

Supplementary Information

Supplementary Table 1. LRGASP data table. Contains links, accession numbers, and additional meta-data associated with the LRGASP project long-read and short-read sequencing data.

Spearman Correlation Coefficient

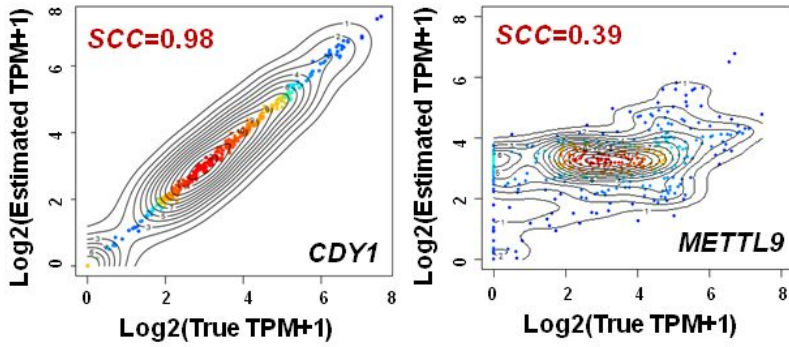
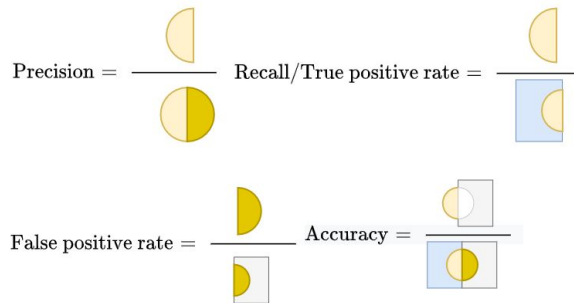
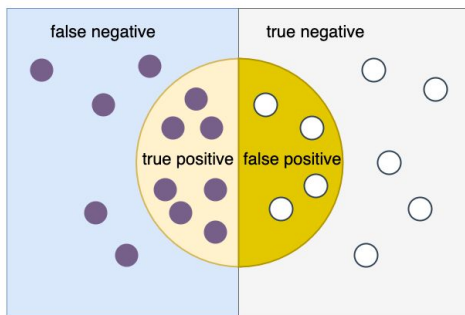


Fig S1. Spearman Correlation Coefficient (SCC) between the estimation and gold standard. The simulation study based on SCC reveals gene *CDY1* can be accurately quantified but not gene *METTL9*.

Fold change based evaluation

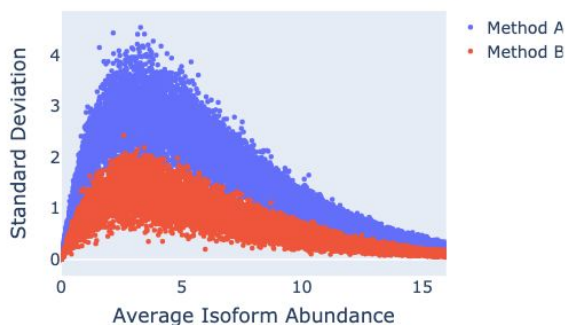


True positive: **actually** differentially expressed and quantified as **differentially expressed**
 False positive: **not** differentially expressed and quantified as **differentially expressed**
 True negative: **not** differentially expressed and quantified as **not** differentially expressed
 False negative: **actually** differentially expressed and quantified as **not** differentially expressed

Fig S2. Log-fold-change-based evaluation metrics. This figure illustrates how ROC statistics such as precision, recall and accuracy are calculated. It measures the performance on detecting real biological changes.

Reproducibility

Standard Deviation vs Average Isoform Abundance

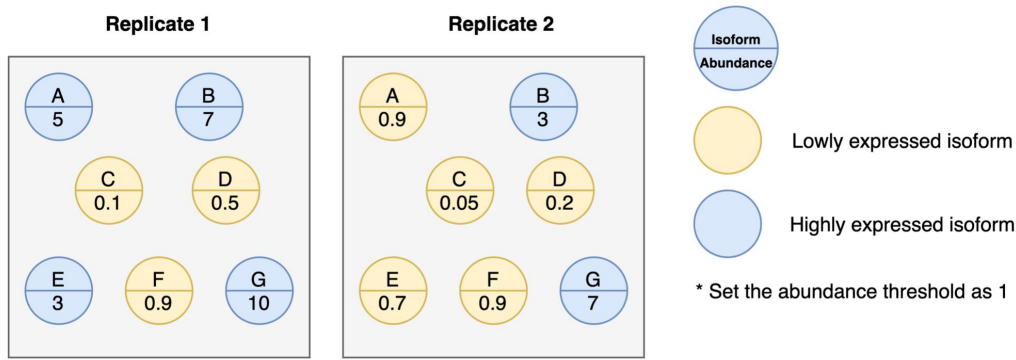


Standard Deviation Curve



Fig S3. Reproducibility among different replicates. By fitting the standard deviation versus average isoform abundance into a smooth curve, it can be shown that Method B has lower standard deviation and higher reproducibility.

