

**Name of Mouse model or mutation:****CX3CL1-FLOX-EM1-B6N****Description:**

Floxed mutant made using CRISPR/Cas9.

**Type of mutation:**

Floxed exon ENSEMBL:ENSMUSE00001312139 of Cx3cl1 gene.

**Sequence details****Cx3cl1 WT**

```
ACAACGCAGGCCGATAGAAACAGAATGCAAAAATGTTTATATCAAATAACTAGGTTTATCCACTCAC
AGAAATCCCTGCCAGTGGCTTCTGCTACGTAGCTATCTGCATGTCCTTGCCCCCTACTTTGCCCCCA
GCTTTTCTCCTGAACTCCTTATCCCAAACCTCATTCTGCAGAACACAGGGGGATCTGCTCAGCAGACAC
TCACACACTTCCCAGGGAGGTAAGCTCCCCTATTCAACACAGACCCTTACCTGATCTATGCACCTGAT
CACATGTGTGTGGAGGAGGAGGATGCATGTGTAGGGTGATTGCTGAGCCCTTGACTTAATTGAGCA
CAAGACAGGGATGAGCAAGGAAGGCAATGCCGTAGGGGTGGAACAGGGCTGGAAGAGCGGGACT
GCACCATCATATCTCCCTAGATTCTCCTCCCGCTATGACTTCATTTTAAAAATTAACGTTTCCAGGC
CAGGAGTTGTGGTCCGCATCTTTAATCCCAGCACTTGGGAGGCAGTGGCAAGCAGATCTTGATGAG
CTCAAAGCCAGTCTGATTGTCTTAATGAGTACCAGGATAGCCAGAGCTACCTAGAGACCTTGTCTCA
AAATAACAACAACAACAACAACAACAACAATGAAATGATTCCCAGGTATCAGGCCAAAATGAGATTTATA
AATAAAGGCTAATTTTTTATCAGTCCAAAACAACCCAGAGGGCTGAGACTCTTGCTGTCTGGCAGGT
TATCACGGGTTGGGTTAGATGTGGATTCTGGCTTGGGAGTATAGCCTCGAGGAGCTGAAATGGGTA
TGGGGCACCAGATGCCAGGGGTCTACTGACTGCTGGCCTCTGTGGATCCAGGTCAGCACCTCGG
CATGACGAAATGCGAAATCATGTGCGACAAGATGACCTCACGAATCCCAGTGGCTTTGCTCATCCGC
TATCAGCTAAACCAGGAGTCCTGCGGCAAGCGTGCCATTGTGTAGGTATCCCTTTCAACCCCCAGAG
GCCCTAGGACATTCCAGCCCTCCCTCTCCCCCTCTCACCTGGGCTTCTGCCAAAGGCCTTAGTAA
GTGTGGGCCTTGTTCAGGCCACAGGCTATCTCCTCGCTGACCTTAAGATCAGTGGTAAACAACAG
AGGTGGGAAATGGTTTCCTGGATGGCTCACCGTGGTTCAGGGCAGCACCCAGGATGGAGCCTCCTT
TCTTCAGCCCCTAGATCCCATGGGGGAGGTTGAGGGAAGTGGCCTACTAGAGCAGGATCCAGACT
CCAGCAGGAGATGGGTAGCAGCAGGGTGAGAGGAGGTGGAGGAGCTAAAATGATTGGCTAACCA
CAAGGATGGTCATACCAAGTGACAGAAGAGCAGGGCCCTGGGAGCCTCTGGGGAGGAAACAGGC
TCAGTCAGTCATTTTCTGCCTGGCCAGTTTGGTCCCTTGTTCCTTCTGGGGCACATGGAGCGTACTGC
CTGTTTGCTCAGTCCCGAGT
```

