Figure 1

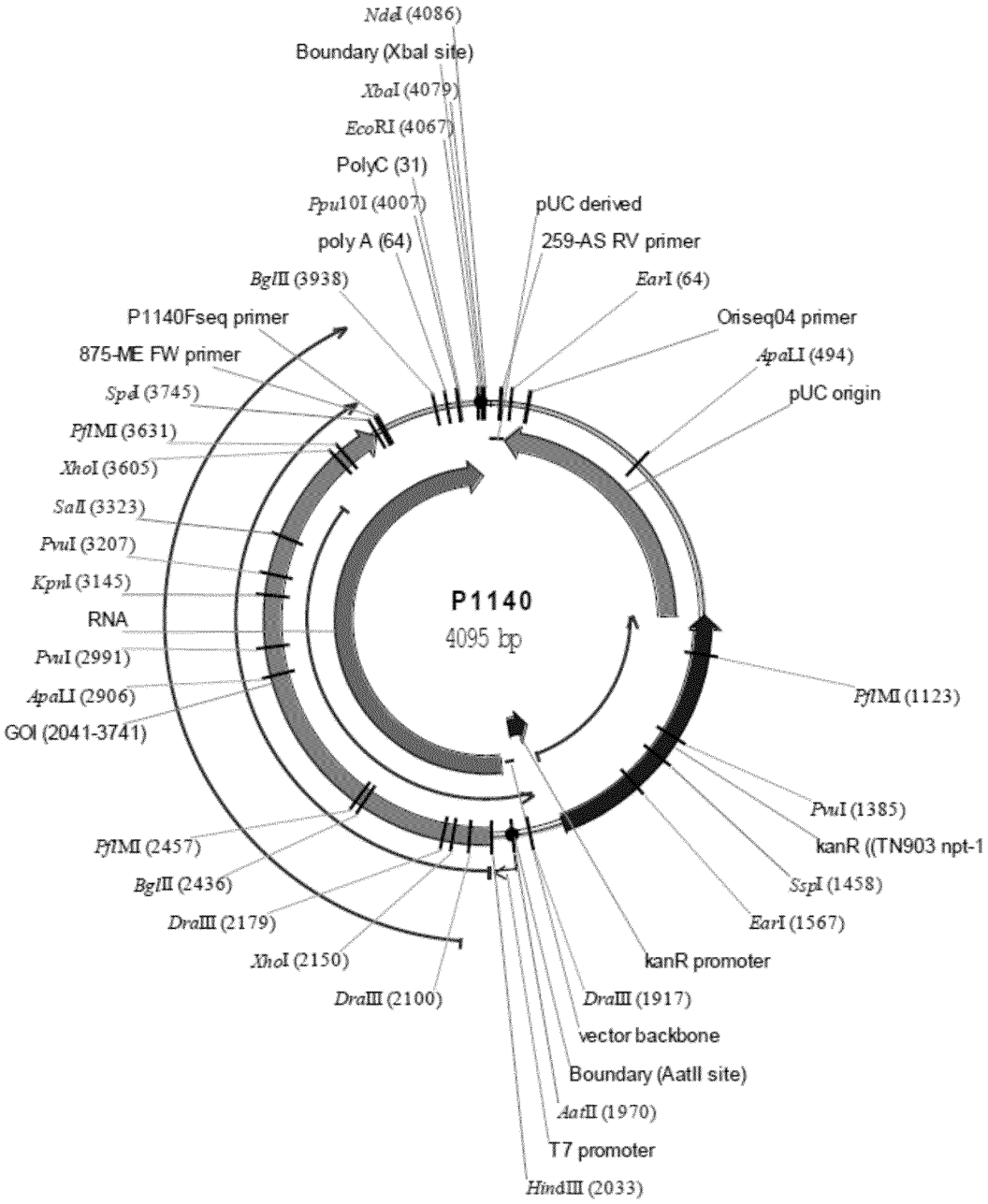

NdeI (4086)
Boundary (XbaI site)
XbaI (4079)
EcoRI (4067)
PolyC (31)
Ppu10I (4007)
poly A (64)
BglII (3938)
P1140Fseq primer
875-ME FW primer
SpeI (3745)
PflMI (3631)
XhoI (3605)
SalI (3323)
PvuI (3207)
KpnI (3145)
RNA
PvuI (2991)
ApaLI (2906)
GOI (2041-3741)
PflMI (2457)
BglII (2436)
DraIII (2179)
XhoI (2150)
DraIII (2100)
pUC derived
259-AS RV primer
EarI (64)
Oriseq04 primer
ApaLI (494)
pUC origin
P1140
4095 bp
PflMI (1123)
PvuI (1385)
kanR ((TN903 npt-1
SspI (1458)
EarI (1567)
kanR promoter
DraIII (1917)
vector backbone
Boundary (AatII site)
AatII (1970)
T7 promoter
HindIII (2033)

