User enters DNA sequence or RefSeq accession number and parameters.

Is user input FASTA or RefSeq accession number?

FASTA

BLASTN alignment of query FASTA sequence vs genomic databases (genome, HTGS)

Valid genomic match to query identified?

Yes

Retrieve mRNA sequence from RefSeq database

Is there an associated genomic contig ID available in LocusLink?

Yes

Retrieve genomic sequence from genomic databases

No

HTML results page: 'genomic sequence not found'

No

Sim4 mRNA to genomic sequence alignment

Extract putative exon/intron locations

Design exon-flanking primers using primer3

HTML results page
- Per-exon primer sequences, statistics and properties
- Genomic sequence with annotated exons and gene flanking sequence
- Graphical representations of predicted exons by Sim4 alignments