

Alternative Splicing Analysis

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Abstract

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Protocol

Filtering of RNA-Seq data using Trimmomatic (version 0.33) with default parameters

Step 1.

☰ SOFTWARE PACKAGE (Linux -)

Trimmomatic, 0.33

Mapping to a given reference genome using HISAT 2 with the following modifications from the default parameters: maximum intron length (4,000); specify strand-specific information (RF); and minimum score (L, -0.1, -0.1)

Step 2.

☰ SOFTWARE PACKAGE (Linux -)

HISAT, 2.0.2

Generating transcripts with Cufflinks (version 2.2.1) .The default parameters were used, except for the following parameters: the minimum isoform fraction (0.05); the small anchor fraction of the spliced reads (0.05); the minimum intron length (20); the maximum intron length (4,000); the library type (fr-firststrand); the corrected frag bias; and the corrected multi-read

Step 3.

☰ SOFTWARE PACKAGE (Linux -)

Cufflinks, 2.2.1

Identifying alternative splicing using ASTALAVISTA (version 4.0) with default parameters

Step 4.

☰ SOFTWARE PACKAGE (Linux -)

ASTALAVISTA, 4.0