Fig. 1

T P L G P A S S L P Q S F L L K
aca ccc ctg ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag

C L E Q V R K I Q G D G A A L Q
tgc tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag

E K L C A T Y K L C H P E E L V
gag aag ctg tgt gcc acc tac aag ctg tgc cac ccc gag gag ctg gtg

L L G H S L G I P W A P L S S C
ctg ctc gga cac tct ctg ggc atc ccc tgg gct ccc ctg agc tcc tgc

P S Q A L Q L A G C L S Q L H S
ccc agc cag gcc ctg cag ctg gca ggc tgc ttg agc caa ctc cat agc

G L F L Y Q G L L Q A L E G I S
ggc ctt ttc ctc tac cag ggg ctc ctg cag gcc ctg gaa ggg ata tcc

P E L G P T L D T L Q L D V A D
ccc gag ttg ggt ccc acc ttg gac aca ctg cag ctg gac gtc gcc gac

F A T T I W Q Q M E E L G M A P
ttt gcc acc acc atc tgg cag cag atg gaa gaa ctg gga atg gcc cct

A L Q P T Q G A M P A F A S A F
gcc ctg cag ccc acc cag ggt gcc atg ccg gcc ttc gcc tct gct ttc

Q R R A G G V L V A S H L Q S F
cag cgc cgg gca gga ggg gtc ctg gtt gct agc cat ctg cag agc ttc

L E V S Y R V L R H L A Q P
ctg gag gtg tcg tac cgc gtt cta cgc cac ctt gcg cag ccc

Fig. 2

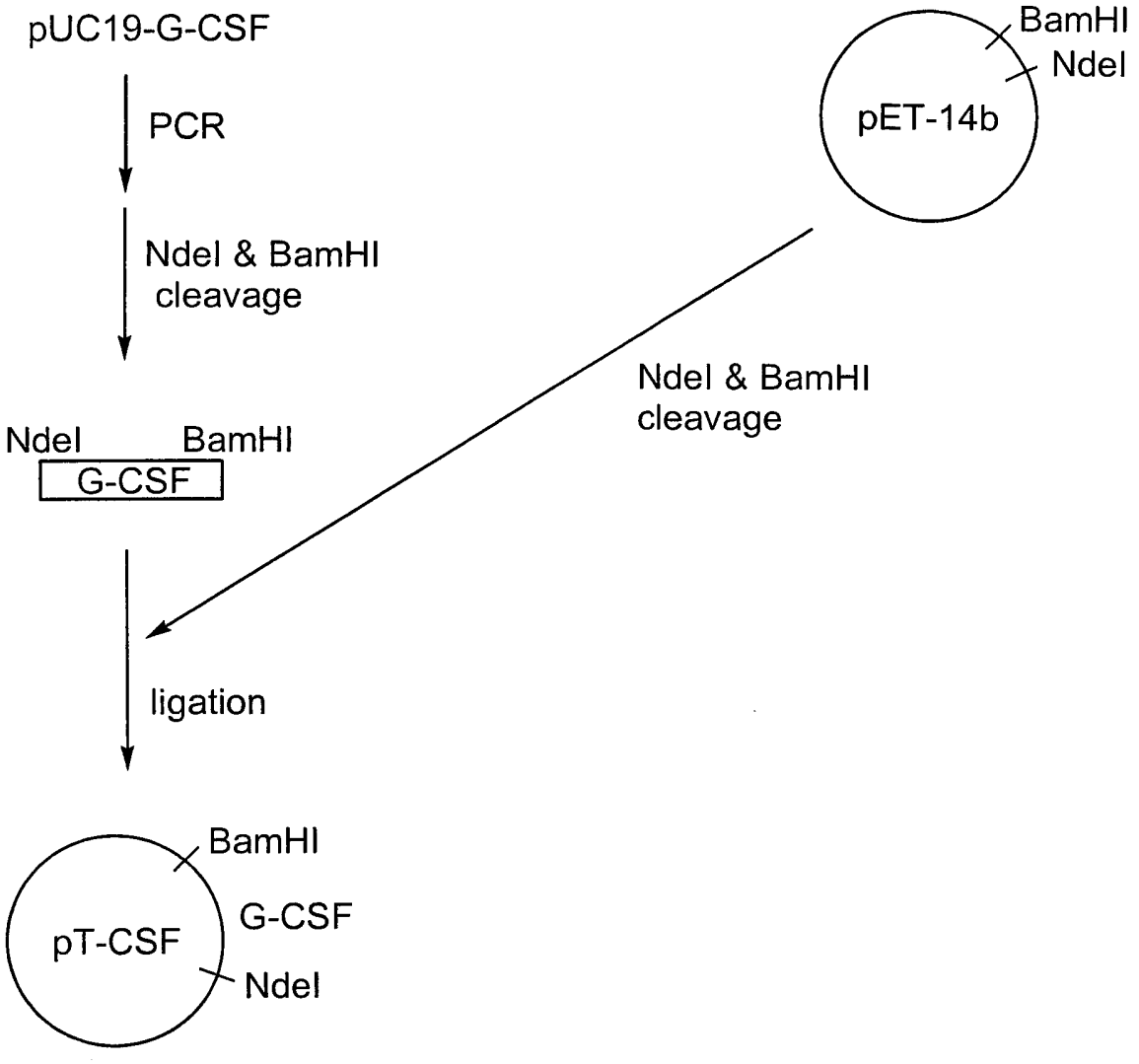

pUC19-G-CSF
PCR
NdeI & BamHI
cleavage
NdeI
BamHI
G-CSF
pET-14b
BamHI
NdeI
NdeI & BamHI
cleavage
ligation
pT-CSF
BamHI
G-CSF
NdeI