Consistency



$$\text{Consistency} = \frac{n(\{B, G\} \cup \{C, D, F\})}{n(\{A, B, C, D, E, F, G\})} = \frac{5}{7}$$

Fig S4. Consistency between replicates. By setting an expression threshold (e.g., 1 in this toy example), we can define which set of genes express (in blue) or not (in yellow). This statistic is to measure the consistency of the expressed gene sets between replicates.

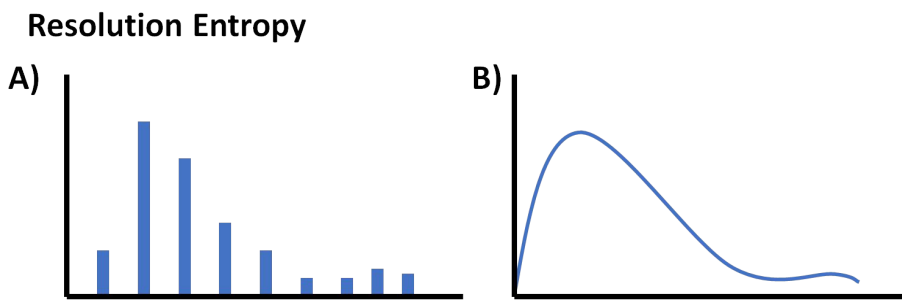


Fig S5. Resolution Entropy. (A) The software output only a few certain discrete values has lower resolution entropy as it cannot capture the continuous and subtle difference of gene expressions. (B) The software with continuous output values has higher resolution entropy.

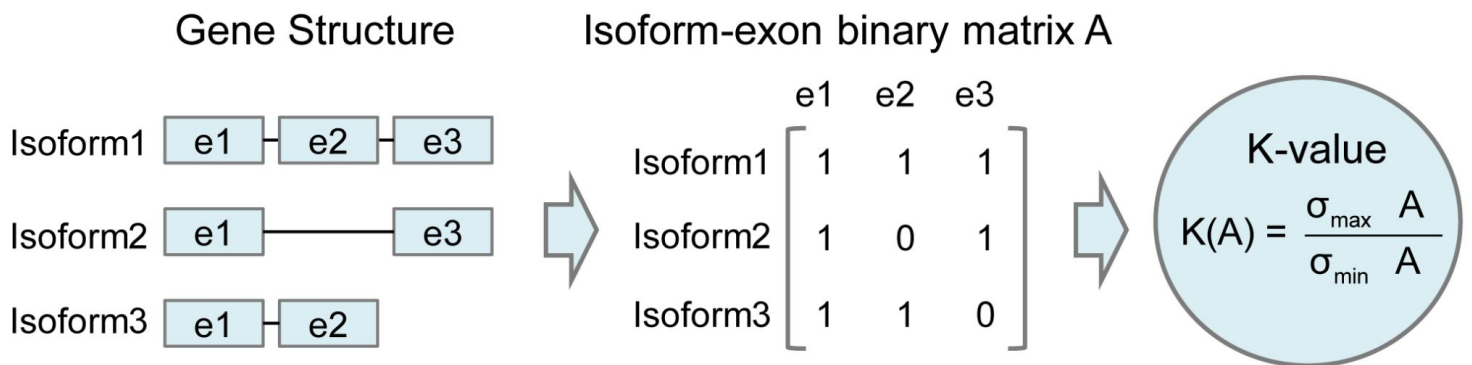


Fig S6. Description of K-value. A measure of the complexity of exon-isoform structures for each gene.

