DART-ID Increases Single Cell Proteome Coverage

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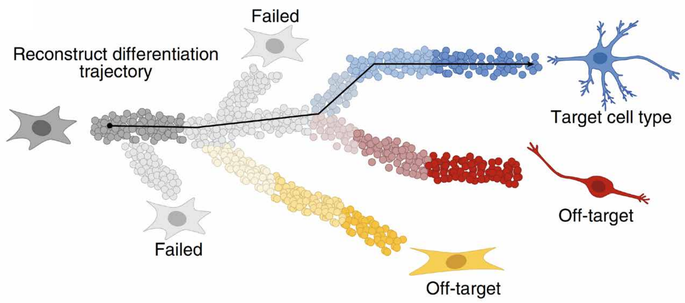
Summary

- Single cell proteomics by mass spectrometry can help answer may biological questions involving cell diversity and heterogeneity
- Peptide/protein identification is a noisy, stochastic process
- Incorporating peptide retention time (RT) within a principled Bayesian framework increases proteome coverage
- Global RT alignment method is more accurate and boosts coverage
- Manuscript: https://doi.org/10.1101/399121
- Code: https://github.com/SlavovLab/DART-ID

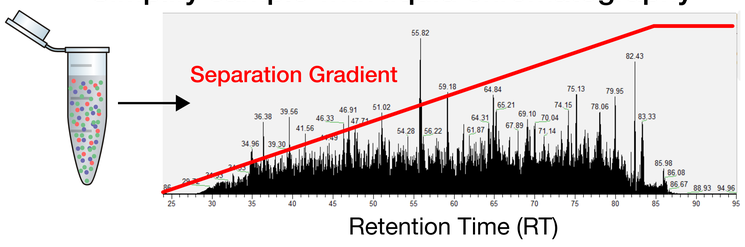
Improve Directed Cell Differentiation



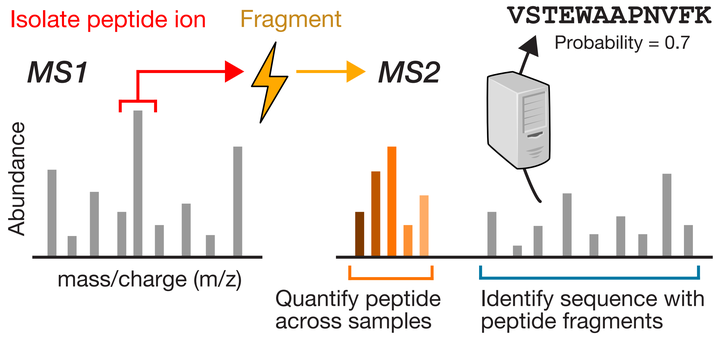
- Use single cell proteomics to profile differentiating cells
- Understand branching points, find biomarkers, and intervene to maximize differentiation efficiency

Peptide Identification by Mass Spectrometry

Simplify sample with liquid chromatography

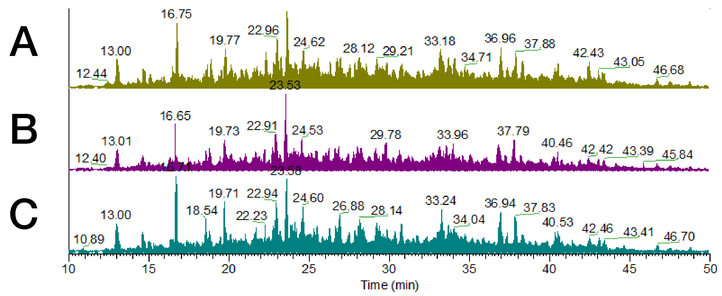


Identify and quantify peptides with fragments



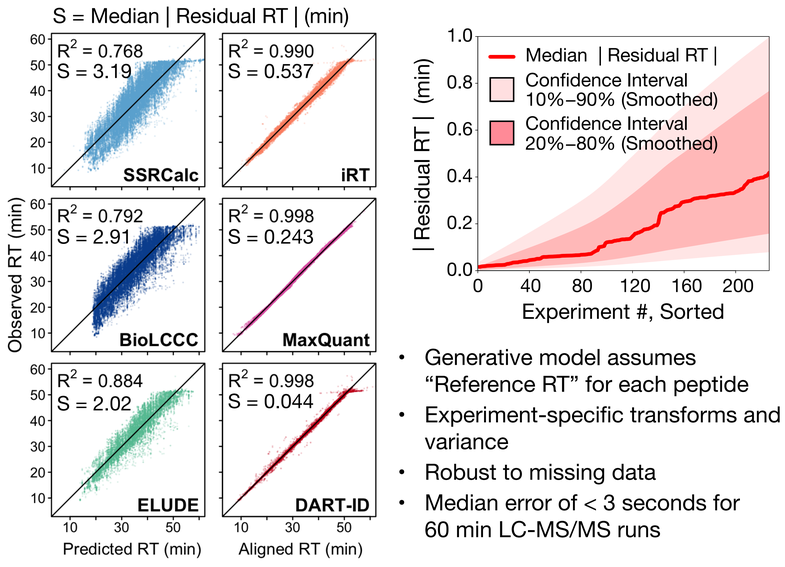
GOAL: Incorporate retention time into identification

