

Kust Gene

5' GAATTC GCGGCCGC T TCTAG ATG --- T ACTAGT A GCGGCCGC CTGCAG 3'
3' CTTAAG CGCCGGCG A AGATC TAG --- A TGATCA T CGCCGGC GACGTC 5'

*Cut with
XbaI and
SpeI*

Kust Gene

5' CTAG ATG --- T A 3'
3' TAG --- A TGATC 5'

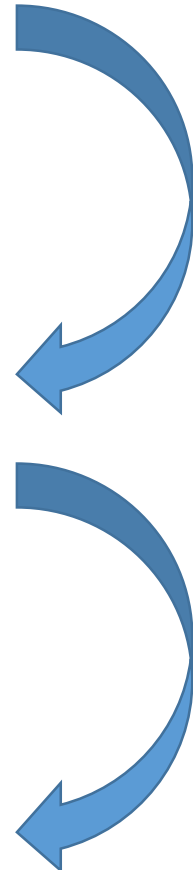
*Extend with
DNA
polymerase
(i.e. Taq)*

*To Gibson
Assembly*

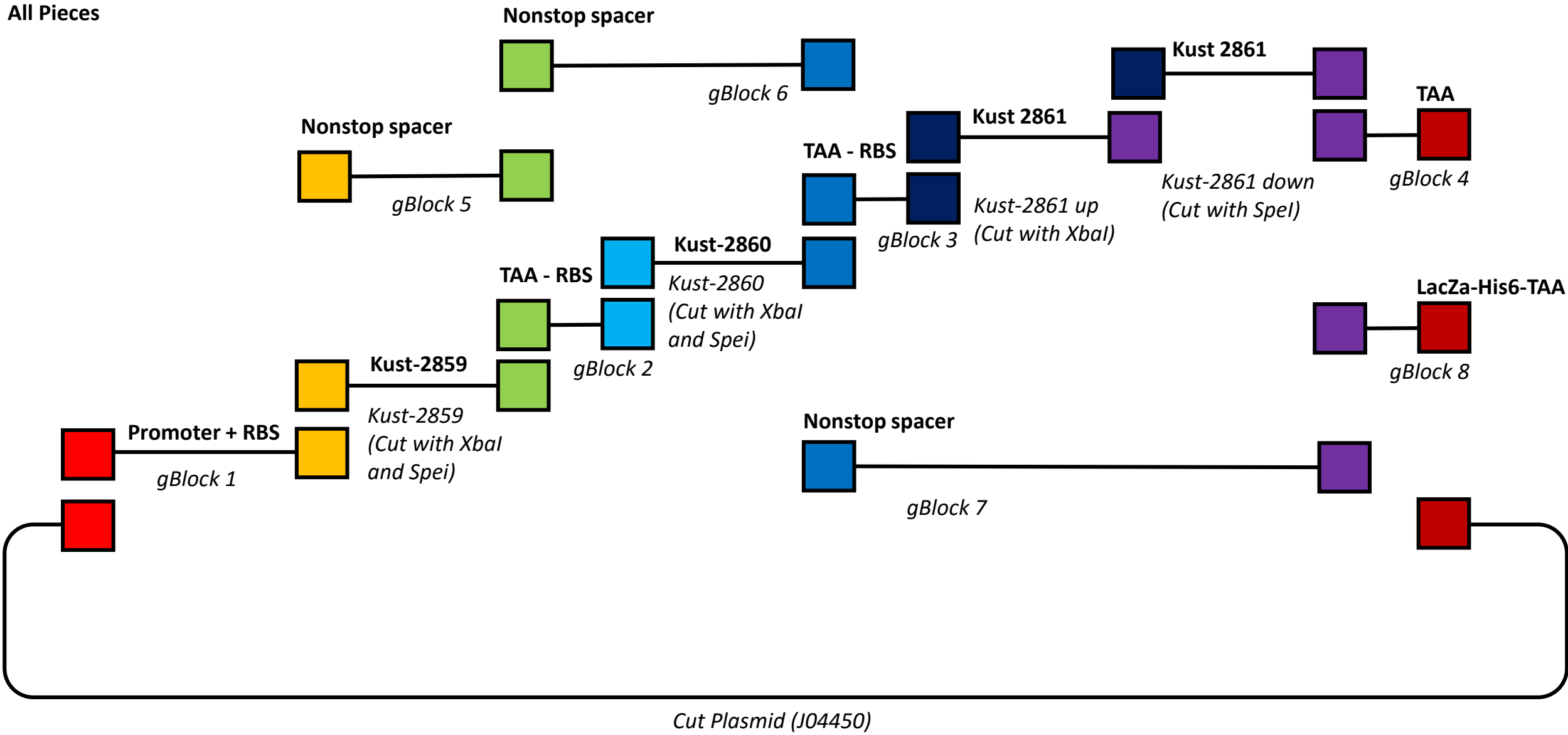
Kust Gene

5' CTAG ATG --- T ACTAG A 3'
3' A GATC TAG --- A TGATC 5'

