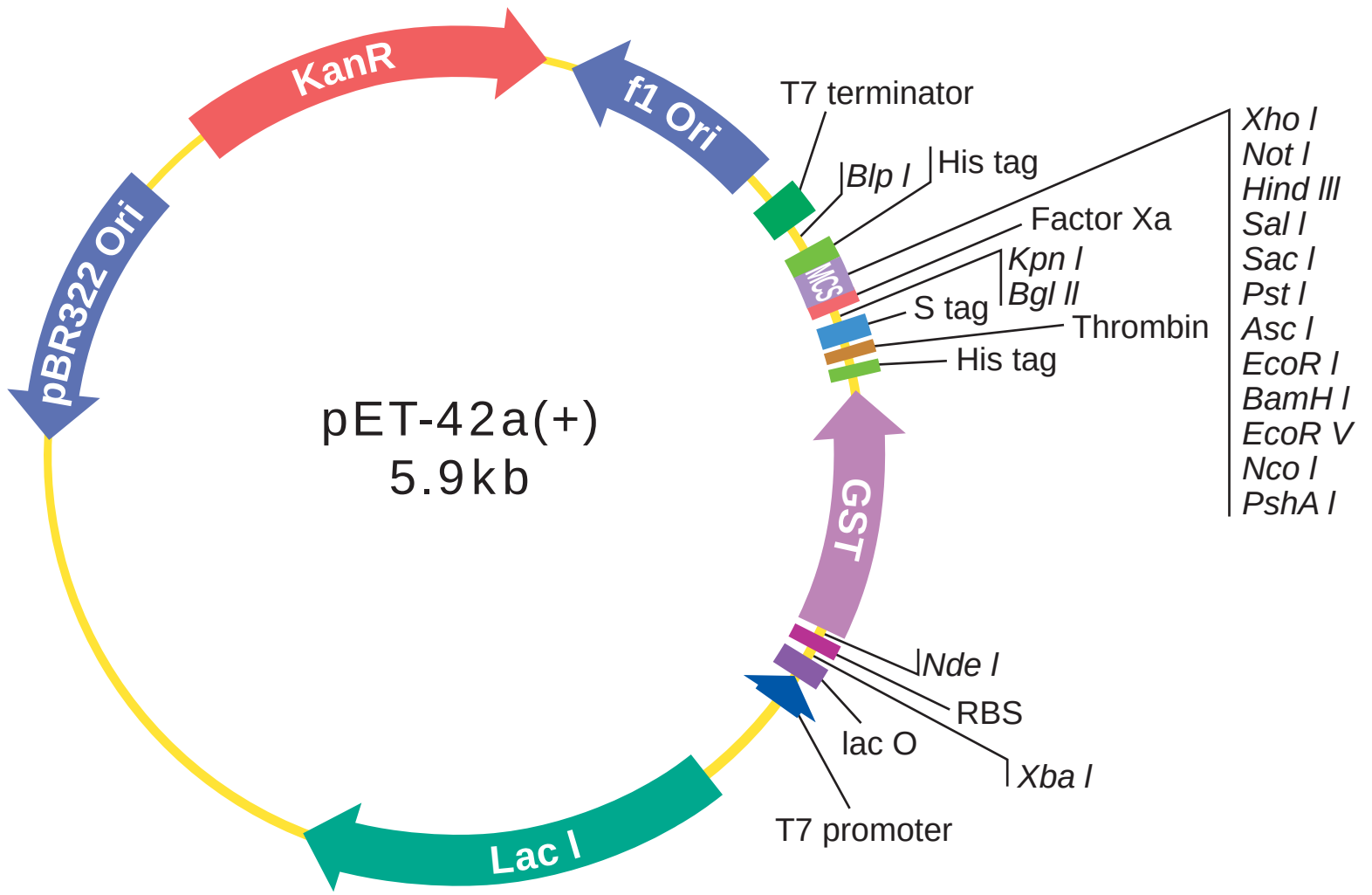

T7 promoter — lac operator — *Xba* I — rbs — *Nde* I — GST tag

--- T AAT ACG ACT CAC TAT AGG GGA ATT GTG AGC GGA TAA CAA TTC CCC TCT AGA AAT AAT TTT GTT TAA CTT TAA GAA GGA GAT ATA CAT ATG TCC CCT ATA CTA GGT

M S P I L G

GST tag — His tag

TAT ... 627bp ... GAC CAT CCT CCA AAA TCG GAT GGT TCA ACT AGT GGT TCT GGT CAT CAC CAT CAC CAT CAC TCC GCG GGT CTG GTG CCA CGC GGT AGT ACT GCA ATT GGT

W ... 209aa ... D H P P K S D G S T S G S G H H H H H H S A G L V P R G S T A I G

thrombin site

S tag — *Bgl* II — *Kpn* I — *PshA* I — *Nco* I — *Eco*R V

ATG AAA GAA ACC GCT GCT GCT AAA TTC GAA CGC CAG CAC ATG GAC AGC CCA GAT CTG GGT ACC GGT GGT GGC TCC GGT ATT GAG GGA CGC GGG TCC ATG GGA TAT CGG

M K E T A A A K F E R Q H M D S P D L G T G G G S G I E G R C S M G Y R

Factor Xa

*Bam*H I — *Eco*R I — *Asc* I — *Pst* I — *Sac* I — *Sal* I — *Hin*d III — *Not* I — *Xho* I — His tag

GGA TCC GAA TTC TGT ACA GGC CTT GGC GCG CCT GCA GGC GAG CTC CGT CGA CAA GCT TGC GGC CGC ACT CGA GCA CCA CCA CCA CCA CCA CCA CCA CTA ATT GAT TAA TAC

G S E F C T G L G A P A G E L R R Q A C G R T R A P P P P P P P L I D Stop

Avr II — *Blp* I — T7 terminator

CTA GGC TGC TAA ACA AAG CCC GAA AGG AAG CTG AGT TGG CTG CTG CCA CCG CTG AGC AAT AAC TAG CAT AAC CCC TTG GGG CCT CTA AAC GGG TCT TGA GGG GTT TTT TG ---