Conventional Pairwise RT Alignments



- Conventional methods align pairwise to a "reference experiment"
- Choice of reference experiment not obvious
- Does not account for measurement errors in reference experiment

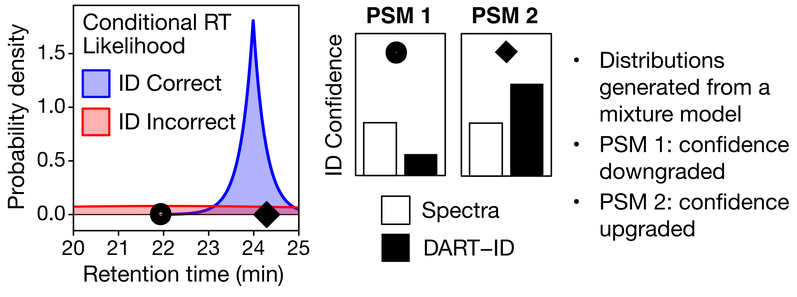
Global RT Alignment Reduces Error



- Generative model assumes "Reference RT" for each peptide
- Experiment-specific transforms and variance
- Robust to missing data
- Median error of < 3 seconds for 60 min LC-MS/MS runs

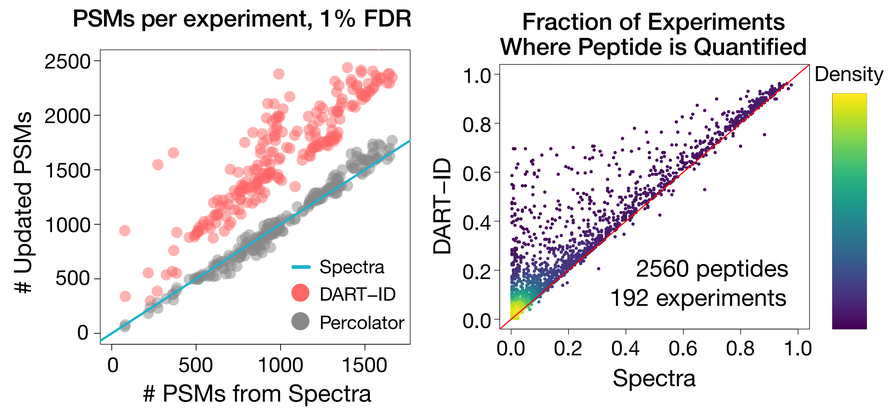
Incorporating RT Inferences into Identification with Bayes Theorem

$$P(\text{ ID correct } | \text{ RT }) = \frac{P(\text{ RT } | \text{ ID correct }) \times P(\text{ ID correct })}{P(\text{ RT })}$$

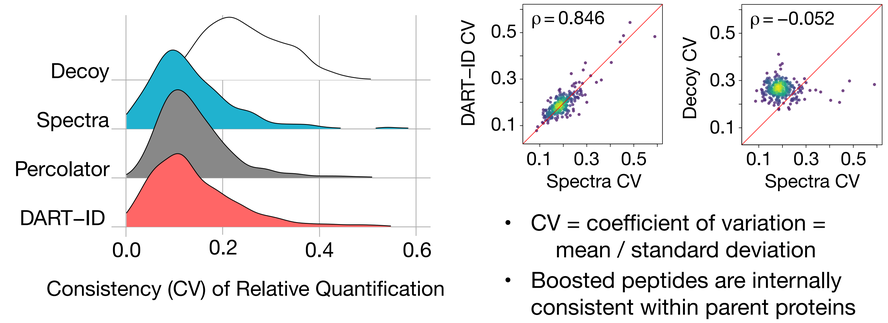


- Distributions generated from a mixture model
- PSM 1: confidence downgraded
- PSM 2: confidence upgraded

DART-ID Identifies More Peptides and Reduces Missing Data

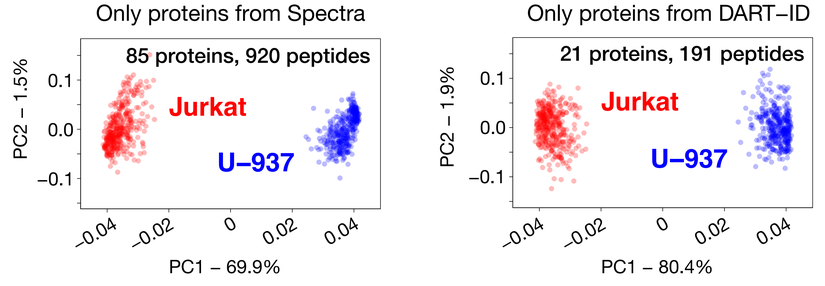


Boosted PSMs Give Consistent Protein Quantification



- CV = coefficient of variation = mean / standard deviation
- Boosted peptides are internally consistent within parent proteins

DART-ID Proteins Separate Cell Types



- Separation of blood cancer cell proteomes by Principal Component Analysis (PCA)

Conclusion

DART-ID takes advantage of reproducible retention times for peptide sequences within sets of LC runs to greatly increase the coverage of single cell proteomes.

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