



## Lexicon Genetics Incorporated – Genentech Project Materials

|                               |          |                               |                                      |
|-------------------------------|----------|-------------------------------|--------------------------------------|
| <b>Genentech ID:</b>          | UNQ1902  | <b>Date of Submission:</b>    | 3-23-05                              |
| <b>Lexicon Contract Name:</b> | DNA187   | <b>Mutation Type:</b>         | X Standard Knock out                 |
| <b>LexVision Name:</b>        | SEC527N1 |                               | <input type="checkbox"/> Conditional |
| <b>Reference accessions:</b>  | AK076169 | <b>Is this gene X-linked?</b> | No                                   |

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**Required Materials:** X pKOS clone DNA(s) \_\_pKOS3, pKOS8, pKOS75\_\_\_\_\_  
X Target Vector DNA \_\_pKOS8TV\_\_\_\_\_  
X Targeted ES Cell DNA \_\_2C10\_\_\_\_\_  
X Genomic Map

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

|                                        | <b><u>5' External</u></b> | <b><u>3' Internal</u></b> |
|----------------------------------------|---------------------------|---------------------------|
| Name of Probe:                         | 25+26                     | Neo2+Neo5                 |
| Restriction Enzyme for Genomic Digest: | <b>BamHI</b>              | <b>AvrII</b>              |
| Predicted Wild-type Band (kb):         | <b>10.3 kb</b>            | -                         |
| Predicted Mutant Band (kb):            | <b>7.6 kb</b>             | <b>15.7 kb</b>            |
| Probe Size:                            | <b>205 bp</b>             | <b>609 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:                                | DNA187-17 | 5' Primer Name:                          | DNA187-17 |
| 3' Primer Name:                                | DNA187-2  | 3' Primer Name:                          | GTIRES    |
| Predicted Wild-type Band (bp):                 | 292 bp    | Predicted Wild-type Band (bp):           | none      |
| Predicted mutant band (bp)                     | none      | Predicted mutant band (bp)               | 315 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**

DNA187-25     5' – CATCTTCAAAAAAGTGGGAAAG

DNA187-26     5' – CATAGATTTAGCAAGGGGGTG

Neo2            5' – CCTCAGAAGAACTCGTCAAG

Neo5            5' – GGCAGCGCGGCTATCGTG

**PCR Genotyping**

DNA187-17     5' – CTGATTGGCTCTGGAGCGAGG

DNA187-2       5' – GCGAACTCCTGCAGATTGCTG

GTIRES         5' – CCCTAGGAATGCTCGTCAAGA

**Genomic Sequence Deleted:**

CTTCTCCTCTACTCGCAAAGATGACGCTTGCCGTCCTGGCCCTGCGGCTGGTGGTCTGTACCCTGGCGCTTCCCATGTT  
TCTGCTGAACCTTCTAGGCTTGTTGGAGCTGGGTATGCAAAAAGTGCTTTCCCTACTTCCTGAAGCGATTACCGTGATGT  
ACAATGAGCAGATGGCGAGCCAAAAGCGGGAGCTCTTCAGCAATCTGCAGGAGTTCGCCGGCCCCCTCGGGGAAGCTGA  
CTCTGCTGGAGGTGGGCTGCGGCACCGGAGCCAACTTCAAGTTCTATCCCCCGGGTGCAGGGTCACTTGTATCGACCC  
CAACCCCAACTTCGAGAAGTCTTGTTCAAGAGCGTCGCAGAGAACC GG CAGCTGCAGTTCGAGCGCTTCGTGGTGGC  
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GAACCAGGAGAAGATTCTGCGTGAGGTGTGCCGAGTGCTGAAGCCGGTGAGTGGCAGGAGTGAGGACTGGCTAGCTGC  
AGCTAGGAGTGAGGGTGCTCCTATCAGTTTCGGGTGGATTCTAAACCAAAACAAACAAACAAACAAACAAAGCAA  
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GAGGGTCACCCACTCATTCAACACAAAACAAAACAACTTCCAAGTCCCGGCAGGCAATGGGAAGCAGAGGCAGGT  
GGATCTCTGAAGCTCGAGGGTCTCGTTGCCAGCATGCACAGAGACCATTGTCTCCAAATAAAACAGCAAAATTTA  
CAGGTCTAGTAAGGATACTGGTCTGTGTTTTGAAATGGGTCAAGGAAAAAGATGAGCAACTGGGCAGGAACCAAGATA  
TAAACTAAAGCAGACAATCATGAGACAGACTGTAAGGCCGTGTTT

**Genomic Locus:** *(The deleted sequence represents nt 8577-9638 in the sequence below. KOS8 used to generate the TV represents nt 2836-12578 in the sequence below.)*

ATGCTTCATGTATGTGCACATCATGGTATGACAAATAACAACCTTTTAGAGTGGAGAGCCAGTGAGATAGCTCAGCAGGTT  
TTTGCAGCCAAGACCATGACAGCCTGATTATGATCCCTGGGACCCACGTAGTAAAGGAGACAAGTGACTCCACAGTT  
GTCCTCTGACCTCTACACATGTATCCTGGCATGTATGCCTGTGTTCTACACACACACACACACACACACACACAC  
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[illegible]

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### **Selection Cassette:**

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### **Targeted Locus:**

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