Figure 2

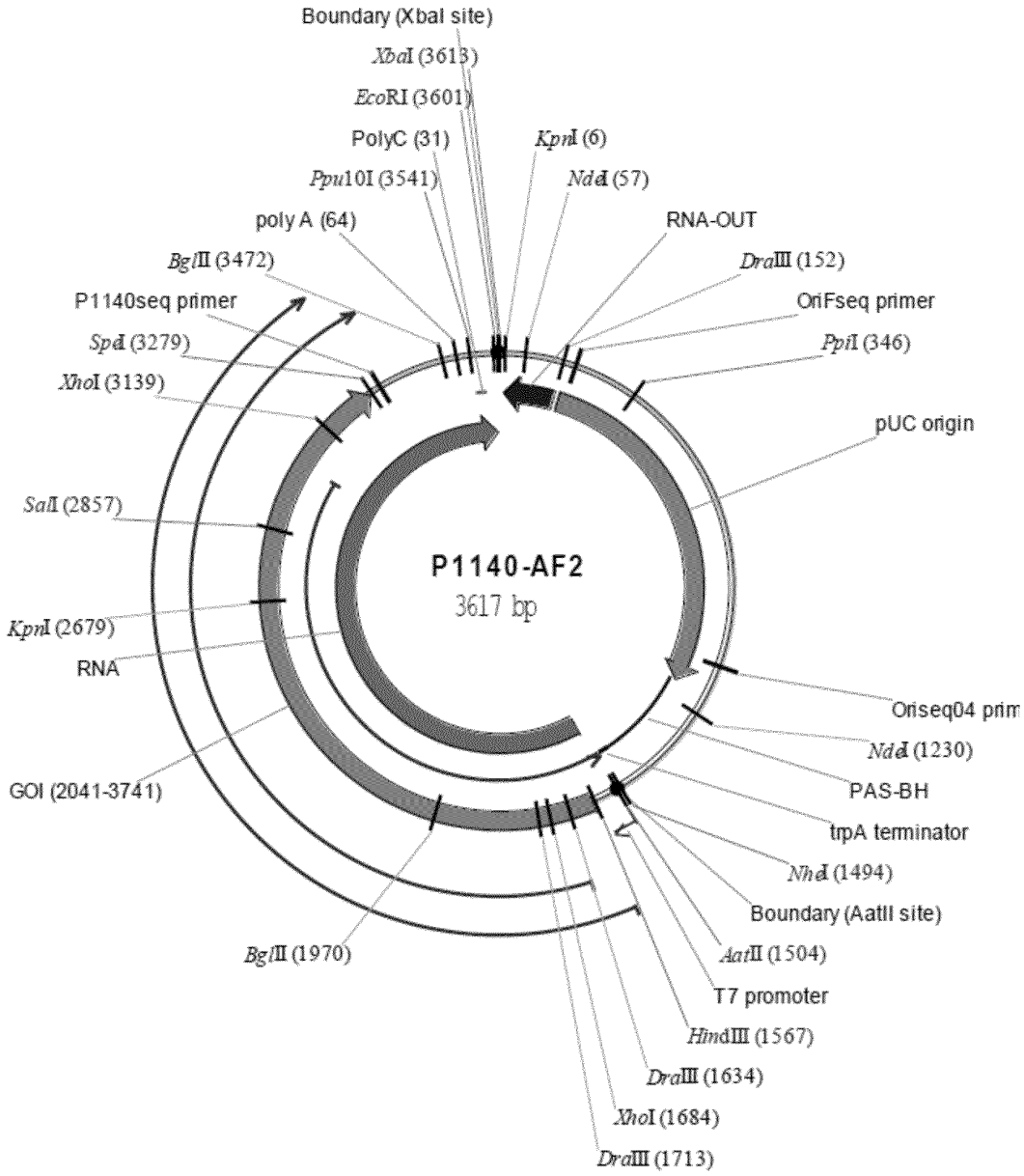

Boundary (XbaI site)
XbaI (3613)
EcoRI (3601)
PolyC (31)
KpnI (6)
Ppu10I (3541)
NdeI (57)
poly A (64)
RNA-OUT
BglII (3472)
DraIII (152)
P1140seq primer
OriFseq primer
SpeI (3279)
PpiI (346)
XhoI (3139)
pUC origin
SalI (2857)
P1140-AF2
3617 bp
KpnI (2679)
RNA
Oriseq04 prim
NdeI (1230)
PAS-BH
GOI (2041-3741)
trpA terminator
NheI (1494)
Boundary (AatII site)
BglII (1970)
AatII (1504)
T7 promoter
HindIII (1567)
DraIII (1634)
XhoI (1684)
DraIII (1713)

