# Scikit-ribo - Accurate A-site prediction and robust modeling of translational control

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Genome Informatics

# Acknowledgments


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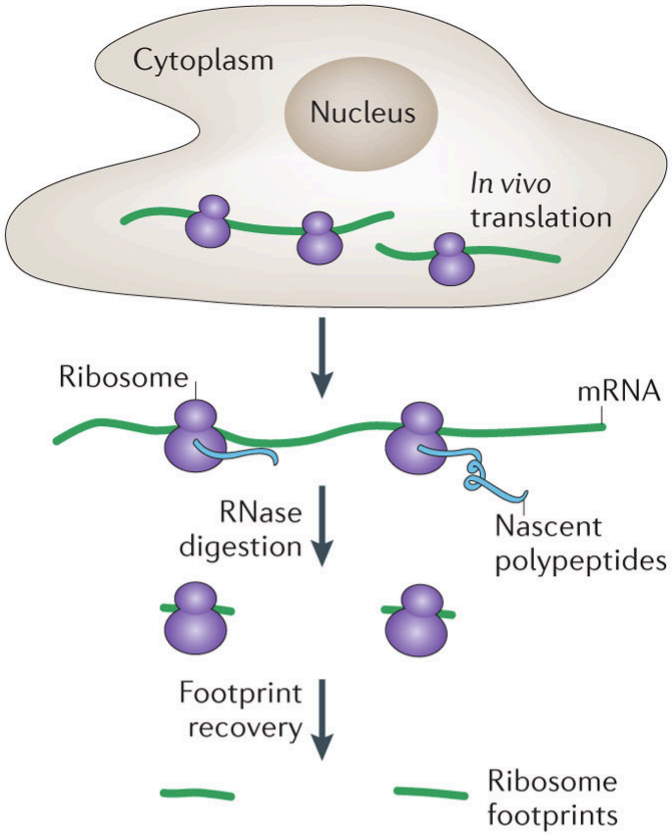
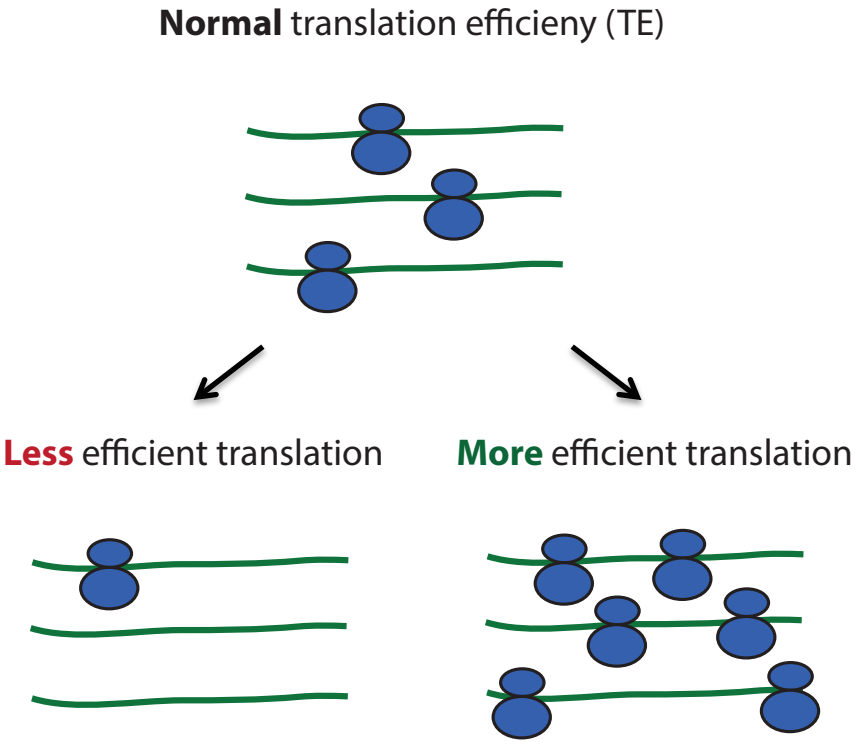
# Central dogma of biology – Classic view

# What is ribosome profiling (Riboseq)?

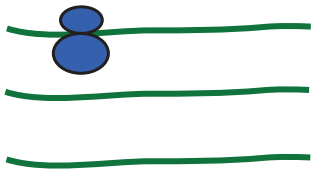



Ingolia. *Science.* (2009)
Ingolia. *Nat Rev Genet.* (2014)

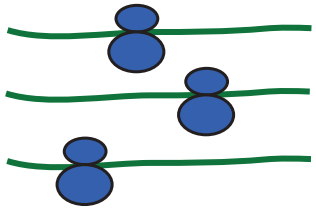
# Calculate translational efficiency (TE)

