4.77 -34.35 -173.32 2 -116.18 0.00 -49.55 -2.75 70.92 16.40

@ 6 -3.71 -4.45 -42.73 -164.52 2 -148.80 0.00 -20.92 0.00 56.04 21.19

@ 7 -3.69 -4.61 -30.98 -165.63 2 -105.87 0.00 -52.15 0.00 71.19 16.40

@ 8 -3.63 -4.33 -41.06 -160.20 2 -142.91 0.00 -21.78 0.00 51.43 21.51

@ 9 -3.59 -4.29 -40.67 -158.73 2 -141.56 0.00 -21.61 0.00 51.64 21.44

@ 10 -3.50 -4.21 -45.98 -159.36 2 -159.34 0.00 -8.34 -2.68 41.22 19.87

REFERENCE COORDINATE DATA

999.90 999.90 0.00 0.00

INFORMATION> ALL EXPERIMENT FINISHED

INFORMATION> COMPOUND RANKING 1

COMPOUND NAME SCORE(/100) dG HIT MTS

1 Q2U.mol2 -5.09 -6.33 -50.30 -228.37

TOTAL EXPERIMENT NUMBER : 1

TOTAL TRIAL NUMBER : 750

TOTAL CPU TIME(S) : 6.238247

INPUT : 1.550246

GRID : 9.8199844E-03

CONF : 1.9330144E-02

DOCK : 4.0799975E-02

MIN : 4.435637