Inference in generalized bilinear models

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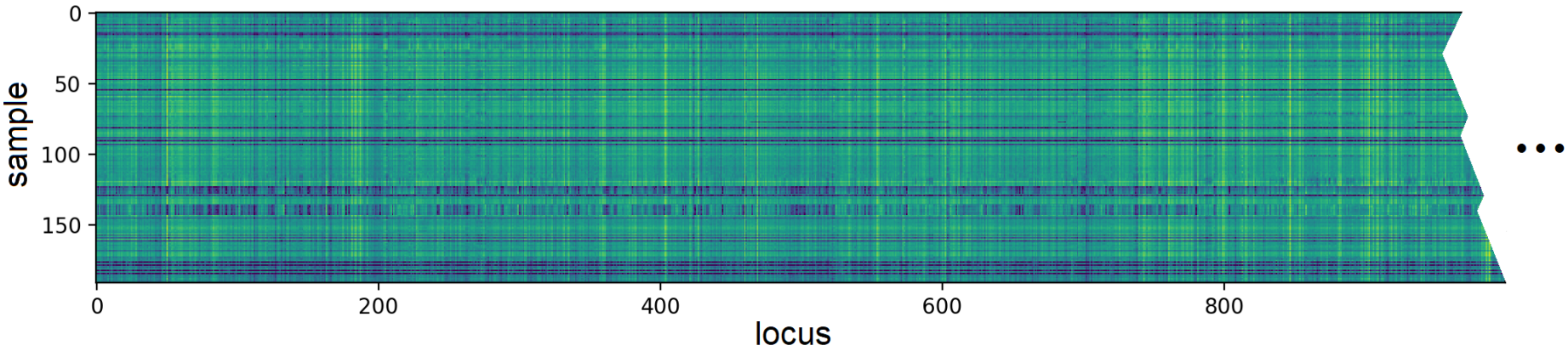
Preprint: https://arxiv.org/abs/2010.04896
Slides: http://jwmi.github.io/talks/ness2022.pdf

Background

Modern high-throughput sequencing yields large matrices of counts.

- Copy ratio estimation in cancer genomics
  - whole-exome or whole-genome sequencing data
- Copy number variation in genetics
  - whole-exome or whole-genome sequencing data
- Gene expression analysis in biology/medicine
  - RNA-seq data for transcript abundance

log counts for a whole-exome seq data set of 191 samples $\times$ 171523 loci



Background

- Latent factor models are widely used to discover and adjust for hidden variation in these applications and many others.
- Estimation and inference in latent factor models is challenging.
- Consequently, most methods do not fully account for uncertainty in the latent factors, which can lead to miscalibrated inferences such as overconfident p-values.

This talk

- Generalized bilinear models (GBMs) are a flexible extension of generalized linear models (GLMs) to include latent factors as well as row covariates, column covariates, and interactions.
- We propose fast and accurate methods for GBM estimation and inference (i.e., uncertainty quantification).
- We introduce *delta propagation*, a novel technique for propagating uncertainty among model components using the delta method.
- We present simulation studies assessing performance.
- We apply GBMs to copy ratio estimation and RNA-seq analysis.

Outline

1. Generalized bilinear models (GBMs)
2. Previous work
3. Estimation
4. Inference (uncertainty quantification)
5. Application
   - RNA-seq gene expression analysis

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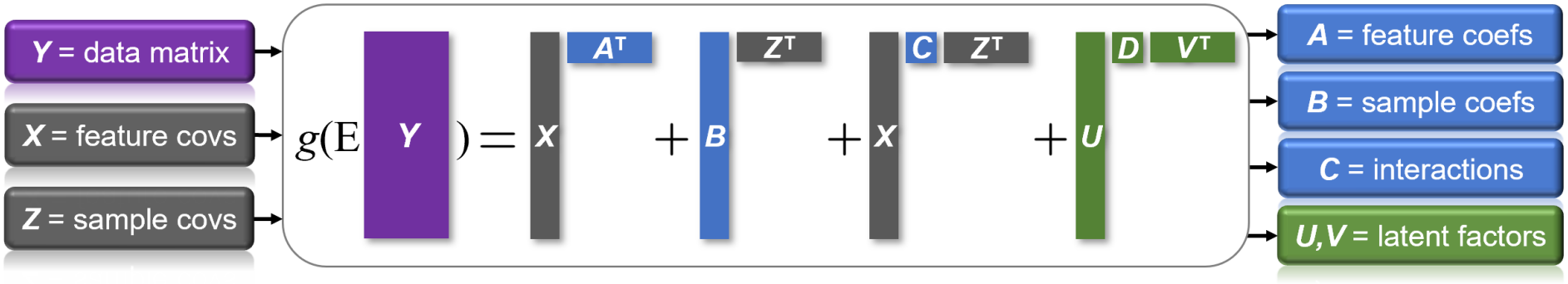
Generalized bilinear models (GBMs)



- Suppose the data matrix $\boldsymbol{Y} = (Y_{ij}) \in \mathbb{R}^{I \times J}$ satisfies

$$g(\mathrm{E}(\boldsymbol{Y})) = XA^{\mathrm{T}} + BZ^{\mathrm{T}} + XCZ^{\mathrm{T}} + UDV^{\mathrm{T}}$$

  where the link function g is applied element-wise.

- We refer to this as a *generalized bilinear model* (Choulakian, 1996).

- The "bilinear" part UDV^{T} is a low-rank matrix that captures latent effects due, for example, to unobserved covariates such as batch.

Outcome distributions

- We consider discrete exponential dispersion families (EDFs).
- Specifically, we suppose $Y_{ij} \sim f(y \mid \theta_{ij}, r_{ij})$ where

$$f(y \mid \theta, r) = \exp(\theta y - r\kappa(\theta))h(y, r).$$

- For any discrete EDF,

$$\mu = \mathrm{E}(Y) = r\kappa'(\theta)$$
$$\sigma^2 = \mathrm{Var}(Y) = r\kappa''(\theta).$$

- For sequencing data, we focus on negative binomial outcomes, which is a special case of discrete EDF.
- We parametrize the dispersions as $1/r_{ij} = \exp(s_i + t_j + \omega)$.