**Less** efficient translation



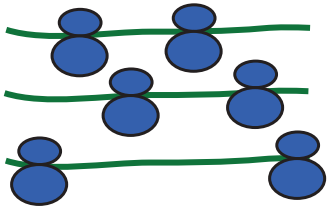
$\log_2(TE) < 0$

**Normal** translation efficieny (TE)



$\log_2(TE) = 0$

**More** efficient translation

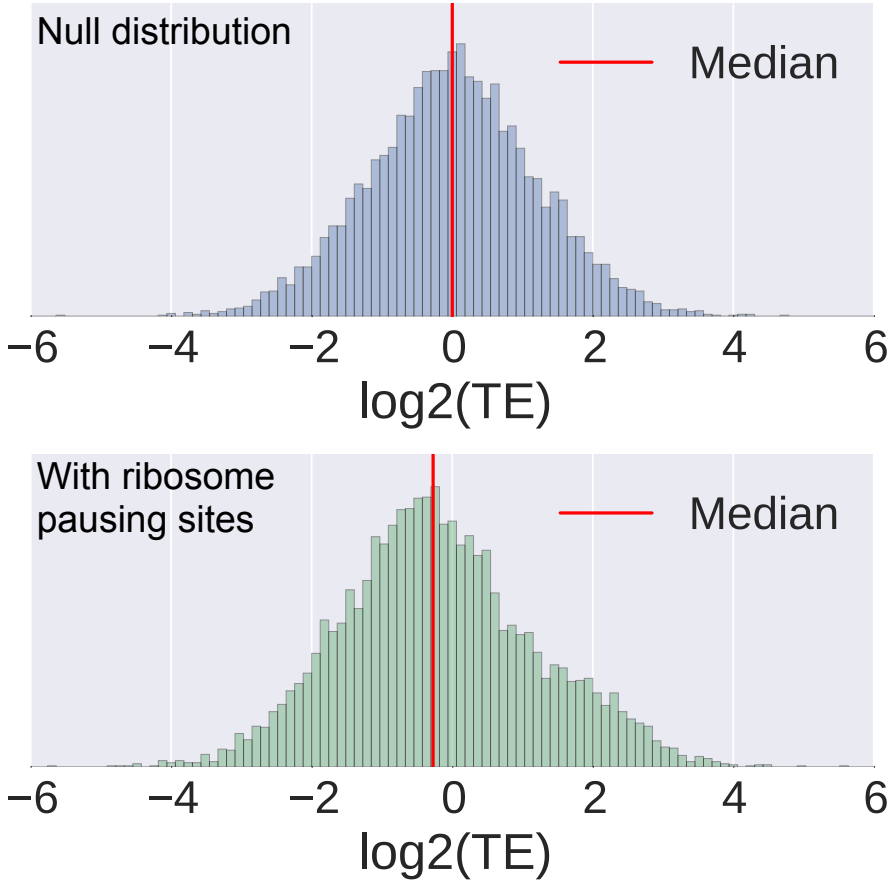


$\log_2(TE) > 0$

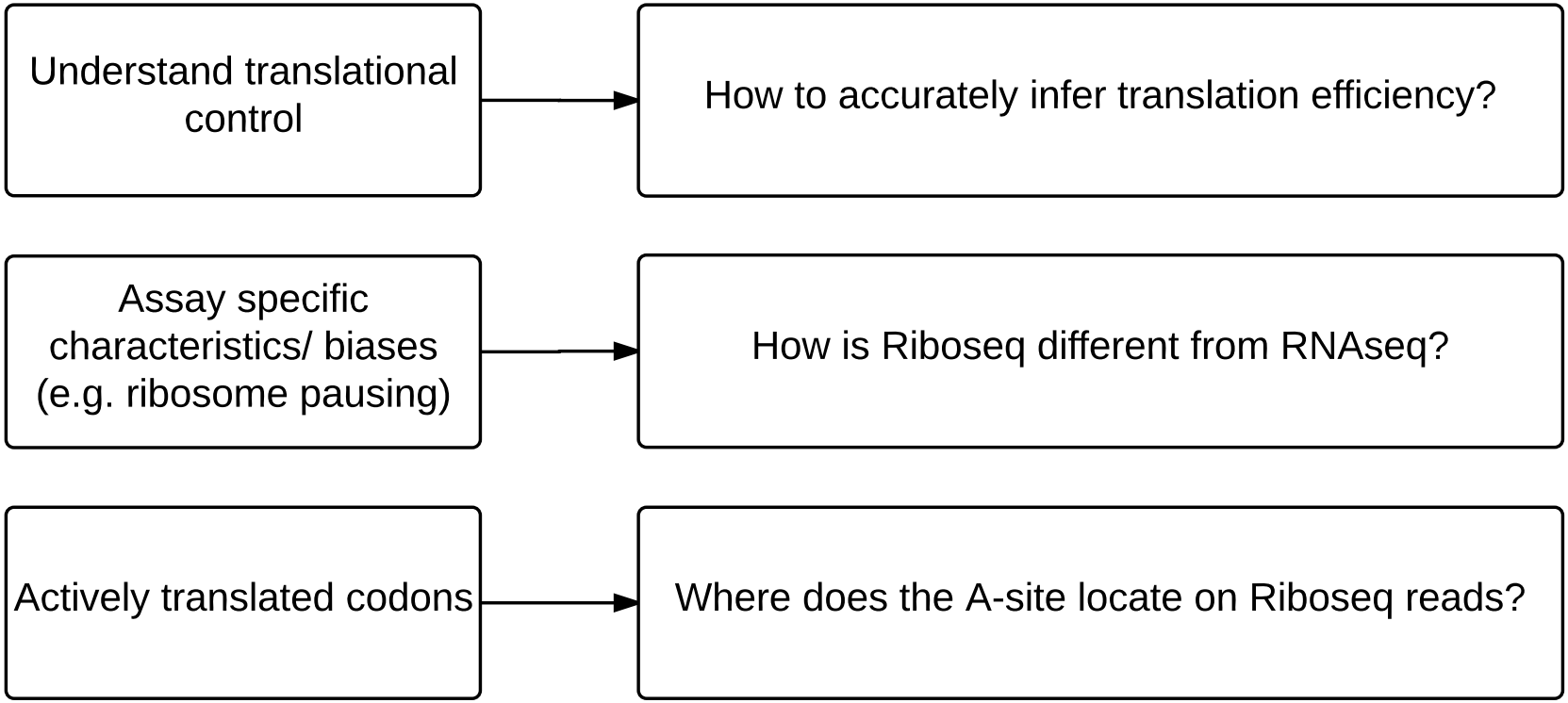
$$TE = \frac{Riboseq\ \ rpkm}{RNAseq\ \ rpkm}$$

