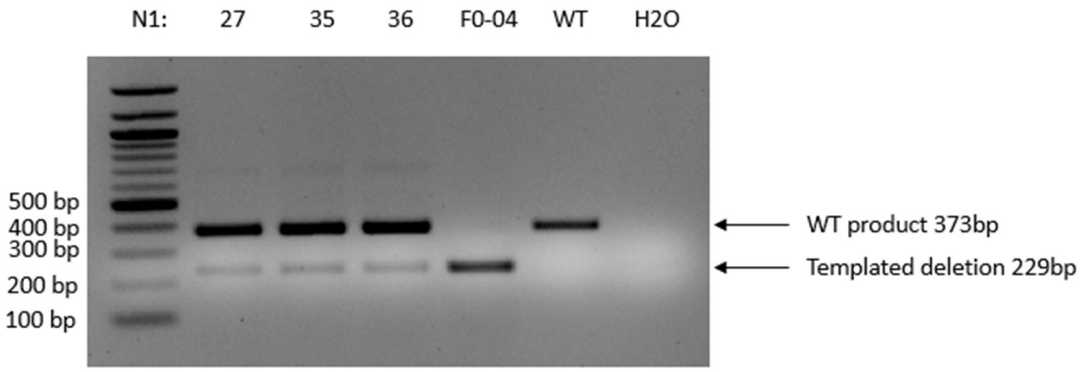
Vanderbilt Genome Editing Resource

GENOTYPING PROTOCOL: Rr546^{em1Mgn}/Gata6Δ1

Primer name	Sequence (5' to 3')	Primer type
Gata6-E4-Rev2	CCTGCTCGGTCATTAATACAG	WT/mutant
Gata6-WT-Specific_FWD	CTCTGCCTTCACCTAGAATTC	WT
Gata6-KO-Specific_FWD	CAAAAGCCAGAGCTCGAG	Mutant

Expected products:
WT = 373 bp
Rr546^{em1Mgn} = Templated Gata6 Rr KO = 229 bp, other deletions may vary in length



Reaction: EconoTaq PLUS 2x Master Mix, Biosearch Technologies

Component	Final Concentration
10 µM forward primer	0.5 µM
10 µM reverse primer	0.5 µM
Genomic DNA template	1 µl of tail lysate or purified DNA < 100 ng/µl
EconoTaq PLUS 2x Master Mix	1x

Thermocycler settings:

Program step	Temperature	Time
1	94°C	2 min
2	94°C	30 sec
3	50°C	30 sec
4	72°C	25 sec
5	go to step 2 for 35 cycles	
6	72°C	5 min
6	4°C	final hold (16°C for overnight)