Fig. 3

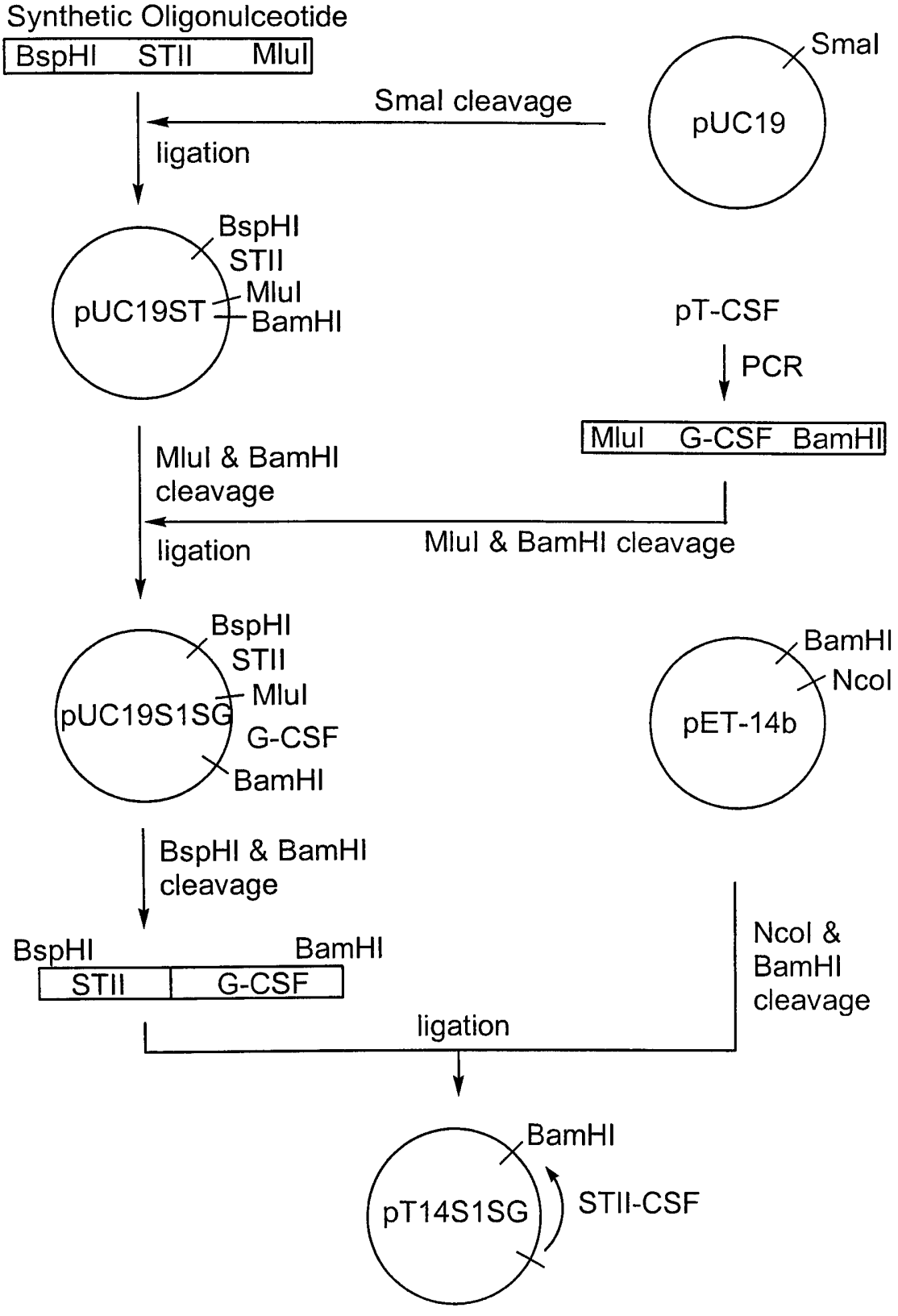

Synthetic Oligonulceotide
BspHI STII MluI
SmaI cleavage
ligation
SmaI
pUC19
BspHI
STII
MluI
pUC19ST
BamHI
pT-CSF
PCR
MluI G-CSF BamHI
MluI & BamHI cleavage
MluI & BamHI cleavage
ligation
BspHI
STII
MluI
pUC19S1SG
G-CSF
BamHI
BamHI
NcoI
pET-14b
BspHI & BamHI cleavage
BspHI
BamHI
STII
G-CSF
NcoI & BamHI cleavage
ligation
BamHI
pT14S1SG
STII-CSF

Fig. 4

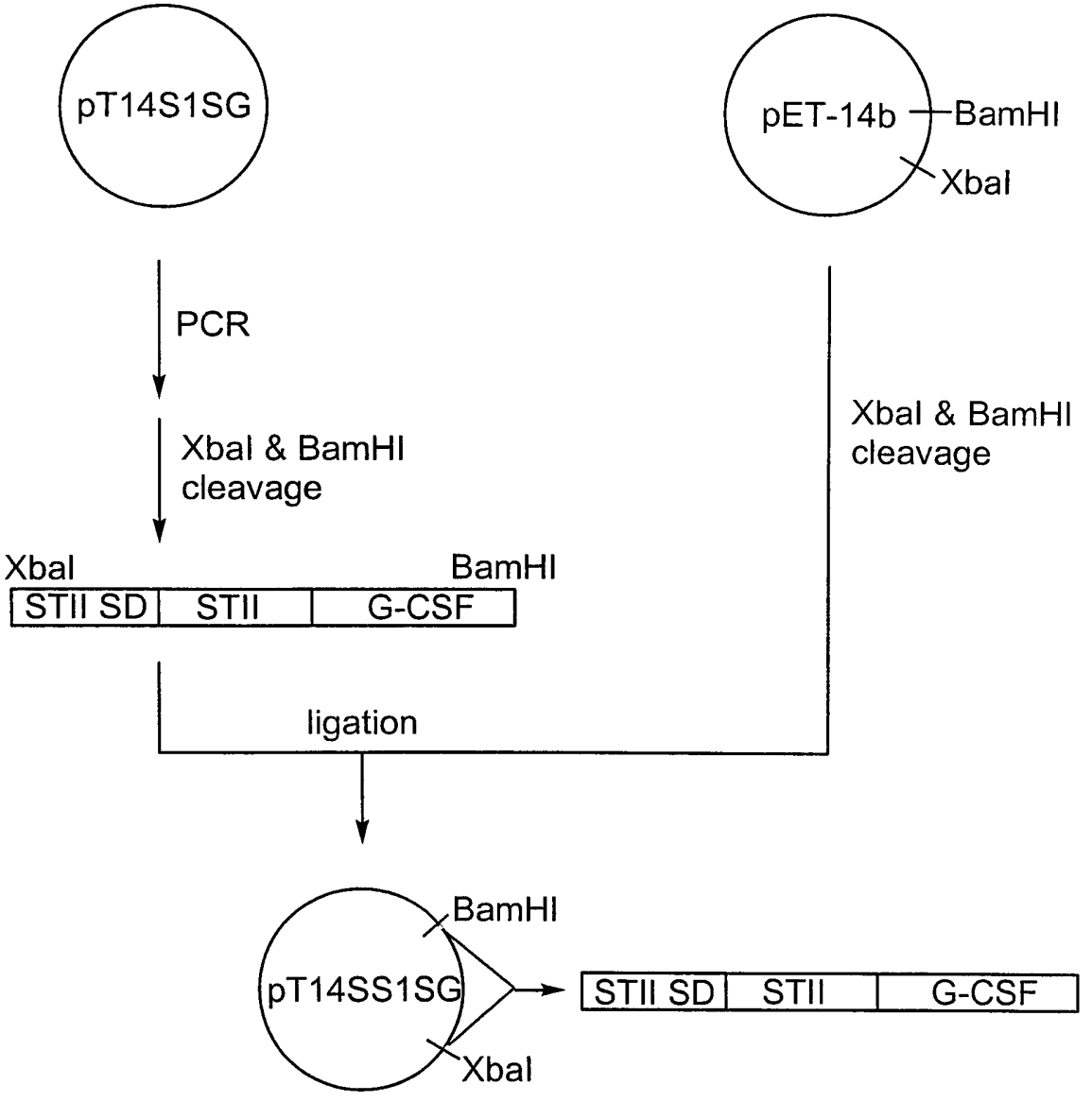

pT14S1SG
pET-14b
BamHI
XbaI
PCR
XbaI & BamHI cleavage
XbaI & BamHI cleavage
XbaI
BamHI
STII SD
STII
G-CSF
ligation
BamHI
pT14SS1SG
XbaI
STII SD
STII
G-CSF

