FIGURE 1A

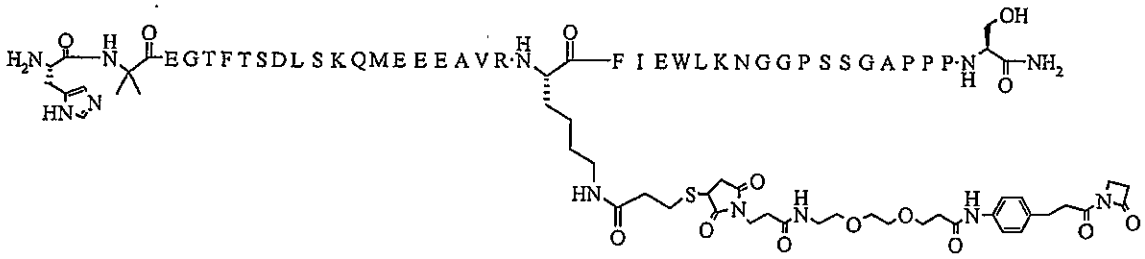


FIGURE 1B

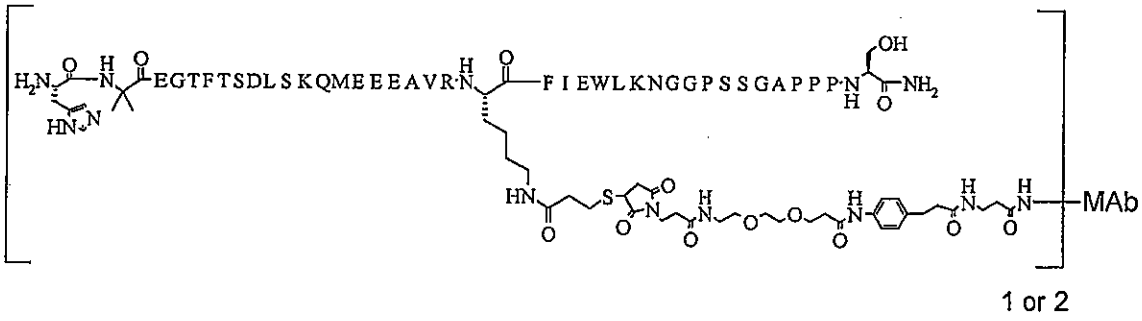


FIGURE 2A

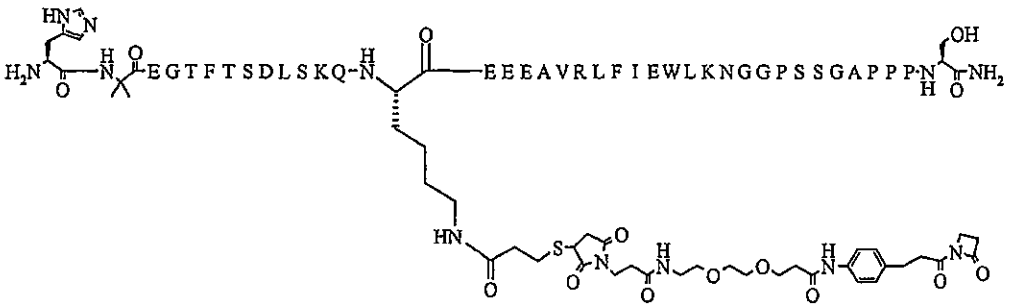


FIGURE 2B

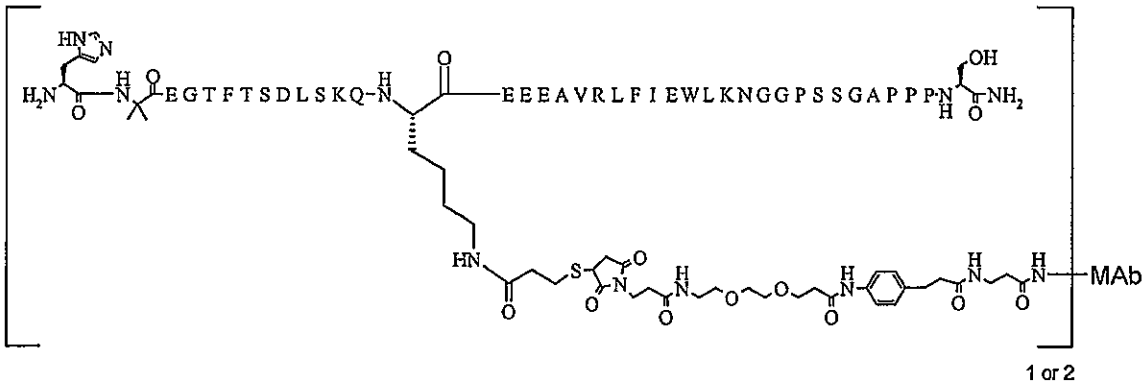


FIGURE 3

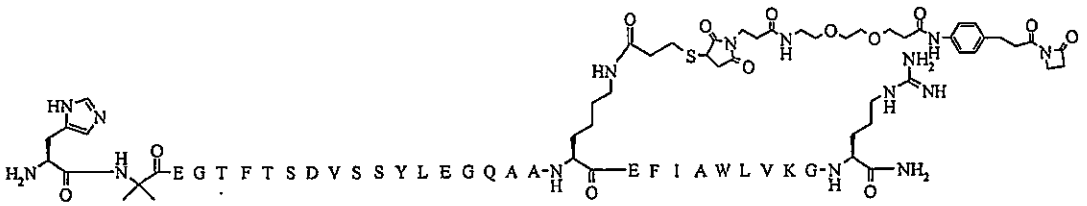

HN
N
H2N
O
H
N
O
E G T F T S D V S S Y L E G Q A A-N
H
O
E F I A W L V K G-N
H
O
NH2
HN
O
S
O
N
O
O
N
H
O
O
O
N
H
NH2
N
H
NH
O
N
O

FIGURE 4

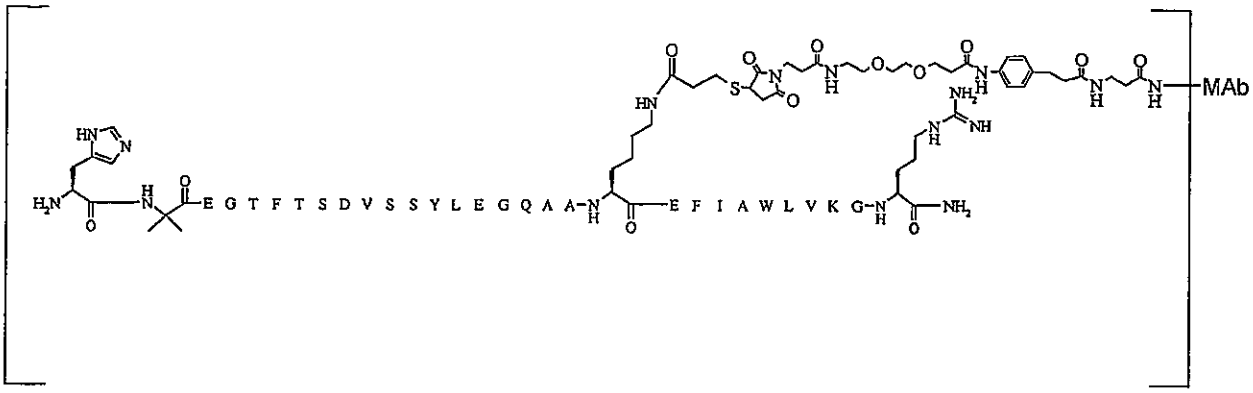


FIGURE 5A

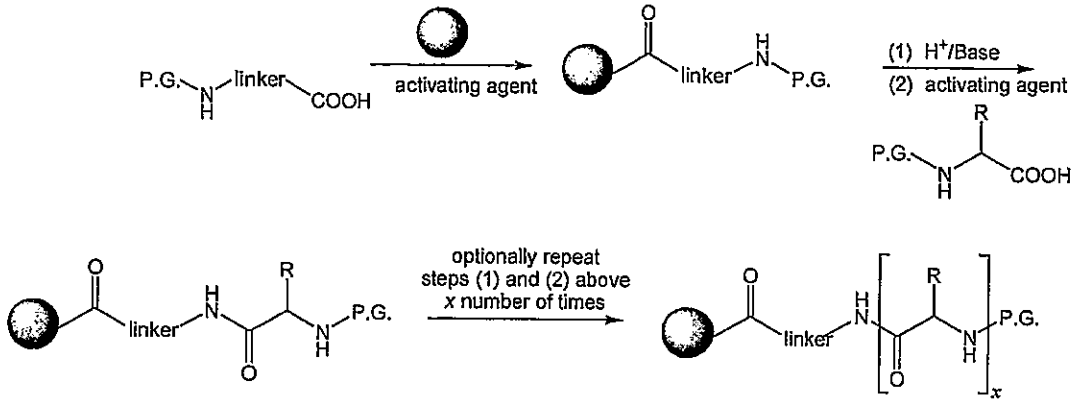


FIGURE 5B

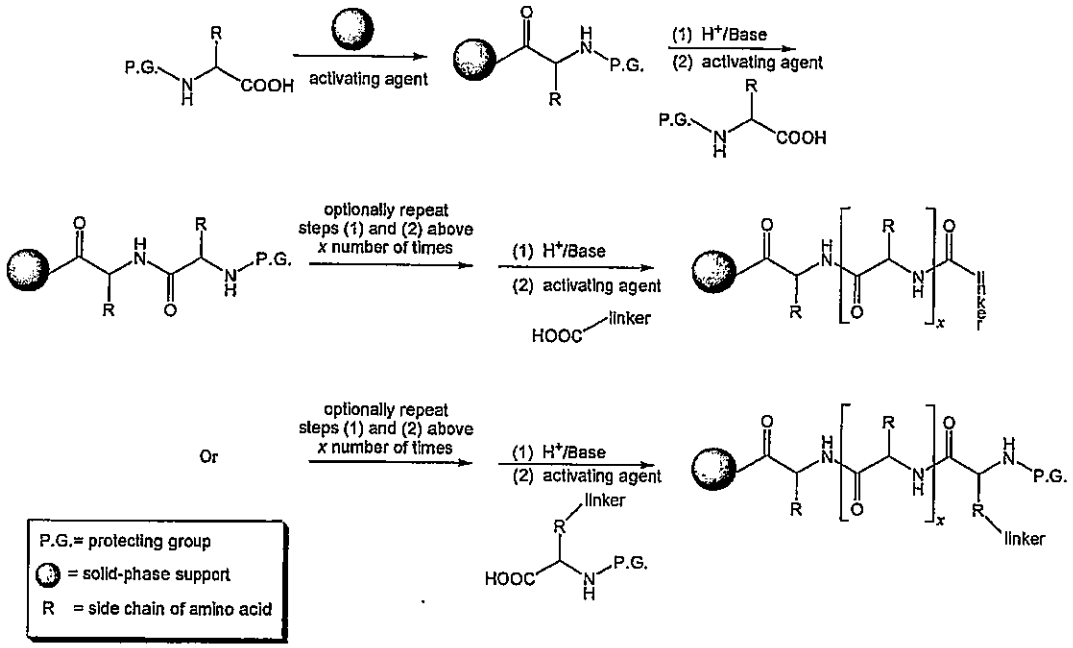


FIGURE 6

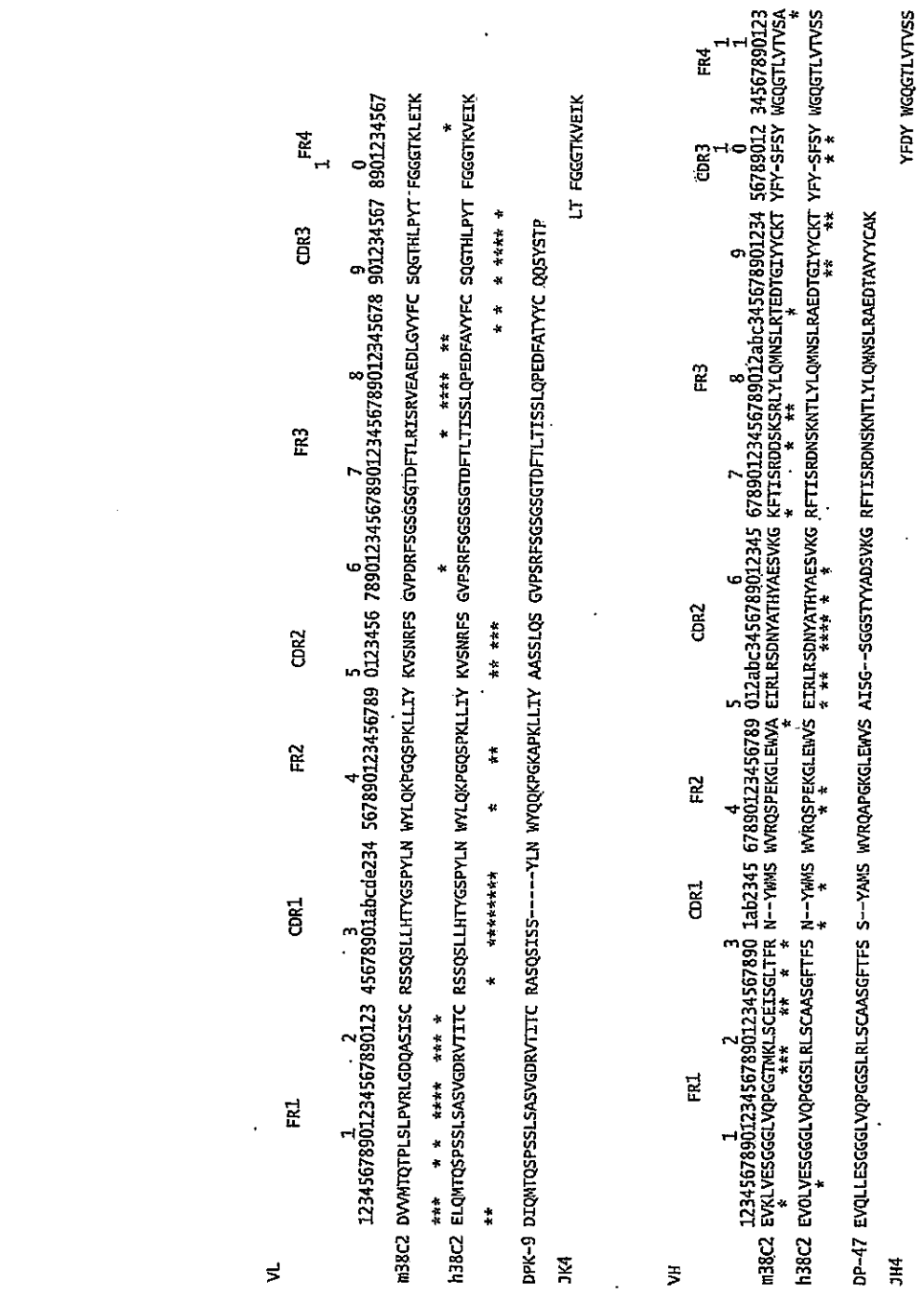
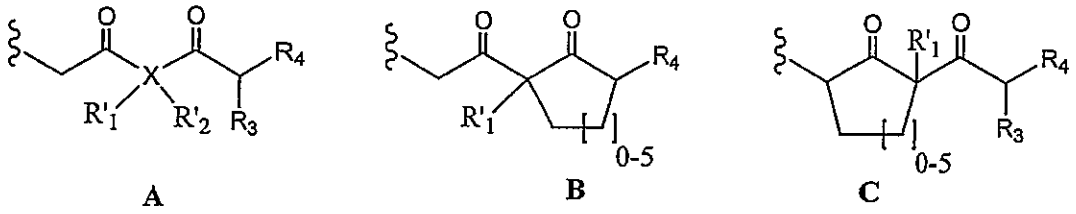
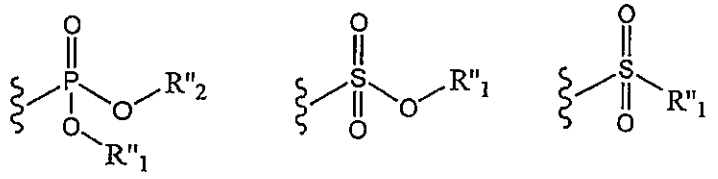
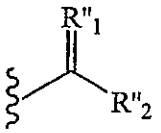

