A Novel Min-Cost Flow Method for Estimating Transcript Expression with RNA-Seq

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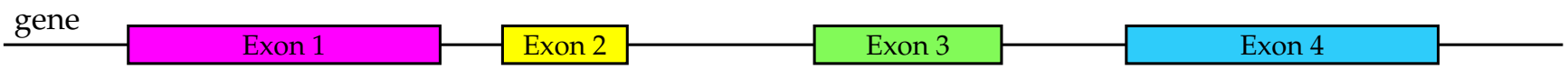
RECOMB-Seq
April 11, 2013

THE BIOLOGICAL PROBLEM



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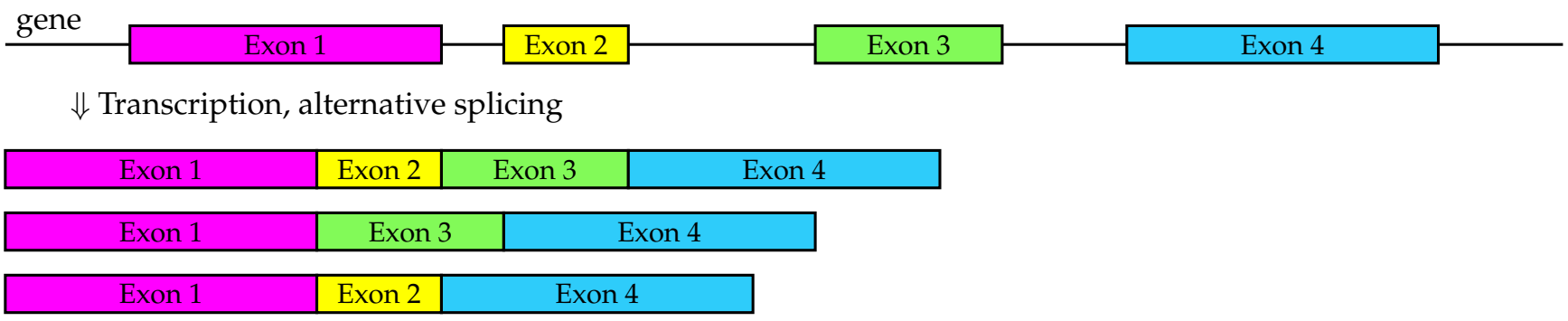

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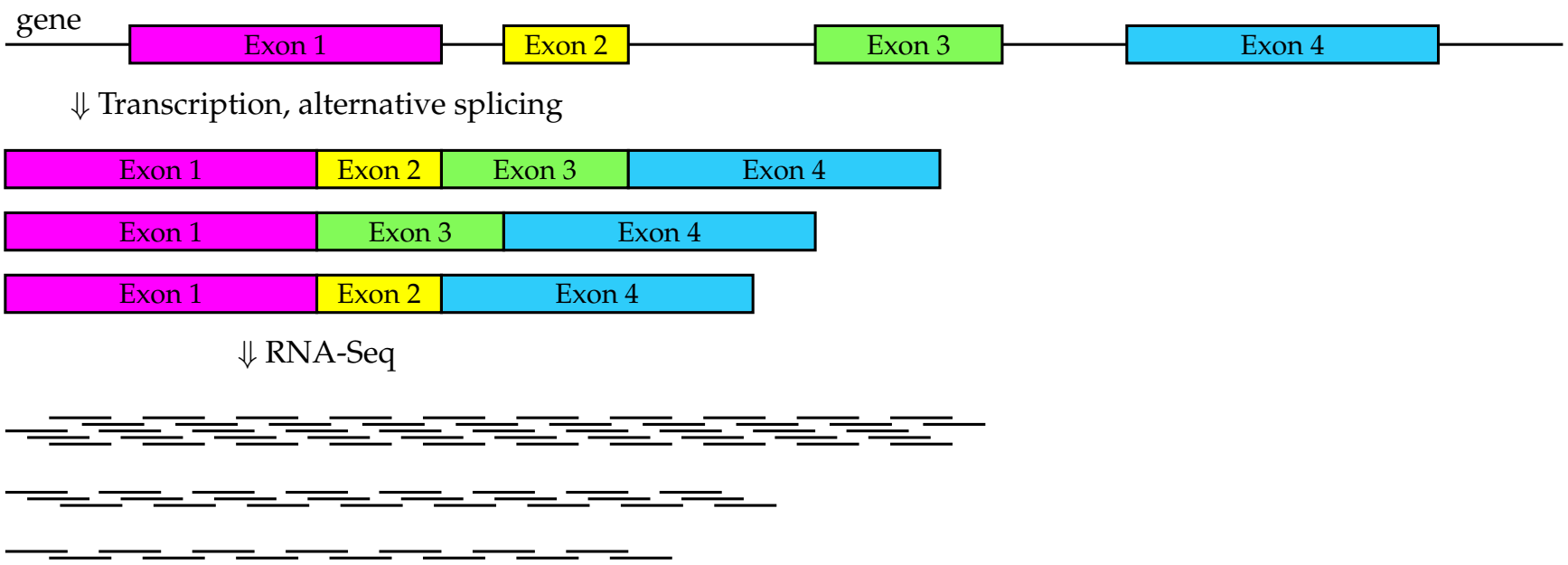