Hypothesis: TE distribution could be skewed by ribosome pausing events.

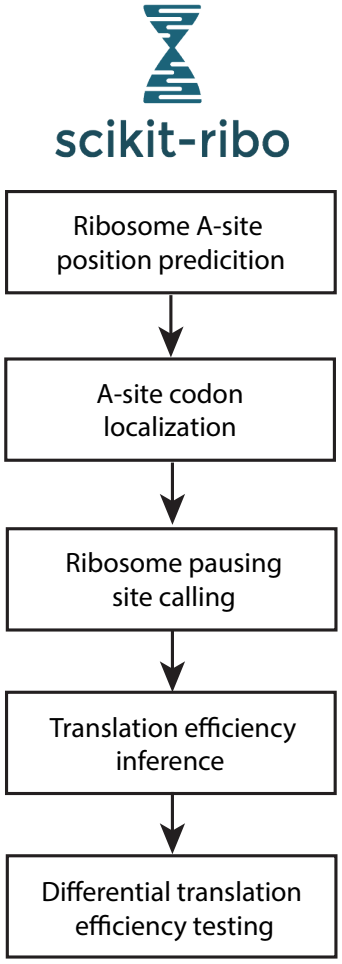
# Simulated *S. cerevisiae* data - TE distribution are negatively-skewed by ribosome pausing events



