



## Lexicon Genetics Incorporated – Genentech Project Materials

|                               |             |                               |                                      |
|-------------------------------|-------------|-------------------------------|--------------------------------------|
| <b>Genentech ID:</b>          | UNQ3003     | <b>Date of Submission:</b>    | 3-31-05                              |
| <b>Lexicon Contract Name:</b> | DNA058      | <b>Mutation Type:</b>         | X Standard Knock out                 |
| <b>LexVision Name:</b>        | KNS504N1    |                               | <input type="checkbox"/> Conditional |
| <b>Reference accessions:</b>  | NM_022416.1 | <b>Is this gene X-linked?</b> | No                                   |

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**Required Materials:**

X pKOS clone DNA(s)    \_\_pKOS60\_\_\_\_

X Target Vector DNA    \_\_pKOS60TV\_\_\_\_

X Targeted ES Cell DNA    \_\_1A4\_\_\_\_

X Genomic Map

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### Southern Blot Analysis: *External/Internal Probe Strategies*

|                                        | <u><b>5' External</b></u> | <u><b>3' External</b></u> |
|----------------------------------------|---------------------------|---------------------------|
| Name of Probe:                         | <b>17+18</b>              | <b>13+14</b>              |
| Restriction Enzyme for Genomic Digest: | <b>EcoRI</b>              | <b>HindIII</b>            |
| Predicted Wild-type Band (kb):         | <b>10.5 kb</b>            | <b>9.4 kb</b>             |
| Predicted Mutant Band (kb):            | <b>5.6 kb</b>             | <b>14.9 kb</b>            |
| Probe Size:                            | <b>534 bp</b>             | <b>242 bp</b>             |

**PCR Strategies:**

*For standard knockouts, give wildtype and mutant-specific strategies*

*For conditionals, give 5' loxP and cre-excision strategies*

| Wild type-specific (absent in targeted allele) |          | Mutation-specific product (absent in wt) |           |
|------------------------------------------------|----------|------------------------------------------|-----------|
| 5' Primer Name:                                | DNA058-7 | 5' Primer Name:                          | DNA058-7  |
| 3' Primer Name:                                | DNA058-8 | 3' Primer Name:                          | IRESUP-LR |
| Predicted Wild-type Band (bp):                 | 384 bp   | Predicted Wild-type Band (bp):           | none      |
| Predicted mutant band (bp)                     | none     | Predicted mutant band (bp)               | 409 bp    |

| 5' loxP strategy               |  | Distinguish Cre-excised and wt |  |
|--------------------------------|--|--------------------------------|--|
| 5' Primer Name:                |  | 5' Primer Name:                |  |
| 3' Primer Name:                |  | 3' Primer Name:                |  |
| Predicted Wild-type Band (bp): |  | Predicted Wild-type Band (bp): |  |
| Predicted mutant band (bp)     |  | Predicted mutant band (bp)     |  |

**Primer sequences:****Southern probes**

DNA058-17 5' – CTATATGAGTTTTTCAGATACCAAG

DNA058-18 5' – GGACATCCACTTCCTTAATATC

DNA058-13 5' – GCCTGCTTCCAAGAGTCCATCA

DNA058-14 5' – CAGATGCACTGGAAAGTCTTTG

**PCR Genotyping**

DNA058-7 5' – CCACATTTAAGTGCTAAATC

DNA058-8 5' – CCACTCACCATAGGTTTCACC

IRESUP-LR 5' – TGGAAAATAACATATAGACAAACGCACACC

### **Genomic Sequence Deleted:**

TGCAGAAGCGAGACACAAAGAAGATGTATGCCATGAAGTACATGAACAAGCAAAAGTGTGTGGAGAGGGATGAGGTG  
CGGAACGTGTTCCGGGAGCTGCAGATCATGCAGGGATTAGAGCACCCCTTCCTGGTGAACCTATGGTGAGTGGGAGGT  
GACTTGGTCCCCAAGAATGGTGTCTTGGCACTGCCAAAAAGGGA

**Genomic Locus:** *(The deleted sequence represents nt 13863-14061 in the sequence below. KOS60 used to generate the TV represents nt 12608-21565 in the sequence below.)*

ATGGTCACCCATGTCTGGCTGATACAAAGCATCATTTCTGCCAGGCAGGACTCAGGATATGTGGTTGCTCTGTCTGGGA  
AATTGTCAGTTAATTAAGTTGTGAAGCAGCGTTCTGTGTGTGAGCTTTGGCTTGCAATTTTGGCCCAACCACAAGCCTCT  
GGGCGAGTTGGCGGACTTCCCCGGCTGCTGTTCTTCTGTGAGAATCCTTTTATAGAAGGCTTAAAGGAGATGCTCGTAT  
GAGCTGCCCTTGTTCTCCAGACATAGCGAGGAAGGATGGGATCCATTTGTTTGCAATGAAATATTTATTCCTTTTCATAT  
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CAGGAGATGCCCCCTTACGAGGCCTCCAACTCTGGCTGGGGCCAAGGACACTTCCTGTCAAAGACGCTTGCTTCTCCA  
ACCAGAAGGCAGGTCTCCCGCTCATCCACATCTCAGTTTCTTCTATAGCGAGCAAGTCCTGGGCTCTGCTCCCAACAT  
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### **Selection Cassette:**

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### **Targeted Locus:**

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