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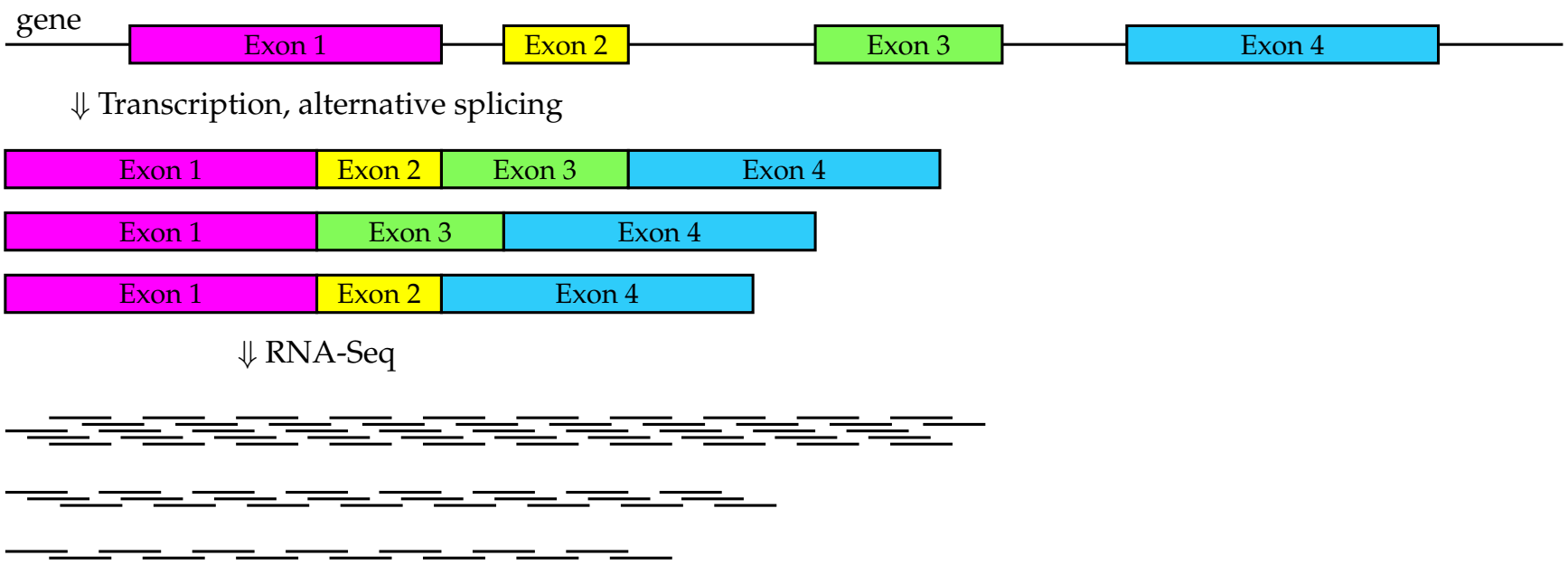

The problem: Assemble the transcripts and estimate their expression levels using only the RNA-Seq read

Existing methods

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Graph models and main existing solutions

