

Gene-specific optimization of data integration improves regression-based Gene Regulatory Network inference in Arabidopsis

NetBio COSI

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Regression-based GRN inference paradigm

Modelling assumption

The expression of regulators hold predictive and descriptive power over the expression of their target genes

Ex : GENIE3 [Huynh-Thu et al., 2010], The Inferelator [Gibbs et al., 2022]



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Limitations

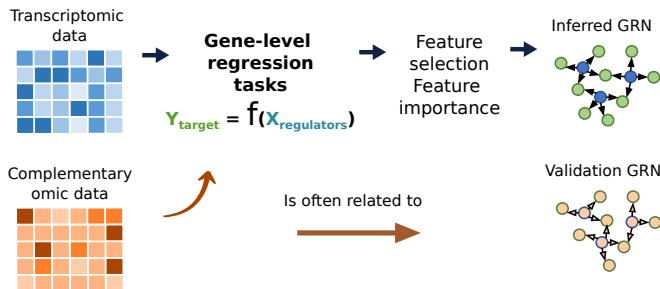
- High dimension
- High correlation among predictors
- Incomplete view of the regulation process

Data integration in regression-based GRN inference

Modelling assumption

Complementary omics can bring more causality to GRN inference

Ex : iRafNet [Petralia et al., 2015], MEN [Greenfield et al., 2013], LASSO-Stars [Miraldi et al., 2019]

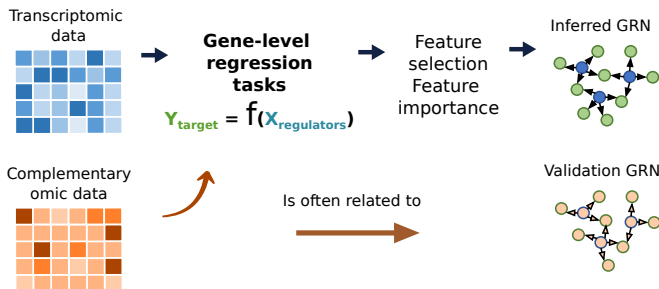


Data integration in regression-based GRN inference

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Current limitations

Omics contributions are **rarely finely tuned**.
They usually **rely on a close gold standard**.

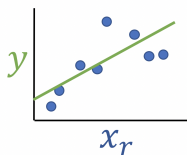
Objectives

- 1 Can we define a new criterion to **robustly estimate the optimal strength of data integration** based on available data?
- 2 What is the benefit of optimising data integration at the **gene level**?

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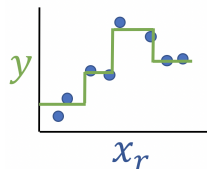
↳ Investigated for **two common forms of integrative regression**



$$Y_{\text{target}} = f(X_{\text{regulators}})$$

Linear weightedLASSO
 Inspired from LASSO-Stars

Non linear weightedRF
 Inspired from iRafNet



[Miraldi et al., 2019]

[Petralia et al., 2015]

Modelling the root response to nitrate induction in *Arabidopsis thaliana*

RNASeq data : Y, X

N conditions



Temporal response to nitrate induction

1426 genes, 201

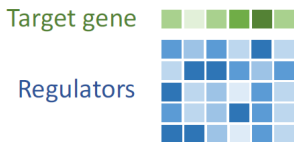
regulators, $N = 45$

samples [Varala et al., 2018]