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Previous work: Normal bilinear models with covariates

- Consider the following special case:

$$\boldsymbol{Y} = XA^{\mathsf{T}} + BZ^{\mathsf{T}} + XCZ^{\mathsf{T}} + UDV^{\mathsf{T}} + \boldsymbol{\varepsilon}$$

  where $\boldsymbol{\varepsilon}$ is a matrix of residuals with $\varepsilon_{ij} \sim \mathcal{N}(0, \sigma^2_{ij})$.

- Work on this model was inspired by Tukey (1962), who suggested combining regression with factor analysis.

- Estimation for this model, assuming $\sigma^2_{ij} = \sigma^2$:

  Gabriel (1978), Takane and Shibayama (1991).

- Hypothesis testing and confidence regions, assuming $\sigma^2_{ij} = \sigma^2$:

  Perry and Pillai (2013) show how to perform inference for univariate linear projections of A and B.

Previous work: GBMs with covariates

- Consider the general case:

$$g(\mathrm{E}(\boldsymbol{Y})) = XA^{\mathsf{T}} + BZ^{\mathsf{T}} + XCZ^{\mathsf{T}} + UDV^{\mathsf{T}}.$$

- Previous authors have considered models of this form:

  Choulakian (1996), Gabriel (1998), de Falguerolles (2000), Townes (2019).

- Townes (2019) develops a fast estimation algorithm using diagonal approximations to Fisher scoring updates for ℓ_2-penalized estimation.

- Limitations of previous work:
  - uncertainty quantification is not addressed,
  - a single common dispersion parameter is assumed, and
  - identifiability constraints are not explicitly enforced during estimation.

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Estimation algorithm

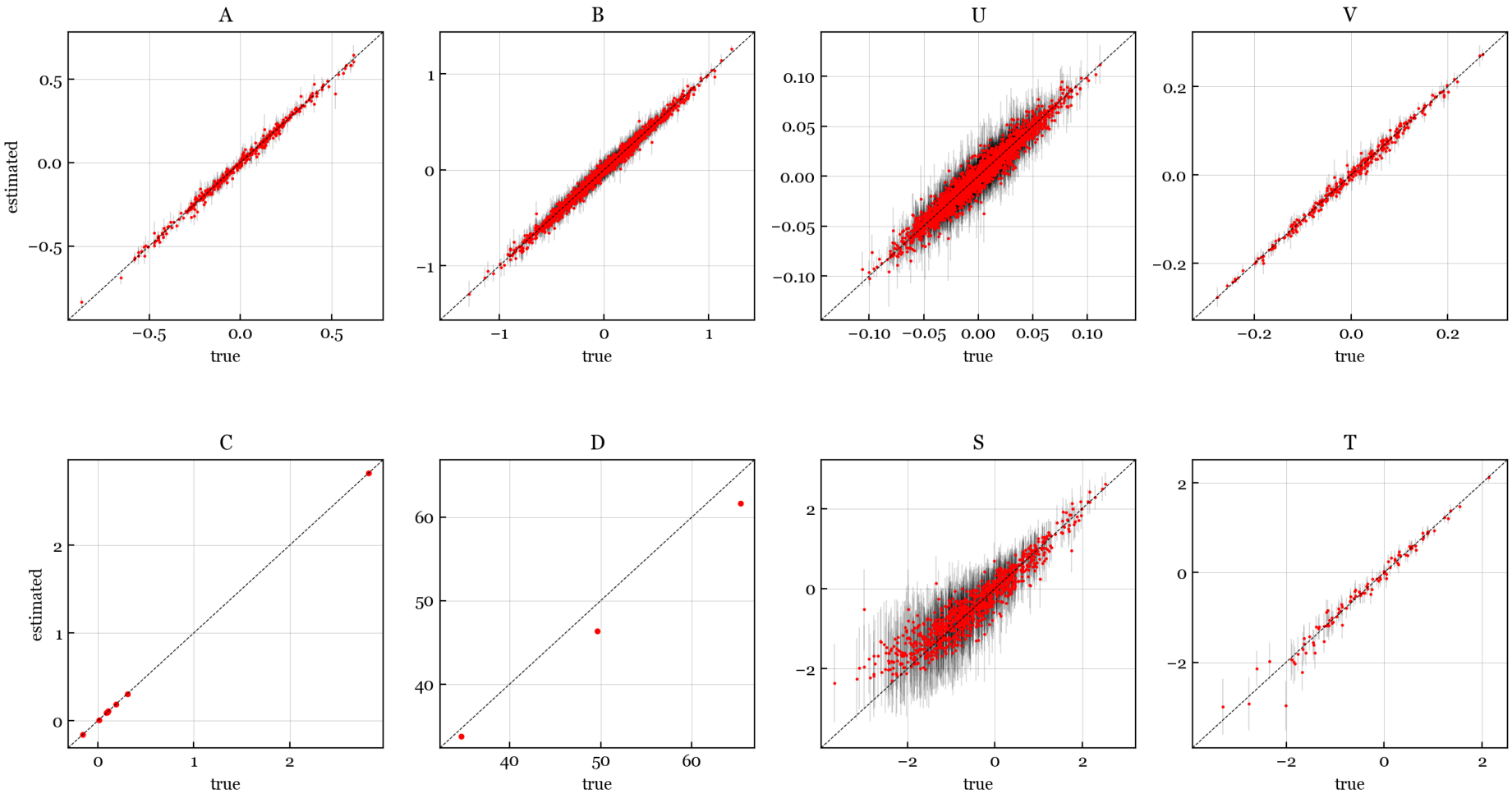
- We provide an algorithm for *maximum a posteriori* GBM estimation that extends previous work by:
  - estimating row- and column-specific dispersion parameters,
  - improving numerical stability, and
  - explicitly enforcing identifiability constraints during estimation.

- Basic idea: Iteratively cycle through the components of the model, updating each in turn using an optimization-projection step.

- "Optimization-projection" = unconstrained optimization step and a likelihood-preserving projection onto the constrained parameter space.