1. Overlap graph (Cufflinks)
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   - detect exon boundaries from the spliced alignments
   - every node stands for an exon
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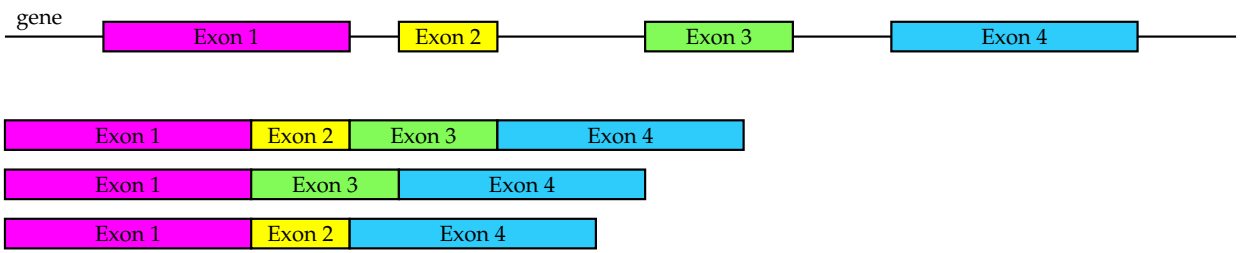

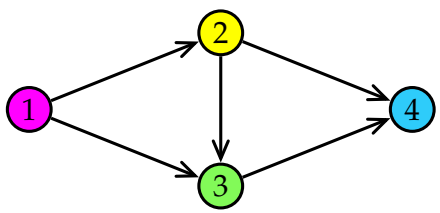

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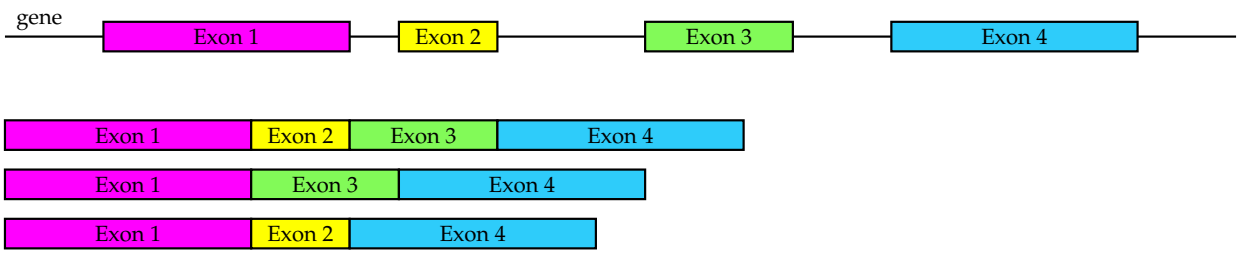

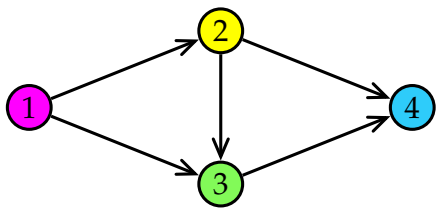

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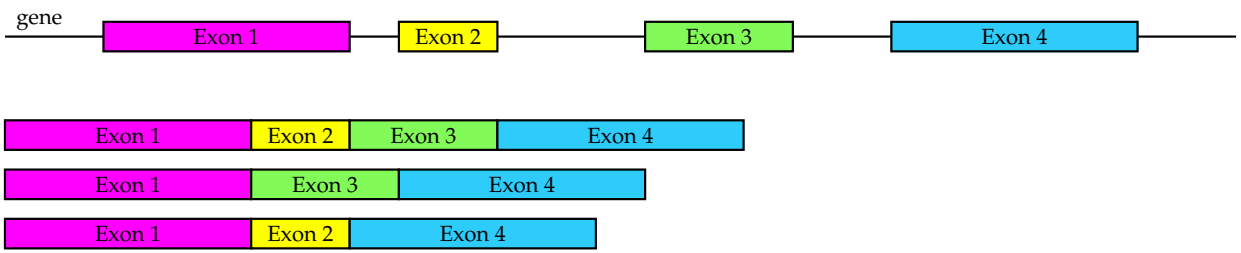