VL
FR1
CDR1
FR2
CDR2
FR3
CDR3
FR4
m38C2 DVVMTQTPLSLPVRLGDQASISC RSSQSLLHTYGSPYLN WYLQKPGQSPKLLIY KVSNRFS GVPDRFSGSGSGTDFTLRISRVEAEDLGVYFC SQGTHLPYT FGGGTKLEIK
h38C2 ELQMTQSPSSLSASVGDRVTITC RSSQSLLHTYGSPYLN WYLQKPGQSPKLLIY KVSNRFS GVPSRFSGSGSGTDFTLTISSLQPEDFAVYFC SQGTHLPYT FGGGTKVEIK
DPK-9 DIQMTQSPSSLSASVGDRVTITC RASQSISS----YLN WYQQKPGKAPKLLIY AASSLQS GVPSRFSGSGSGTDFTLTISSLQPEDFATYYC QQSYSTP
JK4 LT FGGGTKVEIK
VH
FR1
CDR1
FR2
CDR2
FR3
CDR3
FR4
m38C2 EVKLVESGGGLVQPGGTNKLSCEISGLTFR N--YWMS WVRQSPEKGLEWVA EIRLRSDNYATHYAESVKG KFTISRDDSKSRLYLQMNSLRTEDTGIYYCKT YFY-SFSY WGQGTLVTVSA
h38C2 EVQLVESGGGLVQPGGSLRLSCAASGFTFS N--YWMS WVRQSPEKGLEWVS EIRLRSDNYATHYAESVKG RFTISRDNSKNTLYLQMNSLRAEDTGIYYCKT YFY-SFSY WGQGTLVTVSS
DP-47 EVQLLESGGGLVQPGGSLRLSCAASGFTFS S--YAMS WVRQAPGKGLEWVS AISG--SGGSTYYADSVKG RFTISRDNSKNTLYLQMNSLRAEDTAVYYCAK
JH4 YFDY WGQGTLVTVSS

FIGURE 7

Linker Reactive Groups

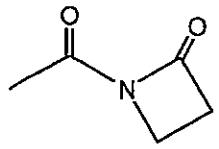




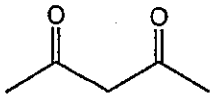


G

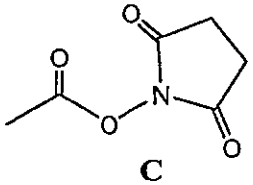
FIGURE 8



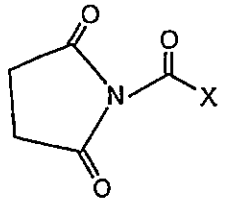
A



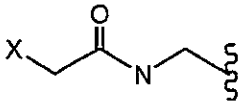
B



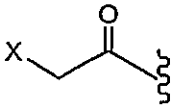
C



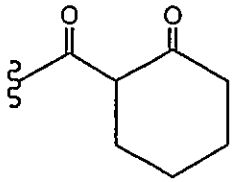
D



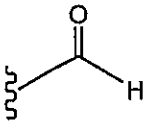
E



F



G



H

FIGURE 9

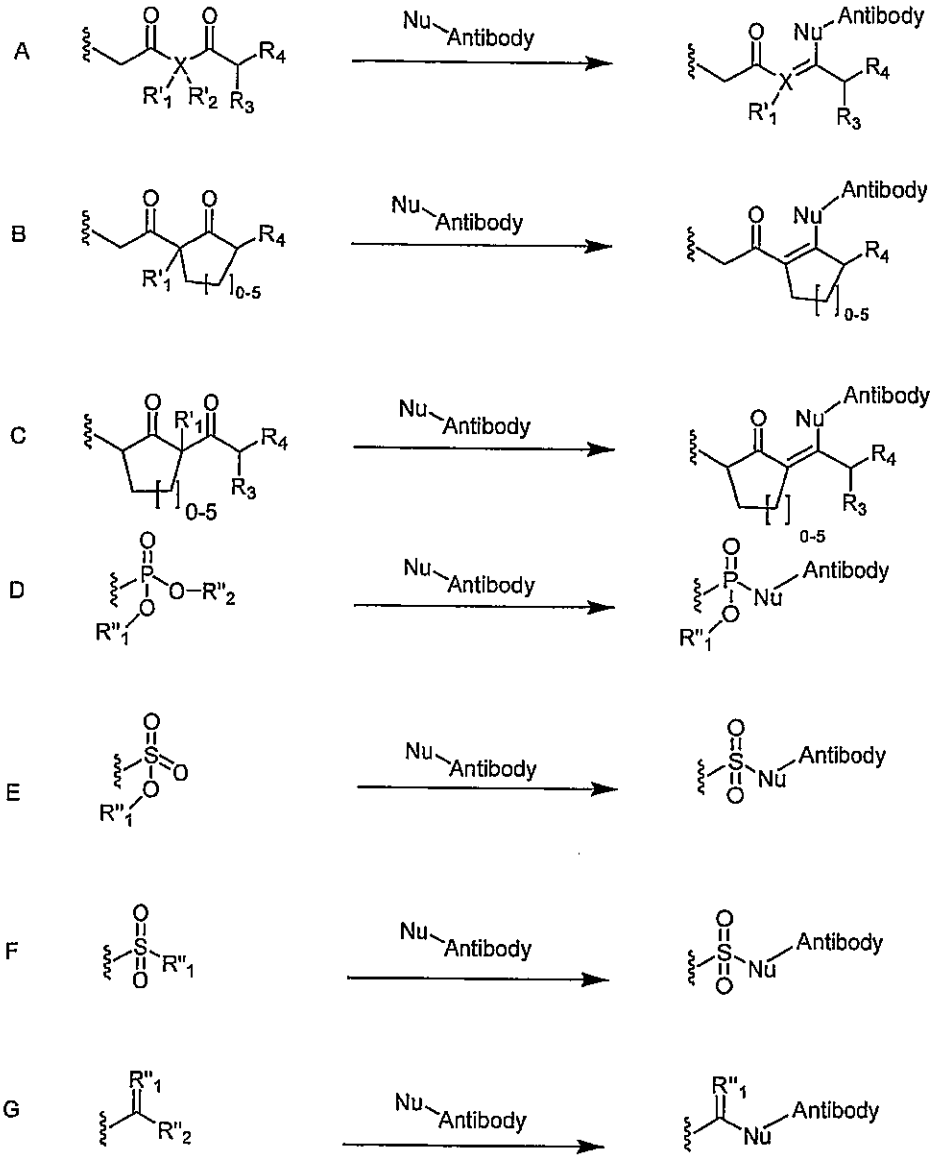


FIGURE 10

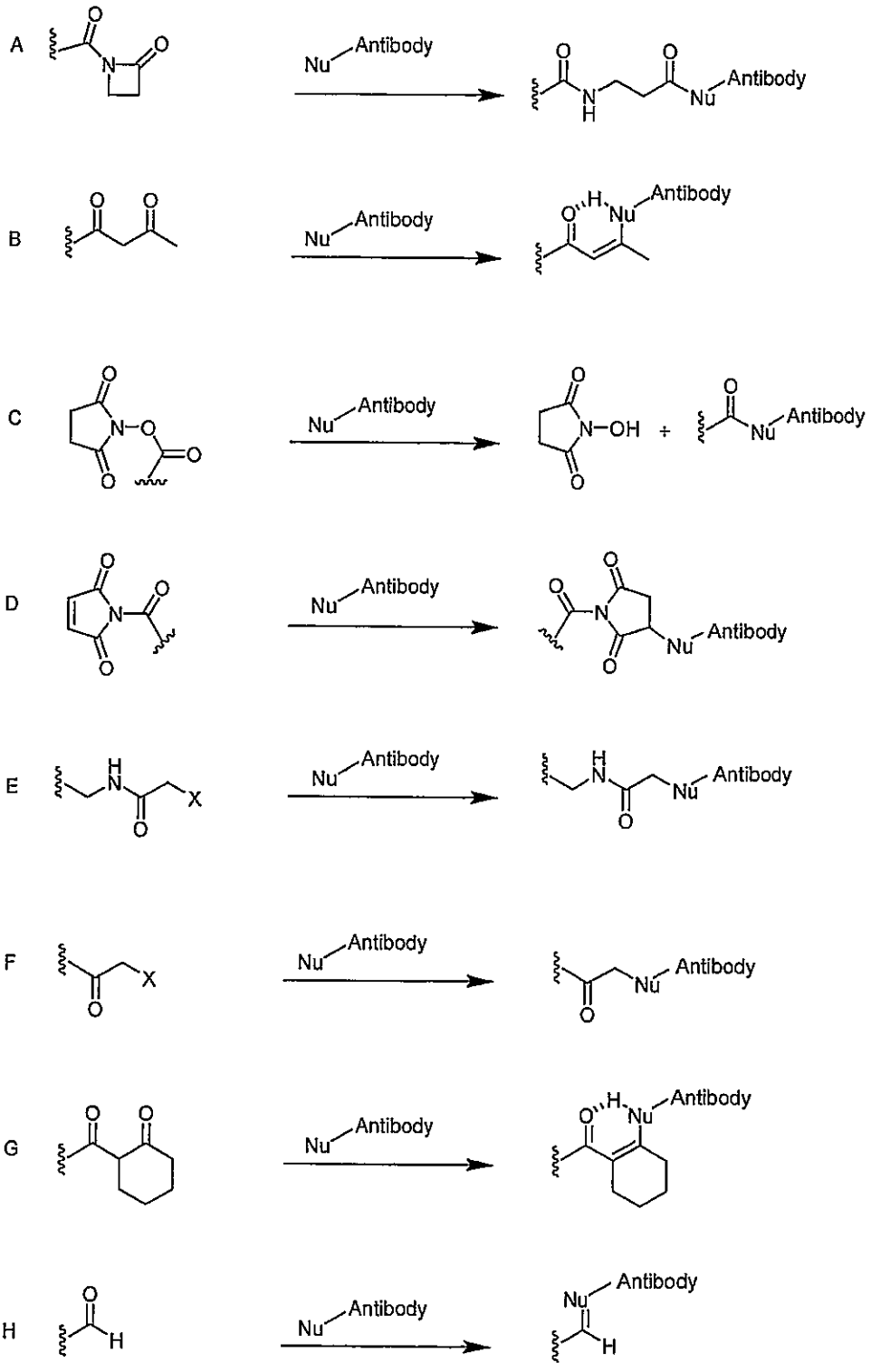

A
Nu
Antibody
Nu
Antibody
B
Nu
Antibody
O
H
Nu
Antibody
C
Nu
Antibody
N–OH
+
Nu
Antibody
D
Nu
Antibody
Nu
Antibody
E
Nu
Antibody
Nu
Antibody
F
Nu
Antibody
Nu
Antibody
G
Nu
Antibody
O
H
Nu
Antibody
H
Nu
Antibody
Nu
Antibody

FIGURE 11

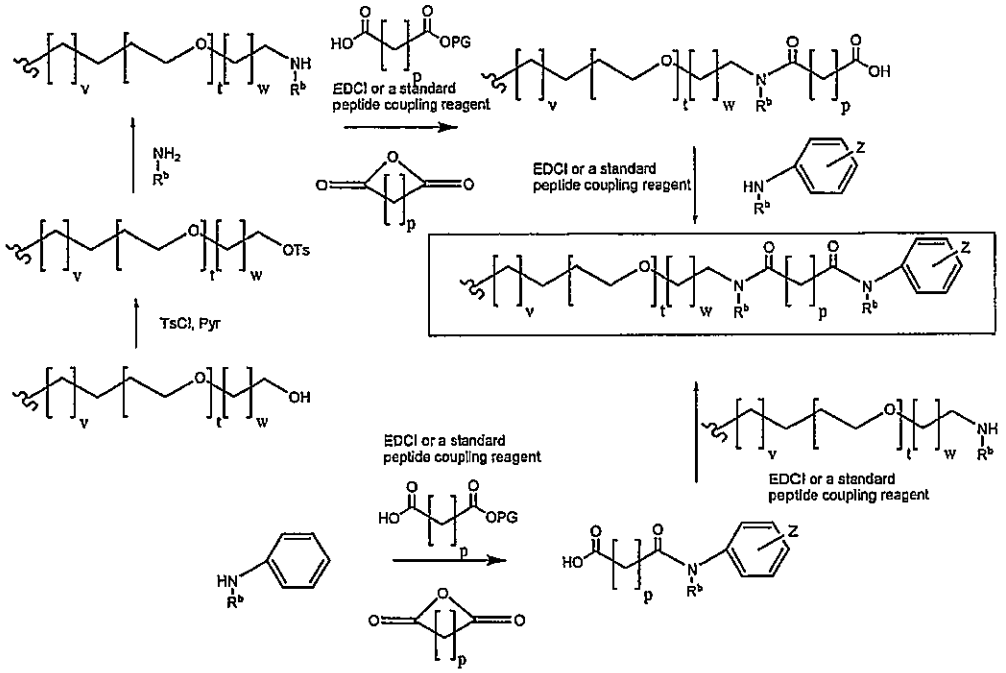

EDCI or a standard peptide coupling reagent
EDCI or a standard peptide coupling reagent
TsCl, Pyr
EDCI or a standard peptide coupling reagent
EDCI or a standard peptide coupling reagent