Estimation: Typical example

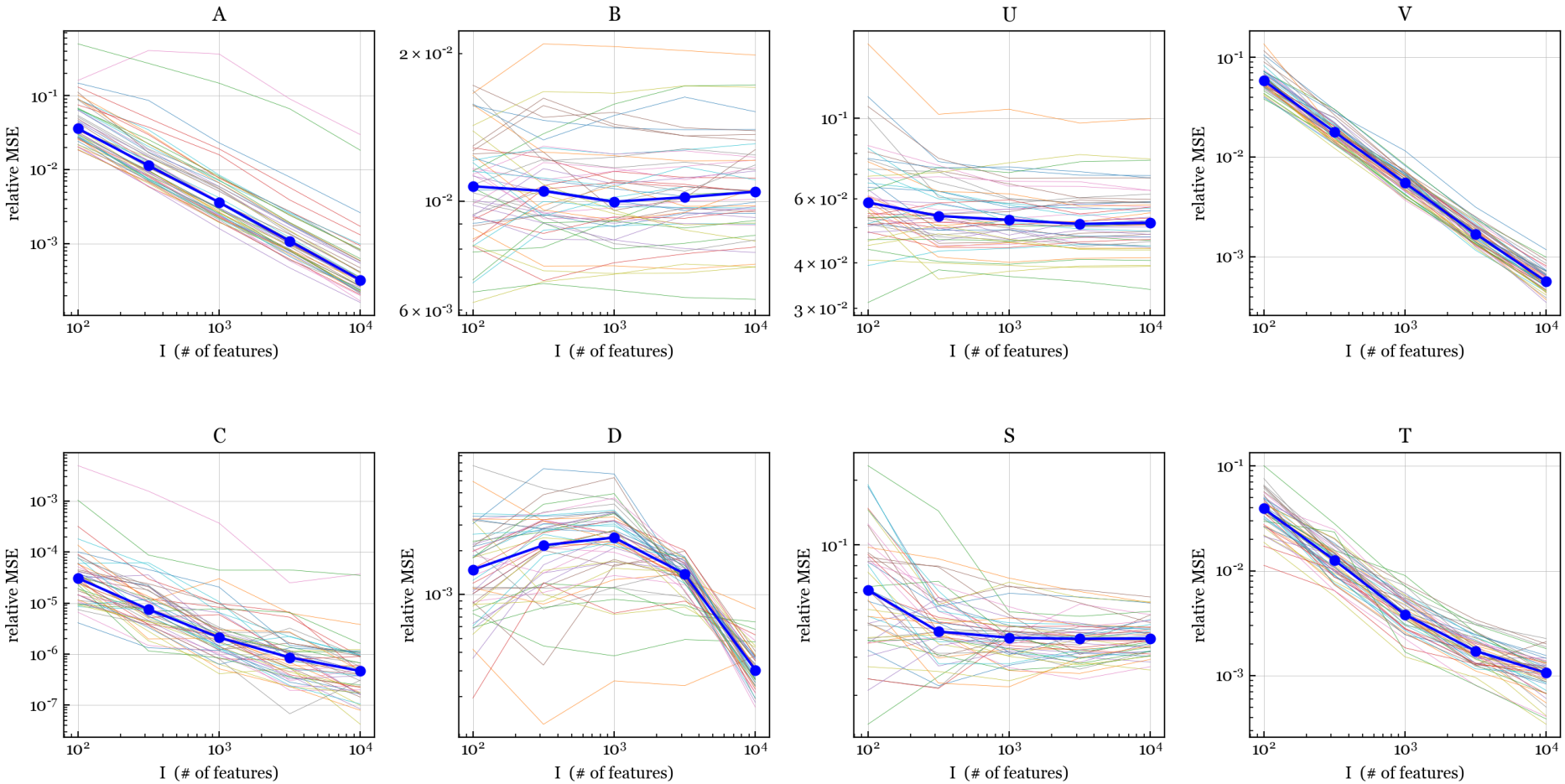
Scatterplots of estimated versus true parameters for a typical simulated data matrix (`NB/Normal/Normal`, 1000 rows, 100 cols, 4 feature covs, 2 samples covs, and 3 factors)



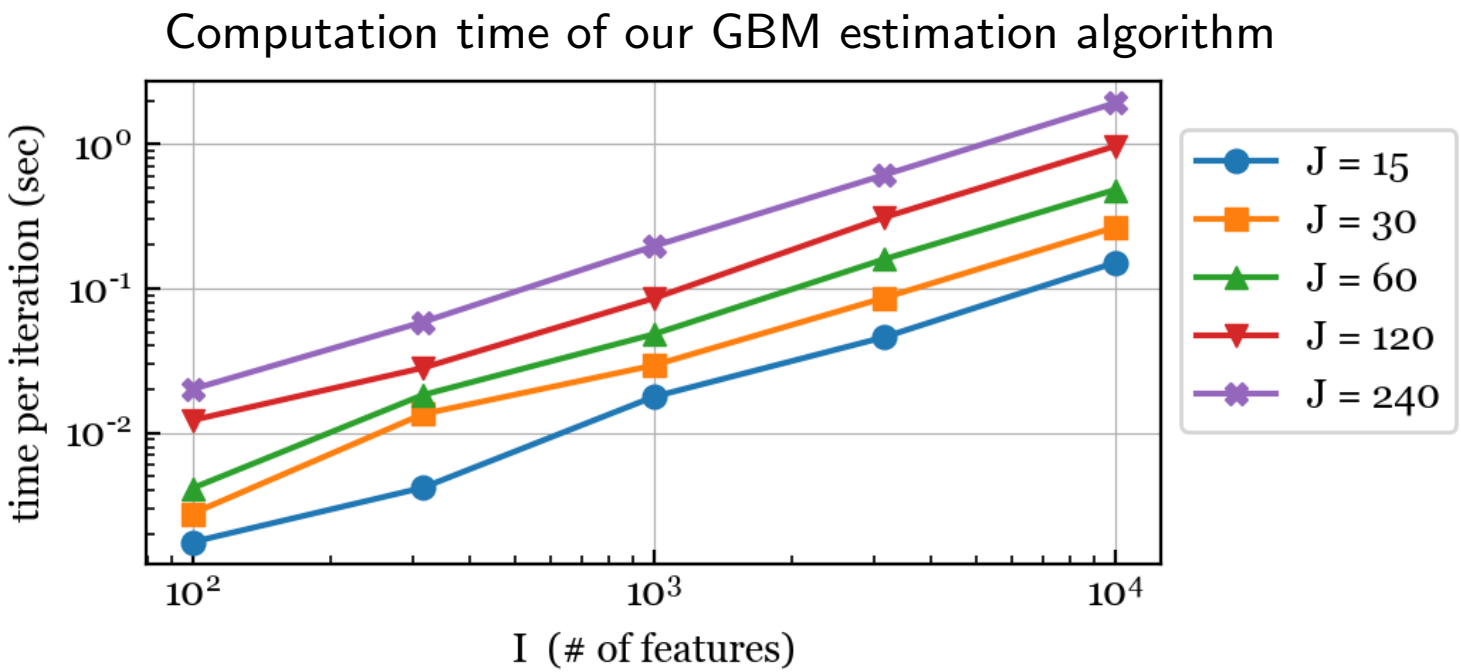
Estimation: Error tends to zero with increasing data