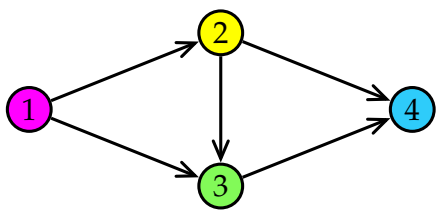

   - the splicing graph is a DAG
   - nodes and edges have observed coverages
   - exhaustively enumerate all possible paths
   - choose the most likely ones based on their coverage using an ILP, QP, QP + LASSO, statistical methods

A unified problem formulation

Problem (Unannotated Transcript Expression Cover)

INPUT: *a splicing DAG* $G = (V, E)$*, and for all* $v \in V$ *and* $(u, v) \in E$*,*

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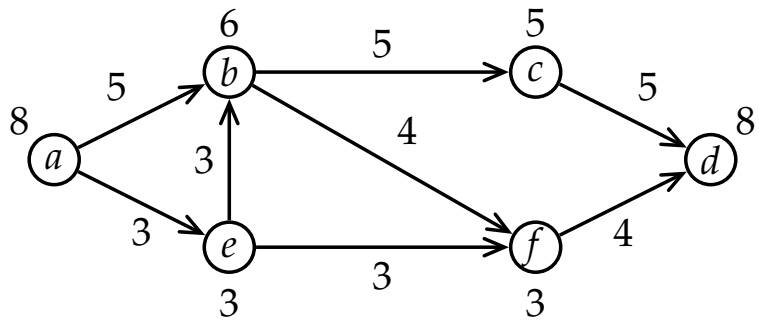
For example, if for all nodes v and edges (u, v),

- $f_v(x) = x, f_{uv}(x) = x \Rightarrow$ least sum of absolute differences model [CLIIQ]
- $f_u(x) = x^2, f_{uv}(x) = x^2 \Rightarrow$ least sum of squares model [IsoLasso, SLIDE]

Example $f_v(x) = x^2, f_{uv}(x) = x^2$

$$\sum_{v \in V} \left(cov(v) - \sum_{P \in \mathcal{P} : v \in P} e(P) \right)^2 + \sum_{(u,v) \in E} \left(cov(u, v) - \sum_{P \in \mathcal{P} : (u,v) \in P} e(P) \right)^2$$



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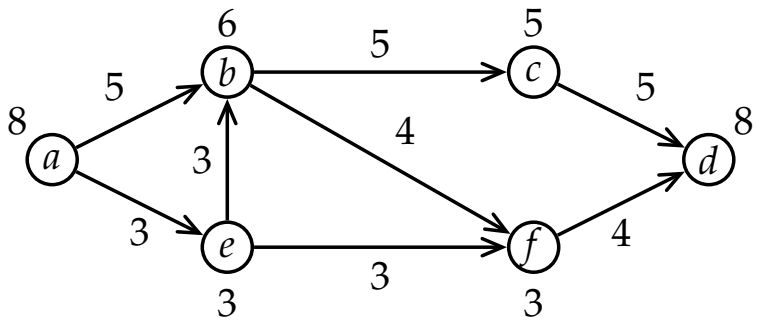

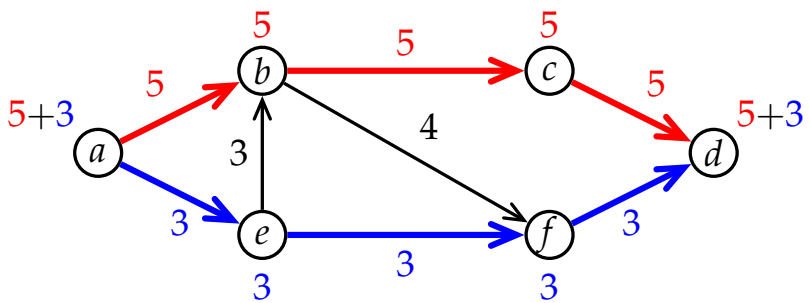

- [Left] A non-optimal tuple of paths with cost $1 + 1 + 3^3 + 4^2 = 27$, from b, (f, d), (e, b), (b, f)