**Mutant CX3CL1-FLOX-EM1-B6N**

ACAACGCAGGCCGATAGAAACAGAATGCAAAAATGTTTATATCAAATAACTAGGTTTATCCACTCAC  
AGAAATCCCTGCCAGTGGCTTCTGCTACGTAGCTATCTGCATGTCCTTGCCCCCTACTTTGCCCCCA  
GCTTTTCTCCTGAACTCCTTATCCCAAACCTATTCTGCAGAACACAGGGGGATCTGCTCAGCAGACAC  
TCACACACTTCCCAGGGAGGTAAGCTCCCCTATTCAACACAGACCCTTACCTGATCTATGCACCTGAT  
CACATGTGTGTGGAGGAGGAGGATGCATGTGTAGGGTGATTGCTGAGCCCTTGACTTAATTGAGCA  
CAAGACAGGGATGAGCAAGGAAGGCAATGCCGTAGGGGTATCCGGGGGTACCGCGTCGAGGCCGA  
TCGCATAACTTCGTATAGCATAATTATACGAAGTTATAATTAACGTTTCCAGGCCAGGAGTTGTG  
GTCCGCATCTTTAATCCCAGCACTTGGGAGGCAGTGGCAAGCAGATCTTGATGAGCTCAAAGCCAGT  
CTGATTGTCTTAATGAGTACCAGGATAGCCAGAGCTACCTAGAGACCTTGCTCAAATAACAACAA  
CAACAACACAACAAAATGAAATGATTCCCAGGTATCAGGCCAAAATGAGATTTATAAATAAAGGCTAA  
TTTTTTATCAGTCCAAAACAACCCAGAGGCTGAGACTCTTGCTGTCTGGCAGGTTATCACGGGTTG  
GGTTAGATGTGGATTCTGGCTTGGGAGTATAGCCTCGAGGAGCTGAAATGGGTATGGGGCACCAG  
ATGCCAGGGGTCTACTGACTGCTGGCCTCTGTGGATCCAGGTCAGCACCTCGGCATGACGAAAT  
GCGAAATCATGTGCGACAAGATGACCTCACGAATCCCAGTGGCTTTGCTCATCCGCTATCAGCTAAA  
CCAGGAGTCCTGCGGCAAGCGTGCCATTGTGTAGGTATCCCTTTCAACCCCCAGAGGCCCTAGGACA  
TTCCAGCCCTCCCTCTCCCCCTCCTCACCTGGGCTTCTGCCAAAGGCCTTAGTAAGTGTGGGCCTT  
GTTTCAGGCCACAGGCTATCTCCTCGCTGACCTAAGATCAGTGGTAAACAACAGAGGTGGGAAAT  
GGTTTCCTGGATGATAACTTCGTATAGCATAATTATACGAAGTTATCGCCGGCGGGTCTGAGCTCG  
CCATCAGTTAGATCCCATGGGGGAGGTTGAGGGAAGTGGCCTACTAGAGCAGGATCCAGACTCCAG  
CAGGAGATGGGTAGCAGCAGGGTGAGAGGAGGTGGAGGAGCTAAAATGATTGGCTAACCACAAG  
GATGGTCATCACCAAGTGACAGAAGAGCAGGGCCCTGGGAGCCTCTGGGGAGGAAACAGGCTCAG  
TCAGTCATTTTCTGCCTGGCCAGTTTGGTCCCTTGTTCTTCTGGGGCACATGGAGCGTACTGCCTGT  
TTGCTCAGTCCCGAGT