# Analytical Challenges



# Introducing scikit-ribo



# What and where is the ribosome A-site?

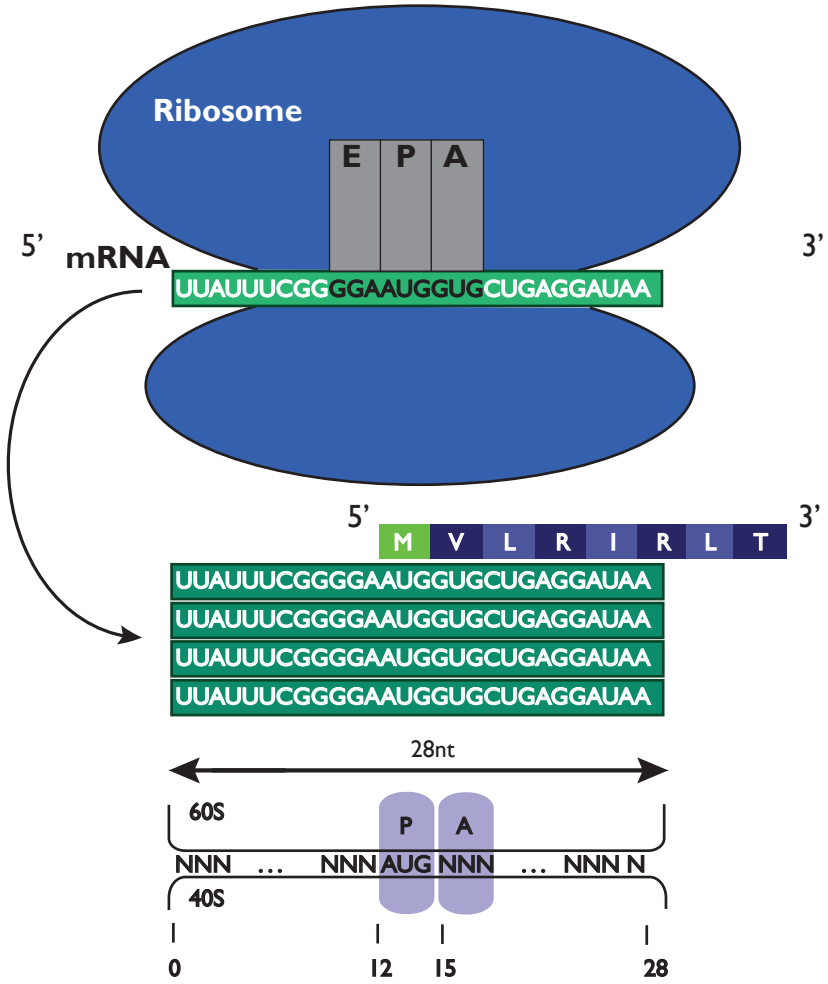
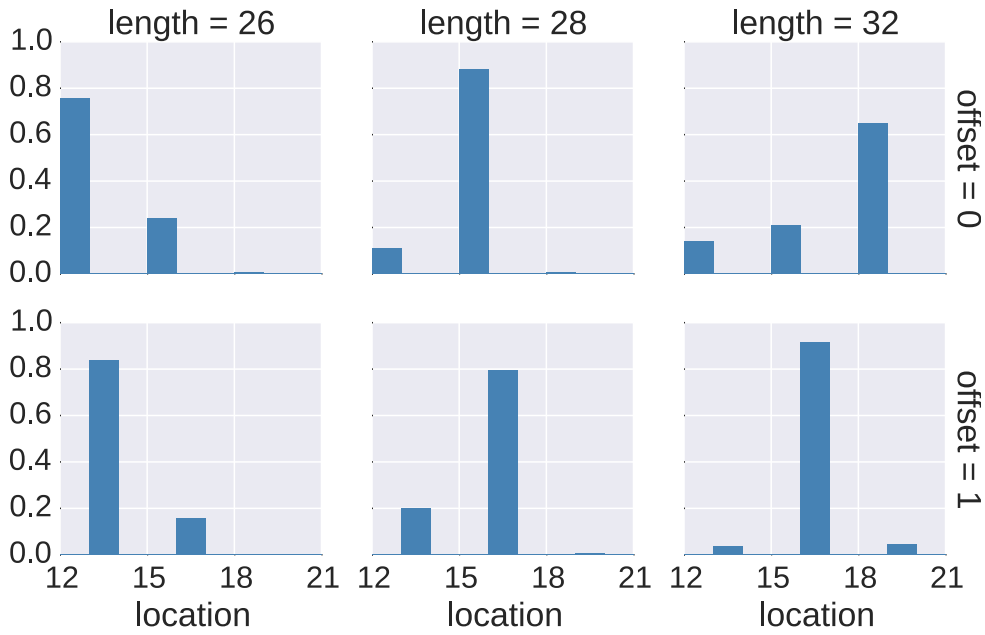
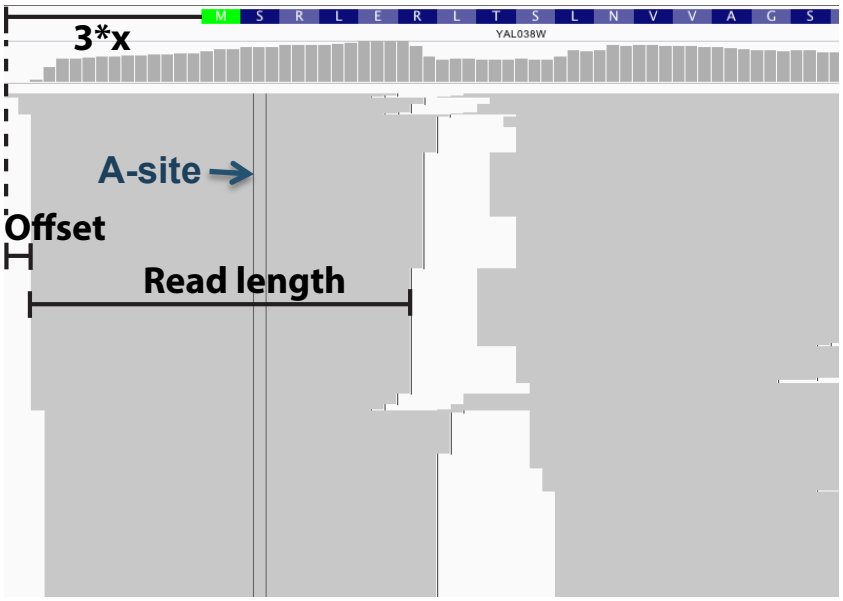



Figure adapted from Ingolia et al. *Science* (2009)



# How to predict A-site?

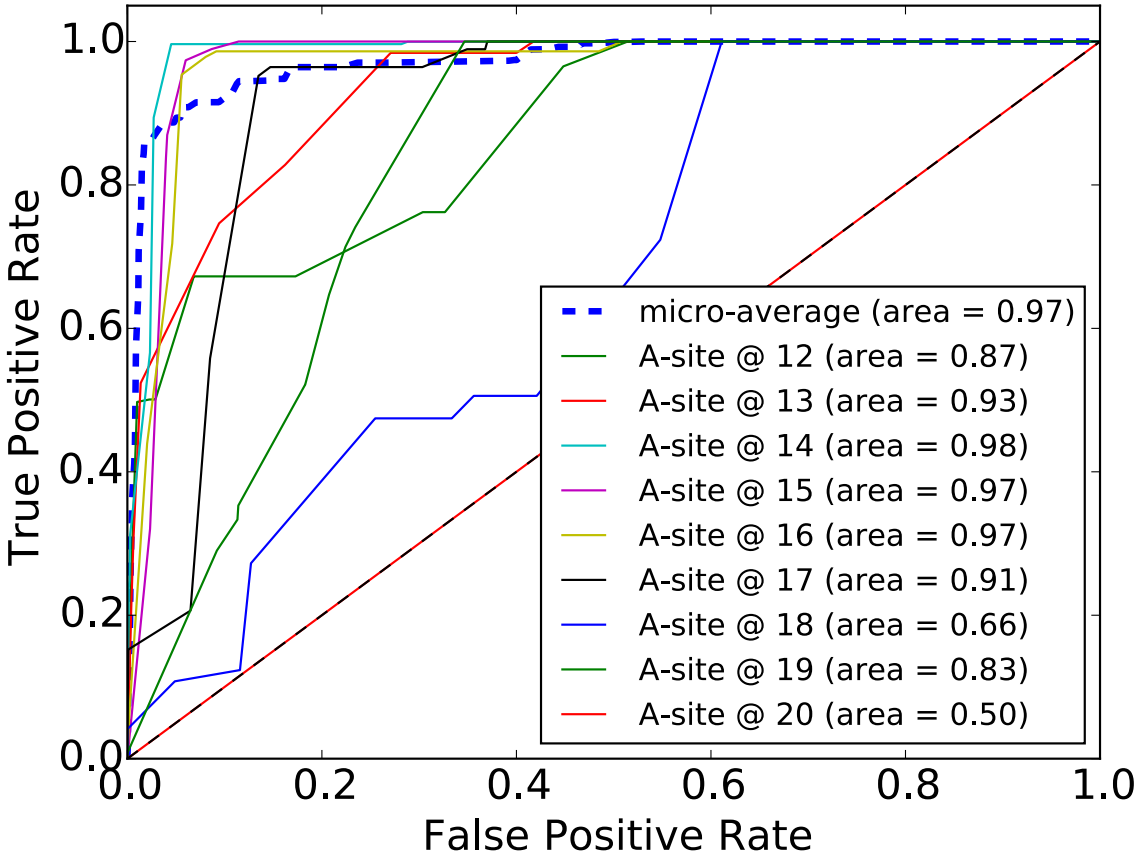
Training data and features:




Classifier and model tuning:

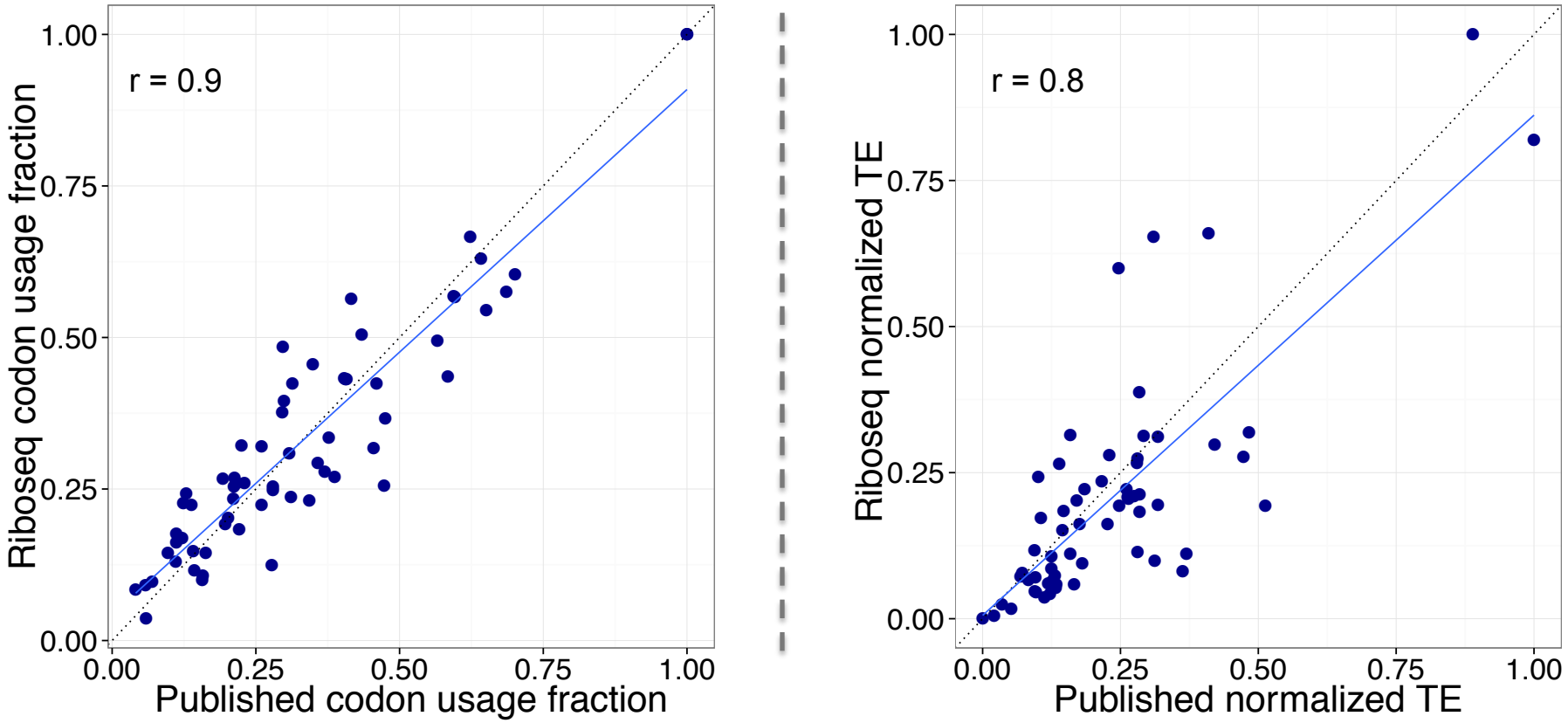
- SVM with RBF kernel (scikit-learn)
- 10 fold cross-validation for grid search
- Make predictions on all reads genome-wide