Figure 3

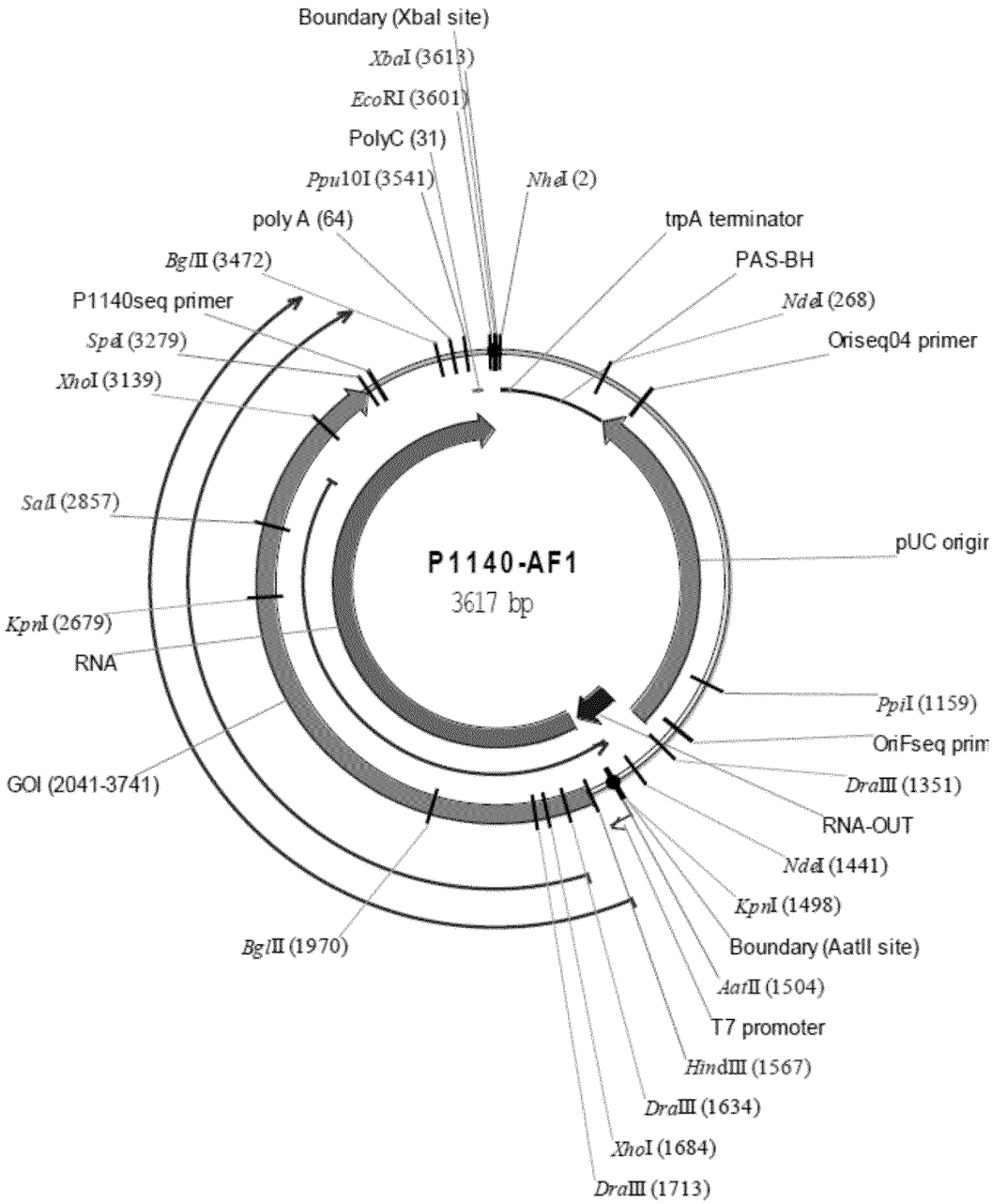

Boundary (XbaI site)
XbaI (3613)
EcoRI (3601)
PolyC (31)
Ppu10I (3541)
NheI (2)
poly A (64)
trpA terminator
BglII (3472)
PAS-BH
P1140seq primer
NdeI (268)
SpeI (3279)
Oriseq04 primer
XhoI (3139)
SalI (2857)
pUC origin
P1140-AF1
3617 bp
KpnI (2679)
RNA
PpiI (1159)
OriFseq prim
DraIII (1351)
GOI (2041-3741)
RNA-OUT
NdeI (1441)
KpnI (1498)
Boundary (AatII site)
BglII (1970)
AatII (1504)
T7 promoter
HindIII (1567)
DraIII (1634)
XhoI (1684)
DraIII (1713)