FIGURE 12

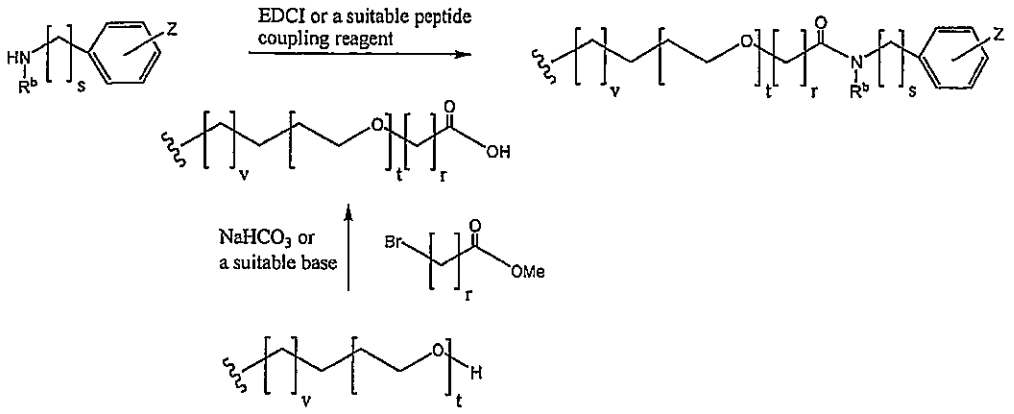

EDCI or a suitable peptide coupling reagent
NaHCO3 or a suitable base
OH
Br
OMe

FIGURE 13

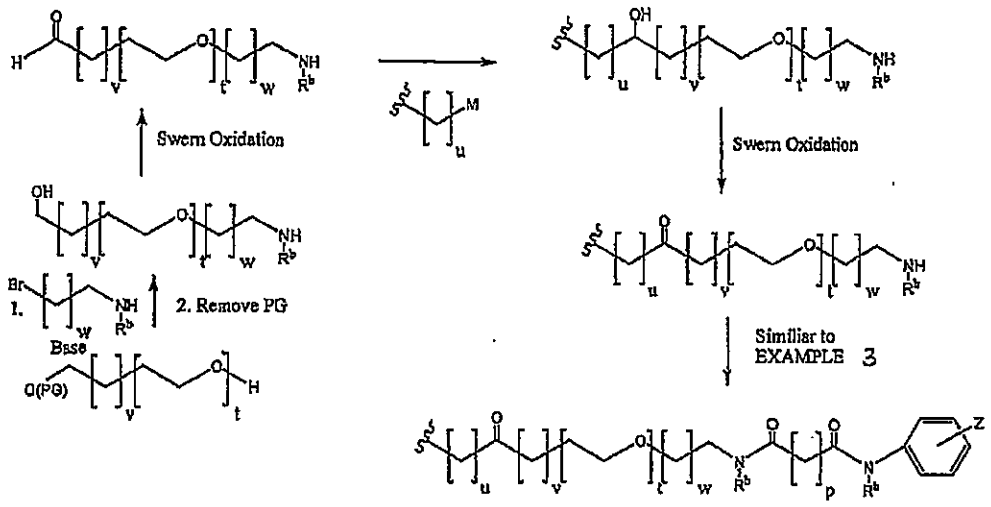

Swern Oxidation
Swern Oxidation
2. Remove PG
Base
Similiar to
EXAMPLE 3

FIGURE 14

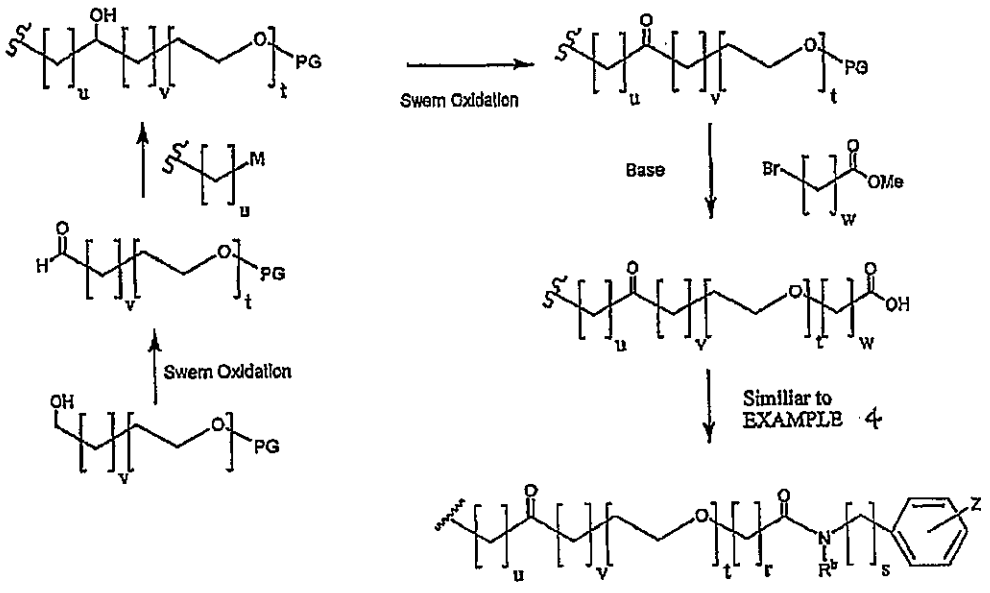

Swern Oxidation
Base
Swern Oxidation
Similiar to EXAMPLE 4

FIGURE 15

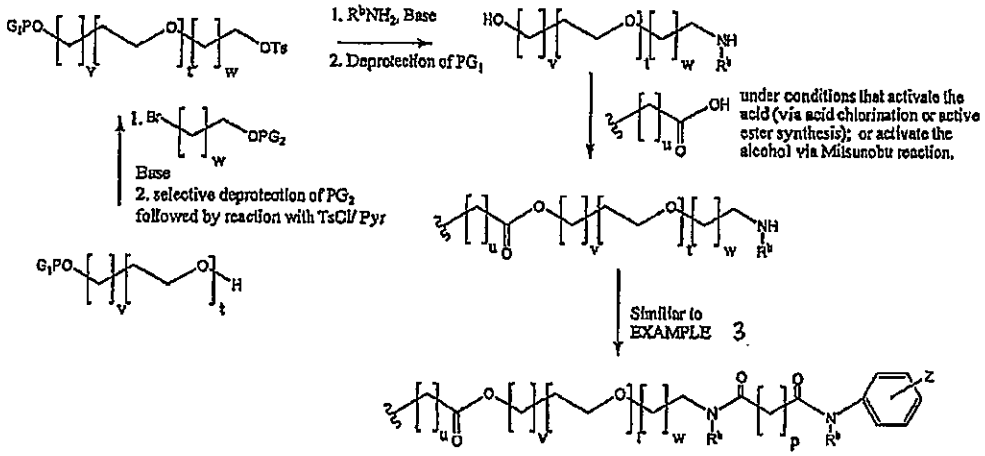

1. R^bNH_2, Base
2. Deprotection of PG_1
under conditions that activate the acid (via acid chlorination or active ester synthesis); or activate the alcohol via Mitsunobu reaction.
1. Br OPG2
Base
2. selective deprotection of PG_2 followed by reaction with TsCl/ Pyr
Similar to EXAMPLE 3

FIGURE 16

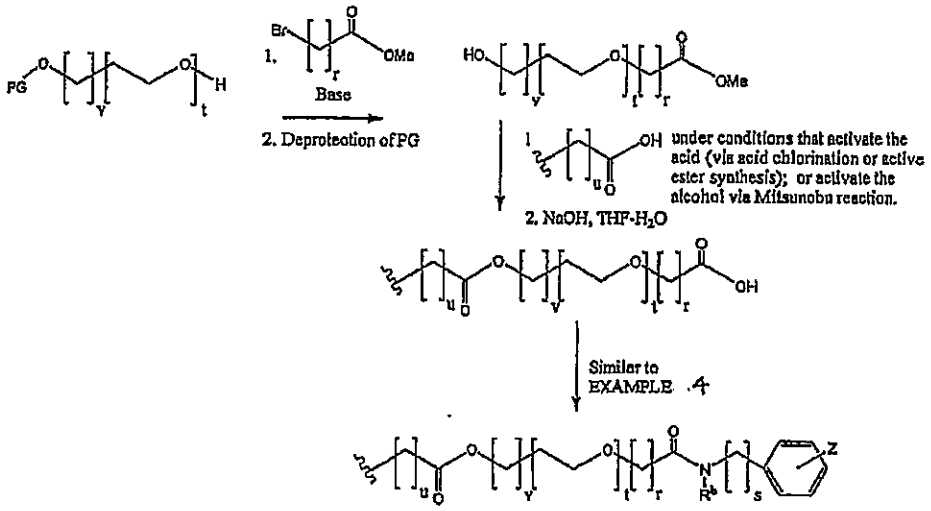

1.
Base
2. Deprotection of PG
under conditions that activate the acid (via acid chlorination or active ester synthesis); or activate the alcohol via Mitsunobu reaction.
2. NaOH, THF-H2O
Similar to EXAMPLE 4

FIGURE 17

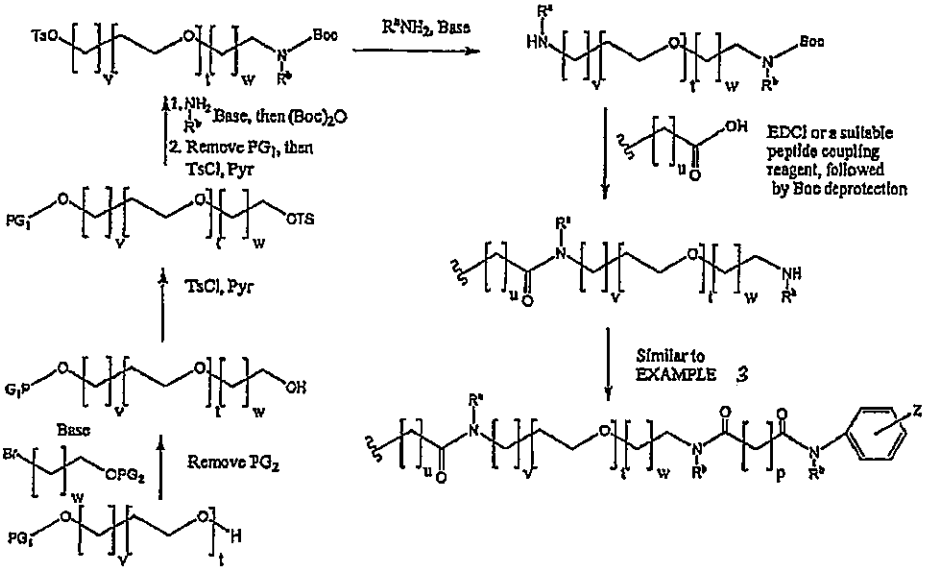

TsO
Boc
R^aNH_2, Base
1. NH_2 R^b Base, then $(Boc)_2O$
2. Remove PG_1, then TsCl, Pyr
PG_1
OTS
TsCl, Pyr
G_1O
OH
Base
Br
OPG_2
Remove PG_2
PG_1
H
EDCI or a suitable peptide coupling reagent, followed by Boc deprotection
Similar to EXAMPLE 3
NH
Z

FIGURE 18

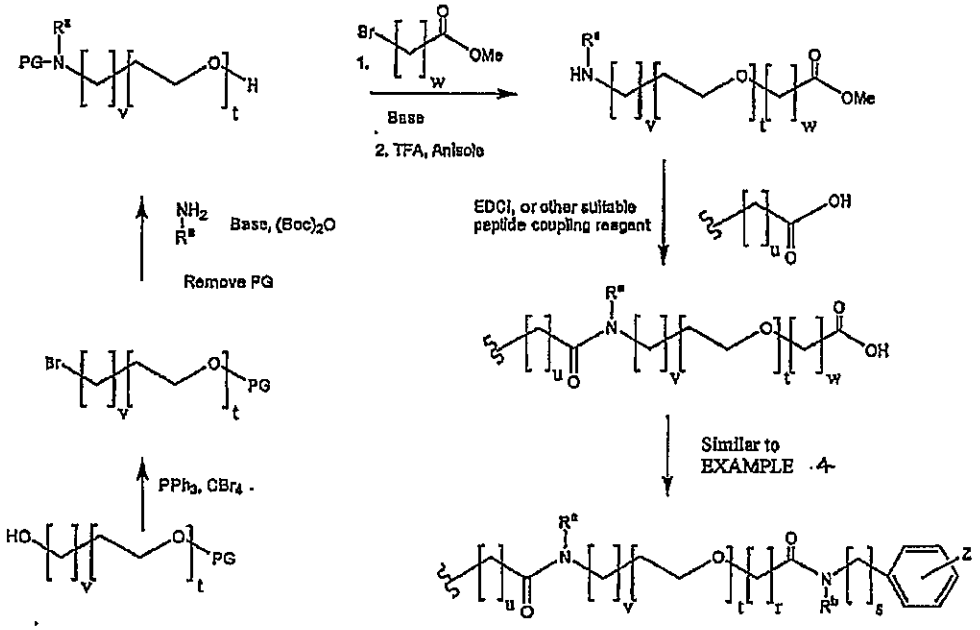

1.
Base
2. TFA, Anisole
EDCI, or other suitable peptide coupling reagent
Similar to EXAMPLE 4
Remove PG
Base, (Boc)2O
PPh3, CBr4

FIGURE 19

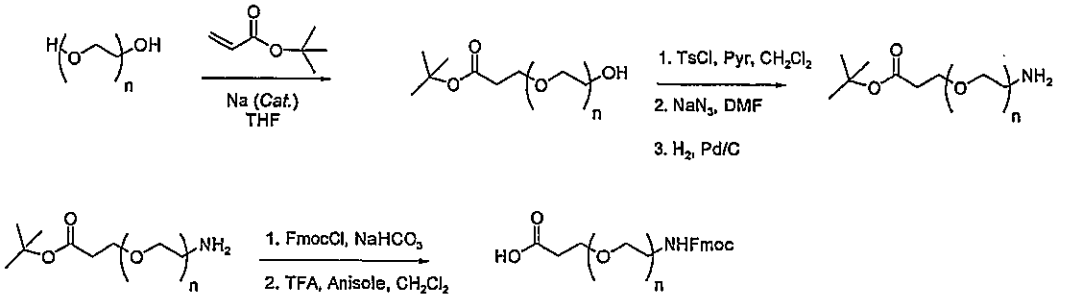

Na (Cat.)
THF
1. TsCl, Pyr, CH2Cl2
2. NaN3, DMF
3. H2, Pd/C
1. FmocCl, NaHCO3
2. TFA, Anisole, CH2Cl2

FIGURE 20

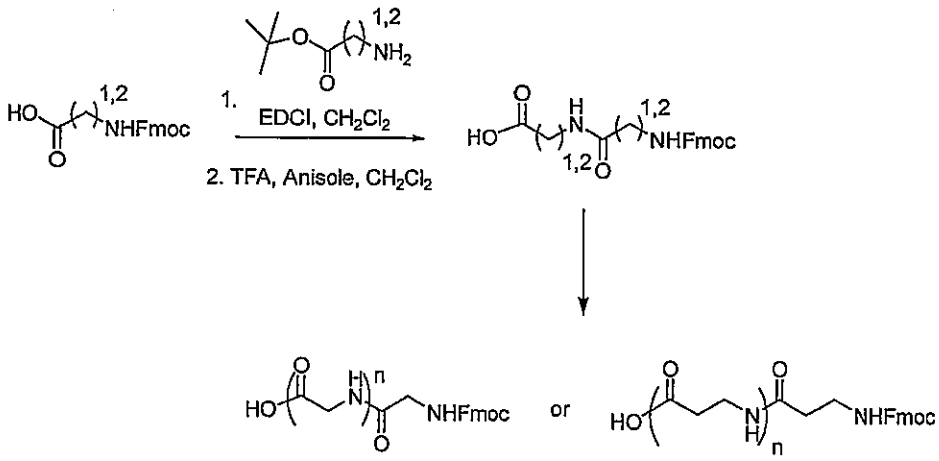

1,2
NH2
1,2
HO
NHFmoc
1.
EDCI, CH2Cl2
2. TFA, Anisole, CH2Cl2
HO
H
N
1,2
1,2
NHFmoc
HO
H
N
n
NHFmoc
or
HO
N
H
n
NHFmoc