Relative mean-squared error between estimated and true parameter values

(50 runs of `NB/Normal/Normal`, 100 cols, 4 feature covs, 2 samples covs, and 3 factors)



Estimation: Computation time is linear in size of matrix



- Computation time grows linearly with I (# rows) and J (# cols).
- Each dot is the average over 10 runs of the `NB/Normal/Normal` scheme with 4 feature covs, 2 samples covs, and 3 factors.
- The empirical results agree with the theory.

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Inference (uncertainty quantification)

- Most latent factor methods do not fully account for uncertainty.
- To remove batch effects in gene expression, several methods estimate UDV^{T} and then treat V as known, handling uncertainty only in U.

  Leek and Storey (2007, 2008), Sun et al. (2012), Risso et al. (2014)
- CNV detection methods often fit UDV^{T} and just subtract it off.

  Fromer et al. (2012), Krumm et al. (2012), Jiang et al. (2015)
- Bayesian inference provides full uncertainty quantification, but MCMC is slow in large parameter spaces with strong dependencies.
- Variational Bayes is faster, but relies on factorized approximations that tend to underestimate uncertainty.

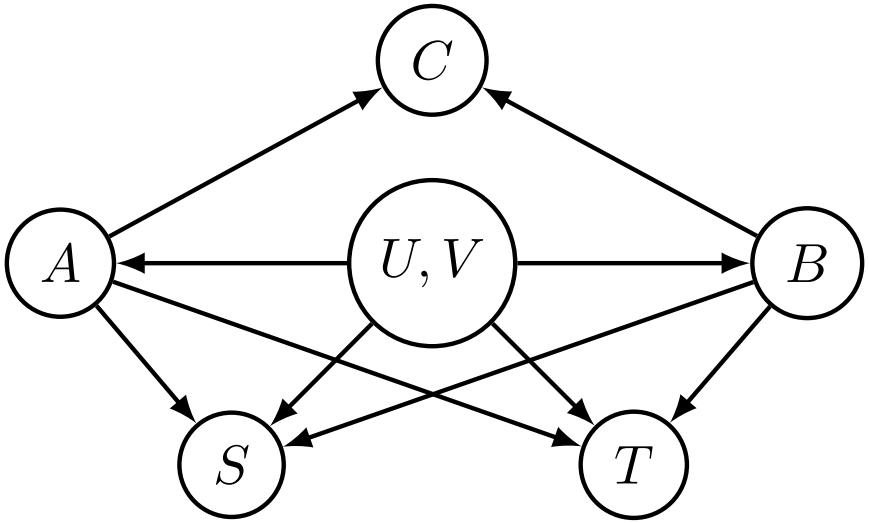
  Stegle et al. (2010), Buettner et al. (2017), Babadi et al. (2018)

Inference: Novel method – "delta propagation"

- We provide a fast, accurate method for GBM uncertainty quantification.
- In particular, we introduce *delta propagation*, a general technique for propagating uncertainty among model components using the delta method.
- Delta propagation can be done analytically using closed-form expressions involving the gradient and the Fisher information.