**Nucleotide Alignment: Orange is ENSMUSE00001312139, Red is LoxP sites, Yellow are universal primer and RE sites**

```

                *      20      *      40      *      60      *      80      *      100     *      120     *      140
Cx3c11_WT   : ACAACGCAGGCCGATAGAAACAGAATGCAAAATGTTTATATCAAATAACTAGGTTTATCCACTCACAGAAATCCCTGCCAGTGGCTTCTGCTACGTAGTATCTGCATGTCCTTGCCCCCTACTTTGCCCCAGCTTTTCTC
Cx3c11_FLOX : ACAACGCAGGCCGATAGAAACAGAATGCAAAATGTTTATATCAAATAACTAGGTTTATCCACTCACAGAAATCCCTGCCAGTGGCTTCTGCTACGTAGTATCTGCATGTCCTTGCCCCCTACTTTGCCCCAGCTTTTCTC

                *      160     *      180     *      200     *      220     *      240     *      260     *      280     *
Cx3c11_WT   : CTGAACCTCCTTATCCCAAACCTATTCTGCAGAACACAGGGGGATCTGCTCAGCAGACTCACACACTTCCAGGGAGGTAAGCTCCCTATTCAACACAGACCCTTACCTGATCTATGCACCTGATCAGATGTGTGGAGGAG
Cx3c11_FLOX : CTGAACCTCCTTATCCCAAACCTATTCTGCAGAACACAGGGGGATCTGCTCAGCAGACTCACACACTTCCAGGGAGGTAAGCTCCCTATTCAACACAGACCCTTACCTGATCTATGCACCTGATCAGATGTGTGGAGGAG

                300      *      320      *      340      *      360      *      380      *      400      *      420      *
Cx3c11_WT   : GAGGATGCATGTAGGGTGATGCTGAGCCCTTGACTTAATTGAGCACAAGACAGGGATGAGCAAGGAAGGCAATGCCGTAGGGGTGGAACAGGGGCTGGAAGAGCGGGGACTGCACCATCATATCTCCCTAGATTCCTCTCCCCG
Cx3c11_FLOX : GAGGATGCATGTAGGGTGATGCTGAGCCCTTGACTTAATTGAGCACAAGACAGGGATGAGCAAGGAAGGCAATGCCGTAGGGGT-----ATCCGGGGGTACCGCGTCGAGGCGATCGCATAAAGCTTCGATAGCATAATTA

                440      *      460      *      480      *      500      *      520      *      540      *      560      *      580
Cx3c11_WT   : CTATGACTTTCATTTTAAATAATTAAACGTTTCCAGGCCAGGAGTGTGGTCCGCATCTTTAATCCAGCACTTGGGAGGCAGTGGCAAGCAGATCTTGATGAGCTCAAAGCCAGTCTGATTGTCTTAATGAGTACCAGGATAGCCA
Cx3c11_FLOX : TACGAAGTTAT-----AATTAAACGTTTCCAGGCCAGGAGTGTGGTCCGCATCTTTAATCCAGCACTTGGGAGGCAGTGGCAAGCAGATCTTGATGAGCTCAAAGCCAGTCTGATTGTCTTAATGAGTACCAGGATAGCCA

                *      600      *      620      *      640      *      660      *      680      *      700      *      720
Cx3c11_WT   : GAGCTACCTAGAGACCTTGCTCAAAATAACAACAACAACAACAACAACAATAAATGAAATGATTCCAGGTATCAGGCCAAAATGAGATTTATAAATAAAGGCTAATTTTTTATCAGTCCAAAACAACCCAGAGGCTGAGACTCTTG
Cx3c11_FLOX : GAGCTACCTAGAGACCTTGCTCAAAATAACAACAACAACAACAACAACAATAAATGAAATGATTCCAGGTATCAGGCCAAAATGAGATTTATAAATAAAGGCTAATTTTTTATCAGTCCAAAACAACCCAGAGGCTGAGACTCTTG