Fig. 5

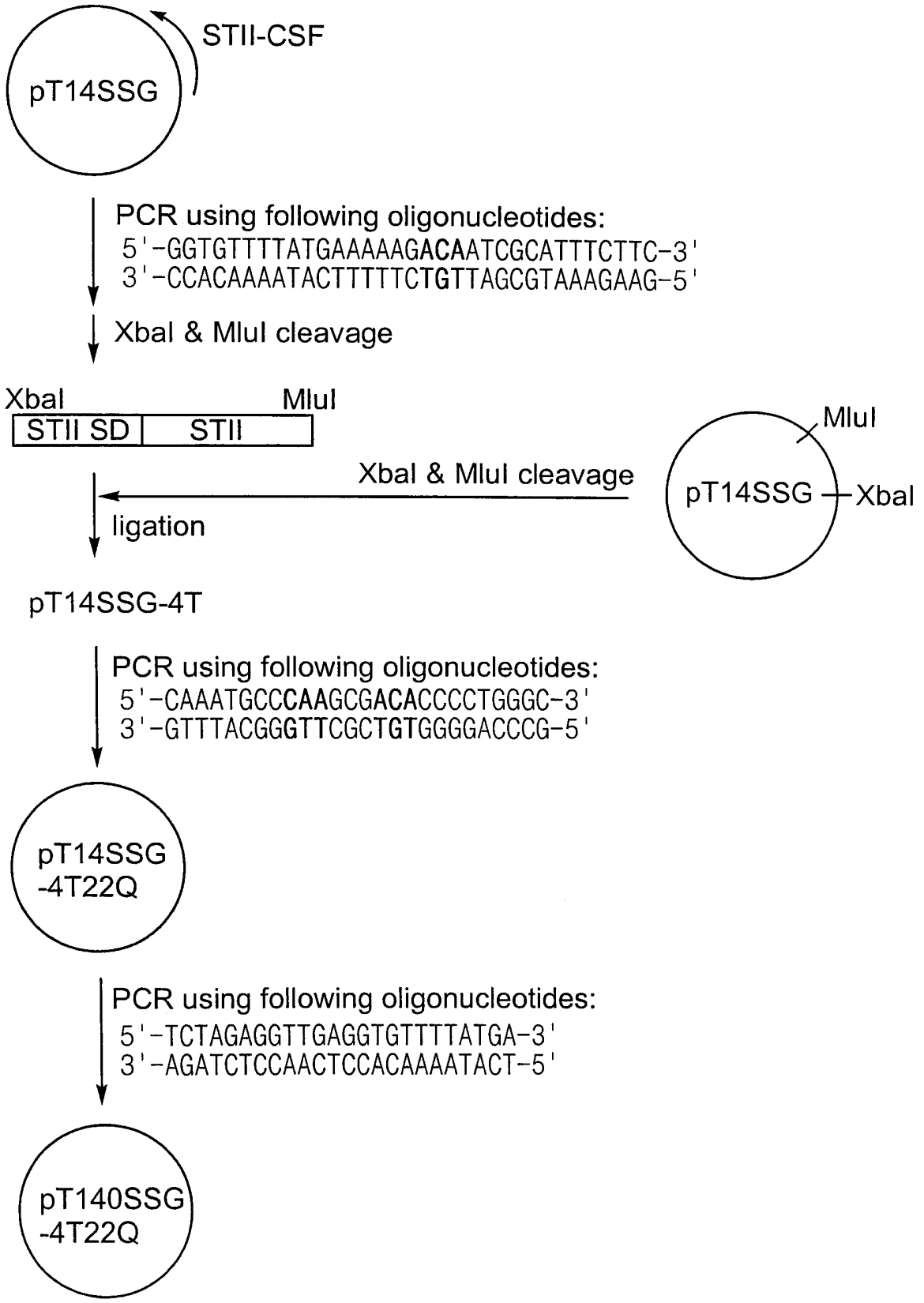

STII-CSF
pT14SSG
PCR using following oligonucleotides:
5'-GGTGTTTTATGAAAAAGACAATCGCATTTCTTC-3'
3'-CCACAAAATACTTTTTCTGTTAGCGTAAAGAAG-5'
XbaI & MluI cleavage
XbaI
MluI
STII SD
STII
XbaI & MluI cleavage
MluI
pT14SSG
XbaI
ligation
pT14SSG-4T
PCR using following oligonucleotides:
5'-CAAATGCCCAAGCGACACCCCTGGGC-3'
3'-GTTTACGGGTTCGCTGTGGGGACCCG-5'
pT14SSG
-4T22Q
PCR using following oligonucleotides:
5'-TCTAGAGGTTGAGGTGTTTTATGA-3'
3'-AGATCTCCAACTCCACAAAATACT-5'
pT140SSG
-4T22Q

Fig. 6


pT14SS1SG
G-CSF
+5
+10
+15
SerProLeuGlyProAlaSerSerLeuProGlnSerPheLeuLeuLys
TCTCCCCTGGGCCCTGCCAGCTCCCTGCCCCAGAGCTTCCTGCTCAAG
AGAGGGGACCCGGGACGGTCGAGGGACGGGGTCTCGAAGGACGAGTTC
ApaI site
+20
+25
+30
CysLeuGluGlnValArgLysIleGlnGlyAspGlyAlaAlaLeuGln
TGCTTAGAGCAAGTGAGGAAGATCCAGGGCGATGGCGCAGCGCTCCAG
ACGAATCTCGTTCACTCCTTCTAGGTCCCGCTACCGCGTCGCGAGGTC
BstXI site
ApaI & BstXI cleavage
followed by recovery of vector fragment
annealing oligonucleotides
5'-CAGCCTCTTCTCTTCCACAATCTTTCCTTCTTAAGTCTCTTGAACAA
3'-CCGGGTCGGAGAAGAGAAGGTGTTAGAAAGGAAGAATTCAGAGAACTTGTT
GTTAGAAAGATCCAAGGCG-3'
CAATCTTTCTAGGTT-5'
ligation
pT14SS1S17SEG