# Prediction performance by cross validation



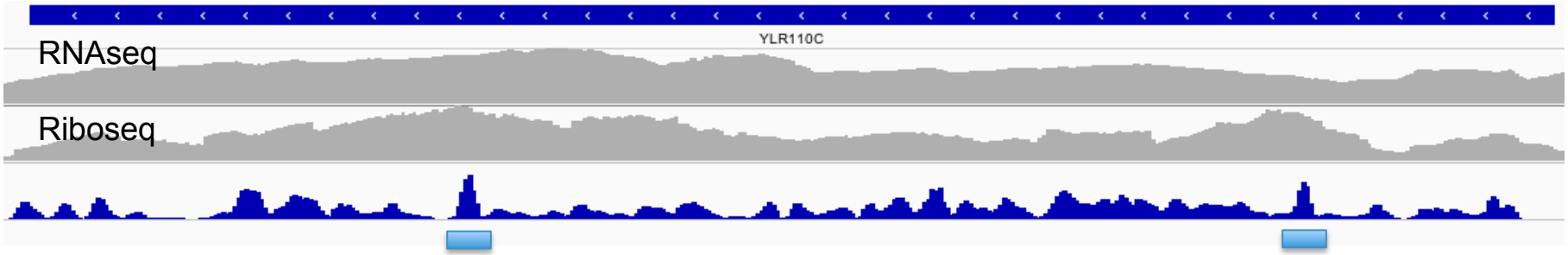

Scikit-ribo has much higher accuracy of identifying A-site than the previous method (0.86 vs. 0.64, 10-fold CV).

# Scikit-ribo accurately predicted codon usage fraction and codon normalized TE



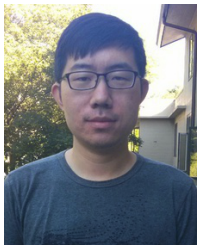

Evolutionary conservation of codon optimality reveals hidden signatures of cotranslational folding, Pechmann, Frydman (2013).

# Finding ribosome pausing sites (peaks) is hard. But it is easier after knowing the A-site location.




Q: how to robustly identify ribosome pausing sites while accounting for over-dispersion?

# Ribosome pausing site identification by negative binomial mixture model



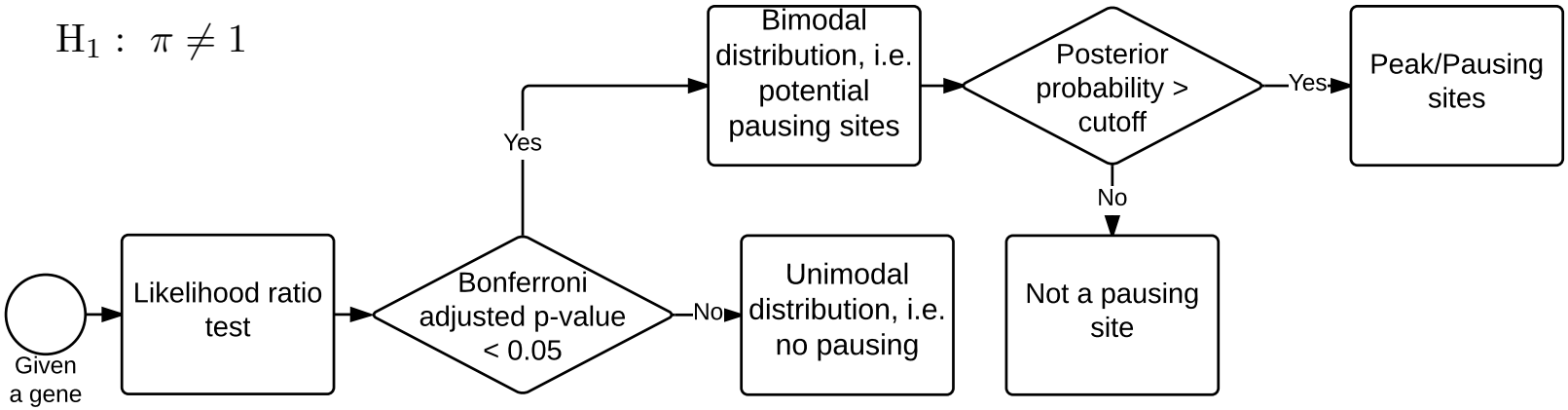
Yifei Huang

$$P(\mathbf{X}_i|\pi_i,\mu_i,k_i,r_i) = \prod_j \pi_i \mathcal{NB}(X_{ij}|\mu_i,r_i) + (1-\pi_i)\mathcal{NB}(X_{ij}|k_i\mu_i,r_i),$$