Inference: Outline of GBM inference algorithm

Diagram of uncertainty propagation scheme for GBM inference

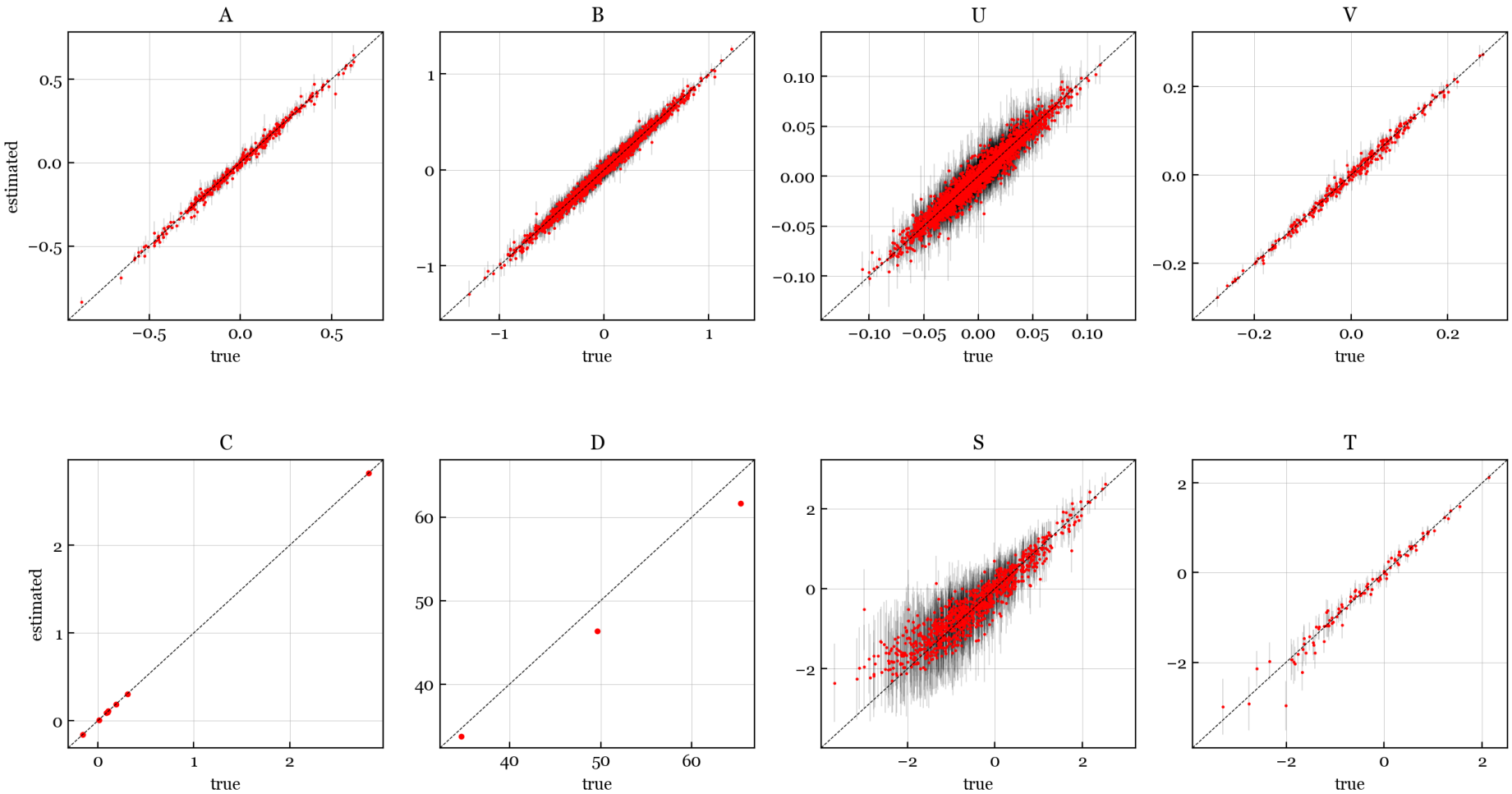


Outline:

1. Compute conditional uncertainty for each parameter matrix/vector.
2. Compute joint uncertainty in (U, V) accounting for constraints.
3. Propagate uncertainty between components using delta propagation.
4. Compute approximate standard errors.

Inference: Typical example

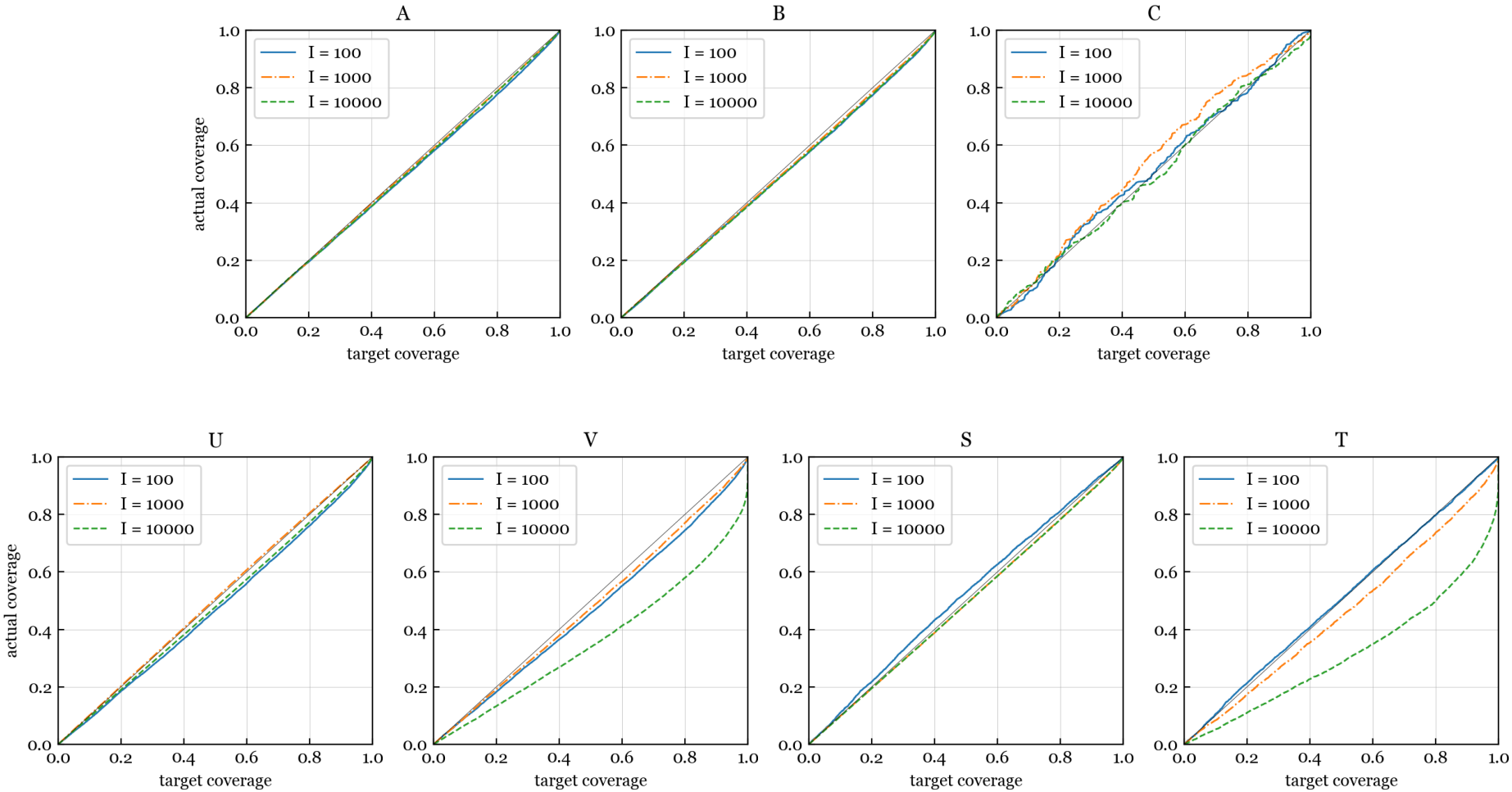
Scatterplots of estimated versus true parameters for a typical simulated data matrix (`NB/Normal/Normal`, 1000 rows, 100 cols, 4 feature covs, 2 samples covs, and 3 factors)