Fig. 7

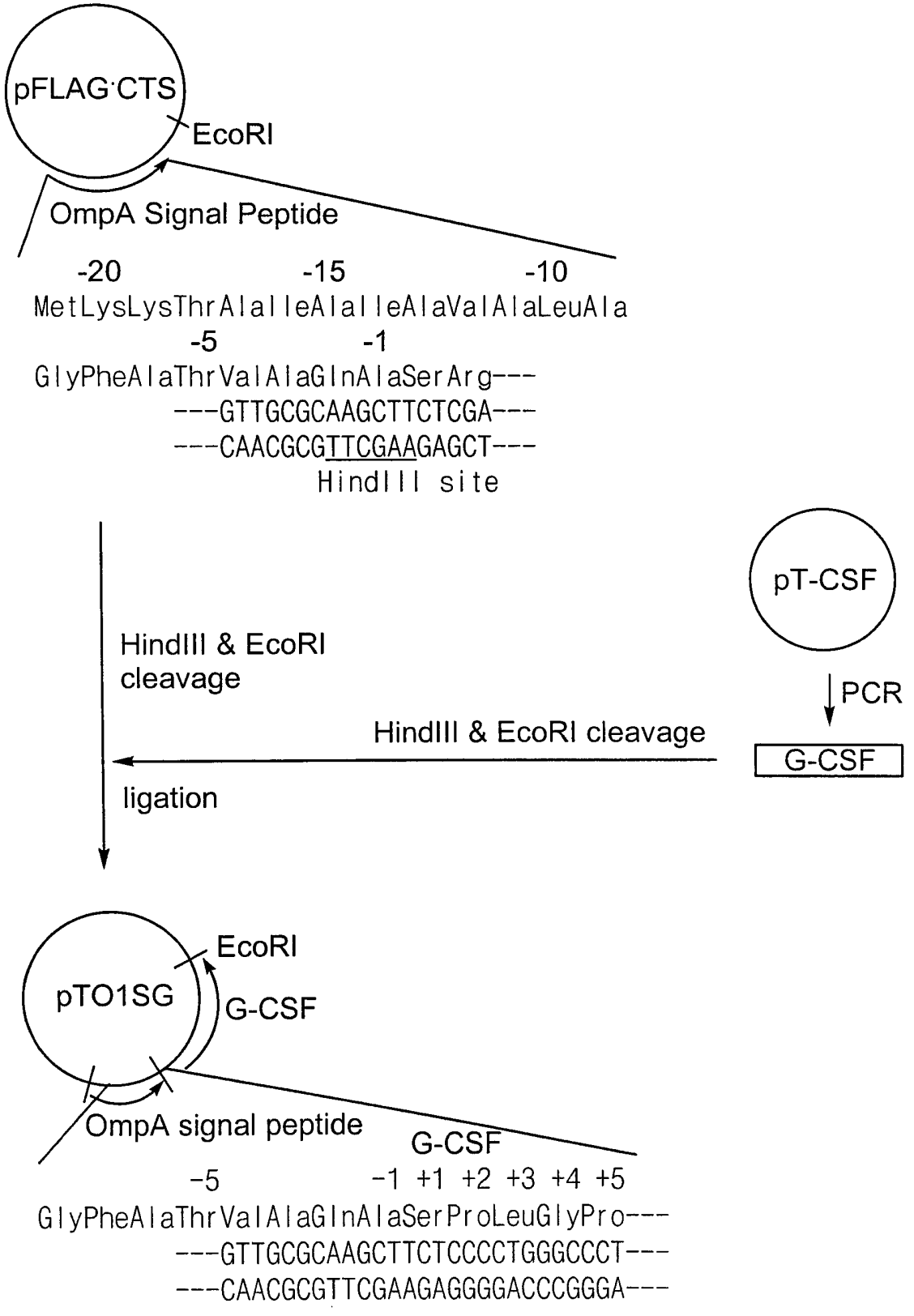

pFLAG·CTS
EcoRI
OmpA Signal Peptide
-20 -15 -10
MetLysLysThrAlaIleAlaIleAlaValAlaLeuAla
-5 -1
GlyPheAlaThrValAlaGlnAlaSerArg---
---GTTGCGCAAGCTTCTCGA---
---CAACGCGTTCGAAGAGCT---
HindIII site
pT-CSF
PCR
G-CSF
HindIII & EcoRI cleavage
HindIII & EcoRI cleavage
ligation
pTO1SG
EcoRI
G-CSF
OmpA signal peptide
G-CSF
-5 -1 +1 +2 +3 +4 +5
GlyPheAlaThrValAlaGlnAlaSerProLeuGlyPro---
---GTTGCGCAAGCTTCTCCCCTGGGCCCT---
---CAACGCGTTCGAAGAGGGGACCCGGGA---

