

pYES2/NT B Multiple Cloning Site

300 ┌──GALI promoter
└──TATA box
 TTAACAGATA TATAAATGCA AAAACTGCAT AACCACTTTA ACTAATACTT TCAACATTTT
 360 CGGTTTGTAT TACTTCTTAT TCAAATGTAA TAAAAGTATC AACAAAAAAT TGTTAATATA
└──start of transcription
 420 ┌──GALI forward priming site └──3' end of GALI promoter
 CCTCTATACT TTAACGTCAA GGAGAAAAAA CCCC GGATCG GACTACTAGC AGCTGTAATA
 480 ┌──T7 promoter/priming site └──Hind III └──Polyhistidine region
 CGACTCACTA TAGGGAATAT TAAGCTTACC **ATG** GGG GGT TCT CAT CAT CAT CAT
Met Gly Gly Ser His His His His
 534 CAT CAT GGT ATG GCT AGC ATG ACT GGT GGA CAG CAA ATG GGT CGG GAT
 His His Gly Met Ala Ser Met Thr Gly Gly Gln Gln Met Gly Arg Asp
 582 ┌──Xpress™ epitope └──Asp718 I └──Kpn I └──BamHI └──BstXI* └──EcoRI
 CTG TAC GAC GAT GAC GAT AAG GTA CCT AAG GAT CCA GTG TGG TGG AAT
 Leu Tyr Asp Asp Asp Asp Lys Val Pro Lys Asp Pro Val Trp Trp Asn
└──Enterokinase recognition site ▲ EK cleavage site
 630 TCT GCA GAT ATC CAG CAC AGT GGC GGC CGC TCG AGT CTA GAGGGCCCTT
 Ser Ala Asp Ile Gln His Ser Gly Gly Arg Ser Ser Leu
└──V5 epitope
 679 CGAA GGT AAG CCT ATC CCT AAC CCT CTC CTC GGT CTC GAT TCT ACG CGT
 Gly Lys Pro Ile Pro Asn Pro Leu Leu Gly Leu Asp Ser Thr Arg
└──Polyhistidine region └──Pme I
 728 ACC GGT CAT CAT CAC CAT CAC CAT TGA GTTTAAACCC GCTGATCCTA
 Thr Gly His His His His His His ***
└──CYC1 reverse priming site
 775 GAGGGCCGCA TCATGTAATT AGTTATGTCA CGCTTACATT CACGCCCTCC CCCACATCC

*Note that there are two *BstX I* sites in the polylinker.