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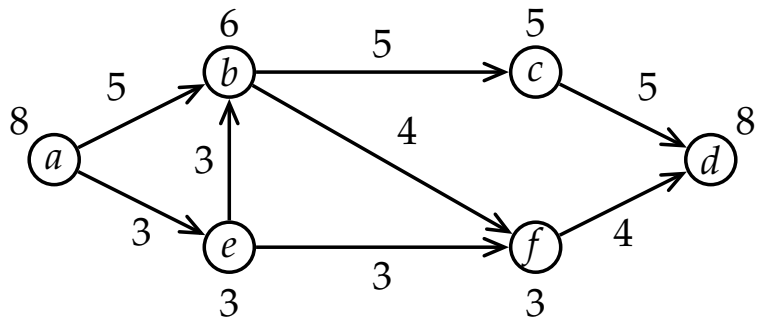

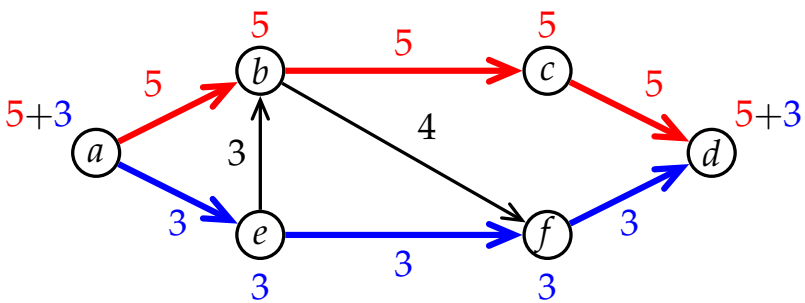

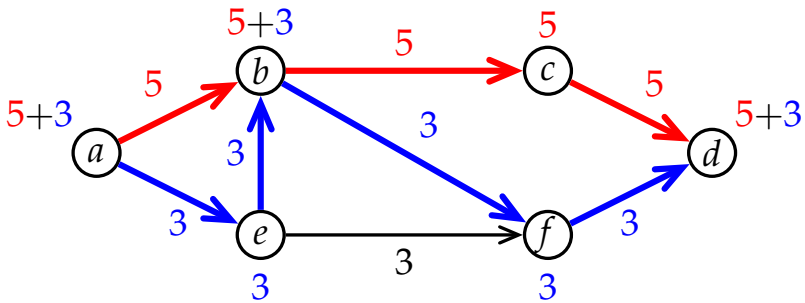

- [Left] A non-optimal tuple of paths with cost $1 + 1 + 3^3 + 4^2 = 27$, from b, (f, d), (e, b), (b, f)
- [Right] The optimal tuple of paths with cost $2^2 + 1 + 1 + 3^2 = 15$, from b, and (b, f), (f, d), (e, f)

SOLUTION: REDUCTION TO NETWORK FLOWS

- A *flow* over a *flow network* $N = (V, E, b)$ is a function x assigning to every arc $(u, v) \in E$ a number $x_{uv} \in \mathbb{N}$ such that
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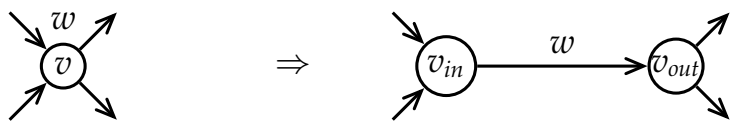
- We first reduce the problem to one having coverages only on edges



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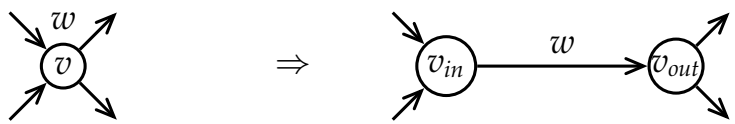

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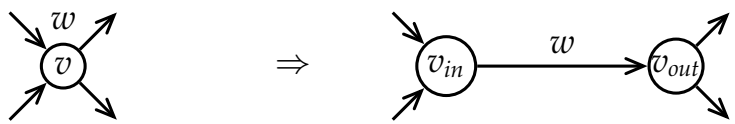

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- We will find an "optimal" flow: the value of the flow on each edge will be the predicted coverage on that edge

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  - a flow value to be pushed from sources to sinks, and
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Theorem