                *      740      *      760      *      780      *      800      *      820      *      840      *      860      *
Cx3c11_WT   : CTGCTGGCAGGTTATCAGGGTTGGGTTAGATGTGGATTCTGGCTTGGGAGTATAGCCTCGAGGAGCTGAAATGGGTATGGGCACCAGATGCCAGGGGTCTACTGACTGCTGGCCTCTGTGGATCCAGTCTCAGCACCTCGG
Cx3c11_FLOX : CTGCTGGCAGGTTATCAGGGTTGGGTTAGATGTGGATTCTGGCTTGGGAGTATAGCCTCGAGGAGCTGAAATGGGTATGGGCACCAGATGCCAGGGGTCTACTGACTGCTGGCCTCTGTGGATCCAGTCTCAGCACCTCGG

                880      *      900      *      920      *      940      *      960      *      980      *      1000     *
Cx3c11_WT   : CATGACGAAATGCGAAATCATGTGCGACAAGATGACCTCAGGAATCCAGTGGCTTTGCTCATCCGCTATCAGCTAAACCAGGAGTCTGCGGCAAGCGTGCCATTGTGTAGGTATCCCTTCAACCCCCAGAGGCCCTAGGACA
Cx3c11_FLOX : CATGACGAAATGCGAAATCATGTGCGACAAGATGACCTCAGGAATCCAGTGGCTTTGCTCATCCGCTATCAGCTAAACCAGGAGTCTGCGGCAAGCGTGCCATTGTGTAGGTATCCCTTCAACCCCCAGAGGCCCTAGGACA

                1020     *      1040     *      1060     *      1080     *      1100     *      1120     *      1140     *      1160
Cx3c11_WT   : TTCCAGCCCTCCCTCTCCCTCCCTCACCCTGGGCTTCCCTGCCAAAGGCCCTAGTAAGTGTGGCCTTGTTTCAGGCCACAGGCTATCTCCTCGCTGACCTTAAGATCAGTGGTAAACAACAGAGGTGGGAAATGGTTTCTGG
Cx3c11_FLOX : TTCCAGCCCTCCCTCTCCCTCCCTCACCCTGGGCTTCCCTGCCAAAGGCCCTAGTAAGTGTGGCCTTGTTTCAGGCCACAGGCTATCTCCTCGCTGACCTTAAGATCAGTGGTAAACAACAGAGGTGGGAAATGGTTTCTGG

                *      1180     *      1200     *      1220     *      1240     *      1260     *      1280     *      1300
Cx3c11_WT   : ATC----GCTCACCGTGGTTTCCAGGCGAGCACCAGGATGGAGCCCTCCTTTCTCAGCCAC----TAGATCCCATGGGGAGGTTGAGGGAAGTGGCCTACTAGAGCAGGATCCAGACTCCAGCAGGAGATGGGTAGCAGCAGGG
Cx3c11_FLOX : ATCATAAAGCTTCGATAGCATAATTAACGAAGTTATCGCCGGCGGGTCTGAGCTCGCCATCAGTTAGATCCCATGGGGAGGTTGAGGGAAGTGGCCTACTAGAGCAGGATCCAGACTCCAGCAGGAGATGGGTAGCAGCAGGG

                *      1320     *      1340     *      1360     *      1380     *      1400     *      1420     *      1440     *
Cx3c11_WT   : TGAGAGGAGGTTGGAGGAGCTAAAATGATTTGGCTAACACACAGGATGGTCAATCACCAGTGCACAGAAGAGCAGGGCCCTGGGAGGCTCTGGGAGGAAACAGGCTCAGTCAGTCAATTTCTGCTGGCCAGTTTGGTCCCTTGTTC
Cx3c11_FLOX : TGAGAGGAGGTTGGAGGAGCTAAAATGATTTGGCTAACACACAGGATGGTCAATCACCAGTGCACAGAAGAGCAGGGCCCTGGGAGGCTCTGGGAGGAAACAGGCTCAGTCAGTCAATTTCTGCTGGCCAGTTTGGTCCCTTGTTC

                1460     *      1480     *
Cx3c11_WT   : CTTCTGGGGCACATGGAGCGTACTGCCTGTTTGGCTCAGTCCCGAGT
Cx3c11_FLOX : CTTCTGGGGCACATGGAGCGTACTGCCTGTTTGGCTCAGTCCCGAGT