Fig. 8

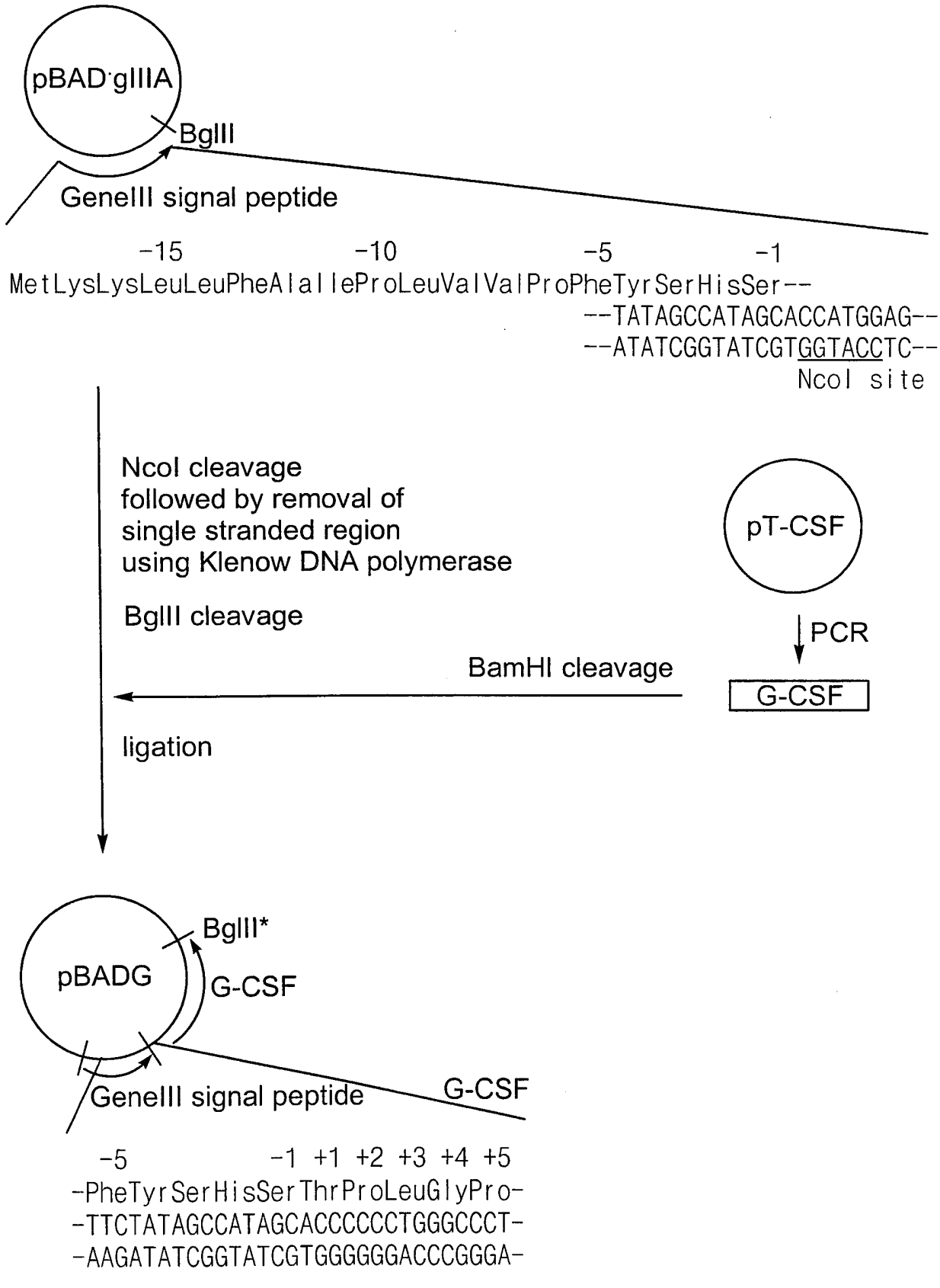

pBAD·gIIIA
BglII
GeneIII signal peptide
-15 -10 -5 -1
MetLysLysLeuLeuPheAlaIleProLeuValValProPheTyrSerHisSer--
--TATAGCCATAGCACCATGGAG--
--ATATCGGTATCGTGGTACCTC--
NcoI site
NcoI cleavage
followed by removal of
single stranded region
using Klenow DNA polymerase
BglII cleavage
pT-CSF
PCR
BamHI cleavage
G-CSF
ligation
BglII*
pBADG
G-CSF
GeneIII signal peptide
G-CSF
-5 -1 +1 +2 +3 +4 +5
-PheTyrSerHisSerThrProLeuGlyPro-
-TTCTATAGCCATAGCACCCCCCTGGGCCCT-
-AAGATATCGGTATCGTGGGGGGACCCGGGA-

Fig. 9

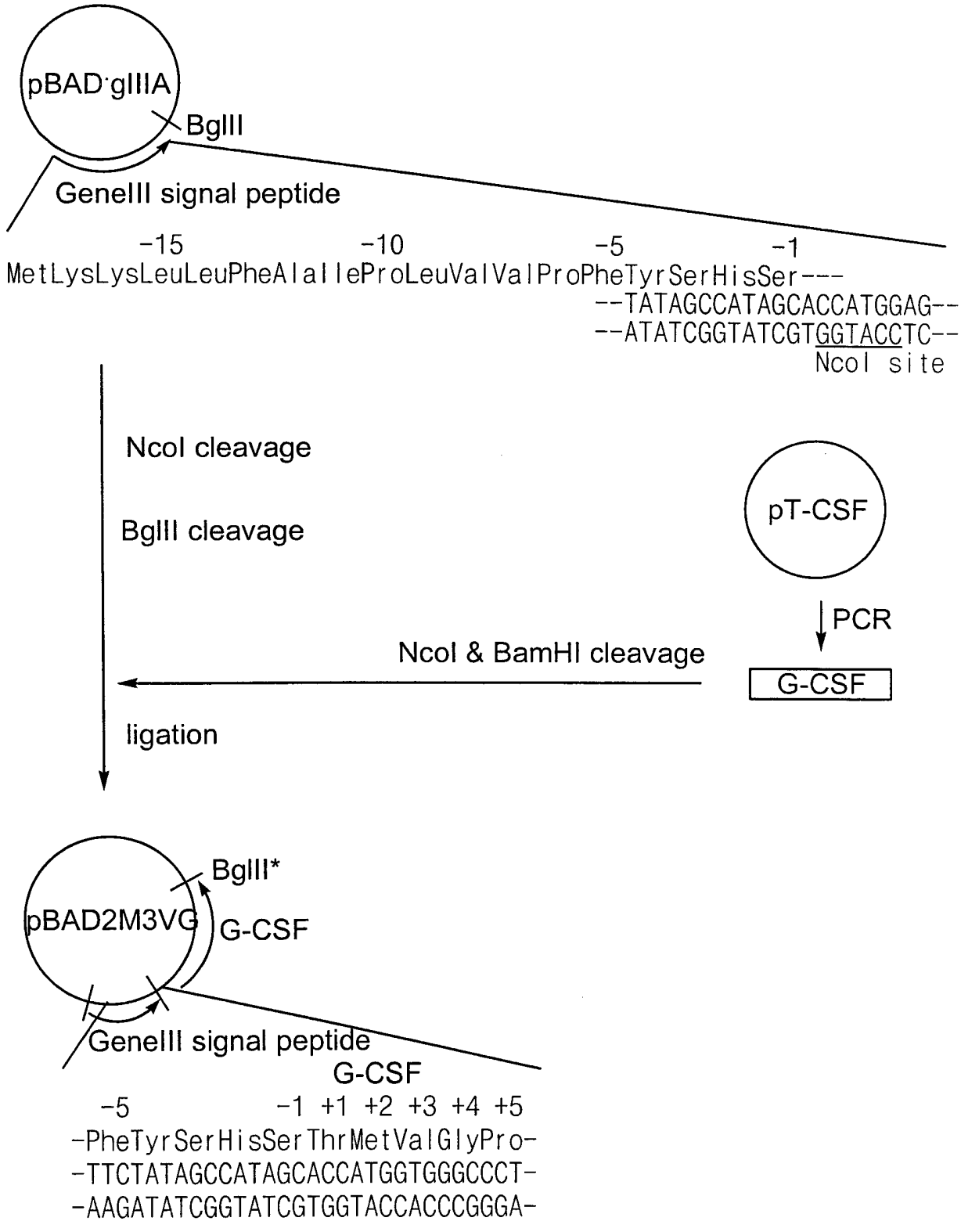

pBAD·gIIIA
BglII
GeneIII signal peptide
-15 -10 -5 -1
MetLysLysLeuLeuPheAlaIleProLeuValValProPheTyrSerHisSer---
--TATAGCCATAGCACCATGGAG--
--ATATCGGTATCGTGGTACCTC--
NcoI site
NcoI cleavage
BglII cleavage
pT-CSF
PCR
NcoI & BamHI cleavage
G-CSF
ligation
BglII*
pBAD2M3VG
G-CSF
GeneIII signal peptide
G-CSF
-5 -1 +1 +2 +3 +4 +5
-PheTyrSerHisSerThrMetValGlyPro-
-TTCTATAGCCATAGCACCATGGTGGGCCCT-
-AAGATATCGGTATCGTGGTACCACCCGGGA-