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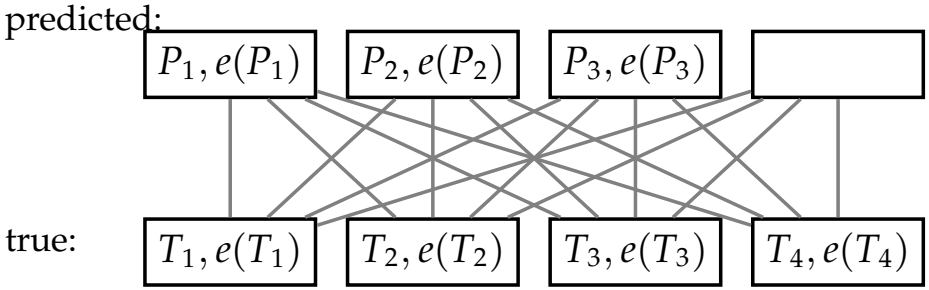
- Splitting a flow into the minimum number of paths is NP-hard
- We currently apply a heuristic iteratively selecting the path of maximum bandwidth

Validation

- construct a bipartite graph with predicted and true transcripts



- the edge weight between $P_i, e(P_i)$ and $T_j, e(T_j)$ is a combined measure between (cf. Normalized Compression Distance)
  - *bitscore* := $\frac{\text{\#bits needed to encode } T_j \text{ given } P_i}{\text{\#bits needed to encode } T_j \text{ just by inserts}}$
  - *relative expression level difference* := $\frac{|e(T_j) - e(P_i)|}{e(T_j)}$

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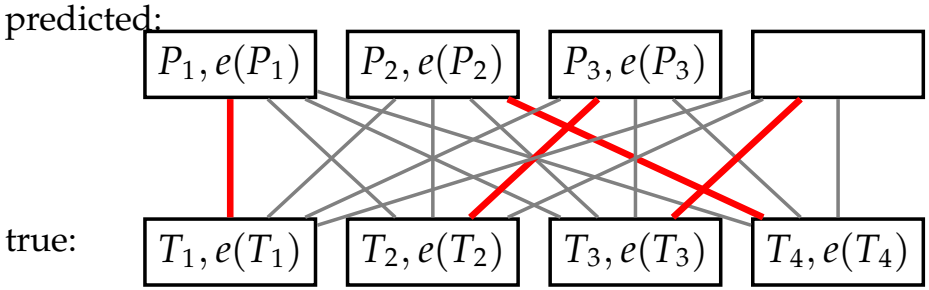

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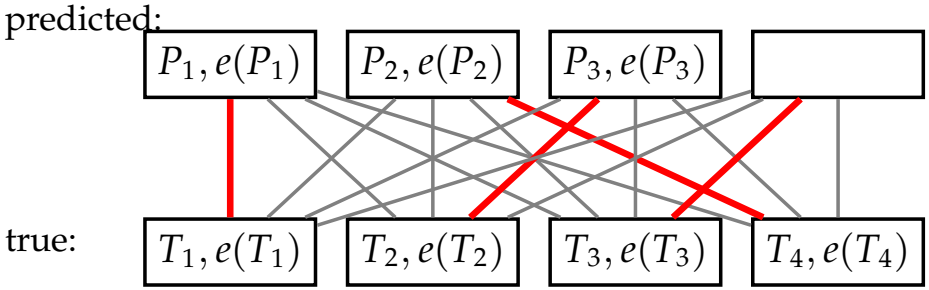