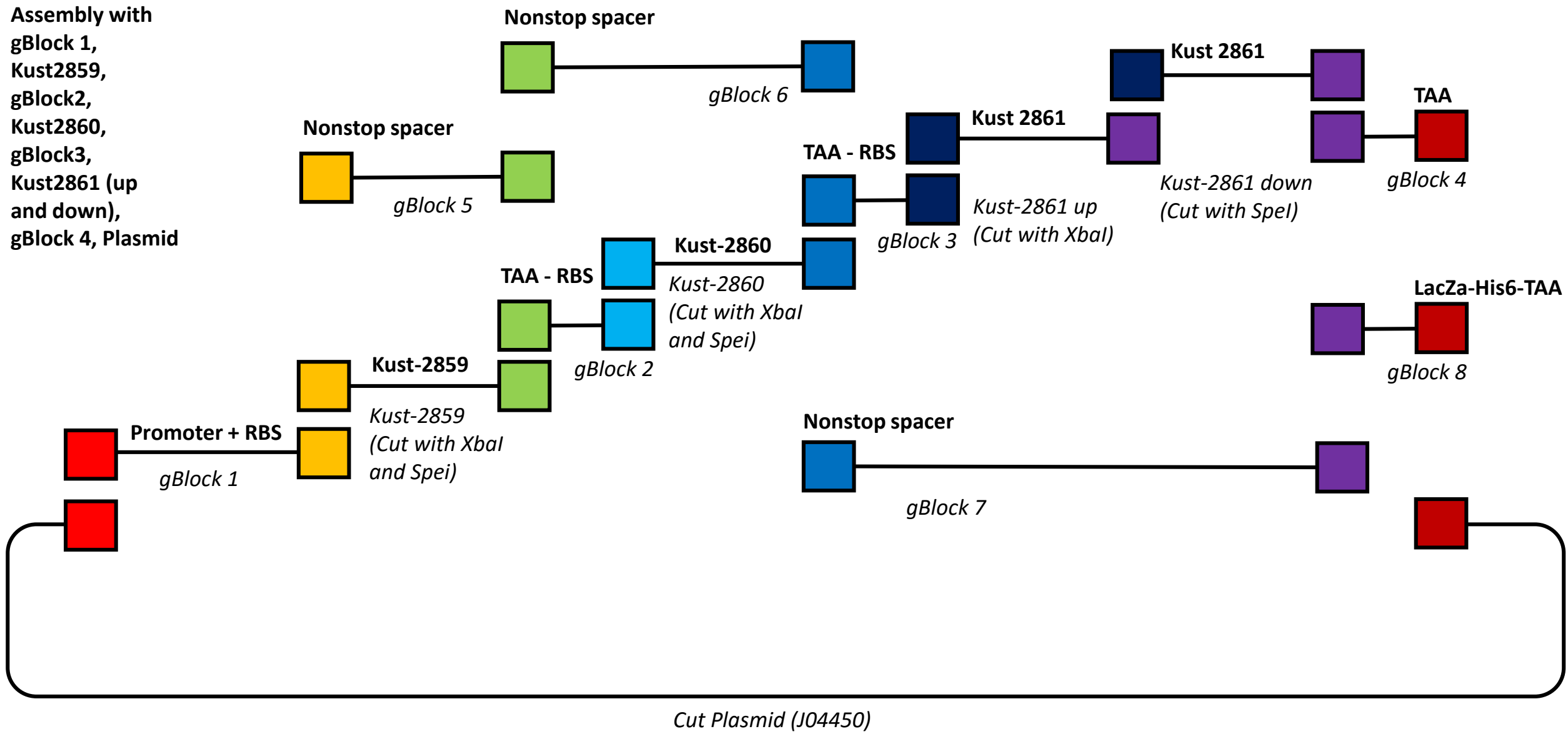
*Taq can add 3'
overhangs of A*



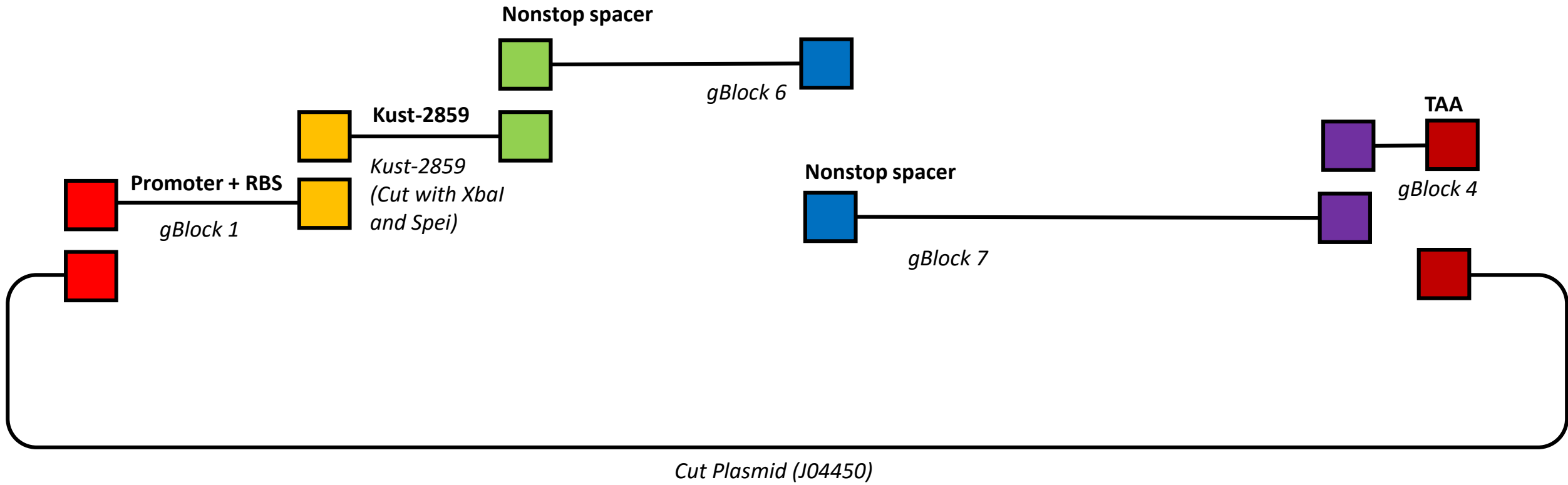
All Pieces