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TFBM prior matrix : Π

PWM occurrence score
in the target's promoter

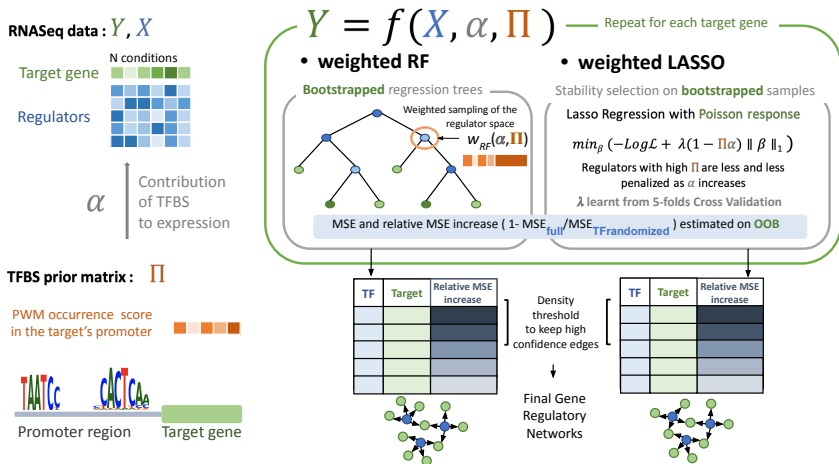


TF binding sites from **JASPAR** and the **Plant Cistrome Databases**

[Castro-Mondragon et al., 2021,
O'Malley et al., 2016]

$$\Pi_{r,t} = \begin{cases} 0 & \text{if the motif of } r \text{ is not in the promoter of } t \\ 1 & \text{if the motif of } r \text{ is in the promoter of } t \\ \frac{1}{2} & \text{if the motif of } r \text{ is missing} \end{cases}$$

Integrative regression-based GRN inference methods



iRafNet [Petralia et al., 2015]

LASSO-Stars [Miraldi et al., 2019]

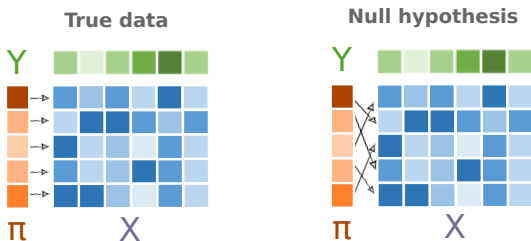
A gene-wise criterion to optimise data integration

Modelling assumption

We want to integrate **TFBMs** while **controlling that the prediction of gene expression is not deteriorated**.

A simulated null hypothesis

↳ Breaks the link between expression profiles and PWM scores, a case where **data integration is uninformative**



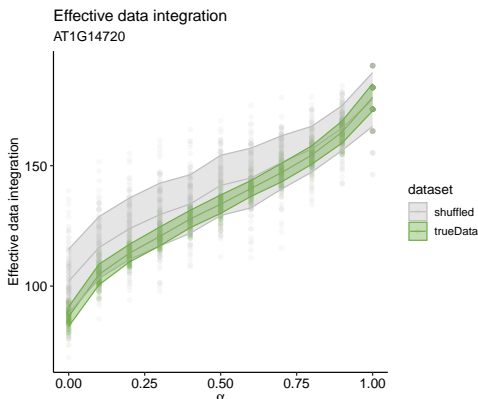
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Effective data integration

Average rank of PWM-supported regulators based on their importance



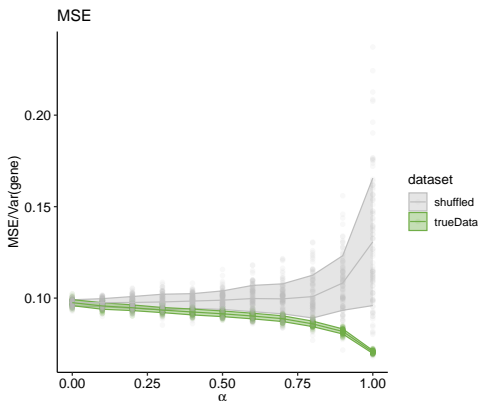
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Prediction error (MSE)

Error committed by the regression model in predicting the target gene expression on test conditions