FIGURE 21

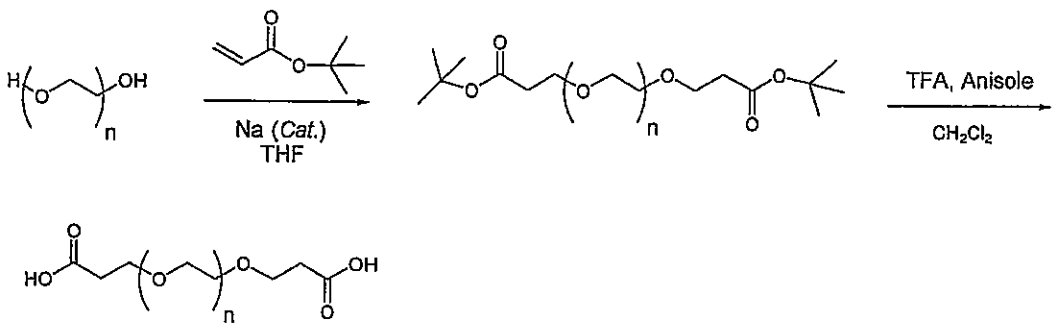


FIGURE 22

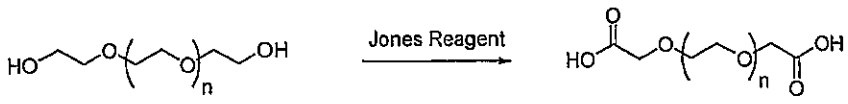


FIGURE 23

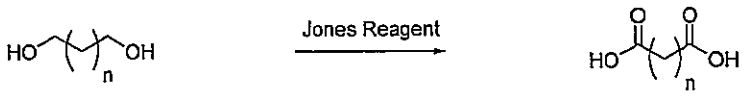


FIGURE 24

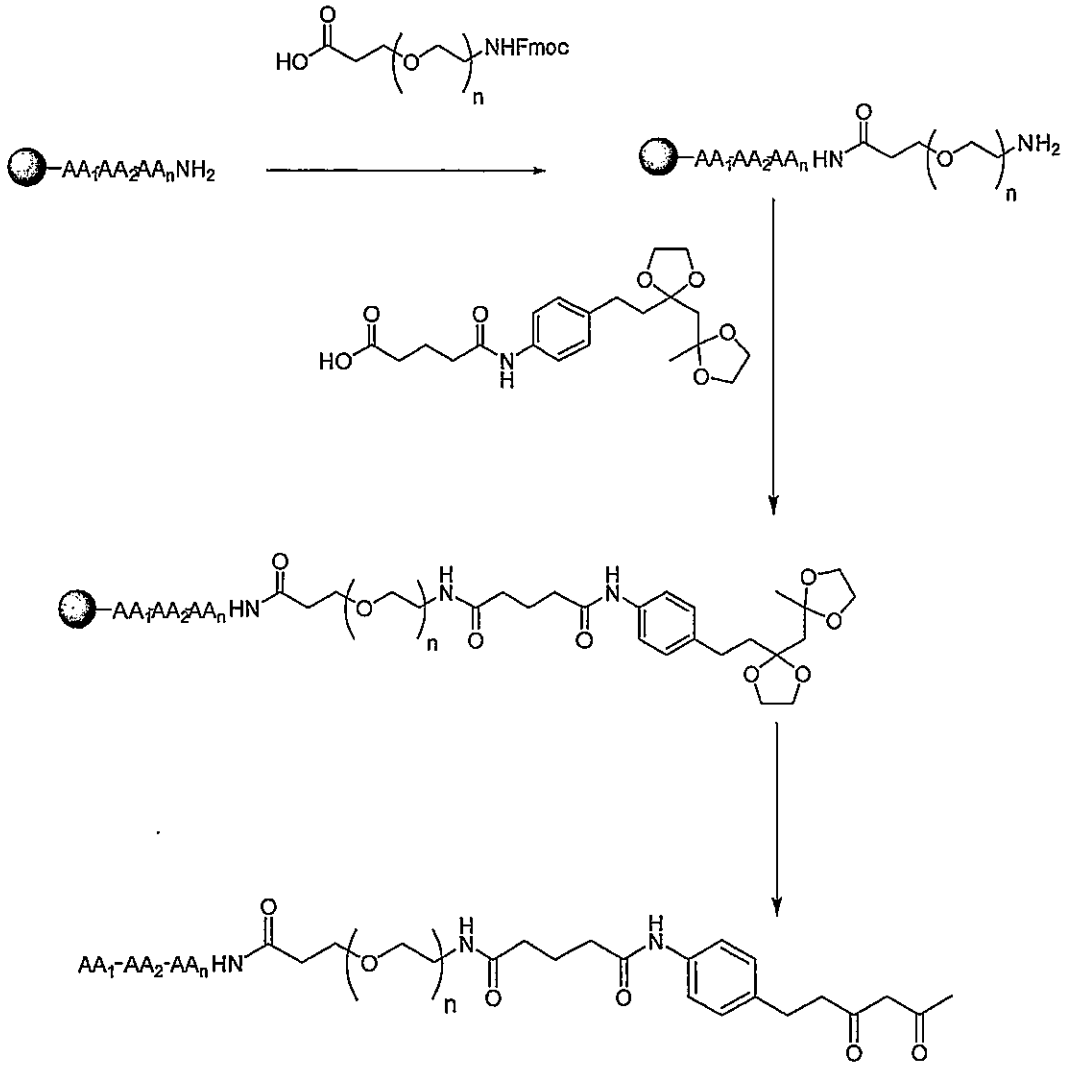

HO
NHFmoc
n
AA1AA2AAnNH2
AA1AA2AAn HN
NH2
HO
N
H
AA1AA2AAn HN
AA1-AA2-AAn HN

FIGURE 25

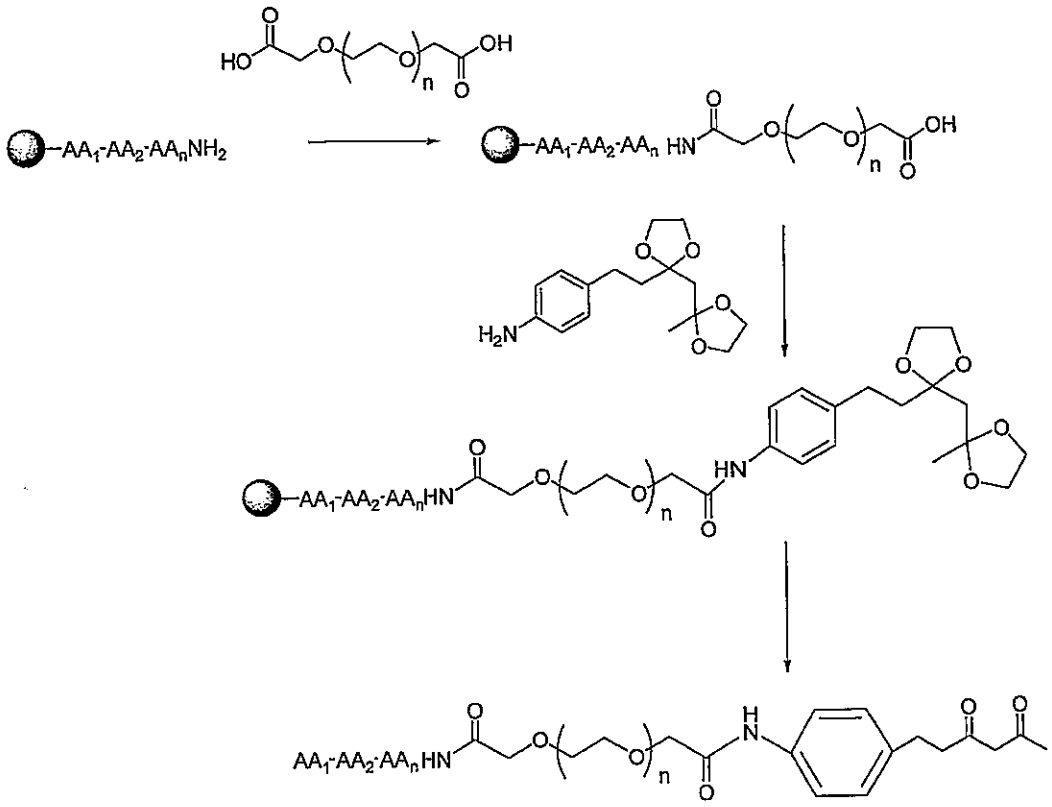

HO
OH
n
AA1-AA2-AAnNH2
AA1-AA2-AAn HN
H2N
AA1-AA2-AAnHN
HN
AA1-AA2-AAnHN
H
N

FIGURE 26

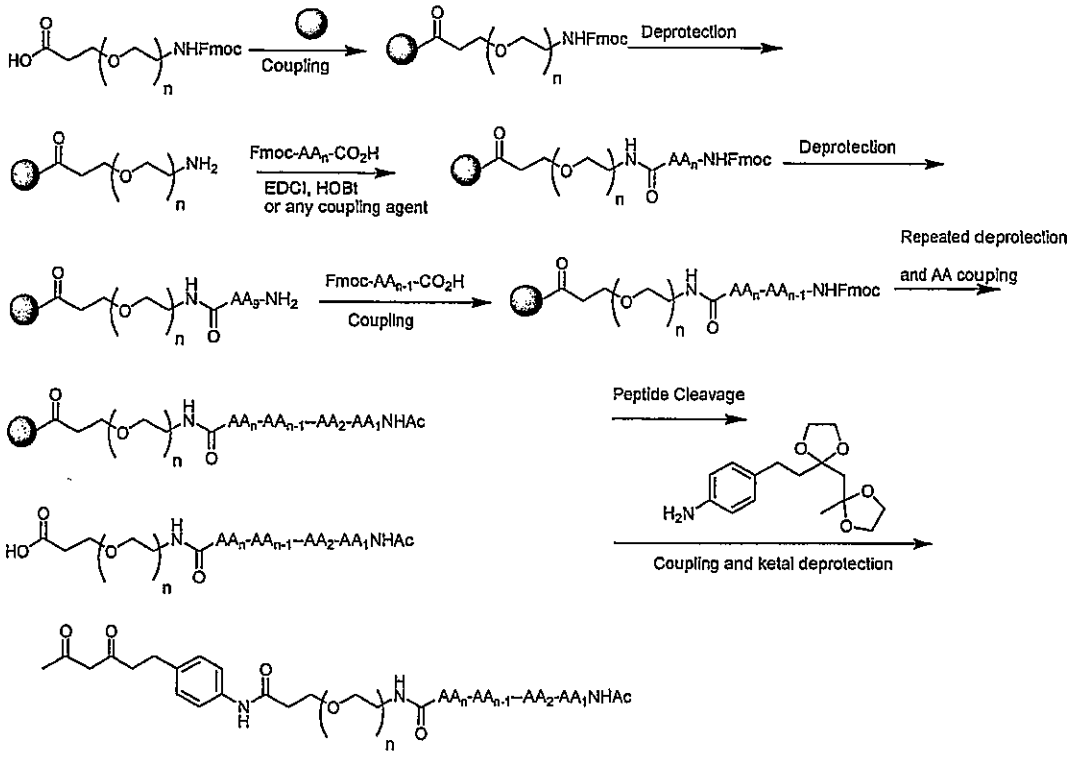

Coupling
Deprotection
Fmoc-AAn-CO2H
EDCI, HOBt
or any coupling agent
Repeated deprotection
and AA coupling
Fmoc-AAn-1-CO2H
Peptide Cleavage
Coupling and ketal deprotection

FIGURE 27

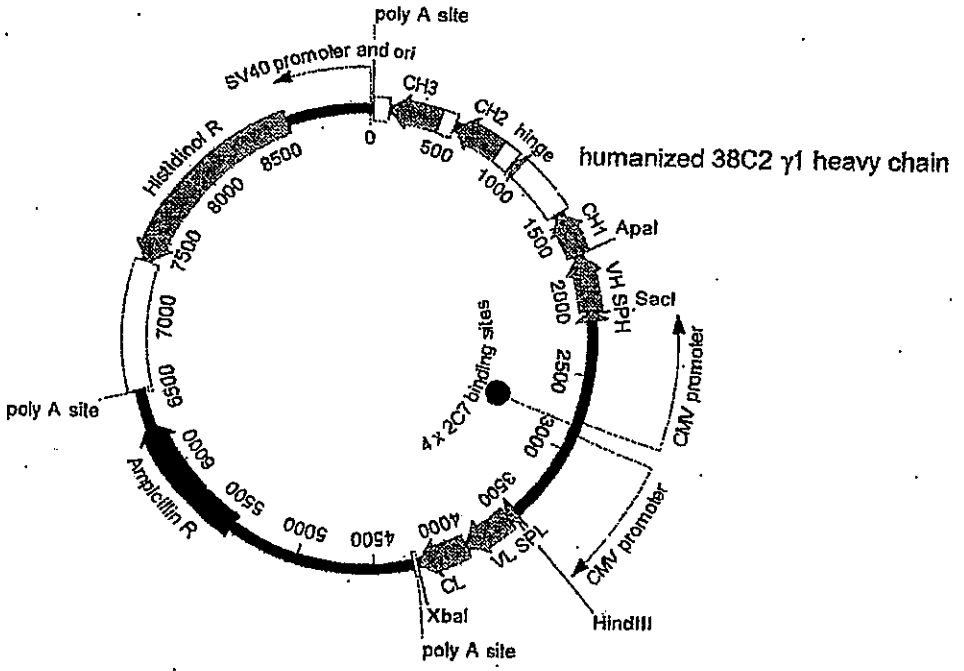

poly A site
SV40 promoter and ori
CH3
CH2
hinge
humanized 38C2 γ1 heavy chain
Histidinol R
0
500
1000
1500
2000
2500
3000
3500
4000
4500
5000
5500
6000
6500
7000
7500
8000
8500
CH1
ApaI
VH SPH
SacI
CMV promoter
4 x 2C7 binding sites
poly A site
Ampicillin R
CMV promoter
VL SPL
CL
XbaI
HindIII
poly A site
humanized 38C2 κ light chain

FIGURE 28

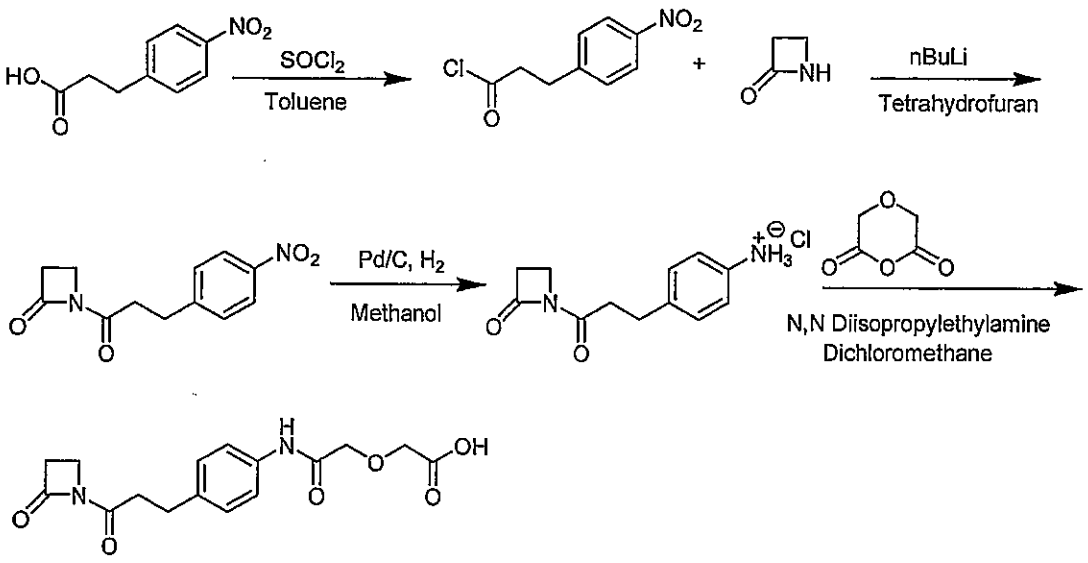

NO2
HO
O
SOCl2
Toluene
Cl
O
NO2
+
O
NH
nBuLi
Tetrahydrofuran
O
N
O
NO2
Pd/C, H2
Methanol
+NH3 ⊖Cl
O
N
O
O
O
O
O
N,N Diisopropylethylamine
Dichloromethane
H
N
O
O
O
OH
O
N
O