```

**QC strategy employed at Harwell to check the edited allele:**

Genomic DNA was extracted from ear clip biopsies and amplified in a PCR reaction using the following conditions/primer sequences:

**Universal PCR:**

Geno_Cx3cl1 _F1	ACAACGCAGGCCGATAGAAA
Geno_Cx3cl1 _R1	ACTCGGGACTGAGCAAACAG
Taq Polymerase used	Roche Expand Long Range DNTPack
Annealing Temperature (°C)	58
Elongation time (min)	1
WT product size (bp)	1488
Mutant product size (bp)	1483
Notes	Sequence with internal primers Geno_Cx3cl1 _F2 (GAACACAGGGGGATCTGCTC) and Geno_Cx3cl1 _R3 (ACTGACTGAGCCTGTTTCCTC)

**LoxP PCR:**

LoxPF	ATCCGGGGGTACCGCGTCGAG
LoxPR	ACTGATGGCGAGCTCAGACC
Taq Polymerase used	Roche Expand Long Range DNTPack
Annealing Temperature (°C)	61
Elongation time (min)	3
Mutant product size (bp)	835
Notes	This is a generic LoxP PCR hence longer extension time than required for amplicon size.

All amplicons were sent for Sanger sequencing to check for integration of the donor oligo sequence at the target site. F1 sequences should be heterozygous unless on Y chromosome.

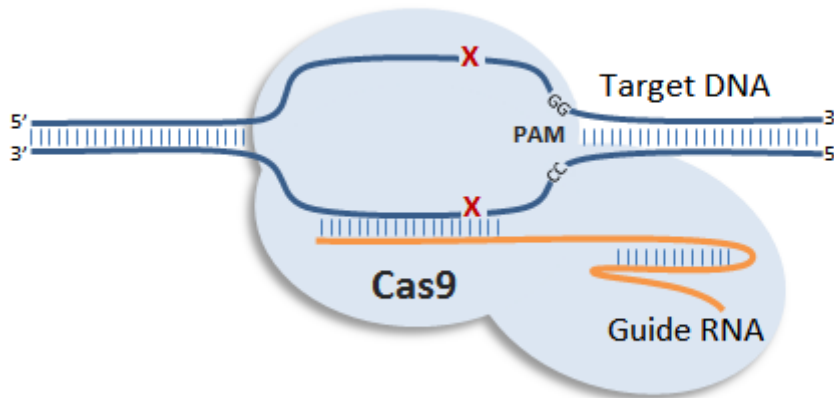
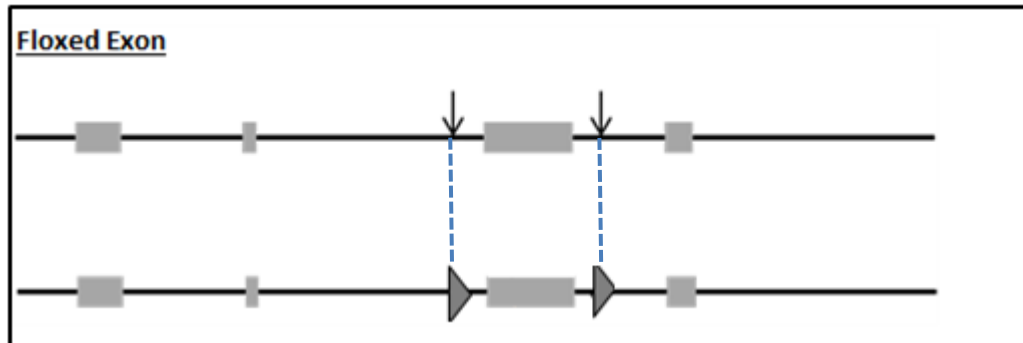
Copy counting of the donor sequence was carried out by ddPCR at the F1 stage to confirm donor oligos were inserted once on target into the genome. The following Taqman assay was used to detect the donor sequence:

Assay name	Cx3cl1-CR-LOA-WT1
Forward Primer	CGGCATGACGAAATGCGAAATC
Reverse Primer	GCAGGACTCCTGGTTTAGCT
Probe	TGTGCGACAAGATGACCTCACGA
Label	FAM-BHQ1

The reference assay for the copy counting assay is a VIC-labelled assay that detects Dot1l on Chr10.