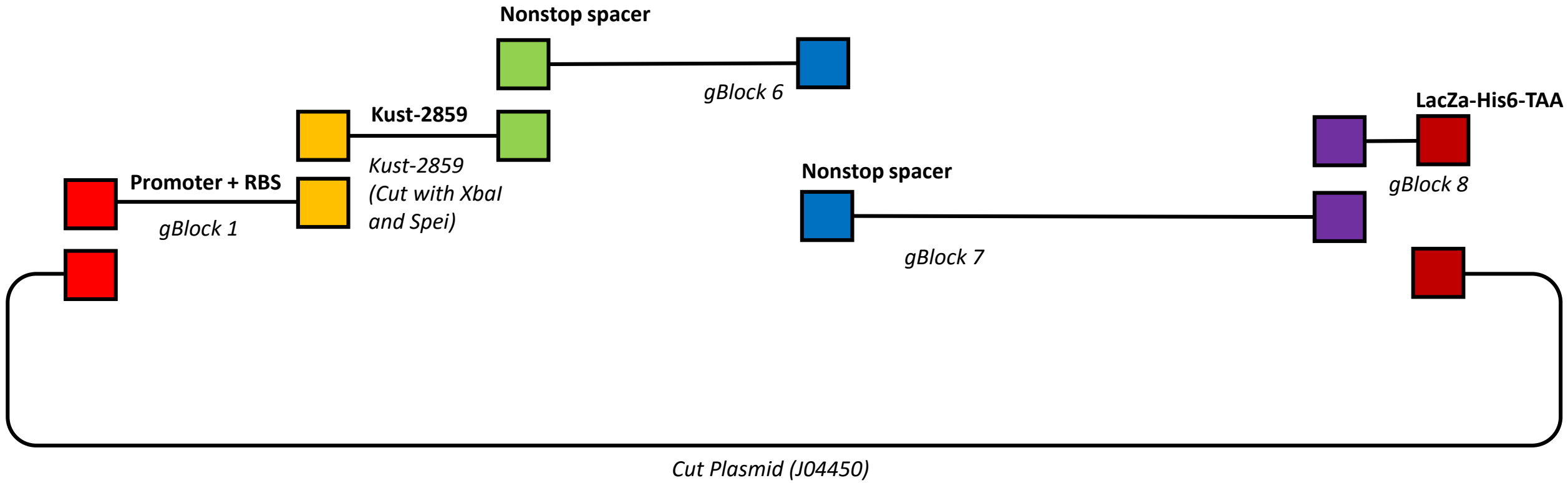
Assembly with
gBlock 1,
Kust2859,
gBlock2,
Kust2860,
gBlock3,
Kust2861 (up
and down),
gBlock 4, Plasmid



