

**STRAIN: EM09880**

**PRIMER** FRT 24 REV

**5'** CCTTCCTCCTACATAGTTGGCAGTG **3'**

**PRIMER** Rnase10

**5'** CCT GAG AAA GCC GTA CTG CAT TTC **3'**

**PRIMER** FRT 24 REV

15pm / total volume 30ul

Tm: 72°C

**PRIMER** Rnase10

15pm / total volume 30ul

Tm: 72°C

**PCR MIX**

DNTPS (2mM)

3ul

PCR BUFFER 10x

3ul

**PRIMER** FRT 24 REV

(0.5pm/ul)

**PRIMER** Rnase10

(0.5pm/ul)

H2O

TAQ POLYMERASE (5U/ul)

0.3ul

DNA

2ul

**TOTAL**

30ul

**PCR PROGRAM:**

- 1) 95°C x 2'00''
- 2) 95°C x 0'30''
- 3) 58°C x 0'30''
- 4) 72°C x 1'00''
- 5) step 2 to step 5 x 50 repeats
- 6) 72°C x 10'00''
- 7) 4°C forever
- 8) End

**MUTATION:** 274 bp