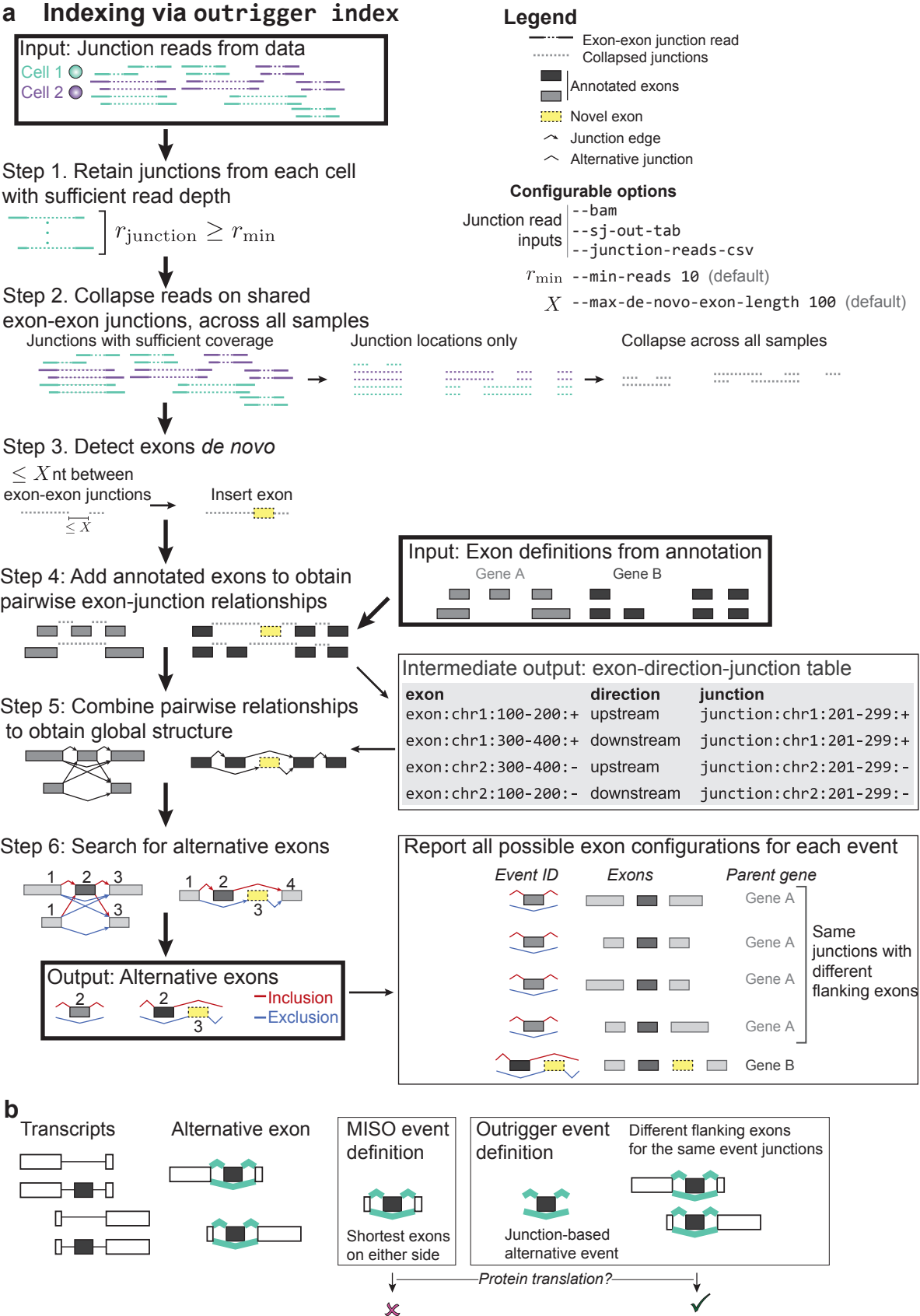
a Indexing via outrigger index
Input: Junction reads from data
Cell 1
Cell 2
Step 1. Retain junctions from each cell with sufficient read depth
$r_{\text{junction}} \geq r_{\min}$
Step 2. Collapse reads on shared exon-exon junctions, across all samples
Junctions with sufficient coverage
Junction locations only
Collapse across all samples
Step 3. Detect exons de novo
$\leq X$ nt between exon-exon junctions
Insert exon
$\leq X$
Step 4: Add annotated exons to obtain pairwise exon-junction relationships
Input: Exon definitions from annotation
Gene A
Gene B
Step 5: Combine pairwise relationships to obtain global structure
Intermediate output: exon-direction-junction table
exon direction junction
exon:chr1:100-200:+ upstream junction:chr1:201-299:+
exon:chr1:300-400:+ downstream junction:chr1:201-299:+
exon:chr2:300-400:- upstream junction:chr2:201-299:-
exon:chr2:100-200:- downstream junction:chr2:201-299:-
Step 6: Search for alternative exons
Output: Alternative exons
Inclusion
Exclusion
Report all possible exon configurations for each event
Event ID
Exons
Parent gene
Gene A
Gene A
Gene A
Gene A
Gene B
Same junctions with different flanking exons
Legend
Exon-exon junction read
Collapsed junctions
Annotated exons
Novel exon
Junction edge
Alternative junction
Configurable options
Junction read inputs
--bam
--sj-out-tab
--junction-reads-csv
$r_{\min}$ --min-reads 10 (default)
X --max-de-novo-exon-length 100 (default)
b
Transcripts
Alternative exon
MISO event definition
Shortest exons on either side
Outrigger event definition
Junction-based alternative event
Different flanking exons for the same event junctions
Protein translation?