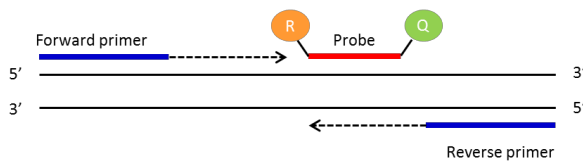
## Cx3cl1-FLOX Genotyping Strategy

Animals have been engineered using the CRISPR/Cas9 technology. Most of the knockout alleles generated through this method will be obtained by deletion of a critical exon or by introduction of indel (insertion/deletion) within the coding sequence of a critical exon. There is also a possibility of inserting sequences like LoxP sites around critical exon to create floxed allele (see picture below).

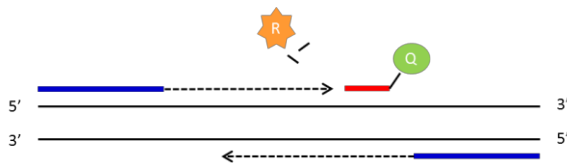


# qPCR genotyping strategy

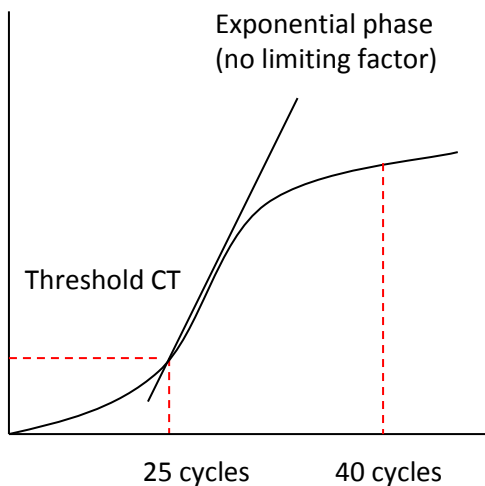
Standard PCR is the amplification of DNA between a pair of primers. Quantitative PCR employs the same principal as standard PCR, although it actually monitors the progress of the DNA synthesis as it occurs. The progress of the reaction is measured by using a Taqman probe. This is a short DNA oligo that is complimentary to part of the DNA sequence between the forward and reverse primers. At the 5' end of the probe there is a fluorescent reporter (R) and at the 3' end a quencher (Q). Whilst they are in close contact with each other there is no fluorescent signal.



As the forward primer is extended the reporter is cleaved from the probe resulting in a fluorescent signal being detected. Once the primer extends enough to release the quencher this signal is blocked. By using probes with different fluorescent signals multiple PCR assays can be multiplexed and run together.



## PCR reaction plot



The number of cycles the PCR takes to reach a set threshold is known as the CT value. This is inversely correlated to the amount of template DNA in the sample.

- e.g. CT 25 = 2 x template DNA
- CT 26 = 1 x template DNA
- CT above 30 = no template represented in the sample

CT value can be used to determine how many copies of a particular allele samples have.

All our qPCR are run in duplicate. A FAM labelled genotyping assay is run in multiplex with a VIC labelled internal control Dot1I.



# Cx3cl1-FLOX Genotyping Strategy

Samples are genotyped with a WT loss of allele (WT-LOA) assay. This is a FAM labelled assay that is designed to detect the critical exon that has been floxed. If the animal contains the modified allele the copy number of this assay should drop by 1. For autosomal genes that have been targeted this means the following

- WT= 2 copies of the LOA assay
- HET = 1 copy of the LOA assay
- HOM = 0 copies of the LOA assay