Figure 4

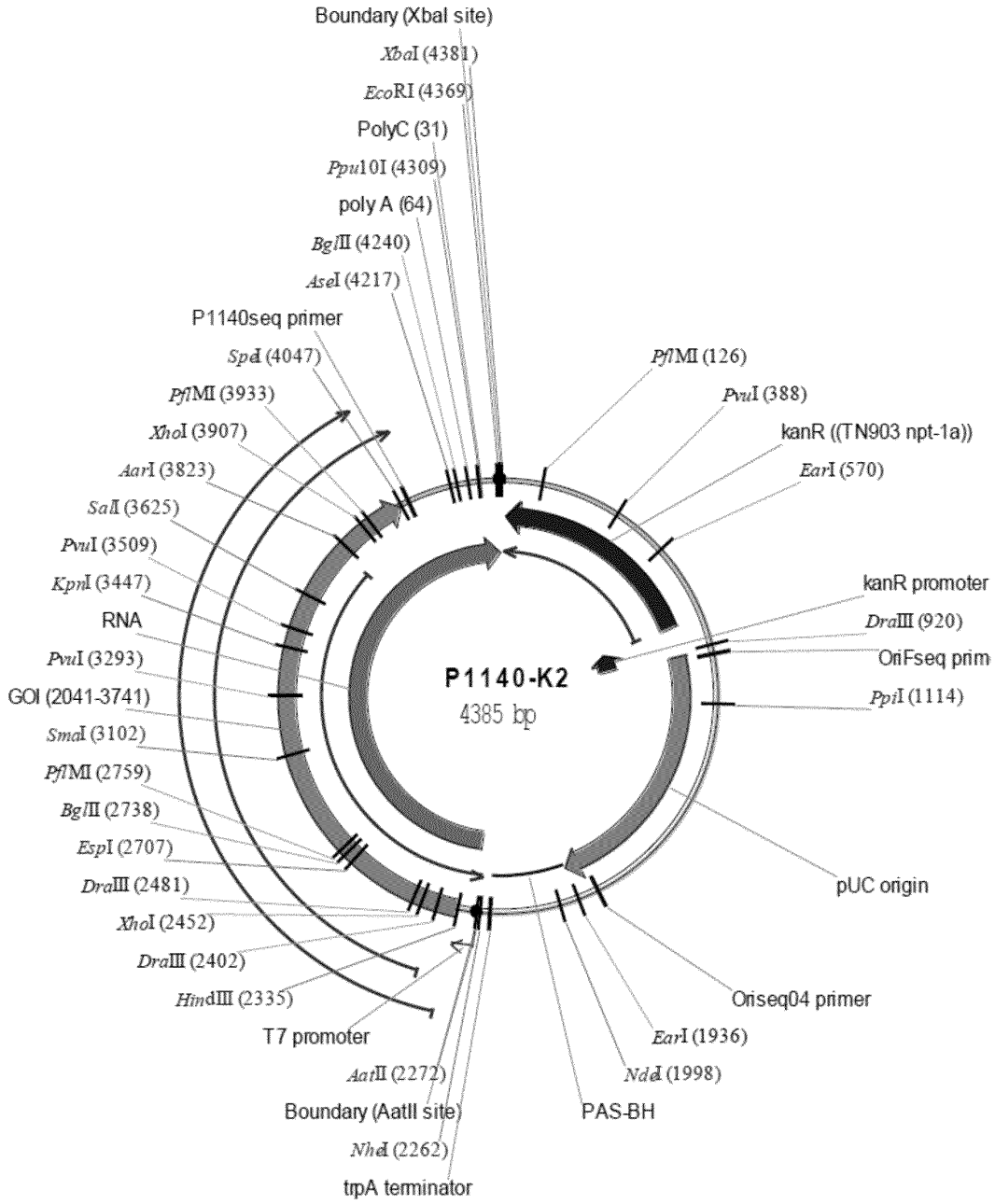

Boundary (XbaI site)
XbaI (4381)
EcoRI (4369)
PolyC (31)
Ppu10I (4309)
poly A (64)
BglII (4240)
AseI (4217)
P1140seq primer
SpeI (4047)
PflMI (3933)
XhoI (3907)
AarI (3823)
SalI (3625)
PvuI (3509)
KpnI (3447)
RNA
PvuI (3293)
GOI (2041-3741)
SmaI (3102)
PflMI (2759)
BglII (2738)
EspI (2707)
DraIII (2481)
XhoI (2452)
DraIII (2402)
HindIII (2335)
T7 promoter
AatII (2272)
Boundary (AatII site)
NheI (2262)
trpA terminator
PflMI (126)
PvuI (388)
kanR ((TN903 npt-1a))
EarI (570)
kanR promoter
DraIII (920)
OriFseq prim
PpiI (1114)
pUC origin
Oriseq04 primer
EarI (1936)
NdeI (1998)
PAS-BH
P1140-K2
4385 bp

