

cei *p63cd*

Information on the mutation

The *p63cd* allele is a maternal recessive mutation induced on the AB/Tuebingen (AB/TU) genetic background. The *p63cd* mutation resides on the TU chromosome (LG14). The molecular nature of the mutation is unknown (Dosch et al., Developmental Cell 6: 771-780, 2004).

Genotyping assay

Genotyping of the *p63cd* mutation is based on a PCR assay using two closely linked SSLP markers (z-markers). These markers flank the mutation and reveal interstrain polymorphisms for the AB and TU genetic backgrounds. An individual fish is identified as homozygous for the *p63cd* mutation when both SSLP markers are homozygous for TU. This genotyping assay can be conducted only if the mutation on the TU background is crossed into the strain (AB) that is polymorphic for the closely linked z-markers on the mutant chromosome.

The flanking SSLP markers to genotype the *p63cd* mutation are: **z14634** (1.9 cM away from the *p63cd* mutation) and **z26570** (9.1 cM away from the *p63cd* mutation).

z14634-specific primers:

z14634_A: 5' TGT GTG TGG TGC ACC TGT AA 3'

z14634_B: 5' GCA CTT GTC TCG GTC TGA CA 3'

PCR program (58_40_40):

1. 94°C for 3 min
2. 94°C for 30 sec
3. **58°C for 40 sec**
4. 72°C for **40 sec**
5. Go to step 2 (above) for 39 cycles
6. 72°C for 5 min
7. 8.0°C hold
8. END

Product size: 195 bp for the TU chromosome
205 bp for the AB chromosome

**z26570-specific primers:****z26570_A:** GAG CCT GAC CGA ATA TGG AA**z26570_B:** AAG TTG ACG AAC ACA CGC AG**PCR program (58_40_40):**

1. 94°C for 3 min
2. 94°C for 30 sec
3. **58°C for 40 sec**
4. 72°C for **40 sec**
5. Go to step 2 (above) for 39 cycles
6. 72°C for 5 min
7. 8.0°C hold
8. END

Product size: 210 bp for the TU chromosome
195 bp for the AB chromosome**Zebrafish International Resource Center (ZIRC)**

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