

## *ihb180Tg/*+ *(AB)* (CZRC Catalog ID: CZ 327)

## Nature of the mutation

The transgenic zebrafish line  $Tg(HSP70:creb1a\_S133A -mcherry)$  with generated by random integration of a fusion mCherry-containing construct with an hsp70-1 (*hsp70l*) promoter to drive zebrafish *creb1a* transgene expression in whole body and eyes. mCherry was fused to the zebrafish *creb1a133S-A* which the protein in changed from S to A at the position 133, then the fusion proteins were cloned downstream of a 1.5 kb fragment of the zebrafish hsp70-1 promoter.

## Genotyping assay

There are two methods for *Tg*(*HSP70:creb1a\_S133A -mcherry*) Genotyping assay.

- Genotyping of the *Tg(HSP70:creb1a\_S133A -mcherry)* allele is based on the fluorescent microscopy. This line expresses jundn-mCherry ubiquitously by heat shock at 24 hpf. Heat shock is performed by transferring fish from 28 water to water preheated to 37 ℃ with subsequent incubation in an air incubator at 39 ℃ for 2 hour. The initial RFP expression in whole body and eyes at 60 hpf.
- ② Tg(HSP70:creb1a\_S133A -mcherry) allele genome was collected using a Tissue DNA Kit (Omega Bio-Tek) and was detected by the forward primer 5' ATGAACAGACGGGGCATTTAC 3' and reverse primer 5' TGATTGCTGGGAACAAGTAT 3' . The forward primer was located at the end of the hsp70l promoter, and the reverse primer was located at the end of C-terminus of zebrafish creb1a. A 984bp fragment was amplified using these two primers.



Figure. A transgenic zebrafish line  $Tg(HSP70:creb1a\_S133A$  -mcherry). The figure show the lateral view of  $Tg(HSP70:creb1a\_S133A$  -mcherry) embryos expresses creb1a\\_S133A -mCherry ubiquitously at 72 hpf after heat shock at 24hpf.

## **Reference**