Fig. 10A

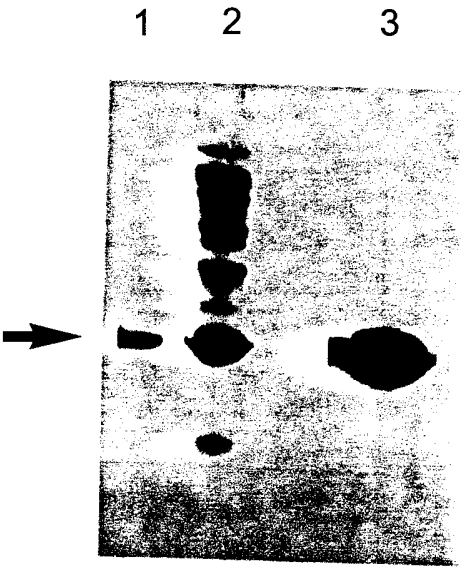


Fig. 10B

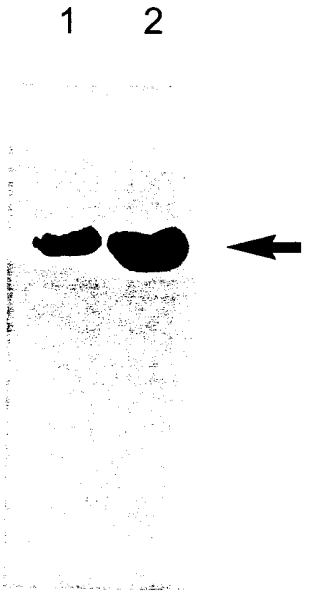
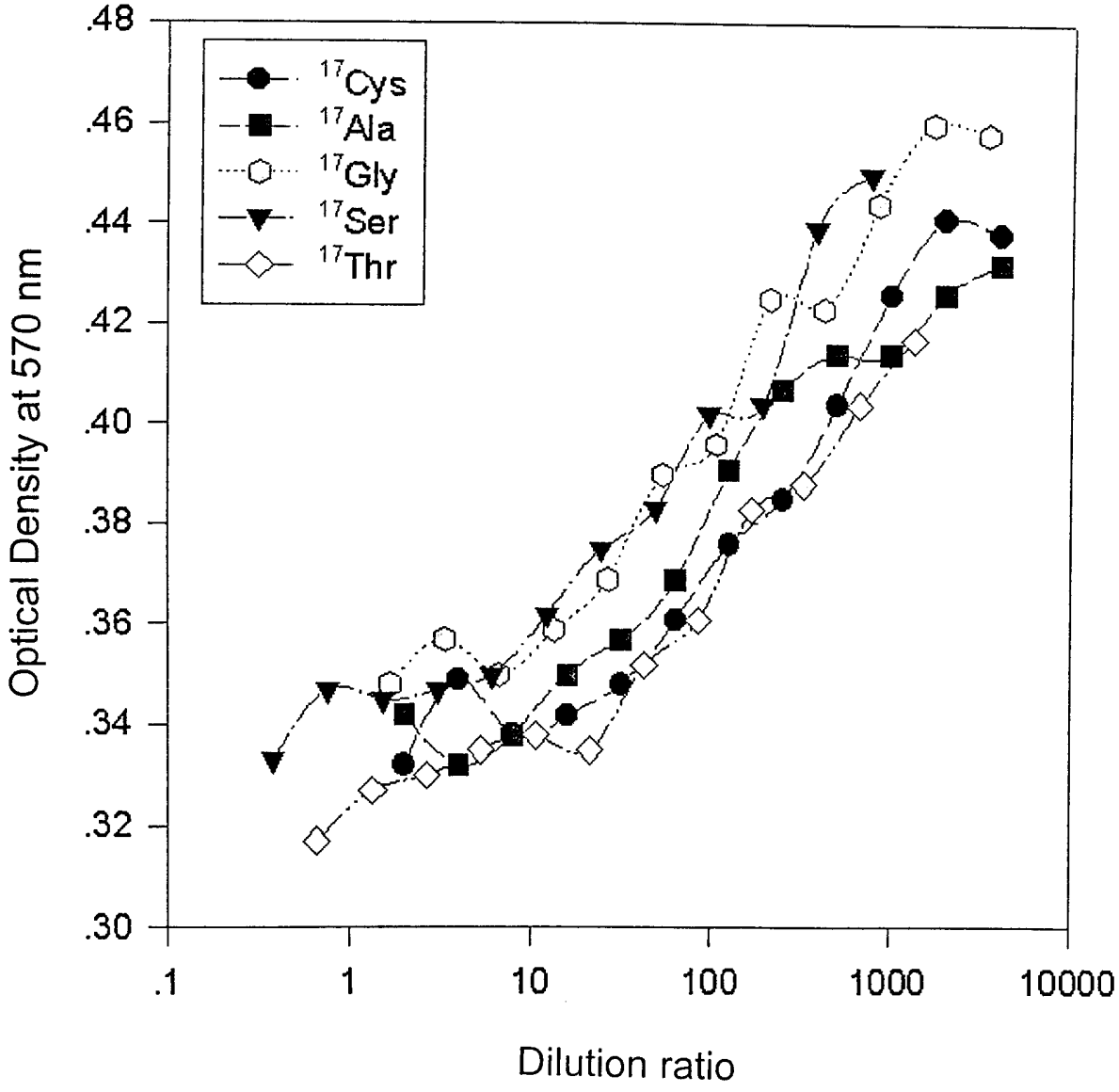


Fig. 11


.48
.46
.44
.42
.40
.38
.36
.34
.32
.30
Optical Density at 570 nm
^{17}Cys
^{17}Ala
^{17}Gly
^{17}Ser
^{17}Thr
.1
1
10
100
1000
10000
Dilution ratio

Sequence Listing

<110> Pharm. Co., Ltd.

<120> Modified human granulocyte-colony stimulating factor and process for producing same

<130> PCA00729/HMY

<160> 71

<170> KOPATIN 1.0

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<211> 522
<212> DNA
<213> *Homo sapiens*

<220>
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<222> (1)..(522)

<400> 1

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  1               5                  10                  15

tgc tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag    96
Cys Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
            20                  25                  30

gag aag ctg tgt gcc acc tac aag ctg tgc cac ccc gag gag ctg gtg   144
Glu Lys Leu Cys Ala Thr Tyr Lys Leu Cys His Pro Glu Glu Leu Val
        35                  40                  45

ctg ctc gga cac tct ctg ggc atc ccc tgg gct ccc ctg agc tcc tgc   192
Leu Leu Gly His Ser Leu Gly Ile Pro Trp Ala Pro Leu Ser Ser Cys
    50                  55                  60

ccc agc cag gcc ctg cag ctg gca ggc tgc ttg agc caa ctc cat agc   240
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Pro Ser Gln Ala Leu Gln Leu Ala Gly Cys Leu Ser Gln Leu His Ser
65 70 75 80

ggc ctt ttc ctc tac cag ggg ctc ctg cag gcc ctg gaa ggg ata tcc 288
Gly Leu Phe Leu Tyr Gln Gly Leu Leu Gln Ala Leu Glu Gly Ile Ser
85 90 95

ccc gag ttg ggt ccc acc ttg gac aca ctg cag ctg gac gtc gcc gac 336
Pro Glu Leu Gly Pro Thr Leu Asp Thr Leu Gln Leu Asp Val Ala Asp
100 105 110

ttt gcc acc acc atc tgg cag cag atg gaa gaa ctg gga atg gcc cct 384
Phe Ala Thr Thr Ile Trp Gln Gln Met Glu Glu Leu Gly Met Ala Pro
115 120 125

gcc ctg cag ccc acc cag ggt gcc atg ccg gcc ttc gcc tct gct ttc 432
Ala Leu Gln Pro Thr Gln Gly Ala Met Pro Ala Phe Ala Ser Ala Phe
130 135 140

cag cgc cgg gca gga ggg gtc ctg gtt gct agc cat ctg cag agc ttc 480
Gln Arg Arg Ala Gly Gly Val Leu Val Ala Ser His Leu Gln Ser Phe
145 150 155 160

ctg gag gtg tcg tac cgc gtt cta cgc cac ctt gcg cag ccc 522
Leu Glu Val Ser Tyr Arg Val Leu Arg His Leu Ala Gln Pro
165 170