for gene i at position j, where k $\geq 5$

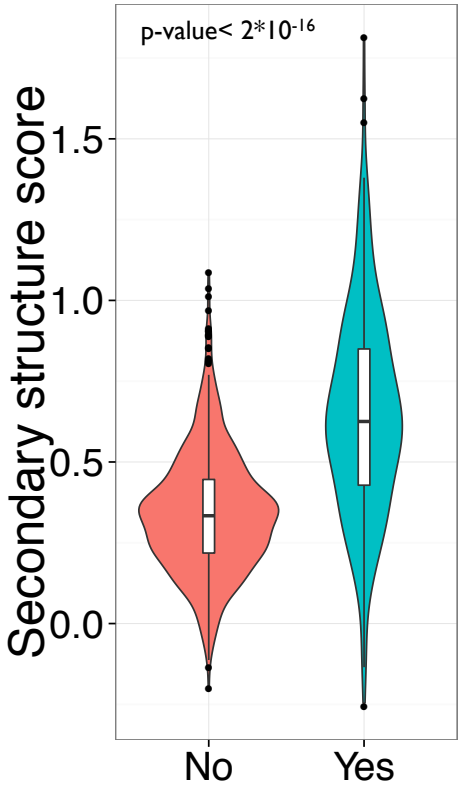
$H_0: \ \pi = 1$

$H_1: \ \pi \neq 1$



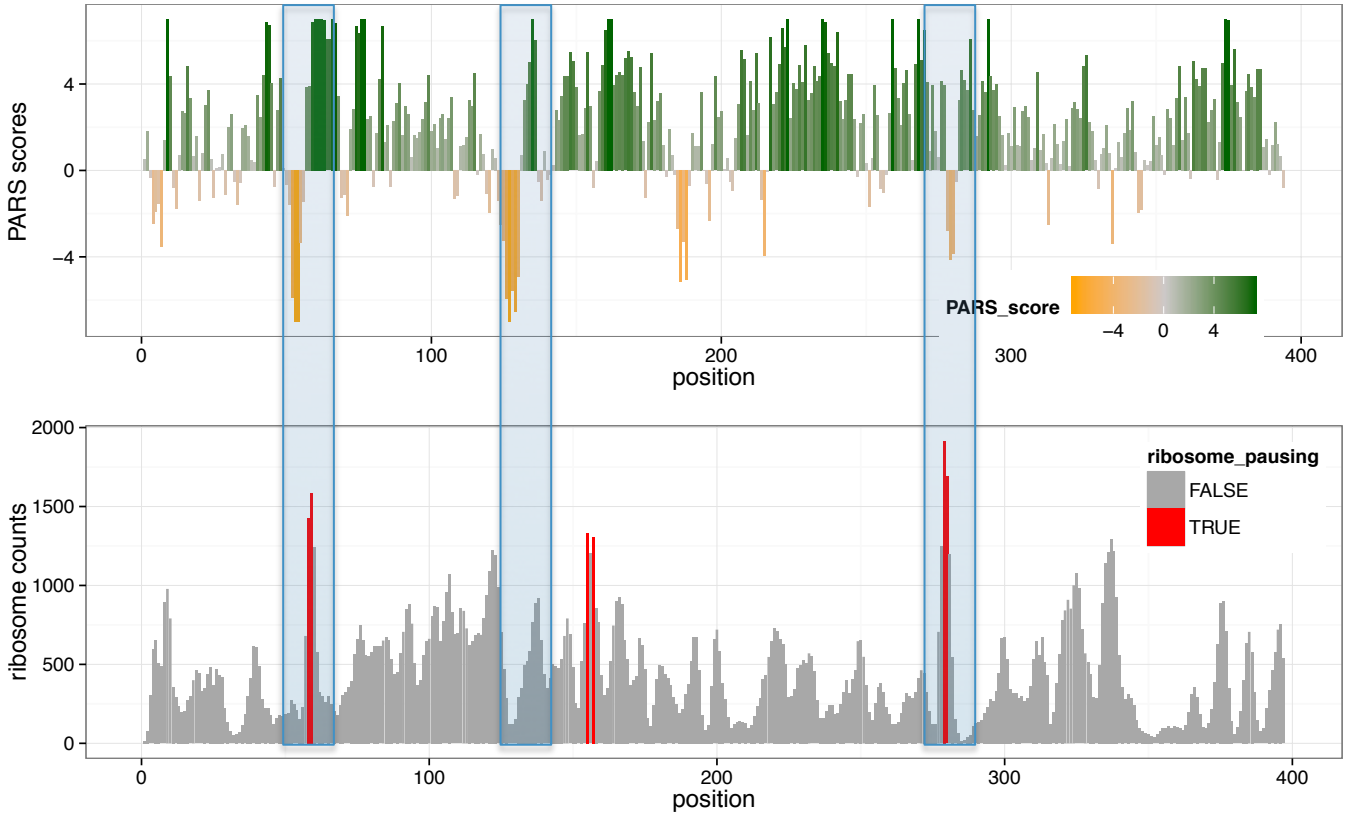

| # genes | # genes (rpkm > 100) | # genes with pausing | # ribosome pausing sites identified |
|---|---|---|---|
| 6664 | 1252 | 94 | 180 |