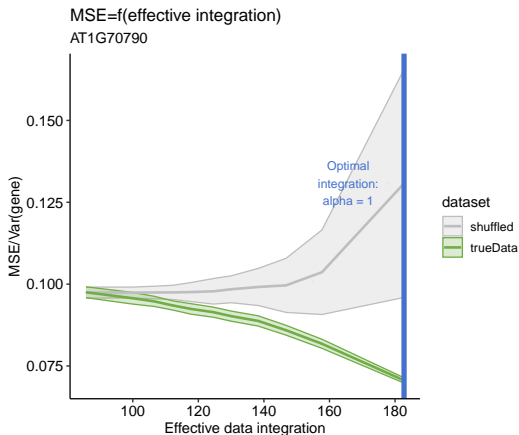
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Ideal case

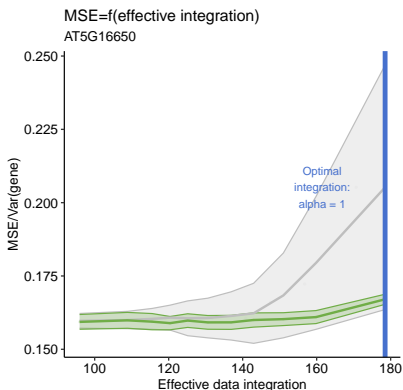
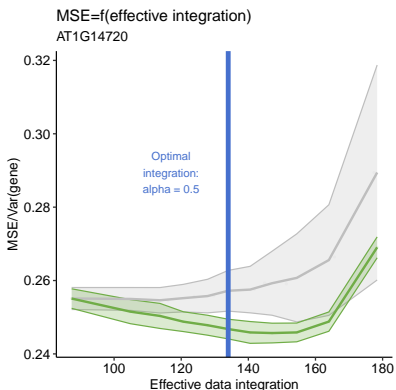
TFBM help selecting robust regulators and improve model generalisation performance



A gene-wise criterion to optimise data integration

Intermediate cases

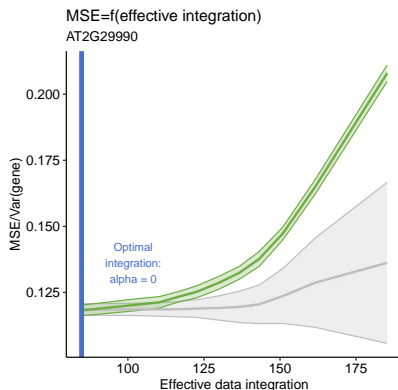
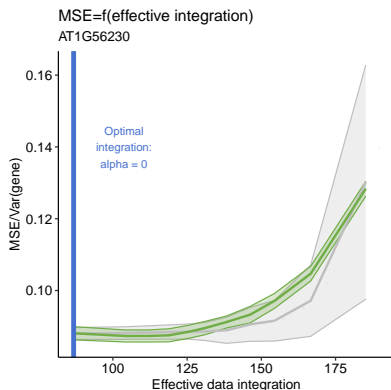
The MSE reaches an optimum, or is improved over chance



A gene-wise criterion to optimise data integration

Cases where data integration should be avoided

The MSE does not differ from chance or is even higher



Gene-specific integration offers low MSE, and good precision/recall

GRN

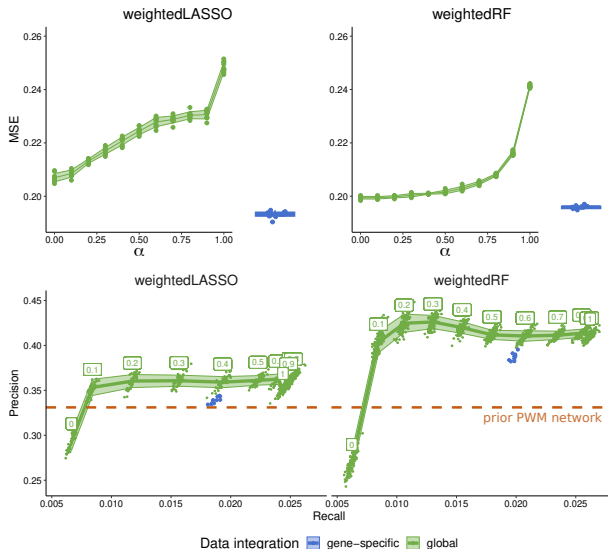
reconstruction

↳ 0.005 density threshold on importance-ranked edges (1432 edges)