## Cx3cl1-FLOX-WT1 assay (FAM labelled probe)

### Cx3cl1 WT

Sequence highlighted in blue letters is the BP-LOA (break point loss of allele) where the two loxp sites have been inserted

```
TGGAGGAGGAGGATGCATGTGTAGGGTGATTGCTGAGCCCTTGACTTAATTGAGCACAAGACAGGGATGAGCAAGGAAGGCCAA
TGCCGTAGGGGTGGAACAGGGCTGGAAGAGCGGGACTGCACCATCATATCTCCCTAGATTCCCTCTCCCCGCTATGACTTCATT
TTAAATAATTAAACGTTTCCAGGCCAGGAGTTGTGGTCCGCATCTTTAATCCCAGCACTTGGGAGGCAGTGGCAAGCAGATCTT
GATGAGCTCAAAGCCAGTCTGATTGTCTTAATGAGTACCAGGATAGCCAGAGCTACCTAGAGACCTTGTCTCAAAAATAACAAC
AACAAACACAACAAAAATGAAATGATTTCCAGGTATCAGGCAAAAATGAGATTTATAAATAAAGGCTAATTTTTTATCAGTCC
AAAACAACCCAGAGGCTGAGACTCTTGTCTGTCTGGCAGGTTATCACGGGTTGGGTTAGATGTGGATTCTGGCTTGGGAGTAT
AGCCTCGAGGAGCTGAAATGGGTATGGGGCACCAGATGCCAGGGGTCTACTGACTGCTGGCCTCTGTGGATCCAGGTCAGC
ACCTCGGCATGACGAAATGCGAAATCATGTGCGACAAGATGACCTCACGAATCCCAGTGGCTTTGTCTCATCCGCTATCAGCTA
AACCAGGAGTCTTGCAGCAAGCGTGCCTTTGTGTAGGTATCCCTTTCAACCCCAAGAGGCCCTAGGACATTCCAGCCCTCCCT
CTCCCTCCTCACCTGGGCTTCCCTGCCAAAGGCCTTAGTAAGTGTGGCCCTTGTTCAGGCCACAGGCTATCTCCTCGCT
GACCTTAAGATCAGTGGTAAACAACAGAGGTGGGAAATGGTTCCTGGATGGCTCACCGTGGTTCAGGGCAGCACCAGGATG
GAGCCTCTTTCTTCAGCCCACTAGATCCCATGGGGGAGGTTGAGGGAAGTGGCCTACTAGAGCAGGATCCAGACTCCAGCAG
```

### Cx3cl1-FLOX-EM1-B6N

Sequence highlighted in RED letters is the mutant sequence replaced by BP-LOA from above

```
TGGAGGAGGAGGATGCATGTGTAGGGTGATTGCTGAGCCCTTGACTTAATTGAGCACAAGACAGGGATGAGCAAGGAAGGCCAA
TGCCGTAGGGGTatccgggggtaccgctcgaggcgcgcataacttcgatagcatacattatacgaagtataAATTAAAC
GTTTCCAGGCCAGGAGTTGTGGTCCGCATCTTTAATCCCAGCACTTGGGAGGCAGTGGCAAGCAGATCTTGATGAGCTCAAAG
CCAGTCTGATTGTCTTAATGAGTACCAGGATAGCCAGAGCTACCTAGAGACCTTGTCTCAAAAATAACAACAACAACAACAAA
CAAAATGAAATGATTTCCAGGTATCAGGCAAAAATGAGATTTATAAATAAAGGCTAATTTTTTATCAGTCCAAAACAACCCAG
AGGCTGAGACTCTTGTCTGTCTGGCAGGTTATCACGGGTTGGGTTAGATGTGGATTCTGGCTTGGGAGTATAGCCTCGAGGAGC
TGAATGGGTATGGGGCACCAGATGCCAGGGGTCTACTGACTGCTGGCCTCTGTGGATCCAGGTCAGCACCTCGGCATGAC
GAAATGCGAAATCATGTGCGACAAGATGACCTCACGAATCCCAGTGGCTTTGTCTCATCCGCTATCAGCTAAACCAGGAGTCCCT
GCGGCAAGCGTGCCATTGTGTAGGTATCCCTTTCAACCCCAAGAGGCCCTAGGACATTCCAGCCCTCCCTCTCCCTCCCTCA
CCCTGGGCTTCCCTGCCAAAGGCCTTAGTAAGTGTGGCCCTTGTTCAGGCCACAGGCTATCTCCTCGCTGACCTTAAGATCA
GTGGTAAACAACAGAGGTGGGAAATGGTTTCTGGATGataacttcgatagcatacattatacgaagttatcgccggcgggt
ctgagctcgccatcagtTAGATCCCATGGGGGAGGTTGAGGGAAGTGGCCTACTAGAGCAGGATCCAGACTCCAGCAG
```