Figure 5

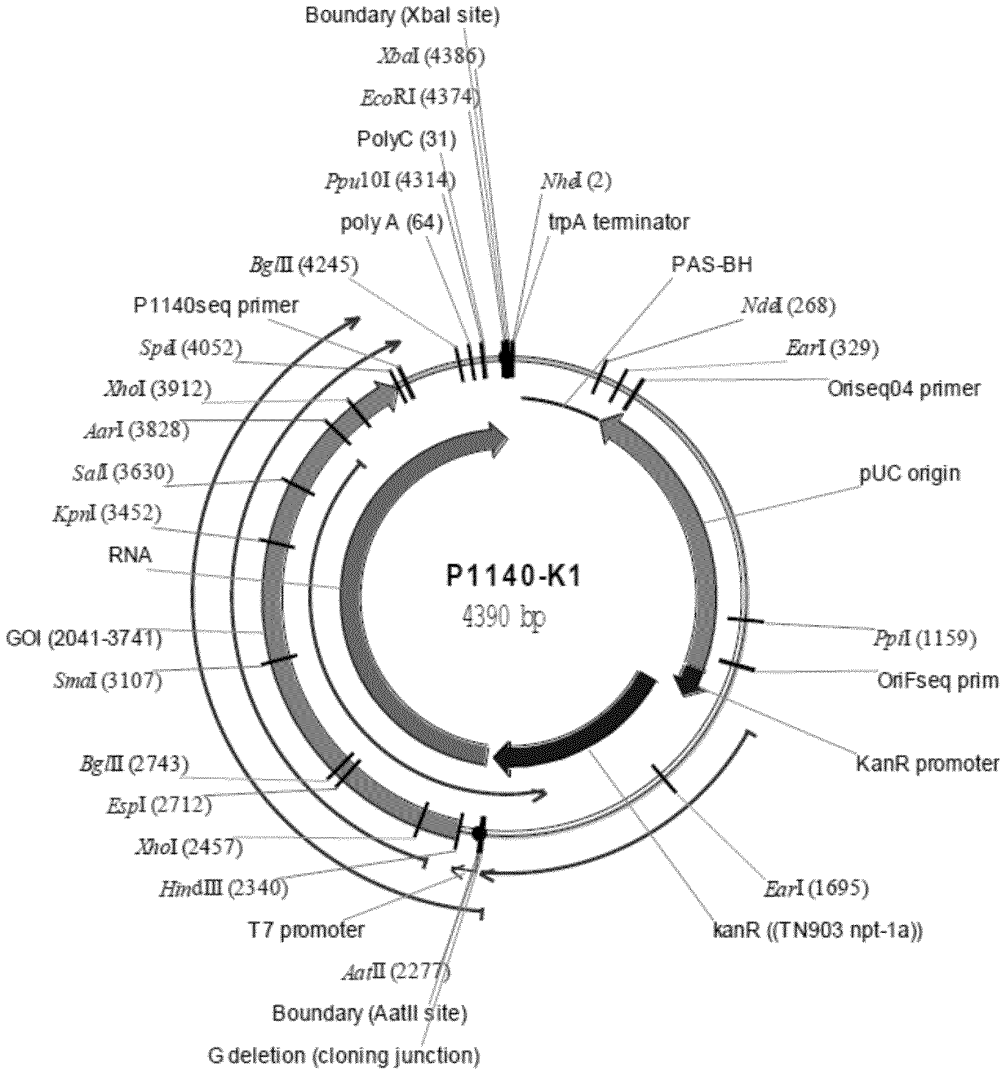

Boundary (XbaI site)
XbaI (4386)
EcoRI (4374)
PolyC (31)
Ppu10I (4314)
NheI (2)
poly A (64)
trpA terminator
BglII (4245)
PAS-BH
P1140seq primer
NdeI (268)
SpeI (4052)
EarI (329)
XhoI (3912)
Oriseq04 primer
AarI (3828)
SalI (3630)
pUC origin
KpnI (3452)
RNA
P1140-K1
4390 bp
GOI (2041-3741)
PpiI (1159)
SmaI (3107)
OriFseq prim
BglII (2743)
KanR promoter
EspI (2712)
XhoI (2457)
EarI (1695)
HindIII (2340)
T7 promoter
kanR ((TN903 npt-1a))
AatII (2277)
Boundary (AatII site)
G deletion (cloning junction)