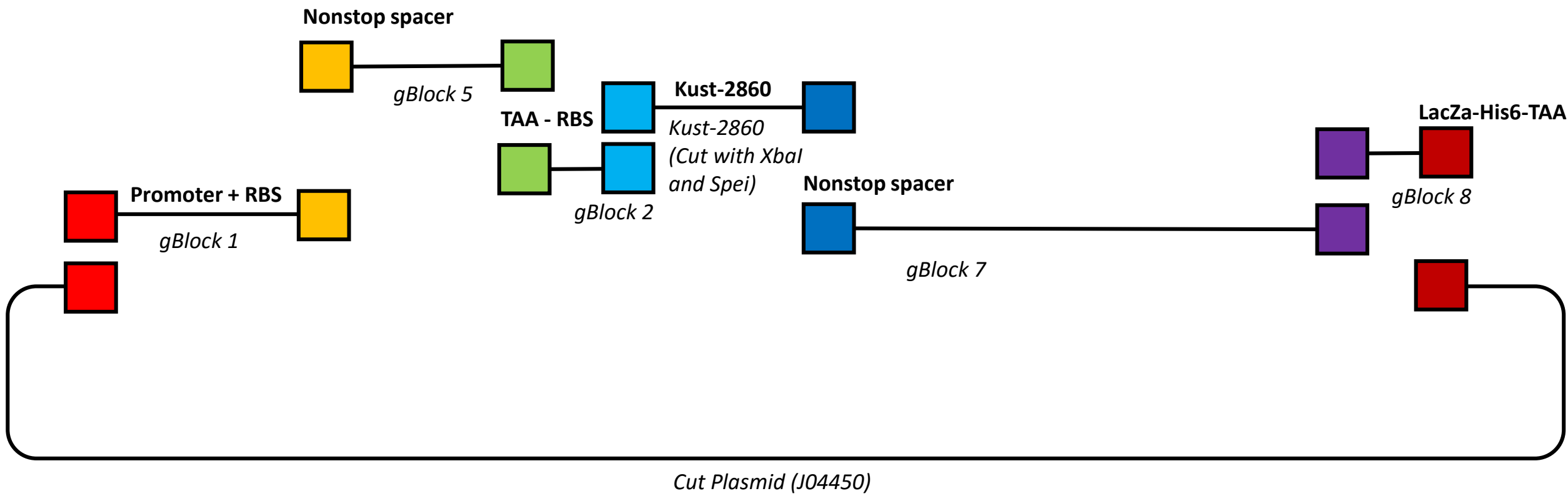
Assembly with
gBlock 1,
Kust2859,
gBlock6, gBlock7
gBlock 4, Plasmid



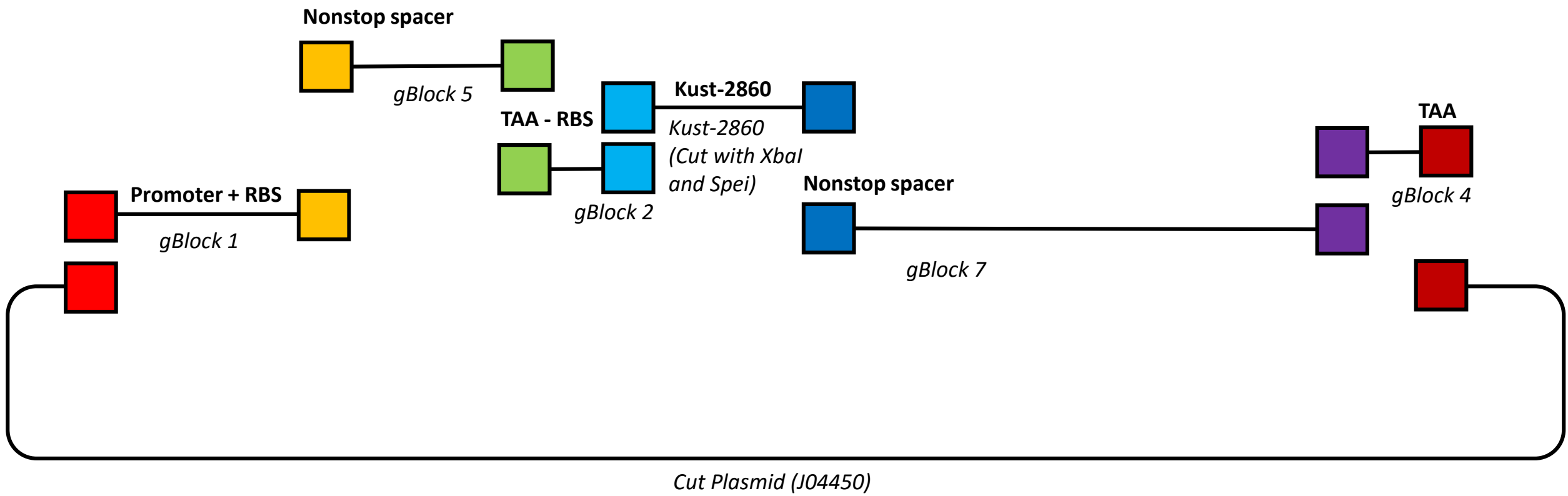
Assembly with
gBlock 1,
Kust2859,
gBlock6, gBlock7
gBlock 8, Plasmid