<210> 2
<211> 174
<212> PRT
<213> *Homo sapiens*

<400> 2
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Cys Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

Glu Lys Leu Cys Ala Thr Tyr Lys Leu Cys His Pro Glu Glu Leu Val
35 40 45

Leu Leu Gly His Ser Leu Gly Ile Pro Trp Ala Pro Leu Ser Ser Cys
50 55 60

Pro Ser Gln Ala Leu Gln Leu Ala Gly Cys Leu Ser Gln Leu His Ser
65 70 75 80

Gly Leu Phe Leu Tyr Gln Gly Leu Leu Gln Ala Leu Glu Gly Ile Ser
85 90 95

Pro Glu Leu Gly Pro Thr Leu Asp Thr Leu Gln Leu Asp Val Ala Asp
100 105 110

Phe Ala Thr Thr Ile Trp Gln Gln Met Glu Glu Leu Gly Met Ala Pro
115 120 125

Ala Leu Gln Pro Thr Gln Gly Ala Met Pro Ala Phe Ala Ser Ala Phe
130 135 140

Gln Arg Arg Ala Gly Gly Val Leu Val Ala Ser His Leu Gln Ser Phe
145 150 155 160

Leu Glu Val Ser Tyr Arg Val Leu Arg His Leu Ala Gln Pro
165 170

<210> 3
<211> 32
<212> DNA
<213> Artificial Sequence

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<223> Oligonucleotide primer for the N-terminal of hG-CSF

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cgccgccata tgacacccct gggccctgcc ag 32

<210> 4
<211> 36
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide primer for the C-terminal of hG-CSF

<400> 4
accgaattcg gatcctcagg gctgcgcaag gtggcg 36

<210> 5
<211> 72
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing *E. coli* enterotoxin II signal peptide

<400> 5
tcatgaaaaa gaatatcgca tttcttcttg catctatgtt cgtttttttct attgctacaa 60

atgcctacgc gt 72

<210> 6
<211> 72
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing *E. coli* enterotoxin II signal peptide

<400> 6
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tctttttcat ga 72

<210> 7
<211> 39
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide primer coding for the N-terminal of [Ser1]hG-CSF

<400> 7
acaaatgcct acgcgtctcc cctgggccct gccagctcc 39

<210> 8
<211> 42
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide primer coding for the C-terminal of [Ser1]hG-CSF

<400> 8
accgaattcg gatcctcagg gctgcgcaag gtggcgtaga ac 42

<210> 9
<211> 65
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide primer coding for *E.coli* enterotoxin II Shine-Dalgarno sequence

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ctatg 65

<210> 10
<211> 45
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide containing BamHI restriction site

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<210> 11
<211> 10
<212> PRT
<213> Artificial Sequence

<220>
<223> Last five amino acids of *E. coli* enterotoxin II signal peptide plus the 1st to the 5th amino acids of hG-CSF

<400> 11
Thr Asn Ala Tyr Ala Thr Pro Leu Gly Pro
1 5 10

<210> 12
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing [Thr1]hG-CSF

<400> 12
acaaatgcct acgcgacacc cctgggccct 30

<210> 13
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 12
<400> 13
agggcccagg ggtgtcgcgt aggcatttgt 30

<210> 14
<211> 8
<212> PRT
<213> Artificial Sequence

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<223> N-terminal sequence of *E. coli* enterotoxin II signal peptide having threonine as the 4th amino acid

<400> 14
Met Lys Lys Thr Ile Ala Phe Leu
1 5

<210> 15
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for substituting the 4th amino acid of *E. coli* enterotoxin II signal peptide with threonine

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ggtgttttat gaaaaagaca atcgcatttc ttc 33

<210> 16
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
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<400> 16
gaagaaatgc gattgtcttt ttcataaaac acc 33

<210> 17
<211> 8
<212> PRT
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<220>
<223> C-terminal sequence of *E. coli* enterotoxin II signal peptide having glutamine as the 22nd amino acid

<400> 17
Asn Ala Gln Ala Thr Pro Leu Gly
1 5

<210> 18
<211> 26
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for substituting the 22nd amino acid of *E. coli* enterotoxin II signal peptide with glutamine

<400> 18
caaatgccca agcgacaccc ctgggc 26

<210> 19

<211> 26
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 18

<400> 19
gcccaggggt gtcgcttggg catttg 26

<210> 20
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for modifying *E. coli* enterotoxin II Shine-Dalgarno sequence
<400> 20
tctagaggtt gaggtgtttt atga 24

<210> 21
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 20

<400> 21
tcataaaaca cctcaacctc taga 24

<210> 22
<211> 66
<212> DNA
<213> Artificial Sequence

<220>
<223> S1 oligomer having *E. coli*-preferred nucleotide sequence coding for the 6th to 26th amino acids of [Ser17]hG-CSF

<400> 22
cagcctcttc tcttccacaa tctttccttc ttaagtctct tgaacaagtt agaaagatcc 60

aaggcg 66

<210> 23
<211> 66
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 22 (AS1 oligomer)

<400> 23
ccgggtcgga gaagagaagg tgttagaaag gaagaattca gagaacttgt tcaatctttc 60

taggtt 66

<210> 24
<211> 21
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<213> *Escherichia coli*

<220>
<221> SIGNAL
<222> (1).. (21)
<223> *E. coli* OmpA signal peptide

<400> 24
Met Lys Lys Thr Ala Ile Ala Ile Ala Val Ala Leu Ala Gly Phe Ala
1 5 10 15

Thr Val Ala Gln Ala
20

<210> 25
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide containing Hind III recognition site

<400> 25
gttgcgcaag cttctcga 18

<210> 26
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 25

<400> 26
tcgagaagct tgcgcaac 18

<210> 27
<211> 39
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for the N-terminal of [Ser1] hG-CSF

<400> 27
gttgcgcaag cttctcccct gggccctgcc agctccctg 39

<210> 28
<211> 39

<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide containing EcoRI restriction site

<400> 28
accgaattct cagggctgcg caaggtggcg tagaacgcg 39

<210> 29
<211> 13
<212> PRT
<213> Artificial Sequence

<220>
<223> *E. coli* OmpA signal peptide plus the 1st to the 5th amino acids of [Ser1]hG-CSF

