



## Lexicon Genetics Incorporated – Genentech Project Materials

|                               |                                  |                               |                                      |
|-------------------------------|----------------------------------|-------------------------------|--------------------------------------|
| <b>Genentech ID:</b>          | UNQ1925                          | <b>Date of Submission:</b>    | 11/02/03                             |
| <b>Lexicon Contract Name:</b> | DNA290                           | <b>Mutation Type:</b>         | X Standard Knock out                 |
| <b>LexVision Name:</b>        | ENZ779N1                         |                               | <input type="checkbox"/> Conditional |
| <b>Reference accessions:</b>  | NM_028117,<br>ENSMUST00000028809 | <b>Is this gene X-linked?</b> | No                                   |

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**Required Materials:** X pKOS clone DNA(s) \_\_KOS6, KOS92\_\_\_\_\_  
X Target Vector DNA \_\_KOS92-FTV\_\_\_\_\_  
X Targeted ES Cell DNA \_\_2H3\_\_\_\_\_  
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>13/14</b>              | <b>17/18</b>              |
| Restriction Enzyme for Genomic Digest: | <b>HindIII</b>            | <b>ScaI</b>               |
| Predicted Wild-type Band (kb):         | <b>9.6</b>                | <b>11.4</b>               |
| Predicted Mutant Band (kb):            | <b>8.1</b>                | <b>16.1</b>               |
| Probe Size:                            | <b>396 bp</b>             | <b>337 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:                                | 7      | 5' Primer Name:                          | Neo3A  |
| 3' Primer Name:                                | 8      | 3' Primer Name:                          | 8      |
| Predicted Wild-type Band (bp):                 | 461 bp | Predicted Wild-type Band (bp):           | none   |
| Predicted mutant band (bp)                     | none   | Predicted mutant band (bp)               | 276 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**

|           |                             |
|-----------|-----------------------------|
| DNA290-13 | 5' – CGGTGTGAGCAGTCAGGTCA   |
| DNA290-14 | 5' – GGTGTTAAAGGAAGGTTTTAG  |
| DNA290-17 | 5' – CTGTCTCCTGTCTTATAGCTGG |
| DNA290-18 | 5' – CACATTGCCTTGGAGTTCAAG  |

**PCR Genotyping**

|          |                             |
|----------|-----------------------------|
| DNA290-7 | 5' – GTGTGCAATTCTACTCCAGACT |
| DNA290-8 | 5' – CCACTGGACCTGTTTAAGCAGG |
| Neo3A    | 5' – GCAGCGCATCGCCTTCTATC   |

### **Genomic Sequence Deleted:**

GTTTCCCCGCCCTCTGACCCCACTGGCTGCCCCGAAAAGCGCGGAGACCCTGGGCCGCACGCCAAGGCGGGCCCCATT  
GGGCCGGGCCCCGGGCTGGGCTCGGGGGGCCCCCTGCTGCTGCCGTCCATGCTGATGTTTCGCTGTAATCGTGGCCTCC  
AGCGGACTGCTGCTCATGATCGAGCGAGGCATCCTATCGGAGATGAAACCCCTTCCCCTGCACCCTCCCAGCCACAAA  
GGCGCGGCCTGGAGCGGGACAGATCCTAAGCCTAGAGGCCTATCCTTGGATGCTGGGGACTCGGACTTGCAAGTGAGG  
GAGGACATCCGAAACCGGACCTTGAGGGCCGTGTGCGGACAACCAGGCATGCCCCGGGACCCCTGGGACTTGCCGGTG  
GGACAGCGGCGCACCCCTGCTGCGCCACATTCTCGTAAGTGACCGCTACCGCTTCTCTACTGCTATGTCCCCAAAGTGG  
CCTGCTCTAACTGGAACGTGTGCTGAAGGTGCTGGCTGGCATCCTGAACAACGTGGATGTCCGCCTCAAGATGGACCA  
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TGTCACCAAGGAAGCCTGTACCAATGACAGTAGGCCAGCACCTTTTGGAGTTTGGGTTTAATGATATCAGCTTTGGGA  
TGTCTTTCAGAGAAACTCCTGGCTCTGGGTGGCTTCTGTTTCTCTAGGTGTCTCCATATCTCAGTGGTAAGGACTGTC  
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GCCCCCTGGCTGCTGGGTCTATGCAGTCCACCTGGTCTGTTGTAATTTAACCTGTGGCCAAATCCCAAATATGACACTA  
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GCTGTGGTGCCTTAGAAATGAAATGTGTGCAATTCTACTCCAGACTTTTACATTTCTCCTCTTGTCTAGGTCTGAATCAT  
TTTTCTAAGGAAAGAGAAACGGAAGTGGGGCCCTTACCTCGAAGCTCTAAAGCCCAGCCCCTCAAGCATCCAAAGACG  
CCTGTGCCTGACCTCTTCTAGGGCTCCTGGAGCATCTTCAATAAGCCTCCCTTCCCTACAAACCTTTGGAGACTATGTG  
AGACTGTATGGCCCATATATCTGGCTGTCATTGTCTAATGCATTTATTTAAATGTGTATATTTAATAGGATCCTTGT  
AAGGGCTGACTTTTAATAAAGCTTTTTCATATACAAAATGAGCAATTCTGTAATTCC

### **Genomic Locus: (the deleted sequence represents nt 10,127-12,077 -in the sequence below)**

CTTCTTAAAACTCAGCCAGGAGCGGGAGCTACCTCTTCGACCTTCTCGTGCTCAAGCAGAAGCAGATGGGCGCTCCACC  
GAGCCCCTGGCACCCAGGATCCTGCGGCCAAAGGTCAACGGCAAGAAGTGTCCCAAGGCTCGGCAAGGAGCTGGCTCT  
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CACCAGTAAGCACTGTGTGCCAAGAGCCGTTCTCAGTGCTCAACACACAGGTCTCAAAGGTGTGTAATTAGGCAGAC  
TCAGCCAAACTACCTGAGTTAAATGTGTGCCCCCCCCCTCCTCATGGCCATTGGATGAGCCAGTAAGTCTGTCTGTT  
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### **Selection Cassette:**

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### **Targeted Locus:**

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[illegible]