FIGURE 29

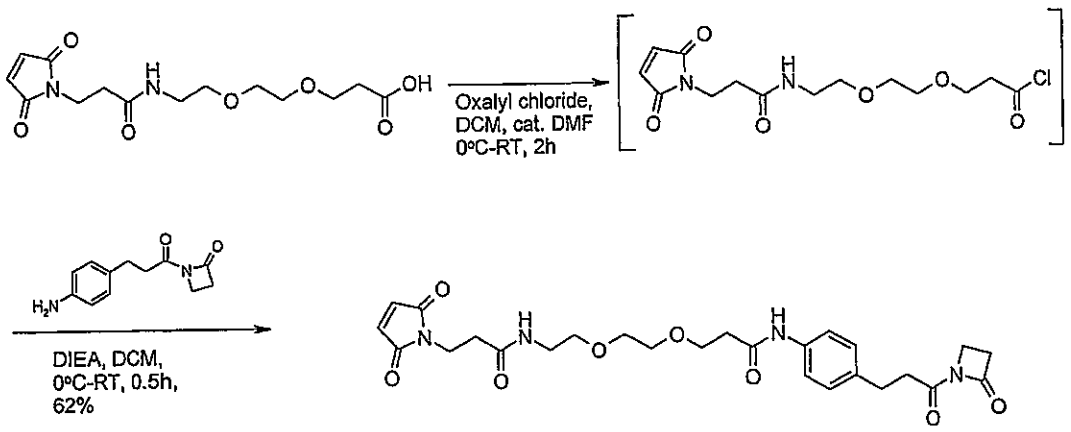


FIGURE 30

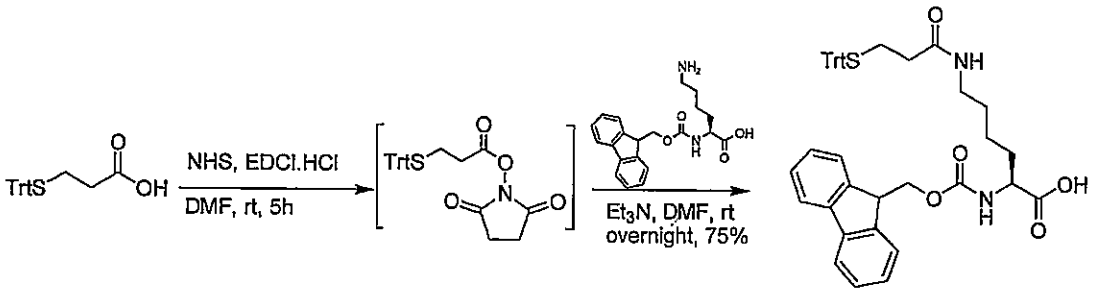


FIGURE 33

Light Chain (219 amino acids):
ELQMTQSPSSLSASVGDRVTITCRSSQSLLHTYGSPYLNWYLQKPGQSPKLLIYKVSNRFSGVPSRF
SGSGSGTDFTLTISSLQPEDFAVYFCSQGTHLPYTFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTA
SVVCLLNNFYPREAKVQWKVDNALQSGNSQESVTEQDSKDSTYSLSSTLTLSKADYEKHKVYAC
EVTHQGLSSPVTKSFNRGEC

Heavy chain (448 amino acids):
EVQLVESGGGLVQPGGSLRLSCAASGFTFSNYWMSWVRQSPEKGLEWVSEIRLRSDNYATHYAE
SVKGRFTISRDNSKNTLYLQMNSLRAEDTGIYYCKTYFYSFSYWGQGTLVTVSSASTKGPSVFPLA
PSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVVTVPSSSLGT
QTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCV
VVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS
NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENN
YKTTPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

FIGURE 31

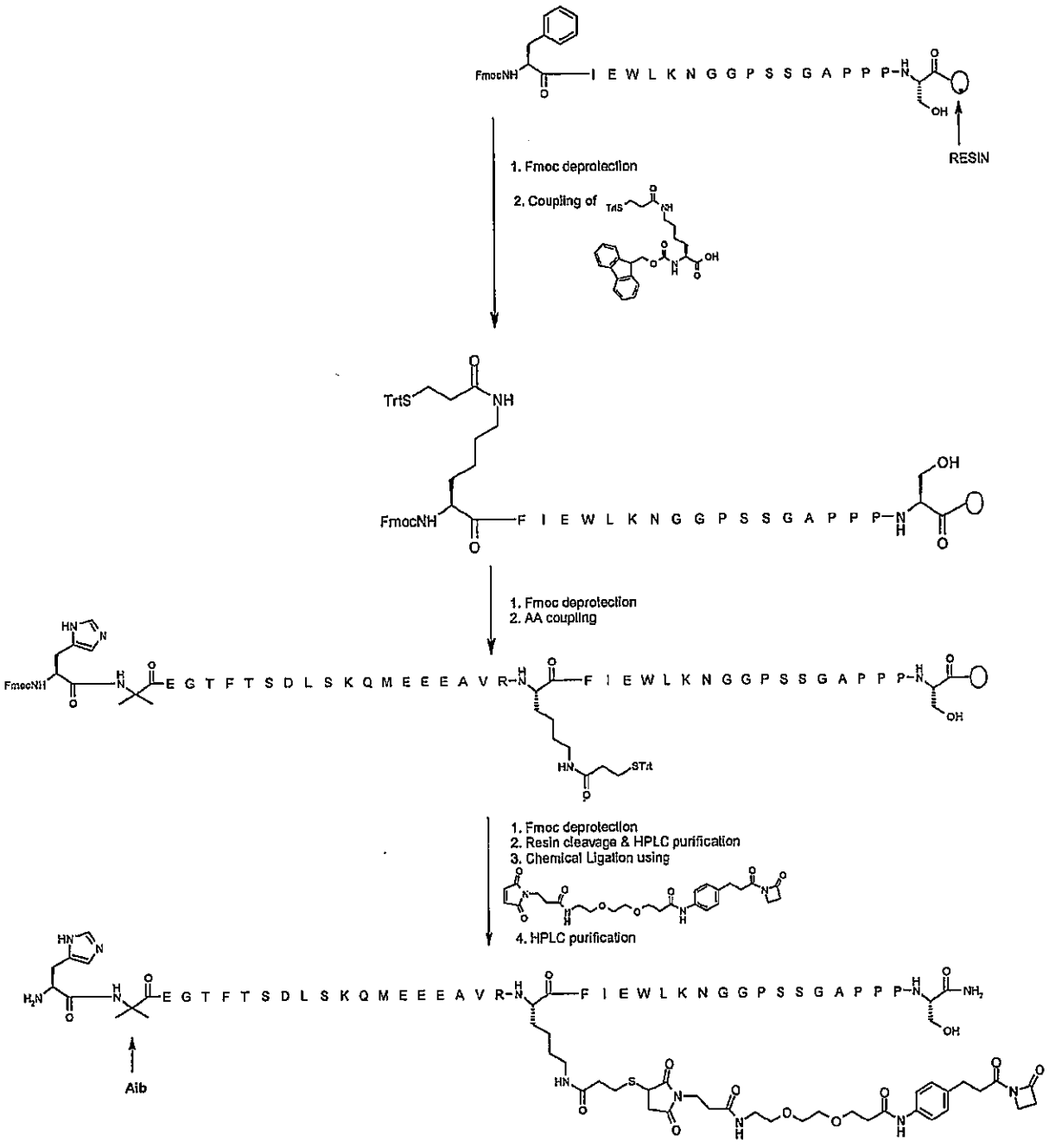
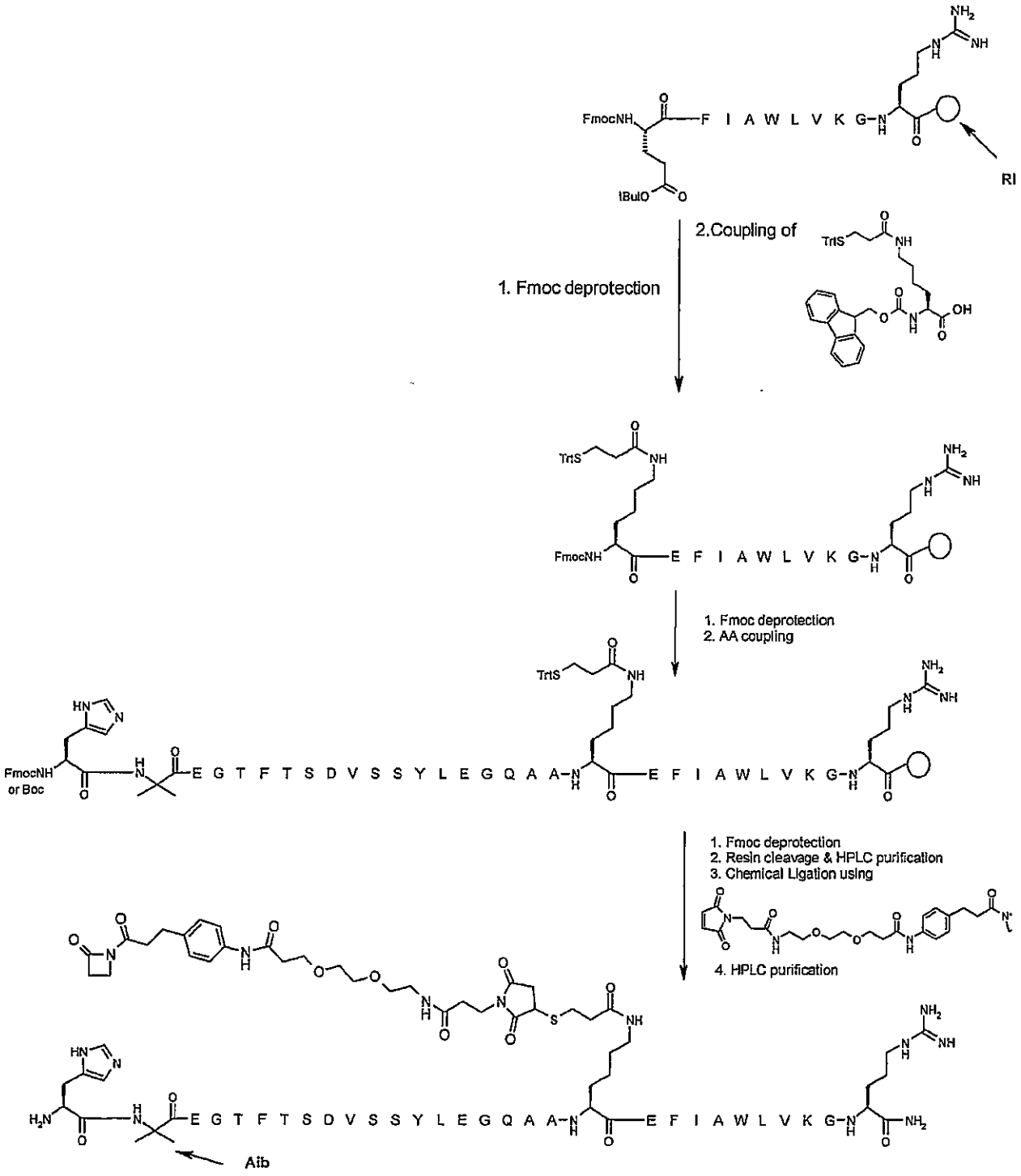

FmocNH
I E W L K N G G P S S G A P P P
OH
RESIN
1. Fmoc deprotection
2. Coupling of
Trt
TrtS
NH
OH
FmocNH
F I E W L K N G G P S S G A P P P
1. Fmoc deprotection
2. AA coupling
FmocNH
E G T F T S D L S K Q M E E E A V R
F I E W L K N G G P S S G A P P P
HN
STrt
1. Fmoc deprotection
2. Resin cleavage & HPLC purification
3. Chemical Ligation using
4. HPLC purification
E G T F T S D L S K Q M E E E A V R
F I E W L K N G G P S S G A P P P
NH2
Aib

FIGURE 32


FmocNH
F I A W L V K G-N
RI
tBuO
1. Fmoc deprotection
2.Coupling of
TrtS
OH
FmocNH
E F I A W L V K G-N
1. Fmoc deprotection
2. AA coupling
FmocNH or Boc
E G T F T S D V S S Y L E G Q A A-N
1. Fmoc deprotection
2. Resin cleavage & HPLC purification
3. Chemical Ligation using
4. HPLC purification
Aib

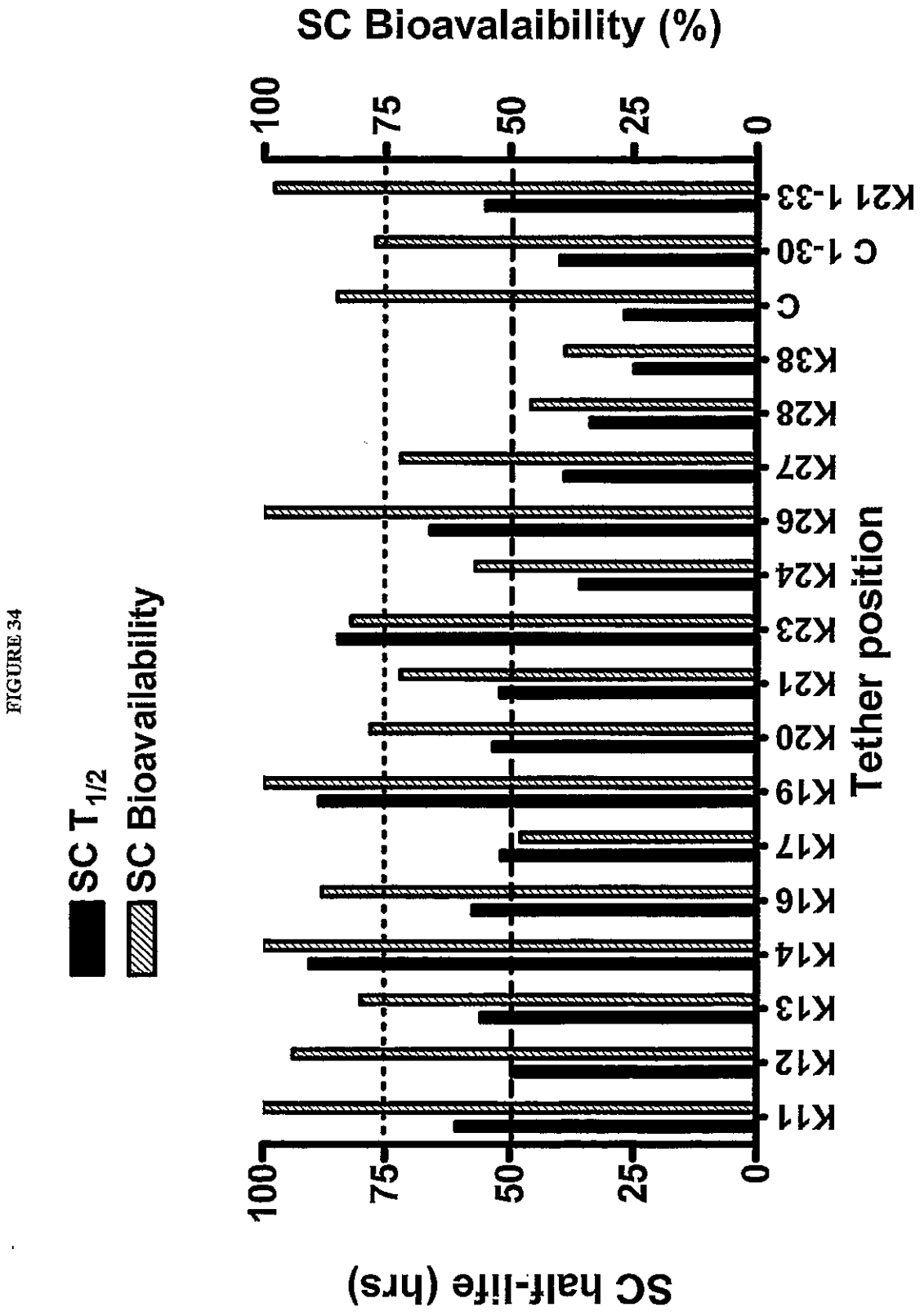

SC Bioavalaibility (%)
100
75
50
25
0
K21 1-33
C 1-30
C
K38
K28
K27
K26
K24
K23
K21
K20
K19
K17
K16
K14
K13
K12
K11
Tether position
SC half-life (hrs)
SC T1/2
SC Bioavailability

