

AUG1 Forward priming site

1081 TAGAAGTTTT ATTTAACATC AGTTTCAATT TACATCTTTA TTTATTAACG AAATCTTTAC

Sac I

1141 GAATTAAGTC AATCAAAACT TTTACGAAAA AAAAATCTTA CTATTAAGAG CTCAAA ATG
Met

α-factor signal sequence

1200 AGA TTT CCT TCA ATT TTT ACT GCT GTT TTA TTC GCA GCA TCC TCC GCA
Arg Phe Pro Ser Ile Phe Thr Ala Val Leu Phe Ala Ala Ser Ser Ala

1248 TTA GCT GCT CCA GTC AAC ACT ACA ACA GAA GAT GAA ACG GCA CAA ATT
Leu Ala Ala Pro Val Asn Thr Thr Thr Glu Asp Glu Thr Ala Gln Ile

1296 CCG GCT GAA GCT GTC ATC GGT TAC TCA GAT TTA GAA GGG GAT TTC GAT
Pro Ala Glu Ala Val Ile Gly Tyr Ser Asp Leu Glu Gly Asp Phe Asp

1344 GTT GCT GTT TTG CCA TTT TCC AAC AGC ACA AAT AAC GGG TTA TTG TTT
Val Ala Val Leu Pro Phe Ser Asn Ser Thr Asn Asn Gly Leu Leu Phe

α-factor Forward priming site

1392 ATA AAT ACT ACT ATT GCC AGC ATT GCT GCT AAA GAA GAA GGG GTA TCT
Ile Asn Thr Thr Ile Ala Ser Ile Ala Ala Lys Glu Glu Gly Val Ser

Xho I Kex2 signal cleavage Pst I EcoR I Pml I Sfi I Xma III* BamH I

1440 CTC GAG AAA AGA GAG GCT GAA GCT GCAGGAATTC ACGTGGCCCA GCCGCGCGTG
Leu Glu Lys Arg Glu Ala▲Glu Ala▲

Ste 13 signal cleavage

Sal I Sac II Not I/Xma III* Spe I V5 epitope

1494 GATCCACGCG TCGTCGACCC GCGGCGGCCG CCAGCTTACT AGTA GGT AAG CCT ATC
Gly Lys Pro Ile
Polyhistidine (6xHis) region

1550 CCT AAC CCT CTC CTC GGT CTC GAT TCT ACG CGT ACC GGT CAT CAT CAC
Pro Asn Pro Leu Leu Gly Leu Asp Ser Thr Arg Thr Gly His His His

1598 CAT CAC CAT TGA T CTAGTATACA ATTCTAGGGC TGCCTGTTTG GATATTTTAA
His His His ***

AUG1 Reverse priming site

1651 TAATTTTTGA GAGTTTGCCA ACTAATGTTT TTCTCTTCTA TGATATTTAT

*The Xma III sites are unique to the polylinker.