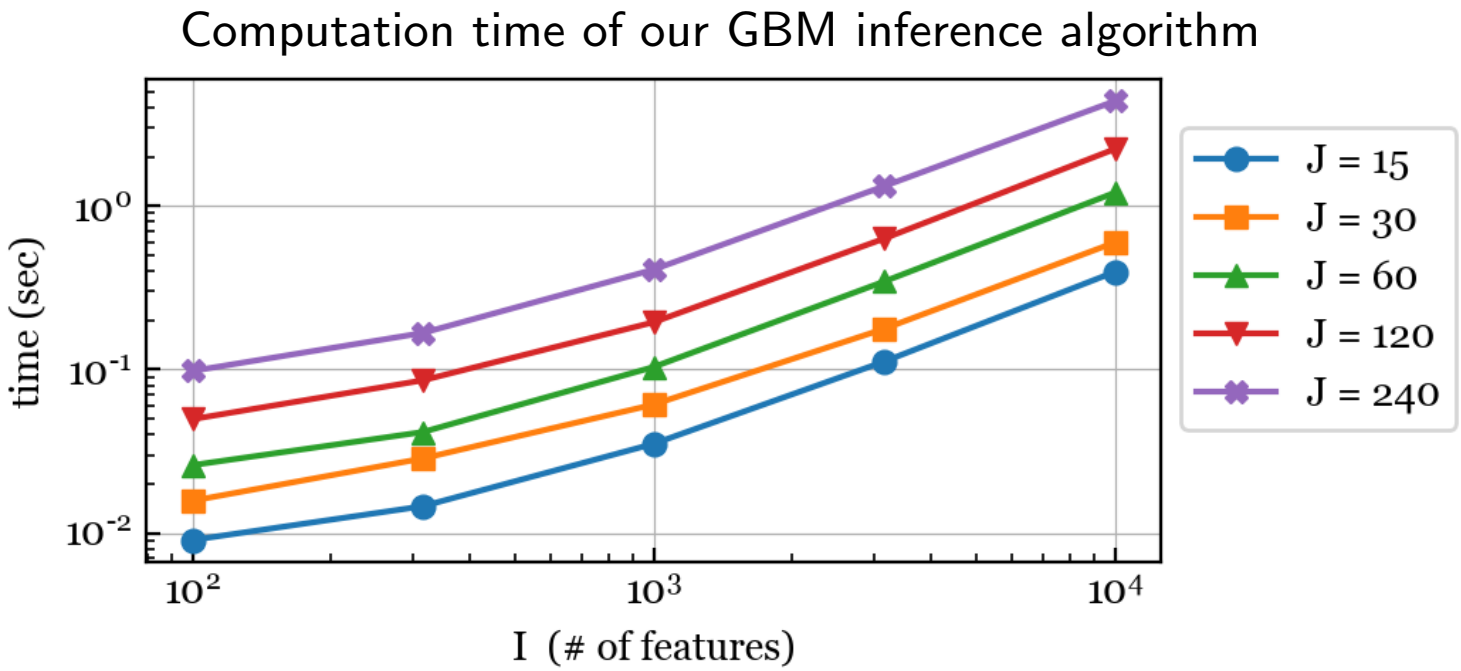
Inference: Coverage is good for most params of interest

Coverage of confidence intervals for the entries of each parameter matrix/vector

(50 runs of `NB/Normal/Normal`, 100 cols, 4 feature covs, 2 samples covs, and 3 factors)



Inference: Empirical assessment of computation time



- Theory indicates that computation time is linear in I (# rows) and quadratic in J (# cols).
- Thus, as I increases, the curves should become linear in I.
- Each dot is the average over 10 runs of the `NB/Normal/Normal` scheme with 4 feature covs, 2 samples covs, and 3 factors.

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RNA-seq: Analyzing GTEx data for aging-related genes