FIGURE 34

FIGURE 35 a

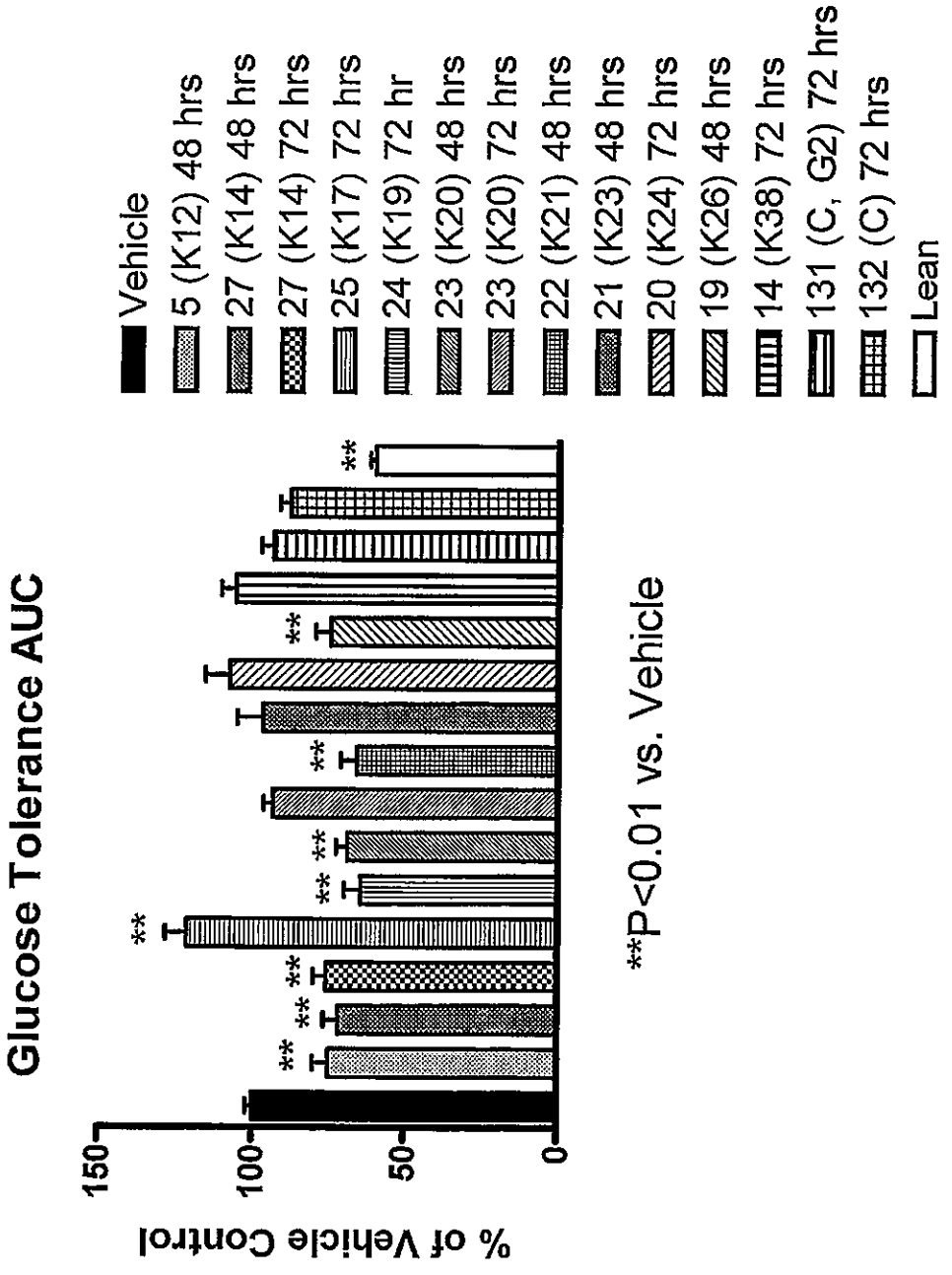

Glucose Tolerance AUC
% of Vehicle Control
150
100
50
0
**P<0.01 vs. Vehicle
Vehicle
5 (K12) 48 hrs
27 (K14) 48 hrs
27 (K14) 72 hrs
25 (K17) 72 hrs
24 (K19) 72 hr
23 (K20) 48 hrs
23 (K20) 72 hrs
22 (K21) 48 hrs
21 (K23) 48 hrs
20 (K24) 72 hrs
19 (K26) 48 hrs
14 (K38) 72 hrs
131 (C, G2) 72 hrs
132 (C) 72 hrs
Lean

FIGURE 35 b

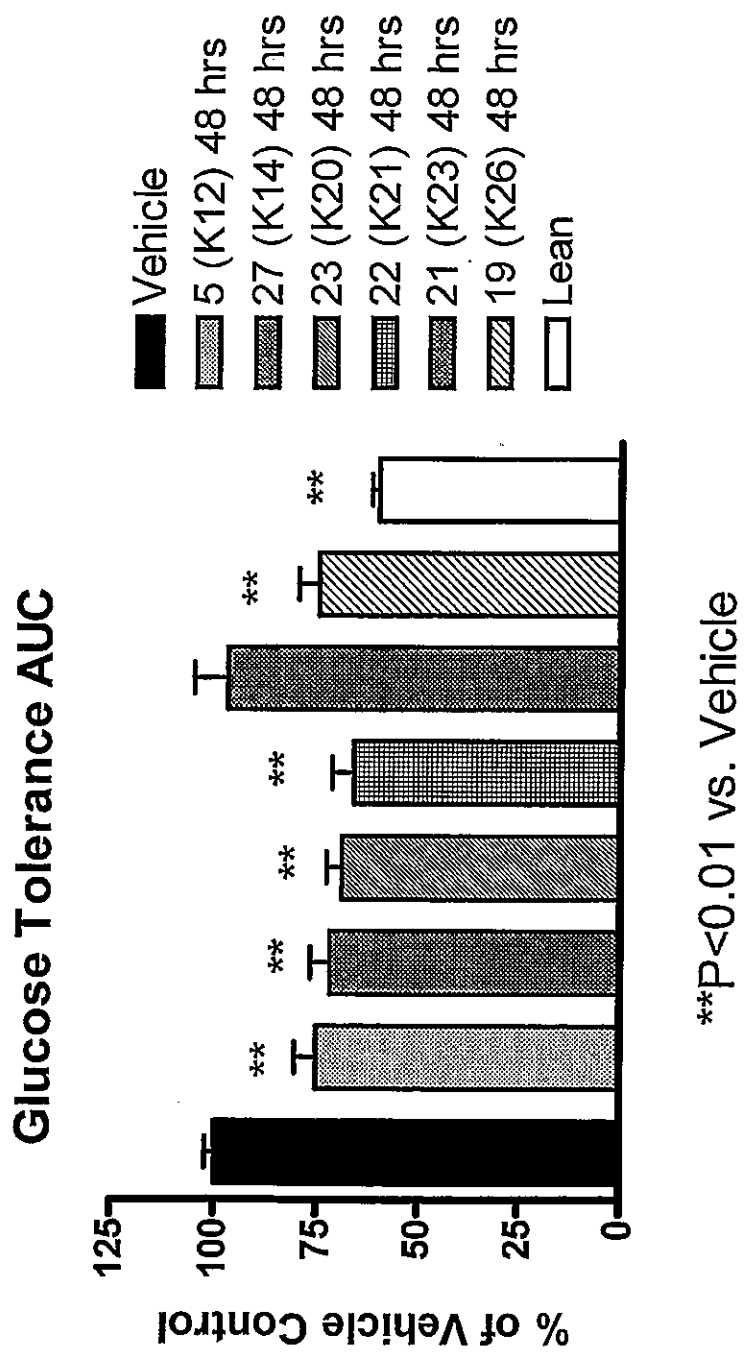

Glucose Tolerance AUC
% of Vehicle Control
125
100
75
50
25
0
**
**
**
**
**
**
Vehicle
5 (K12) 48 hrs
27 (K14) 48 hrs
23 (K20) 48 hrs
22 (K21) 48 hrs
21 (K23) 48 hrs
19 (K26) 48 hrs
Lean
**P<0.01 vs. Vehicle

FIGURE 35 c

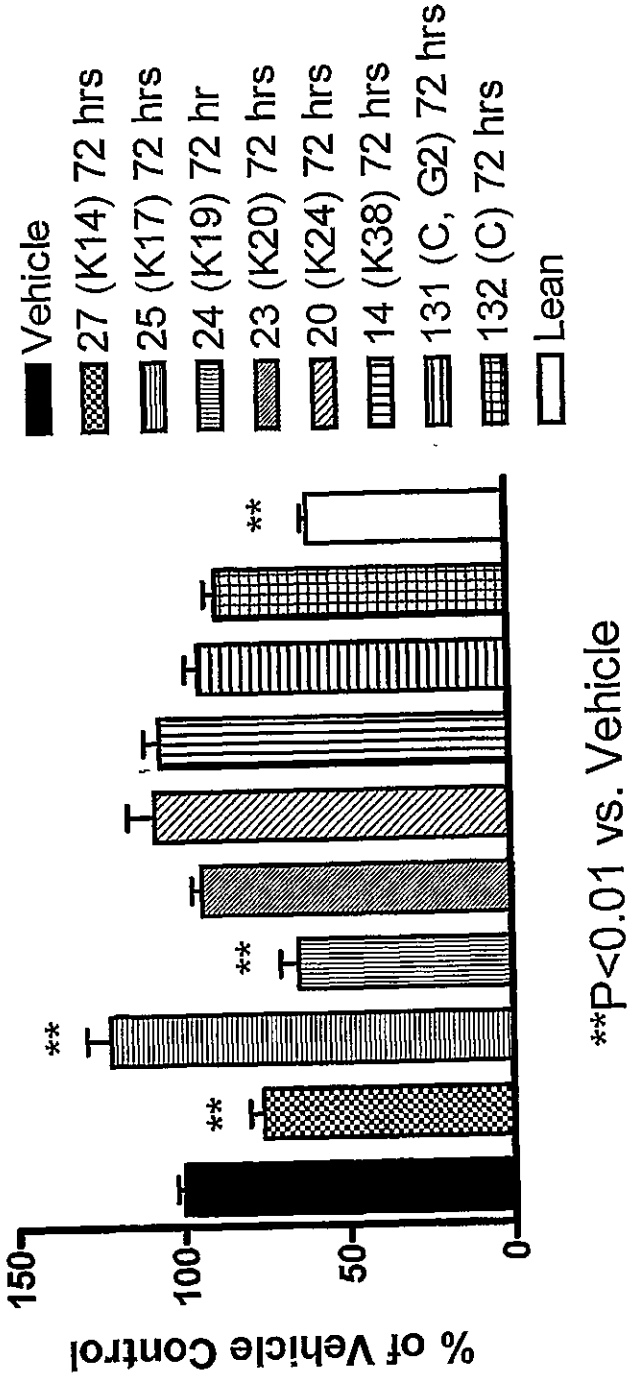

Glucose Tolerance AUC
% of Vehicle Control
150
100
50
0
**
**
**
**
Vehicle
27 (K14) 72 hrs
25 (K17) 72 hrs
24 (K19) 72 hr
23 (K20) 72 hrs
20 (K24) 72 hrs
14 (K38) 72 hrs
131 (C, G2) 72 hrs
132 (C) 72 hrs
Lean
**P<0.01 vs. Vehicle

FIGURE 36 a

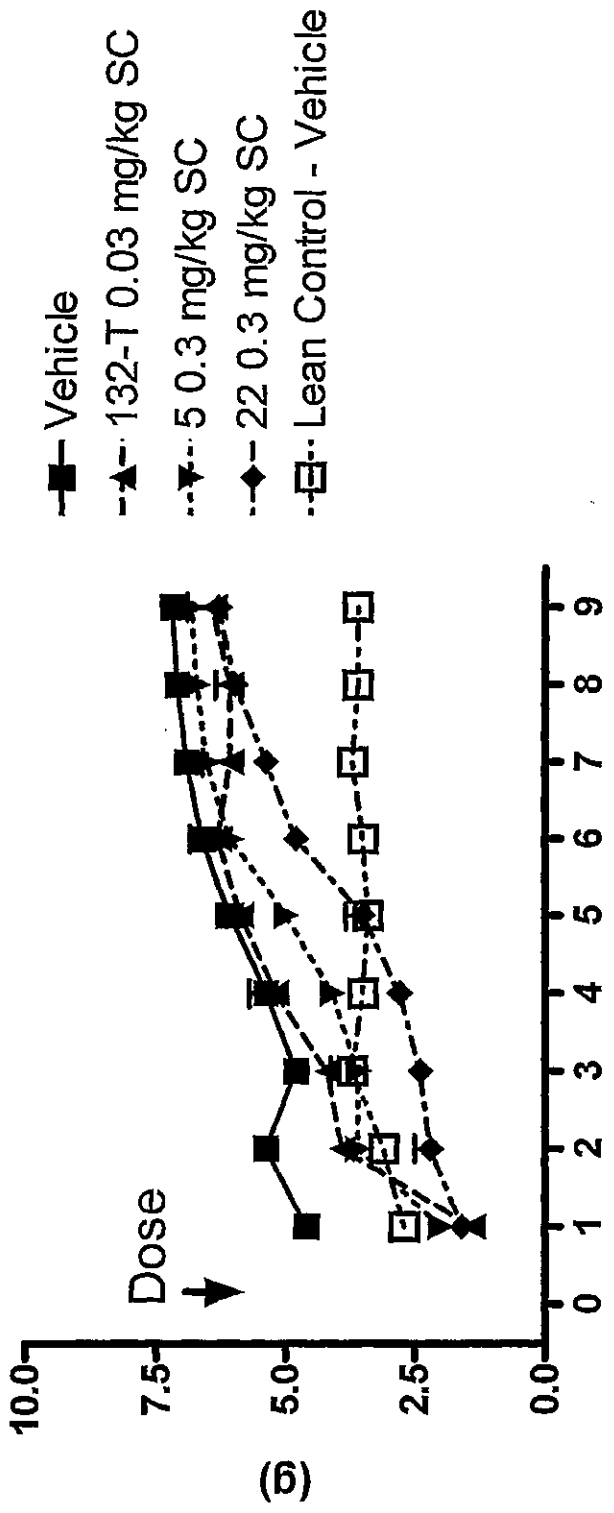

Vehicle
132-T 0.03 mg/kg SC
5 0.3 mg/kg SC
22 0.3 mg/kg SC
Lean Control - Vehicle
Dose
10.0
7.5
5.0
2.5
0.0
0 1 2 3 4 5 6 7 8 9
(б)

FIGURE 36 b

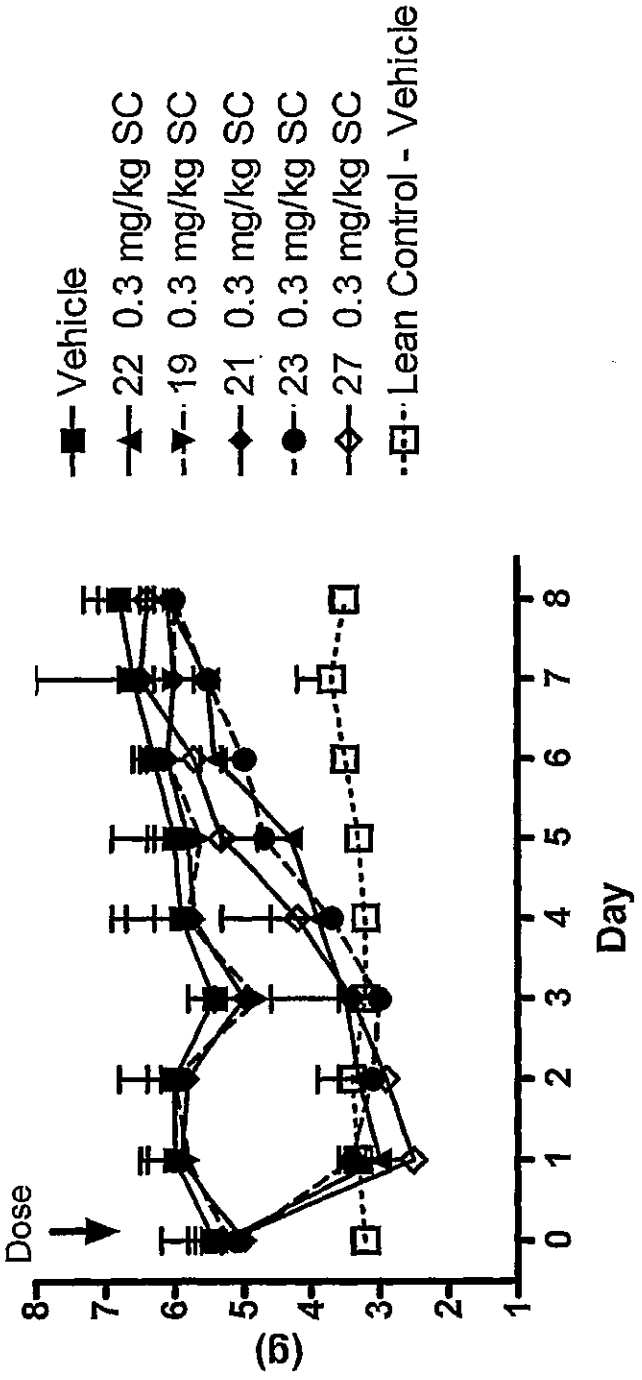

Vehicle
22 0.3 mg/kg SC
19 0.3 mg/kg SC
21 0.3 mg/kg SC
23 0.3 mg/kg SC
27 0.3 mg/kg SC
Lean Control - Vehicle
Dose
Day
(g)
8
7
6
5
4
3
2
1
0
1
2
3
4
5
6
7
8