# mRNA with stronger secondary structure tend to have ribosome pausing events

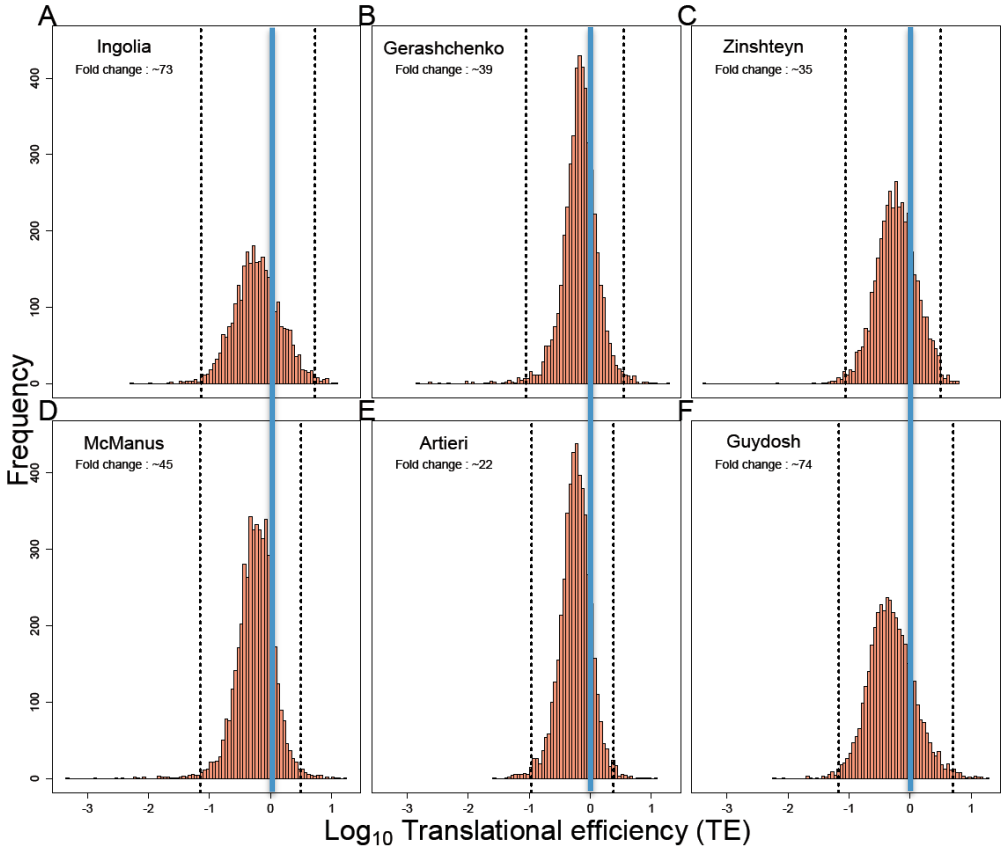


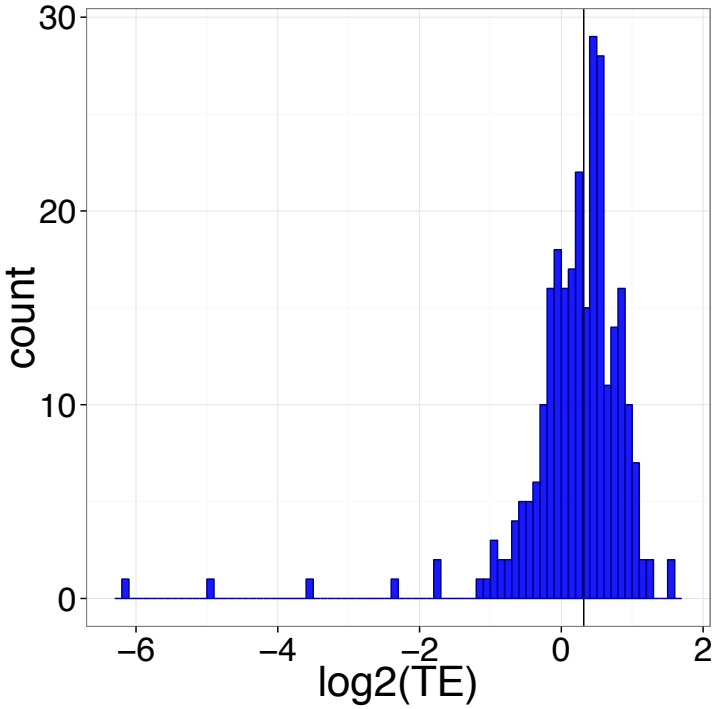

Fisher exact test p-value = 0.001

Kertesz et al. Nature (2010)



# TE distributions are negatively-skewed in many studies. Over-structured mRNA show inflated TE.




Chi Square test p-value < $2*10^{-16}$

Improved ribosome-footprint and mRNA measurements provide insights into dynamics and regulation of yeast translation. Weinberg, Shah et al. (2015)

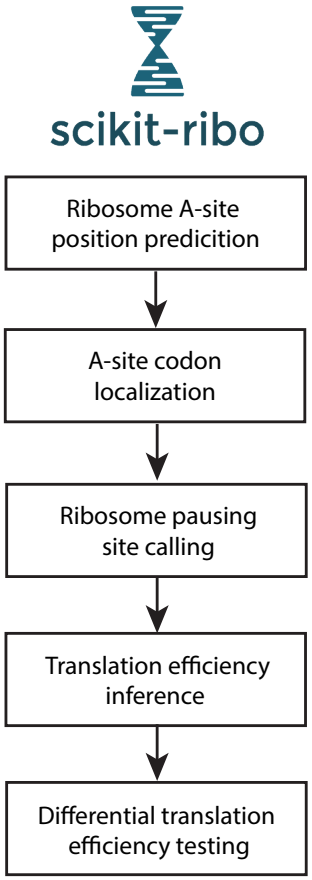
# Summary

**Discussed:**

1) Introduce scikit-ribo for joint analysis of Riboseq & RNAseq data.
2) Learn from data itself to determine ribosome A-site location.
3) Reveal biases in Riboseq data due to ribosome pausing.
4) How Riboseq biases lead to issues with estimating TE.

**Ongoing work:**

1) Adjust for those biases and provide an unbiased estimate of TE.
2) Extend the ribosome pausing calling to a HMM based method.
3) Joint inference of translation initiation and elongation rates.




https://github.com/hanfang/