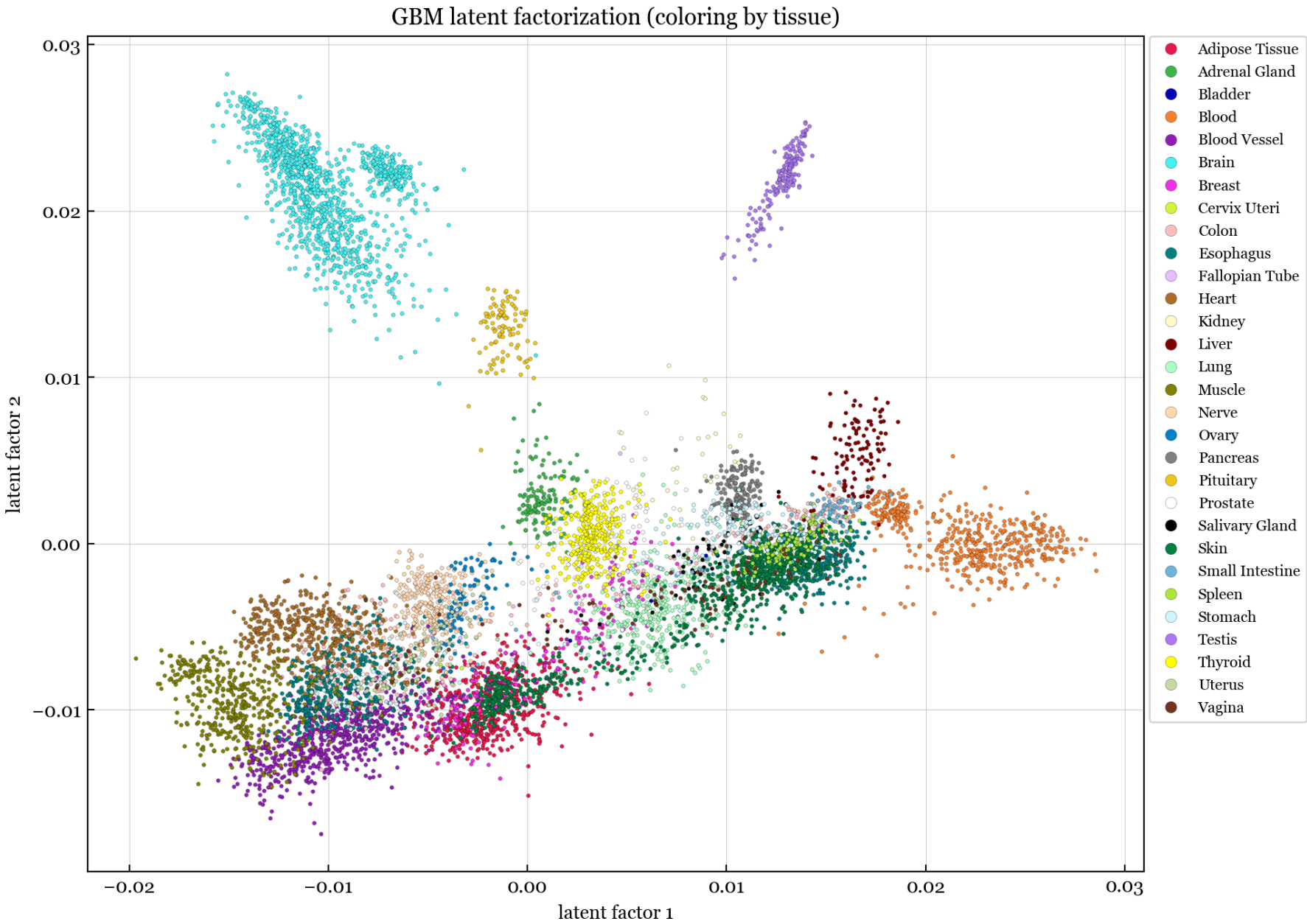
- We consider RNA-seq data from the Genotype-Tissue Expression (GTEx) project (Melé et al., 2015).
- 8,551 samples from 30 tissues in the human body, from 544 subjects.
- We apply the GBM to find genes whose expression changes with age, adjusting for technical biases.

RNA-seq: PCA of GTEx data using log-transformed TPMs

RNA-seq: Visualizing GTEx data using a GBM

- Similar to PCA, we can use the GBM to visualize high-dimensional data by plotting the V matrix.
- First, we take a random subset of 5,000 genes and fit a negative binomial GBM with:
  - two latent factors,
  - no sample covariates, and
  - $\log(\texttt{length}_i)$, gc_i, and $(\text{gc}_i - \overline{\text{gc}})^2$ as gene covariates.
- Model dimensions: $I = 5{,}000$, $J = 8{,}551$, $K = 4$, $L = 1$, and $M = 2$.

RNA-seq: Visualizing GTEx data using a GBM



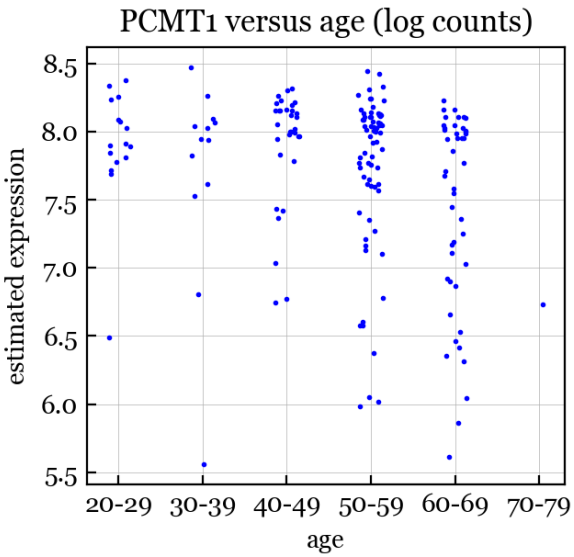
RNA-seq: Analyzing GTEx data for aging-related genes

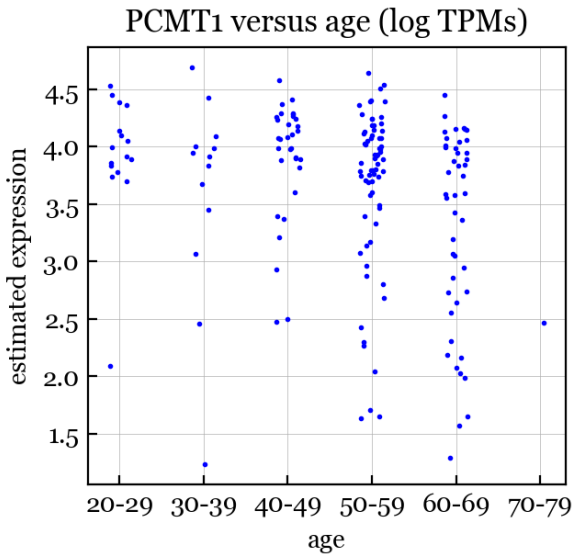
- To find aging-related genes, we add subject age as a sample covariate.
- For illustration, we present results for the "Heart - Left Ventricle" subtissue (Heart-LV).
- We ran the GBM on the 176 Heart-LV samples in the test set, using:
  - the 19,853 genes with nonzero median across these samples,
  - gene covariates: $\log(\texttt{length}_i)$, $\texttt{gc}_i$, and $(\texttt{gc}_i - \overline{\texttt{gc}})^2$,
  - sample covariates: `smexncrt` (exonic rate) and age (subject age),
  - 3 latent factors.
- This choice of subtissue and model was based on the exploratory phase.