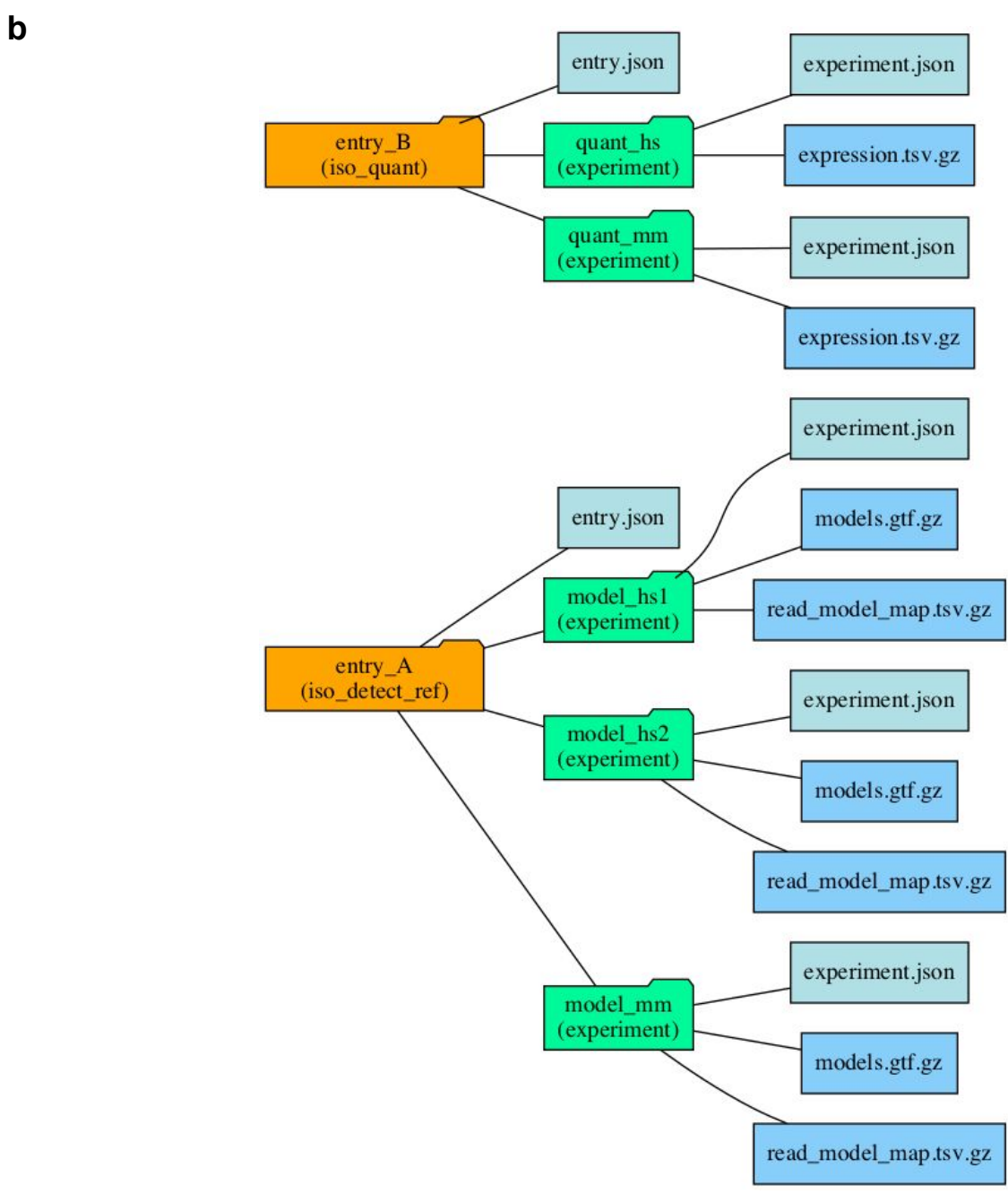
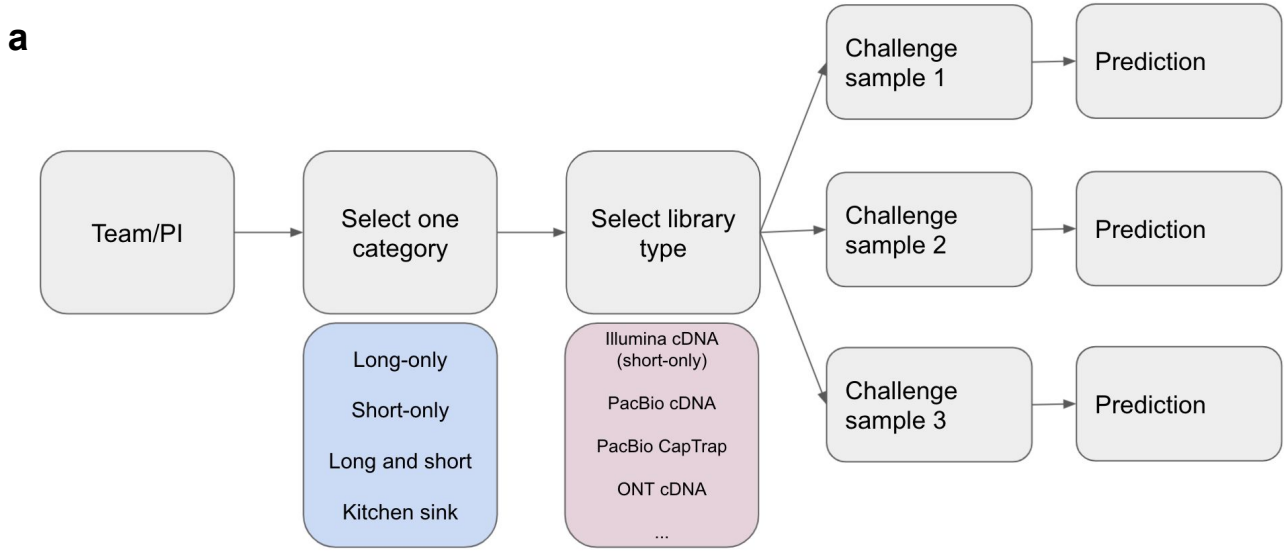


Fig. S7: Challenge submissions. **a**, Overview of challenge submissions. Each entry will be derived from a specific category and library type **b**, Schematic of files that would be included in each entry