Figure 6

GC-optimized mRNA encoding H1N1(Netherlands2009)-HA

GGGGCGCUGCCUACGGAGGUGGCAGCCAUCUCCUUCUCGGCAUCAAGCUUACCAUGAAGGCC
AUCCUGGUGGUCCUCCUGUACACCUUCGCCACCGCGAACGCCGACACGCUGUGCAUCGGCUA
CCACGCCAACAACAGCACCGACACCGUGGACACCGUGCUCGAGAAGAACGUCACGGUGACCC
ACUCCGUGAACCUGCUGGAGGACAAGCACAACGGGAAGCUCUGCAAGCUGCGGGGCGUCGCC
CCGCUGCACCUCGGGAAGUGCAACAUCGCCGGCUGGAUCCUGGGGAACCCGGAGUGCGAGAG
CCUGUCCACCGCGAGCUCCUGGAGCUACAUCGUGGAGACCUCCAGCUCCGACAACGGCACGU
GCUACCCCGGCGACUUCAUCGACUACGAGGAGCUCCGCGAGCAGCUGAGCUCCGUGAGCUCC
UUCGAGCGGUUCGAGAUCUUCCCCAAGACCAGCUCCUGGCCCAACCACGACAGCAACAAGGG
GGUCACCGCCGCCUGCCCGCACGCCGGCGCGAAGUCCUUCUACAAGAACCUGAUCUGGCUCG
UGAAGAAGGGGAACAGCUACCCCAAGCUGUCCAAGAGCUACAUCAACGACAAGGGCAAGGAG
GUGCUGGUCCUCUGGGGGAUCCACCACCCCAGCACCUCCGCCGACCAGCAGAGCCUGUACCA
GAACGCCGACGCCUACGUGUUCGUGGGCUCCAGCCGCUACUCCAAGAAGUUCAAGCCCGAGA
UCGCCAUCCGGCCGAAGGUCCGCGACCAGGAGGGCCGGAUGAACUACUACUGGACGCUGGUG
GAGCCCGGGGACAAGAUCACCUUCGAGGCGACCGGCAACCUCGUGGUCCCCCGCUACGCCUU
CGCCAUGGAGCGGAACGCCGGGAGCGGCAUCAUCAUCUCCGACACCCCCGUGCACGACUGCA
ACACGACCUGCCAGACCCCGAAGGGCGCCAUCAACACCAGCCUGCCCUUCCAGAACAUCCAC
