Method:

First 12nt of sequences were trimmed from SE-RNAseq reads, and first 14nt were trimmed from PE-RNAseq reads. Then trimmed reads were mapped to 175 sequences of transposon elements (TE, dm6.transposon.fa) with bowtie21 (parameters: -D 20 -R 3 -N 1 -L 20 -i S,1,0.50 --local -k 1 --seed 1), which allows multiple mapping, and randomly assigned multiple mapped reads to one of the best matches2. Then mapping results were quantified with featureCounts3. Expression quantification of TE were combined with the count table of regular genes in RNAseq data analysis for more robust normalization. Differential expression (DE) analysis was performed with DESeq24. Within DE analysis, 'ashr' was used to create log2FoldChange (LFC) shrinkage for each comparison5. Significant DE TE were filtered with the criteria FDR < 0.05 and |LFC| > 1. The heatmap was created with ComplexHeatmap6.

Results:

After expression quantification, 159 out of the 175 TE were detected in RNAseq data. In comparison between DelMut and FRT19A, 91 out of the 159 detected TE were differentially expressed.

TE: Transposon Element

DE: Differentially Expressed

LFC: log2FoldChange

FDR: False Discovery Rate.

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Log:

@ Use local mapping in bowtie2, not end\_to\_end:

@ Combine TE count with regular genes for robust normalization (very important)

@ DESeq2