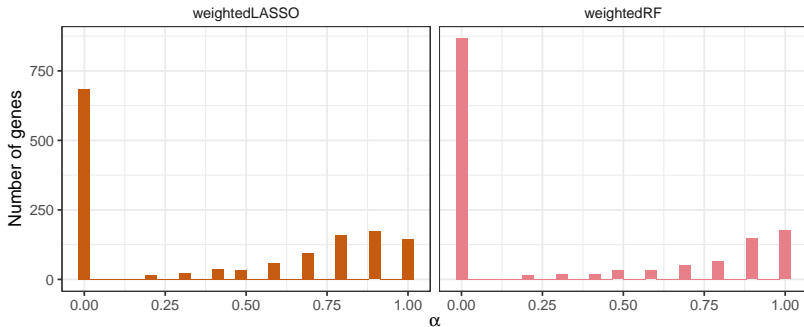
GRN quality

metrics

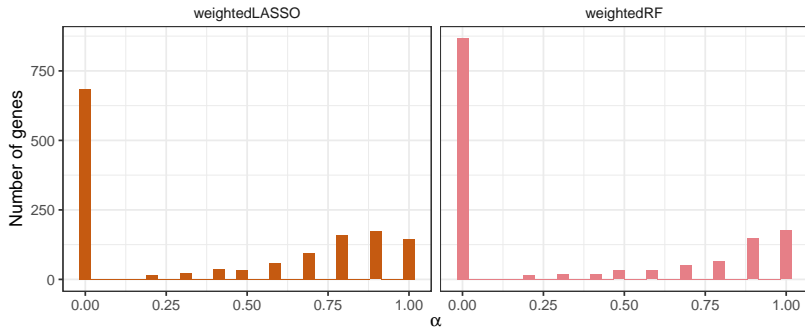
↳ **Median MSE**, and precision-recall curves against **DAP-Seq** in vitro TF binding [O'Malley et al., 2016]



Distributions of optimal integration strengths α_{opt}



Distributions of optimal integration strengths α_{opt}



TFBM integration is not warranted for all genes

↳ Cooperative binding events? Post-transcriptional regulations (RNA stability)?

Key message

Ingredients to tune integrative regression-based GRN inference

- **Tissue specific metrics** available for **all genes** (like the MSE) prioritized over related, incomplete and imperfect gold standards.
- **Simulated null hypotheses** breaking data integration assumptions. **TFBM will be integrated if they improve prediction performance over this baseline.**

Code : https://github.com/OceaneCsn/integrative_GRN_N_induction

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Results

- Desirable **trade-off between MSE and precision/recall.**
- Holds for both our linear and non-linear regression cases.
- Retrieves major players of nitrate nutrition in Arabidopsis.

Perspectives

Limitations

- **Computation time.**
 - ↳ Proof of concept that could be optimized in further works.
- **Missing TFBMs.**
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Future research directions

- ① Explore differences between the weightedLASSO and weightedRF and test the impact of **linearity assumptions**.
- ② Extend it to **other organisms**, potentially with enhancers to scan for TFBMs, and **other types of omics and prior knowledge** to integrate.

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Christophe Vroland, Elliott Butz, Quentin Bouvier



IPSIM : SIRENE team
Antoine Martin

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Effective data integration

Importance of PWM-supported regulators :

$$\frac{\sum_{\Pi_{r,t}=1} \text{Rank}(\text{Importance}_{r,t,\alpha})}{N_{\Pi_{r,t}=1}}$$

Prediction error (MSE)

