



## Lexicon Genetics Incorporated – Genentech Project Materials

|                               |           |                               |                                      |
|-------------------------------|-----------|-------------------------------|--------------------------------------|
| <b>Genentech ID:</b>          | UNQ8923   | <b>Date of Submission:</b>    | 2-25-04                              |
| <b>Lexicon Contract Name:</b> | DNA457    | <b>Mutation Type:</b>         | X Standard Knock out                 |
| <b>LexVision Name:</b>        | SEC241N1  |                               | <input type="checkbox"/> Conditional |
| <b>Reference accessions:</b>  | NM_015775 | <b>Is this gene X-linked?</b> | No                                   |

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**Required Materials:**

- ✓ pKOS clone DNA(s) \_\_pKOS17\_\_
- ✓ Target Vector DNA \_\_DNA457 TV\_\_
- ✓ Targeted ES Cell DNA \_\_3F9\_\_
- ✓ Genomic Map

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

|                                        | <u><b>5' External</b></u> | <u><b>3' Internal</b></u> |
|----------------------------------------|---------------------------|---------------------------|
| Name of Probe:                         | <b>DNA457-10/24</b>       | <b>DNA457-21/22</b>       |
| Restriction Enzyme for Genomic Digest: | <b>BamHI</b>              | <b>EcoRV</b>              |
| Predicted Wild-type Band (kb):         | <b>2.8</b>                | <b>12.4</b>               |
| Predicted Mutant Band (kb):            | <b>1.9</b>                | <b>14.6</b>               |
| Probe Size:                            | <b>459 bp</b>             | <b>168 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:                                | DNA457-5 | 5' Primer Name:                          | Neo3a    |
| 3' Primer Name:                                | DNA457-9 | 3' Primer Name:                          | DNA457-9 |
| Predicted Wild-type Band (bp):                 | 288 bp   | Predicted Wild-type Band (bp):           | none     |
| Predicted mutant band (bp)                     | none     | Predicted mutant band (bp)               | 233 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**

DNA457-10    5' – GCACTAGGATTACAGTTG  
 DNA457-24    5' – CTGCTCTATGATTGTAATTTC  
 DNA457-21    5' – GAGATCAGTGCTTCCAGG  
 DNA457-22    5' – CAATACAATGCCAGGGGAC

**PCR Genotyping**

DNA457-5    5' – CGGACCTTGCTATGAGAACC  
 DNA457-9    5' – GACAAGGTAGGAGAAATAAGAG  
 Neo3a        5' – GCAGCGCATCGCCTTCTATC

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[illegible]

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**Selection Cassette:**

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**Targeted Locus:**

[illegible]

[illegible]

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