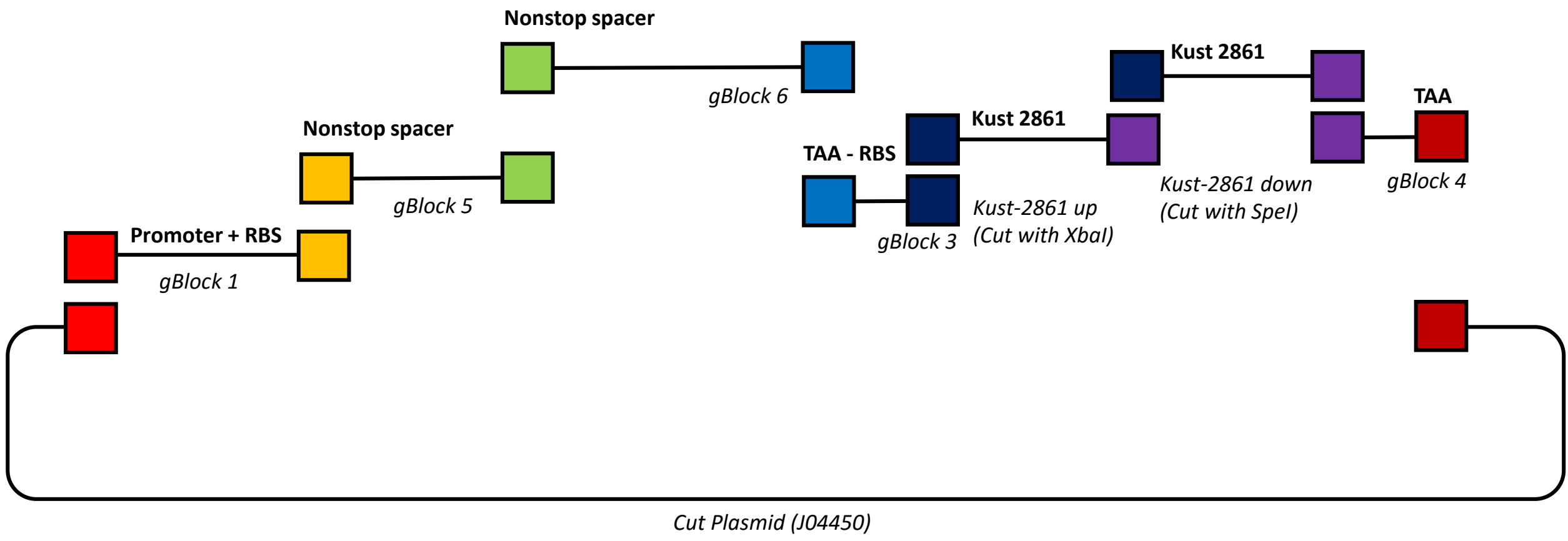
Assembly with
gBlock 1,
gBlock5, gBlock2,
Kust-2860,
gBlock7, gBlock8,
Plasmid



Assembly with
gBlock 1,
gBlock5, gBlock2,
Kust-2860,
gBlock7, gBlock4,
Plasmid



Assembly with
gBlock 1,
gBlock5, gBlock6,
gBlock3,
Kust2861 (up
and down),
gBlock 4, Plasmid



Assembly with
gBlock 1,
gBlock5, gBlock6,
gBlock3,
Kust2861 (up
and down),
gBlock 8, Plasmid

