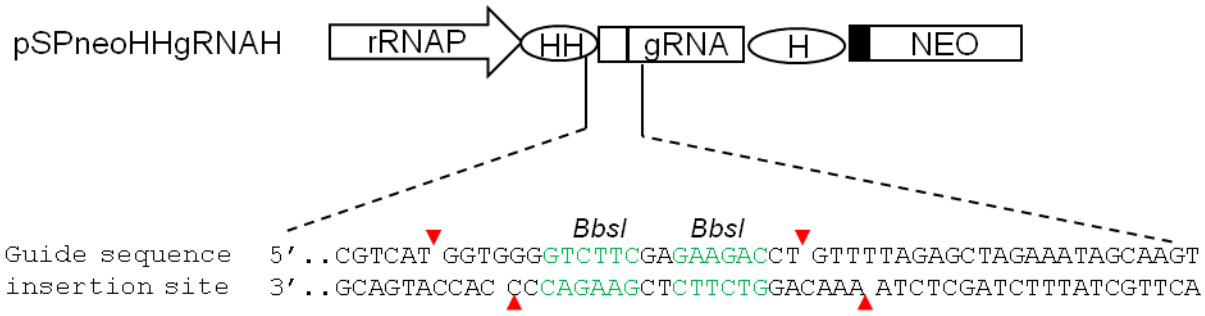


```
Guide sequence          5'GGTGNNNNNNNNNNNNNNNNNNN
        insert                NNNNNNNNNNNNNNNNNNNNCAAA 5'

          gRNA        5'- AUGGUGNNNNNNNNNNNNNNNNNNNNGUUUUAGAGCUA...- 3'
```

Note: the 6 base at the 5'end of gRNA can be matched exactly to the targeting site sequence if the 6 complementary base at the 5'end of Hammerhead sequence are adjusted accordingly.

pSPneoHHgRNAH (Hind III and BamH I fragment):

```
  1 AAGCTTGTGA GTTATGAGGT CTGCGATTGA CGTAGGAGTT GCAAGGGGGA GGGGGTATGA
 61 ACGGGGTGGG TAGAGCTTTT TTAGGTGGAA GTAGTGAGAG GGTGGGCTTG AGAGAATTTG
121 AGGTGTGTTC GTGATGTGTG GATCTTATCG GGGGCTCGGT TGAGTTTTTG GTTTGGTGAT
181 TTTGTAGGAA GCTTAGATCT CACCATCTGA TGAGTCCGTG AGGACGAAAC GAGTAAGCTC
241 GTCATGGTGG GGTCTTCGAG AAGACCTGTT TTAGAGCTAG AAATAGCAAG TTAAAATAAG
301 GCTAGTCCGT TATCAACTTG AAAAAGTGGC ACCGAGTCGG TGCTTTTTTG GCCGGCATGG
361 TCCCAGCCTC CTCGCTGGCG CCGGCTGGGC AACATGCTTC GGCATGGCGA ATGGGACGGA
421 TCC
```