<400> 29
Gly Phe Ala Thr Val Ala Gln Ala Ser Pro Leu Gly Pro
1 5 10

<210> 30
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing [Thr1]hG-CSF

<400> 30
accgttgcgc aagctacacc cctgggccct 30

<210> 31
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 30

<400> 31
agggcccagg ggtgtagctt gcgcaacggt 30

<210> 32
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing [Ser17]hG-CSF

<400> 32
agcttcctgc tcaagtcttt agagcaagtg agg 33

<210> 33
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 32

<400> 33
cctcacttgc tctaaagact tgagcaggaa gct 33

<210> 34
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing [Thr17]hG-CSF

<400> 34
agcttcctgc tcaagacctt agagcaagtg agg 33

<210> 35
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 34

<400> 35
cctcacttgc tctaaggtct tgagcaggaa gct 33

<210> 36
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing [Ala17]hG-CSF

<400> 36
agcttcctgc tcaaggcctt agagcaagtg agg 33

<210> 37
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 36

<400> 37
cctcacttgc tctaaggcct tgagcaggaa gct 33

<210> 38
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing [Gly17]hG-CSF

<400> 38
agcttcctgc tcaagggctt agagcaagtg agg 33

<210> 39
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 38

<400> 39
cctcacttgc tctaagccct tgagcaggaa gct 33

<210> 40
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide for preparing [Asp17]hG-CSF

<400> 40
agcttcctgc tcaaggactt agagcaagtg agg 33

<210> 41
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 40

<400> 41
cctcacttgc tctaagtcct tgagcaggaa gct 33

<210> 42
<211> 18
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<213> *Escherichia coli*

<220>
<221> SIGNAL
<222> (1)..(18)
<223> *E. coli* Gene III signal peptide

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1 5 10 15

His Ser

<210> 43
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide containing Nco I restriction site

<400> 43
tatagccata gcaccatgga g 21

<210> 44
<211> 21
<212> DNA

<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 43

<400> 44
ctccatggtg ctatggctat a 21

<210> 45
<211> 8
<212> PRT
<213> Artificial Sequence

<220>
<223> The 2nd to the 10th amino acids of hG-CSF

<400> 45
Pro Leu Gly Pro Ala Ser Ser Leu
1 5

<210> 46
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Oligonucleotide primer coding for the 2nd to the 10th amino acids of hG-CSF plus an additional cytosine at its 5'-end

<400> 46
ccccctgggc cctgccagct ccctg 25

<210> 47
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 46

<400> 47
cagggagctg gcagggccca ggggg 25

<210> 48
<211> 10
<212> PRT
<213> Artificial Sequence

<220>
<223> *E. coli* Gene III signal peptide plus the 1st to the 5th amino acids of hG-CSF

<400> 48
Phe Tyr Ser His Ser Thr Pro Leu Gly Pro
1 5 10

<210> 49
<211> 9
<212> PRT
<213> Artificial Sequence

<220>
<223> The 1st to the 9th amino acids of [Met2,Val3]hG-CSF

<400> 49
Thr Met Val Gly Pro Ala Ser Ser Leu
1 5

<210> 50
<211> 33
<212> DNA
<213> Artificial Sequence

<220>

<223> Oligonucleotide for preparing [Met2,Val3]hG-CSF

<400> 50
tacgcgtcca tggtgggccc tgccagctcc ctg 33

<210> 51
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Antisense of SEQ ID NO: 50

<400> 51
cagggagctg gcagggccca ccatggacgc gta 33

<210> 52
<211> 10
<212> PRT
<213> Artificial Sequence

<220>
<223> *E. coli* Gene III signal peptide plus the 1st to the 5th amino acids of [Met2,Val3]hG-CSF

<400> 52
Phe Tyr Ser His Ser Thr Met Val Gly Pro
1 5 10

<210> 53
<211> 23
<212> PRT
<213> *Escherichia coli*

<220>
<221> SIGNAL
<222> (1)..(23)

<223> Thermoresistant *E. coli* enterotoxin II signal peptide

<400> 53
Met Lys Lys Asn Ile Ala Phe Leu Leu Ala Ser Met Phe Val Phe Ser
1 5 10 15

Ile Ala Thr Asn Ala Tyr Ala
20

<210> 54
<211> 23
<212> PRT
<213> Artificial Sequence

<220>
<223> Modified thermoresistant *E. coli* enterotoxin II signal peptide

<400> 54
Met Lys Lys Thr Ile Ala Phe Leu Leu Ala Ser Met Phe Val Phe Ser
1 5 10 15

Ile Ala Thr Asn Ala Gln Ala
20

<210> 55
<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to 32nd amino acids of [Ser1, Ser17]hG-CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 55
tct ccc ctg ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Ser Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

tct tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Ser Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 56
<211> 32
<212> PRT
<213> Artificial Sequence

<400> 56
Ser Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Ser Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 57
<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to the 32nd amino acids of [Ser1]hG-CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 57
tct ccc ctg ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Ser Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

tgc tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Cys Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 58
<211> 32
<212> PRT
<213> Artificial Sequence

<400> 58
Ser Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Cys Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 59
<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to the 32nd amino acids of [Ser17]hG-CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 59
aca ccc ctg ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

tct tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Ser Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 60
<211> 32
<212> PRT
<213> Artificial Sequence

<400> 60
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Ser Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 61
<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to the 32nd amino acids of [Thr17]hG--CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 61
aca ccc ctg ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

acc tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Thr Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 62
<211> 32

<212> PRT
<213> Artificial Sequence

<400> 62
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Thr Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 63
<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to the 32nd amino acids of [Ala17]hG-CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 63
aca ccc ctg ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

gcc tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Ala Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 64
<211> 32
<212> PRT
<213> Artificial Sequence

<400> 64

Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Ala Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 65
<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to the 32th amino acids of [Gly17]hG-CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 65
aca ccc ctg ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

ggc tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Gly Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 66
<211> 32
<212> PRT
<213> Artificial Sequence

<400> 66
Thr Pro Leu Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Gly Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln

20 25 30

<210> 67
<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to the 32nd amino acids of [Met2, Val3]hG-CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 67
aca atg gtc ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Thr Met Val Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

tgc tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Cys Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 68
<211> 32
<212> PRT
<213> Artificial Sequence

<400> 68
Thr Met Val Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Cys Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 69

<211> 96
<212> DNA
<213> Artificial Sequence

<220>
<223> Nucleotide sequence coding for the 1st to the 32nd amino acids of [Met2, Val3, Ser17]hG-CSF

<220>
<221> CDS
<222> (1)..(96)

<400> 69
aca atg gtc ggc cct gcc agc tcc ctg ccc cag agc ttc ctg ctc aag 48
Thr Met Val Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

tct tta gag caa gtg agg aag atc cag ggc gat ggc gca gcg ctc cag 96
Ser Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 70
<211> 32
<212> PRT
<213> Artificial Sequence

<400> 70
Thr Met Val Gly Pro Ala Ser Ser Leu Pro Gln Ser Phe Leu Leu Lys
1 5 10 15

Ser Leu Glu Gln Val Arg Lys Ile Gln Gly Asp Gly Ala Ala Leu Gln
20 25 30

<210> 71
<211> 10
<212> DNA
<213> Artificial Sequence

<220>
<223> Modified Shine-Dalgarno sequence

<400> 71
gaggtgtttt 10