

## **STRAIN: EM00023**

**AMOG E1 -up/newFORW**      **5'-GAC/AGC/CCC/CCT/TTC/TAT/CGC/AGT-3'**  
**AMOG E1 -dw/newREV**      **5'-TGG/TCC/CGG/TGC/GCC/CCA/TGA/ACT-3'**  
**NCAM ko -dw/newREV**      **5'-CGG/AGA/ACC/TGC/GTG/CAA/TCC/ATC-3'**

**AMOG E1 -up/newFORW**    15pm / total volume 30ul      Tm: 76°C  
**AMOG E1 -dw/newREV**    1pm / total volume 30ul      Tm: 80°C  
**NCAM ko -dw/newREV**    15pm / total volume 30ul      Tm: 78°C

## **PCR MIX**

DNTPS (2mM)    3ul  
PCR BUFFER 10x    3ul  
**AMOG E1 -up/newFORW** (0.5pm/ul)  
**AMOG E1 -dw/newREV** (0.03pm/ul)  
NCAM ko -dw/newREV (0.5pm/ul)  
H2O  
TAQ POLYMERASE (5U/ul)                                  0.16ul  
DNA    1ul  
**TOTAL**    30ul

## **PCR PROGRAM:**

- 1) 95°C x 2'00''
- 2) 95°C x 0'45''
- 3) 65°C x 1'00''
- 4) 72°C x 1'20''
- 5) step 2 to step 5 x 40 repeats
- 6) 72°C x 10'00''
- 7) 4°C forever
- 8) End

**MUTATION: 650 BP**      **WILD TYPE: 350 BP**