CCCAUCACGAUCGGGAAGUGCCCCAAGUACGUGAAGUCCACCAAGCUGCGCCUCGCGACCGG
CCUGCGGAACGUCCCGAGCAUCCAGUCCCGCGGGCUGUUCGGCGCCAUCGCCGGGUUCAUCG
AGGGCGGCUGGACCGGGAUGGUGGACGGCUGGUACGGGUACCACCACCAGAACGAGCAGGGC
AGCGGGUACGCCGCCGACCUCAAGUCCACGCAGAACGCGAUCGACGAGAUCACCAACAAGGU
GAACAGCGUCAUCGAGAAGAUGAACACCCAGUUCACCGCCGUGGGCAAGGAGUUCAACCACC
UGGAGAAGCGGAUCGAGAACCUGAACAAGAAGGUCGACGACGGCUUCCUCGACAUCUGGACG
UACAACGCCGAGCUGCUGGUGCUCCUGGAGAACGAGCGCACCCUGGACUACCACGACUCCAA
CGUGAAGAACCUCUACGAGAAGGUCCGGAGCCAGCUGAAGAACAACGCCAAGGAGAUCGGGA
ACGGCUGCUUCGAGUUCUACCACAAGUGCGACAACACCUGCAUGGAGUCCGUGAAGAACGGG
ACCUACGACUACCCCAAGUACAGCGAGGAGGCCAAGCUGAACCGCGAGGAGAUCGACGGCGU
GAAGCUCGAGUCCACGCGGAUCUACCAGAUCCUGGCGAUCUACAGCACCGUCGCCAGCUCCC
UGGUGCUCGUGGUCAGCCUGGGGGCCAUCUCCUUCUGGAUGUGCAGCAACGGCUCCCUGCAG
UGCCGCAUCUGCAUCUGACCACUAGUGCAUCACAUUUAAAAGCAUCUCAGCCUACCAUGAGA
AUAAGAGAAAGAAAAUGAAGAUCAAUAGCUUAUUCAUCUCUUUUUCUUUUUCGUUGGUGUAA
AGCCAACACCCUGUCUAAAAAACAUAAAUUUCUUUAAUCAUUUUGCCUCUUUUCUCUGUGCU
UCAAUUAAUAAAAAAUGGAAAGAACCUAGAUCUAAAAAAAAAAAAAAAAAAAAAAAAAAAAA
AAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAUGCAUCCCCCCCCCCCCCCCCCCCCC
CCCCCCCCCAAAGGCUCUUUUCAGAGCCACCAGAAUU