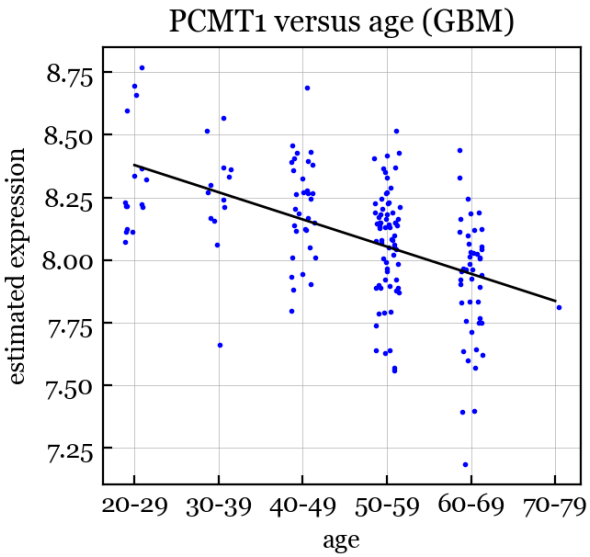
RNA-seq: Analyzing GTEx data for aging-related genes

- In this GBM, each gene has a coefficient describing how its expression changes with age.
- Using our GBM inference algorithm, we compute a p-value for each gene to test whether this coefficient is nonzero.
- 2,444 genes were significantly associated with age in Heart-LV, controlling Type I error at 0.05 using Bonferroni.
- For comparison, simple linear regression on the log-transformed TPMs yields only 1 significant gene.
- This indicates that the GBM has much greater power than a simple standard approach.

RNA-seq: Expression of the top aging-related gene







- The top GBM hit for Heart-LV is PCMT1 (p-value $= 1.1 \times 10^{-47}$).
- PCMT1 is involved in the repair and degradation of damaged proteins, and is a well-known aging gene (Tacutu et al., 2018).
- GBM-estimated expression of PCMT1 exhibits a clear downward linear trend with age.
- The log TPMs for PCMT1 are noisier and the trend is much less clear.

RNA-seq: Top age-related GO terms (Biological Process)

- To test for enrichment of Gene Ontology (GO) term gene sets, we run DAVID on the top 1000 GBM hits for Heart-LV.
- These results are highly consistent with known aging biology (López-Otín et al., 2013).

GO term ID	Description	Count	p-value	Benjamini
GO:0098609	cell-cell adhesion	48	5.1e-12	1.5e-08
GO:0006418	tRNA aminoacylation for protein translation	16	1.4e-09	2.0e-06
GO:0006099	tricarboxylic acid cycle	12	3.7e-07	3.6e-04
GO:1904871	positive regulation of protein localization to Cajal body	7	1.1e-06	6.1e-04
GO:1904851	positive regulation of establishment of protein localization to telomere	7	1.1e-06	6.1e-04
GO:0006607	NLS-bearing protein import into nucleus	10	1.3e-06	6.2e-04
GO:0006914	autophagy	22	1.8e-05	7.6e-03
GO:0016192	vesicle-mediated transport	24	2.6e-05	8.3e-03
GO:0006511	ubiquitin-dependent protein catabolic process	24	2.6e-05	8.3e-03
GO:0006888	ER to Golgi vesicle-mediated transport	24	3.5e-05	1.0e-02
GO:0006886	intracellular protein transport	31	4.3e-05	1.1e-02
GO:1904874	positive regulation of telomerase RNA localization to Cajal body	7	8.3e-05	2.0e-02
GO:0006090	pyruvate metabolic process	8	9.6e-05	2.1e-02
GO:0070125	mitochondrial translational elongation	16	1.1e-04	2.2e-02
GO:0006446	regulation of translational initiation	10	1.5e-04	2.8e-02
GO:0043039	tRNA aminoacylation	5	1.6e-04	3.0e-02
GO:0018107	peptidyl-threonine phosphorylation	10	2.9e-04	4.9e-02
GO:0000462	maturation of SSU-rRNA from tricistronic rRNA transcript	9	3.3e-04	5.4e-02
GO:0006610	ribosomal protein import into nucleus	5	3.7e-04	5.6e-02
GO:0016236	macroautophagy	14	4.0e-04	5.9e-02

RNA-seq: Top age-related GO terms (Cellular Component)

GO term ID	Description	Count	p-value	Benjamini
GO:0016020	membrane	220	9.8e-21	3.7e-18
GO:0005739	mitochondrion	157	1.2e-20	3.7e-18
GO:0070062	extracellular exosome	242	4.3e-16	9.1e-14
GO:0005829	cytosol	282	1.0e-15	1.6e-13
GO:0005913	cell-cell adherens junction	57	9.5e-15	1.2e-12
GO:0005737	cytoplasm	380	2.3e-13	2.4e-11
GO:0043209	myelin sheath	36	4.7e-13	4.2e-11
GO:0005759	mitochondrial matrix	47	5.7e-09	4.5e-07
GO:0005654	nucleoplasm	217	1.1e-08	7.8e-07
GO:0000502	proteasome complex	18	1.4e-08	8.0e-07
GO:0005743	mitochondrial inner membrane	56	1.4e-08	8.0e-07
GO:0042645	mitochondrial nucleoid	14	3.5e-07	1.8e-05
GO:0014704	intercalated disc	14	8.5e-07	4.2e-05
GO:0005832	chaperonin-containing T-complex	7	2.5e-06	1.1e-04
GO:0005643	nuclear pore	16	5.2e-06	2.2e-04
GO:0043231	intracellular membrane-bounded organelle	55	2.7e-05	1.1e-03
GO:0002199	zona pellucida receptor complex	6	2.9e-05	1.1e-03
GO:0043034	costamere	8	5.4e-05	1.9e-03
GO:0043234	protein complex	42	7.8e-05	2.6e-03
GO:0045254	pyruvate dehydrogenase complex	5	1.5e-04	4.6e-03

Conclusion

- GBMs provide a flexible framework for the analysis of matrix data.
- Delta propagation is a novel general technique for uncertainty quantification.
- Our algorithms enable accurate GBM estimation and inference in modern applications.
- Possible directions for future work:
  - extend to more general bilinear model structures,
  - seek theoretical guarantees for delta propagation, and
  - try applying delta propagation to other models.
- Preprint is on arXiv: `https://arxiv.org/abs/2010.04896`

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