FIGURE 36 c

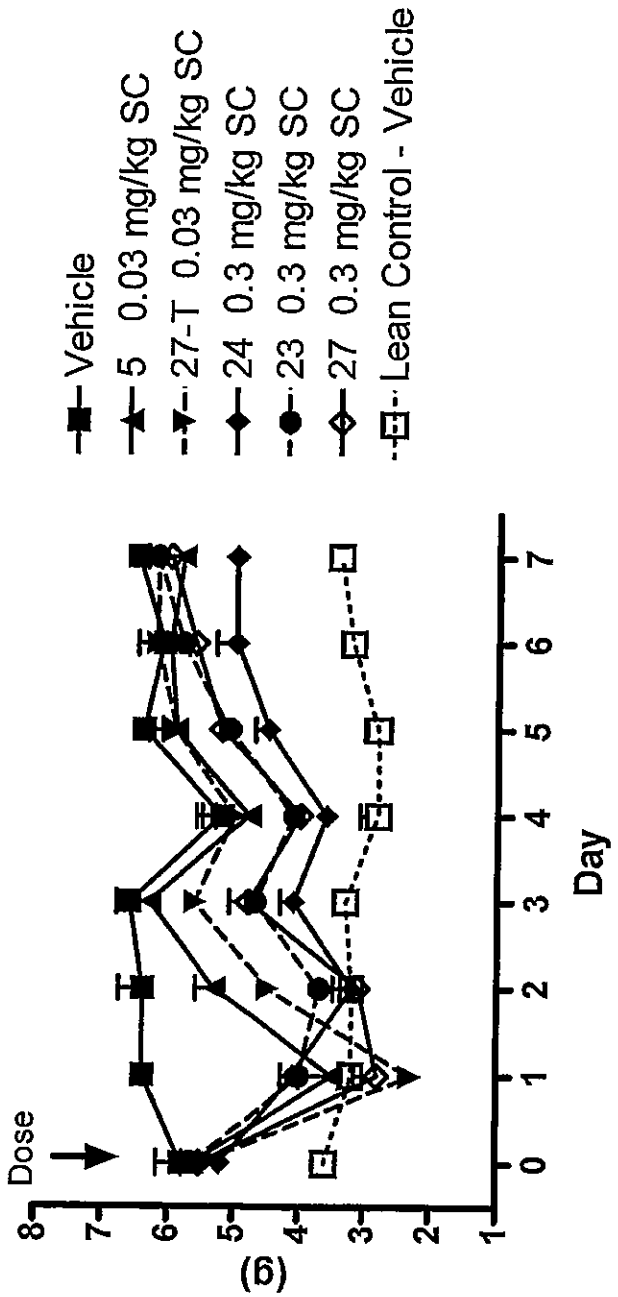

Vehicle
5 0.03 mg/kg SC
27-T 0.03 mg/kg SC
24 0.3 mg/kg SC
23 0.3 mg/kg SC
27 0.3 mg/kg SC
Lean Control - Vehicle
Dose
Day
(б)

FIGURE 36 d

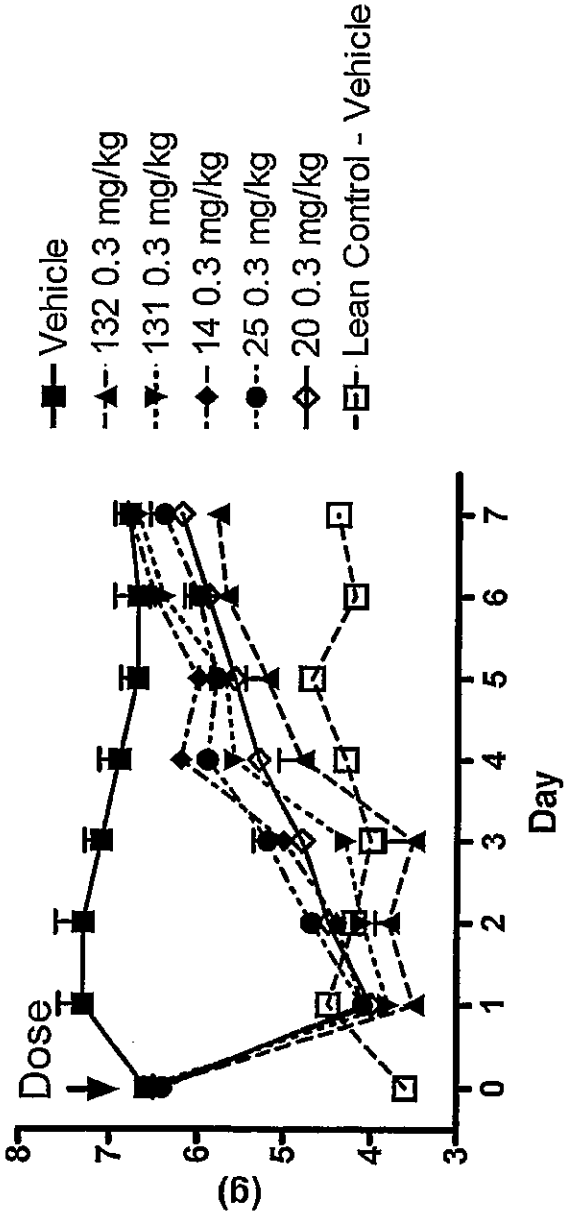

Vehicle
132 0.3 mg/kg
131 0.3 mg/kg
14 0.3 mg/kg
25 0.3 mg/kg
20 0.3 mg/kg
Lean Control - Vehicle
Dose
Day
(g)
8
7
6
5
4
3
0
1
2
3
4
5
6
7

FIGURE 37 a

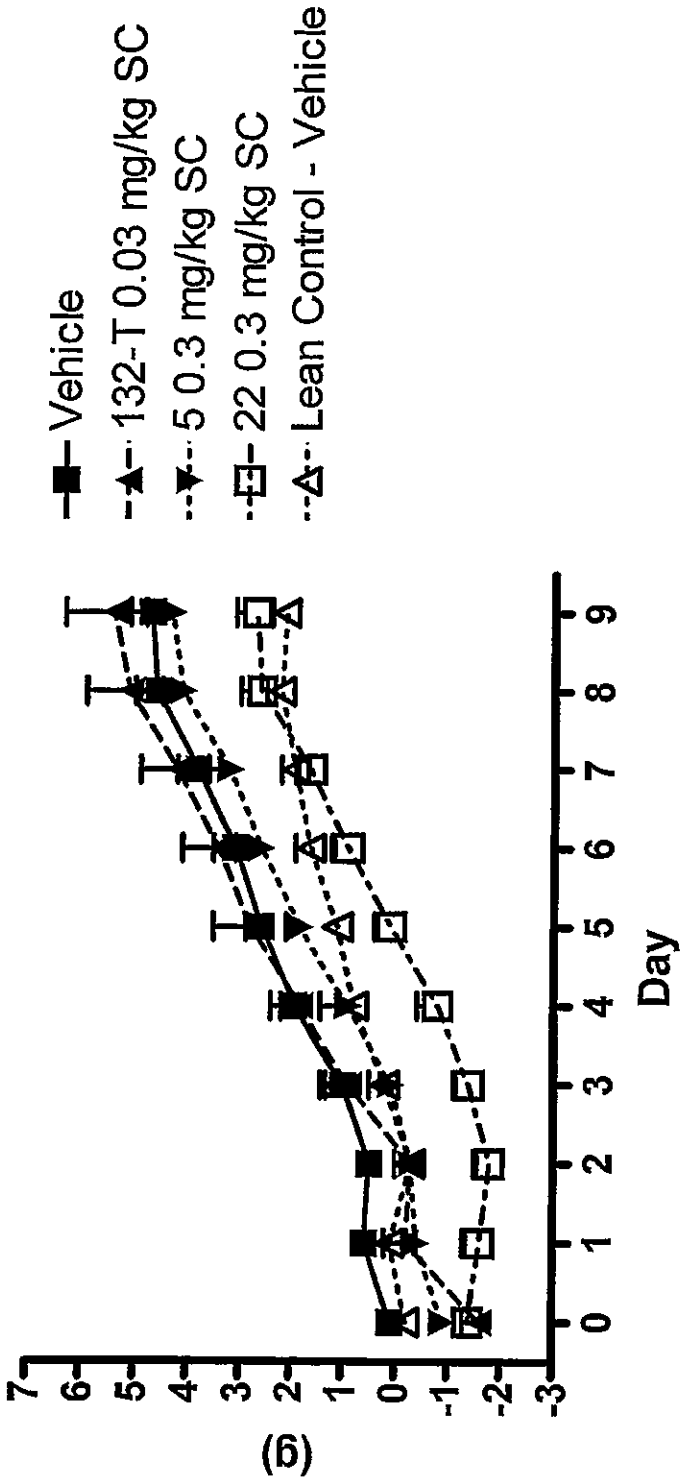

Vehicle
132-T 0.03 mg/kg SC
5 0.3 mg/kg SC
22 0.3 mg/kg SC
Lean Control - Vehicle
7
6
5
4
3
2
1
0
-1
-2
-3
0 1 2 3 4 5 6 7 8 9
Day
(б)

FIGURE 37 b

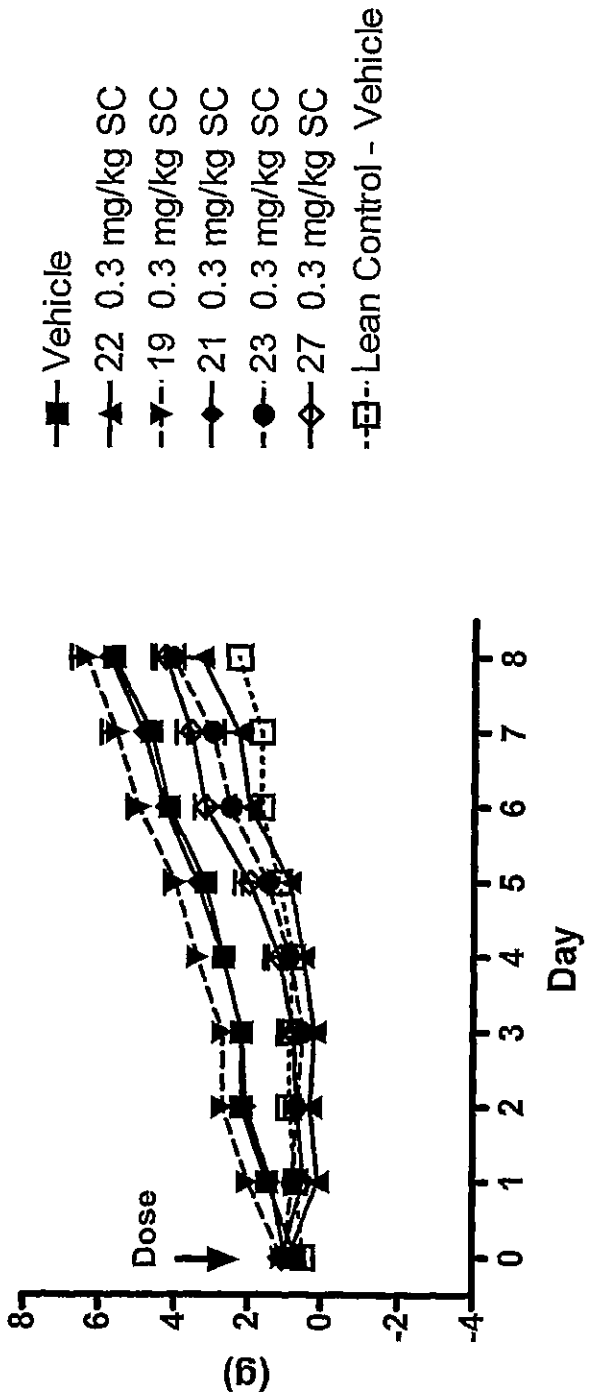

Vehicle
22 0.3 mg/kg SC
19 0.3 mg/kg SC
21 0.3 mg/kg SC
23 0.3 mg/kg SC
27 0.3 mg/kg SC
Lean Control - Vehicle
Dose
Day
8
6
4
2
0
-2
-4
0 1 2 3 4 5 6 7 8
(б)

FIGURE 37 c

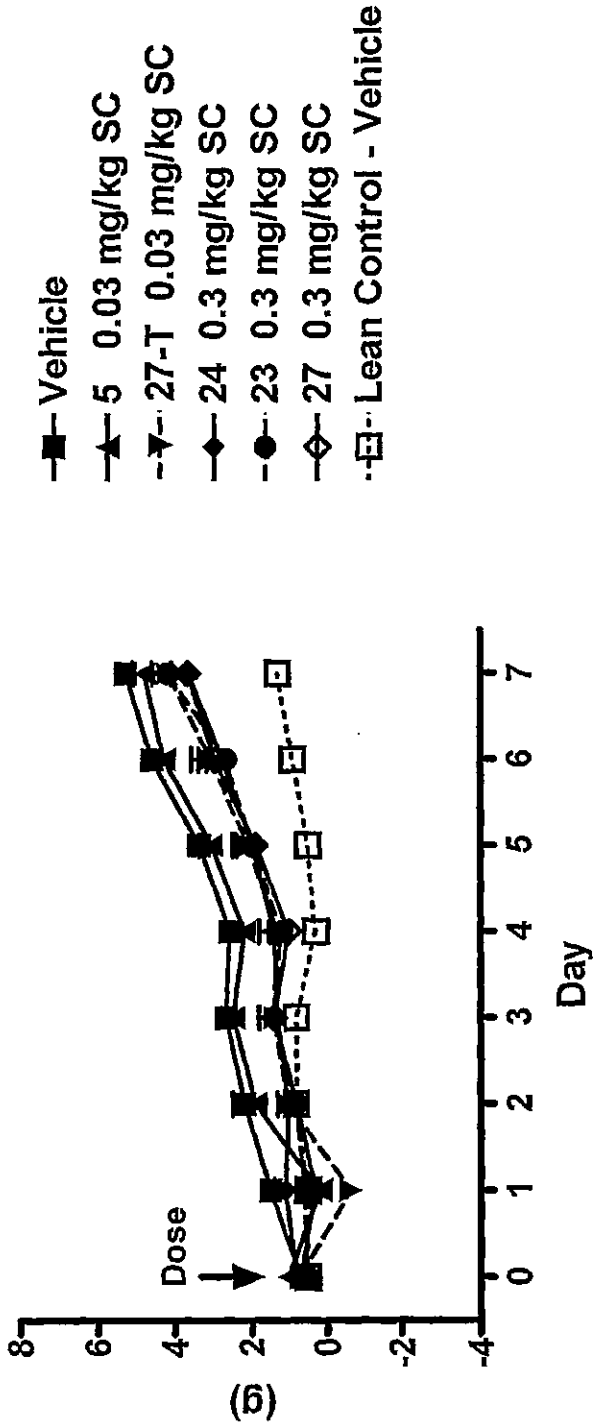

Vehicle
5 0.03 mg/kg SC
27-T 0.03 mg/kg SC
24 0.3 mg/kg SC
23 0.3 mg/kg SC
27 0.3 mg/kg SC
Lean Control - Vehicle
Dose
Day
8
6
4
2
0
-2
-4
0
1
2
3
4
5
6
7
(б)