## Cx3cl1-FLOX-WT1 primers and probe

Primer 1 = AAACAACAGAGGTGGGAAATGGT                      Primer 2 = GGGATCTAGTGGGCTGAAGAAAG  
 Probe = ACCGTGGTTCAGGGCAGCACC

## Dot1l internal control (VIC labelled)

```
TCATAGGGTGACTGGCCAACCCAGGGAAGCCGGAGTGTGCGTCTTCTGTTTCTTCTTCCCTCTAGTCGTTTTTCTG
TTAGTAGTTGGCATCCTTATGCTTCATCTTACAGTCCGACTTGAGAGCTGCCCTGAATGGTTCGTGCTGGGGCAAGGCTTTAAT
TCAGGCGTAGCACACATGGTGGCCAATGGGACTCTGTAGGATCTGCCACACCCATCAG
```

Primer 1 = GCCCCAGCACGACCATT                      Primer 2 = TAGTTGGCATCCTTATGCTTCATC  
 Probe = CCAGCTCTCAAGTCG



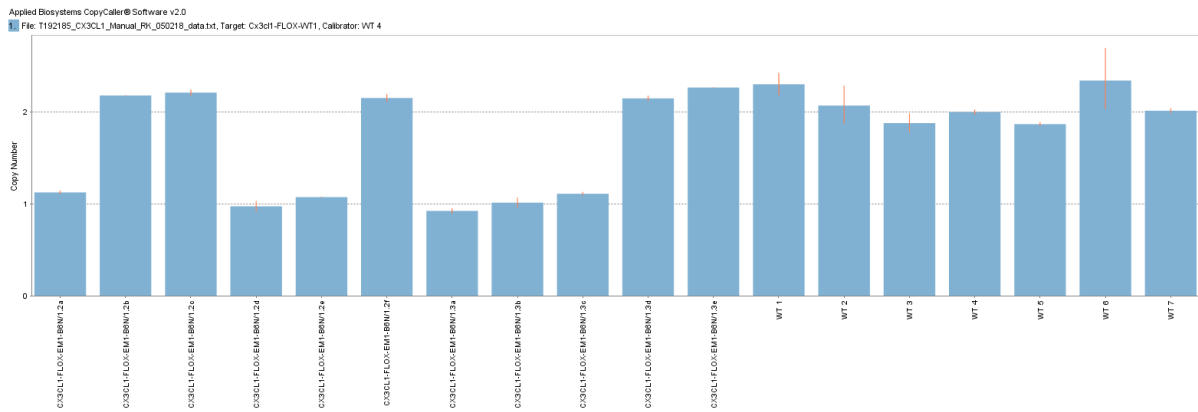


qPCR master mix

ABI GTX Taqman master mix	5µl
Primers Dot1L_2F (20µM)	0.225µl
Primers Dot1L_R (20µM)	0.225µl
Probe DotL_2M (5µM)	0.2µl
FAM Assay (probe 5µM & primers 15µM each)	0.3µl
ddH2O	1.55µl
DNA (1/10 dilution of ABI Sample-to-SNP prep)	2.5µl

Cx3cl1-FLOX LOA copy called result, image showing both replicates and controls

Task 192185 Results



Version No. 1  
Date: 06.02.2018  
Created/Updated by: Ramakrishna Kurapati  
Approved by: Deen Quwailid