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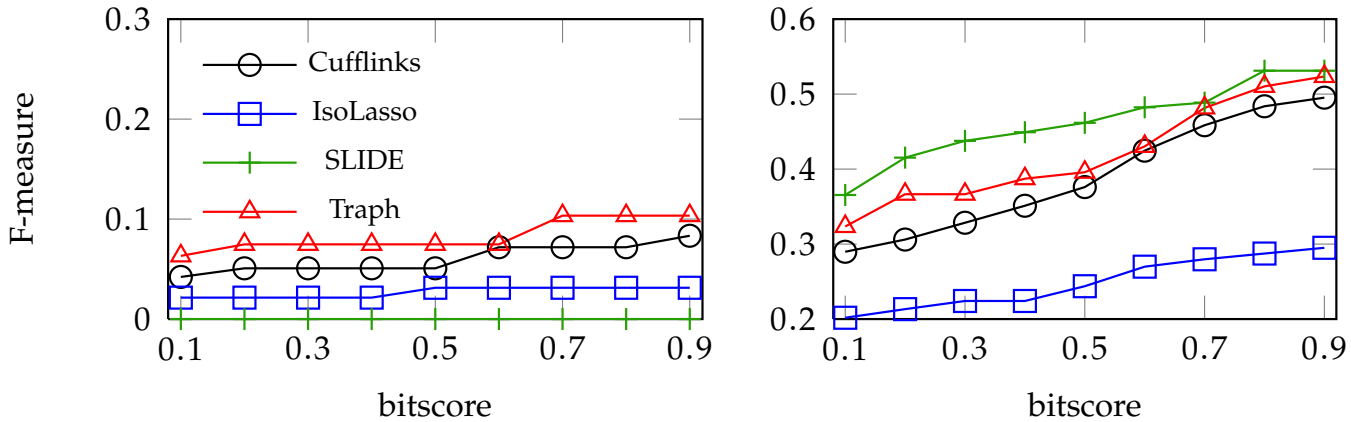
- a True Positive is a match with bitscore and expression difference under given thresholds
- other events define False Positives and False Negatives
- compute precision, recall, F-measure

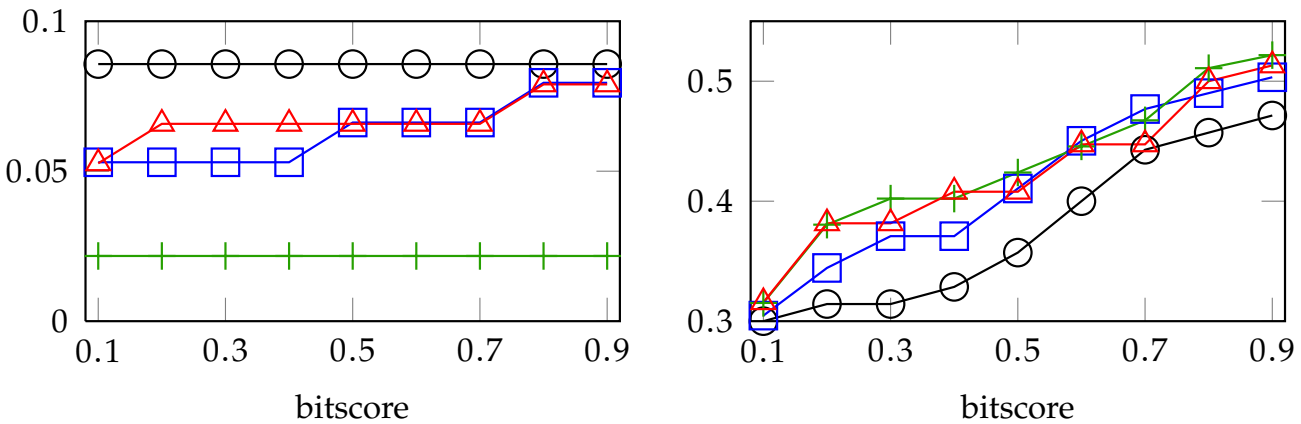
Experimental results on simulated data

- Simulated paired-end reads from the annotated transcripts of 29 genes
- Reads aligned with TopHat

1. Alignments fed to the tools for each gene independently



2. Alignments for all genes combined into one file, fed to the tools



(c) expr. difference threshold 0.1 (d) expr. difference threshold 0.9

Experimental results on real data

- 2 406 339 paired-end reads of length 75bp mapping to human chromosome 2
- 342 genes where all tools made predictions
- 3306 annotated transcripts in total

Tool	Total predicted	Shared with annotation, bitscore under				
		0.1	0.2	0.3	0.4	0.5
Cufflinks	933	128	200	267	342	410
IsoLasso	742	132	199	262	310	361
SLIDE	1131	191	281	381	463	552
Traph	961	157	255	330	398	474

Conclusions cs.helsinki.fi/gsa/traph/

- A novel unified problem formulation for transcript identification and quantification with RNA-Seq
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- A general framework applicable to other multi-assembly problems

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Future work:

- integrate paired-end information
- integrate annotation information
- procure real ground-truth
- if we limit by k the number of paths in an optimal solution
  - the problem becomes NP-hard
  - we can write dynamic programming algorithms which work well assuming k is not too large
  - we get better accuracy
  - $\Rightarrow$ see our poster