FIGURE 37 d

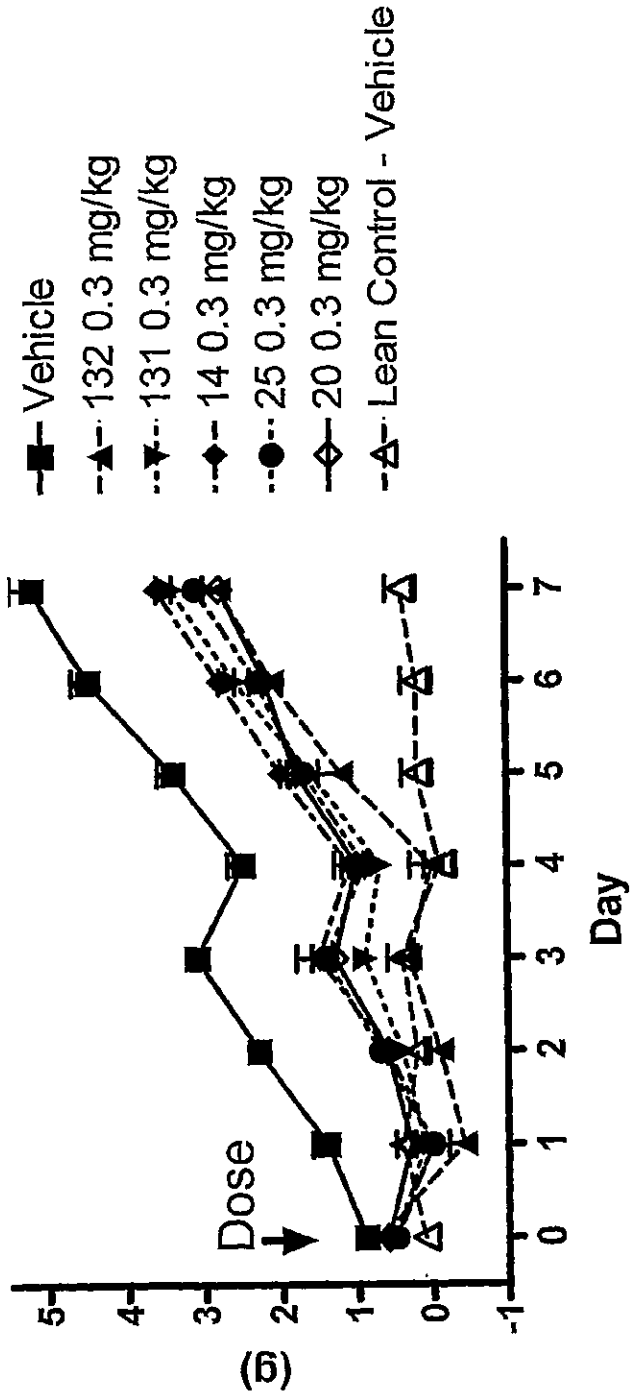

Vehicle
132 0.3 mg/kg
131 0.3 mg/kg
14 0.3 mg/kg
25 0.3 mg/kg
20 0.3 mg/kg
Lean Control - Vehicle
Dose
Day
(б)

FIGURE 38a

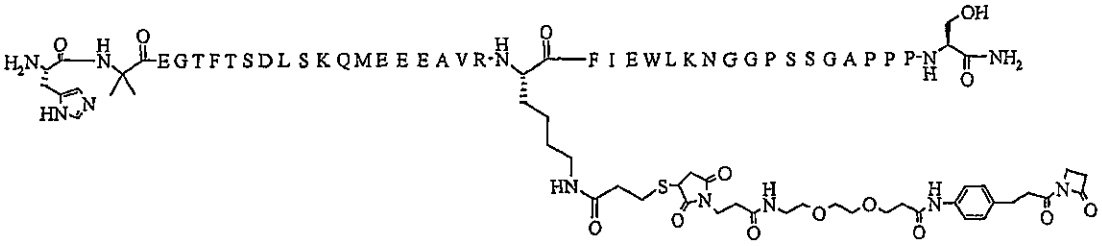


FIGURE 38b

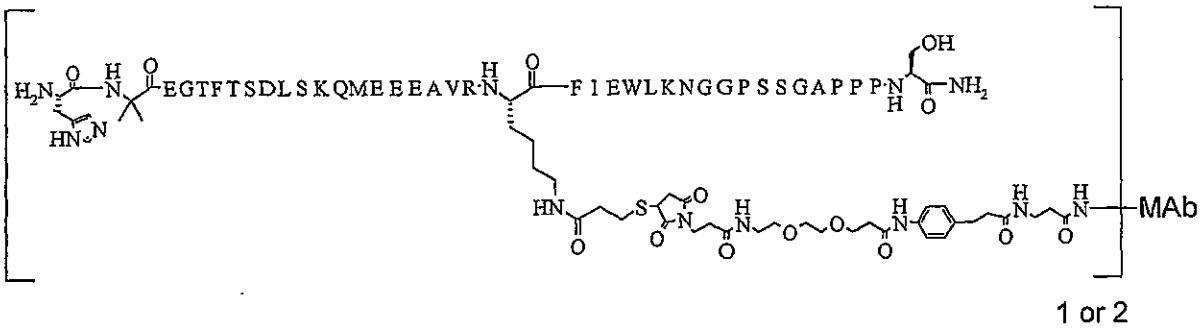


FIGURE 39a

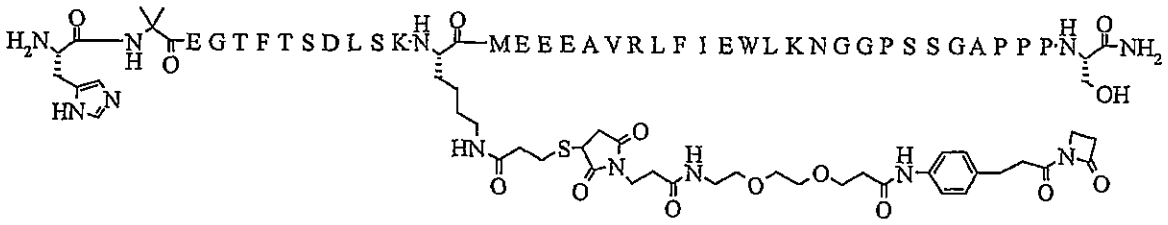


FIGURE 39b

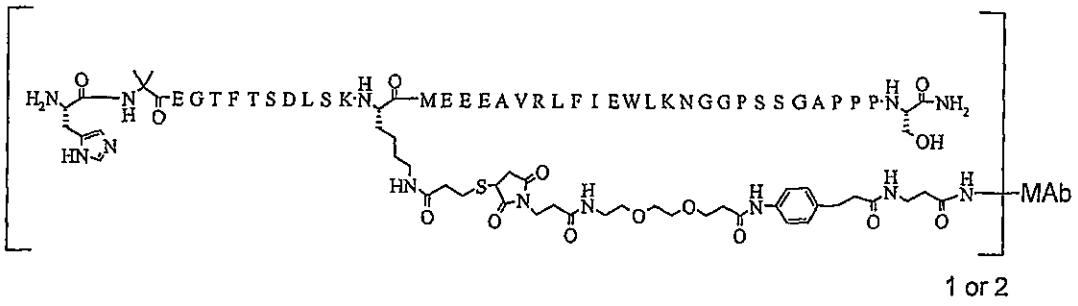


FIGURE 40a

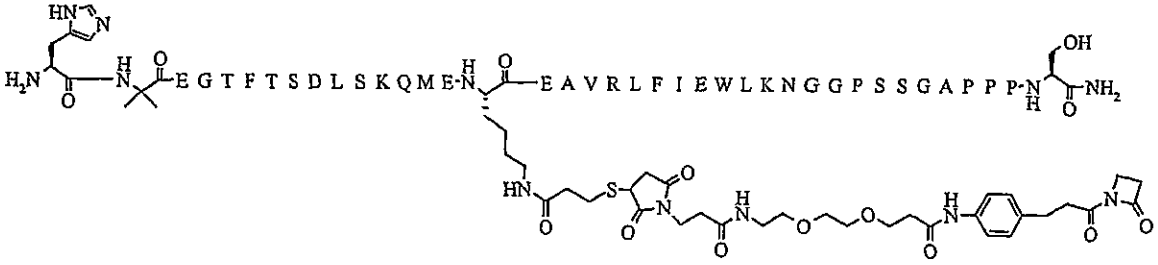


FIGURE 40b

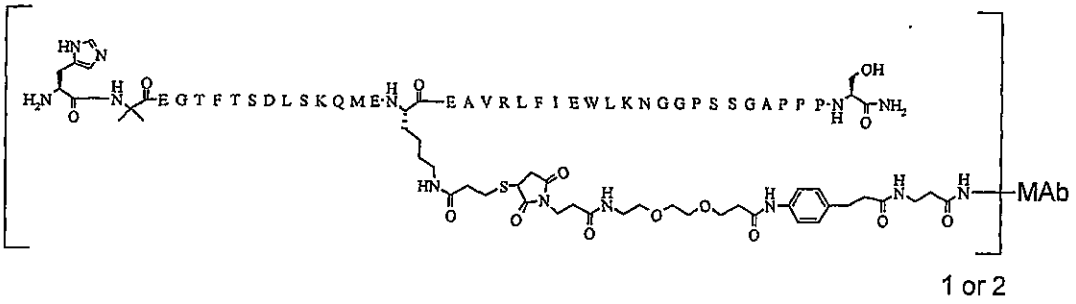


FIGURE 41a

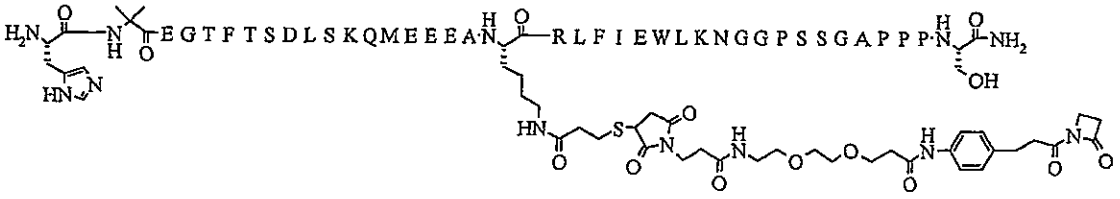


FIGURE 41b

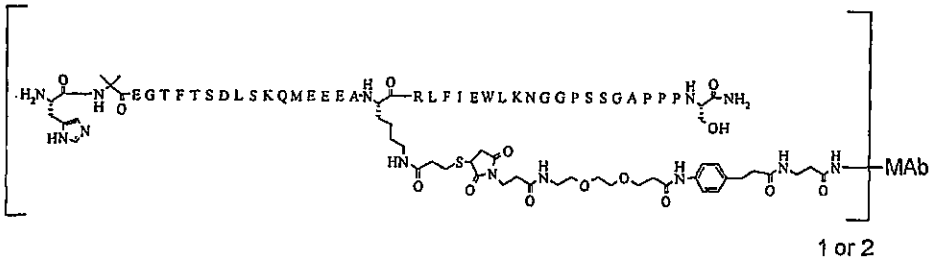


FIGURE 42a

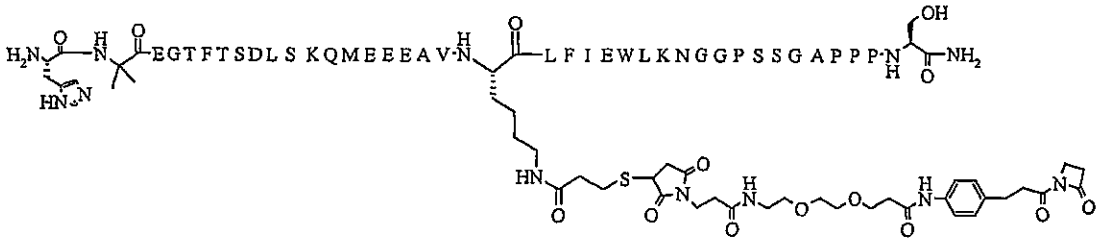


FIGURE 42b

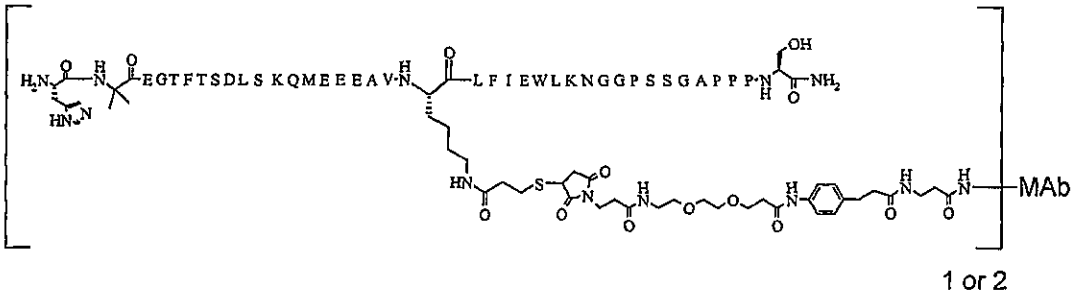


FIGURE 43a

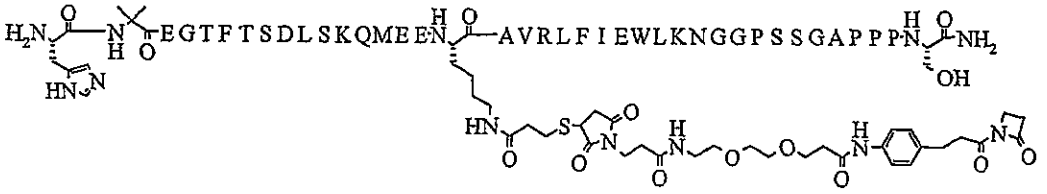


FIGURE 43b

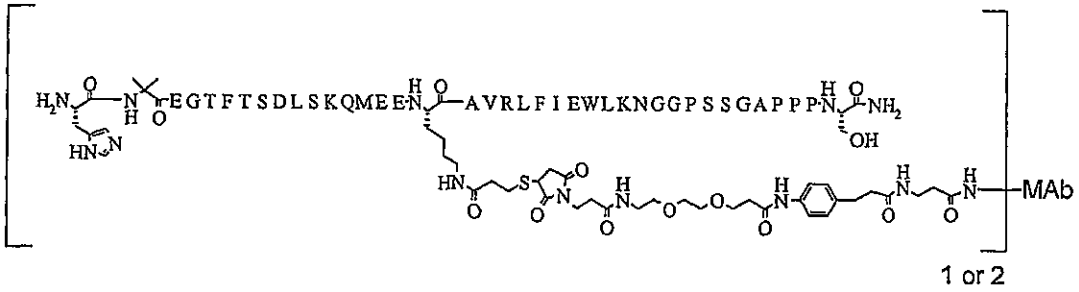


FIGURE 44a

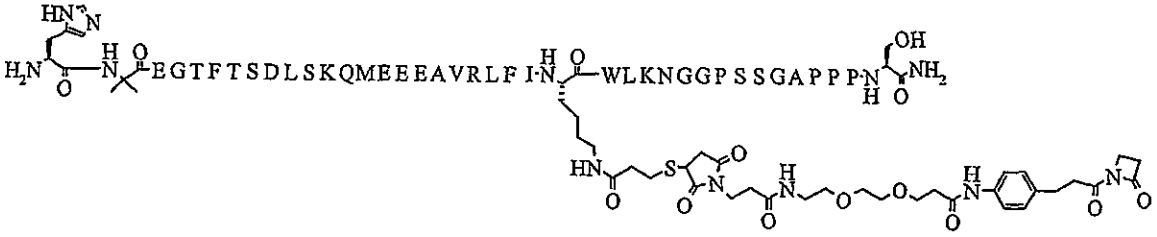


FIGURE 44b