Error committed in predicting the target gene expression :

$$\frac{1}{N_{OOB}} \sum_{i \in OOB} (y_{t,i} - \hat{y}_{m,t,i,\alpha})^2$$

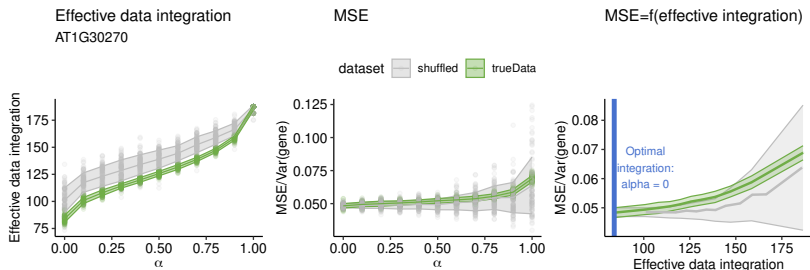
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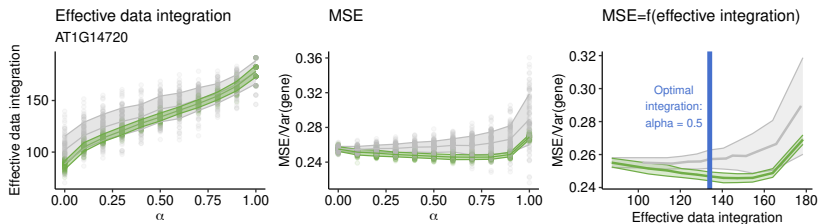
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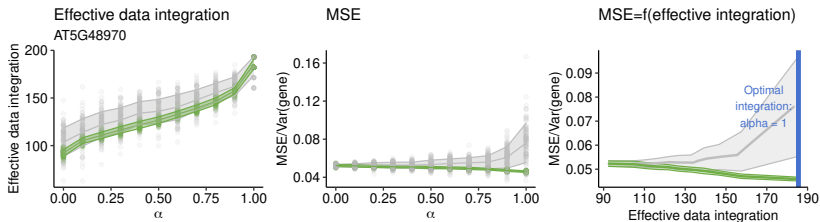
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Criterion formal definition

$$\Delta_{\alpha} = \frac{\mu_{\text{shuffle},\alpha} - \mu_{\text{true},\alpha}}{\sigma_{\alpha}} \quad \alpha_{\text{opt}} = \begin{cases} 0 & \text{if } \max_{\alpha \in [0,1]} (\Delta_{\alpha}) \leq 1 \\ \operatorname{argmax}_{\alpha \in [0,1]} (\Delta_{\alpha}) & \text{otherwise} \end{cases}$$

Deviation measure choice (σ_{α}) can modulate integration stringency:

- $\sigma_{\alpha} = \sigma_{\text{shuffle},\alpha} \rightarrow$ Low, stringent data integration
- $\sigma_{\alpha} = \frac{1}{2}(\sigma_{\text{shuffle},\alpha} + \sigma_{\text{true},\alpha}) \rightarrow$ Moderate data integration
- $\sigma_{\alpha} = \sigma_{\text{true},\alpha} \rightarrow$ Strong, permissive data integration

Modelling of nitrate signalling can be improved

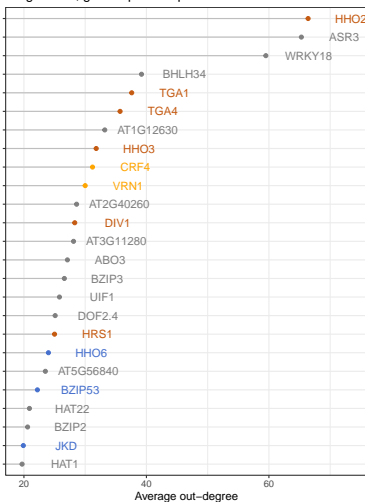
All models capture important nitrate actors

[Bellegarde et al., 2017, Vidal et al., 2020].

Gene specific optimisation of α uniquely retrieves :

- **NPL7** [Marchive et al., 2013, Alvarez et al., 2020] and **PHL1** [Ueda et al., 2020] for **weightedLASSO**
- New candidate TFs of interest for **weightedRF**

weightedRF, gene-specific optimisation



- Candidate nitrate regulator
- Nitrate regulator
- Retrieved by gene specific data integration optimisation