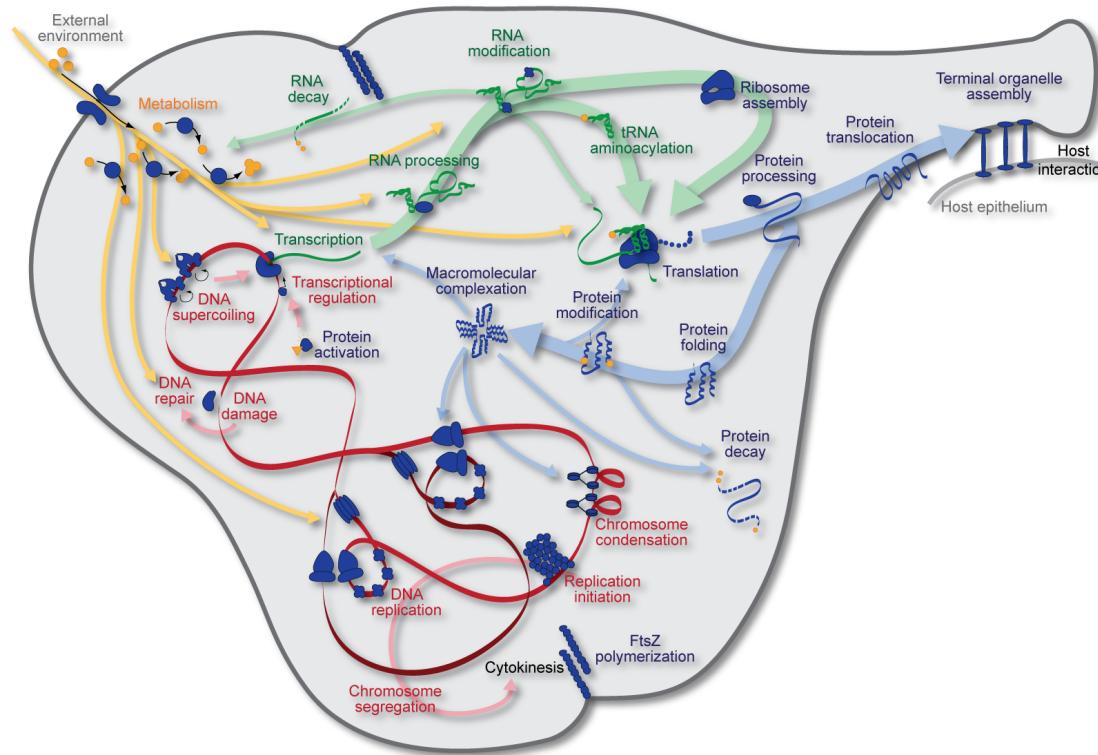
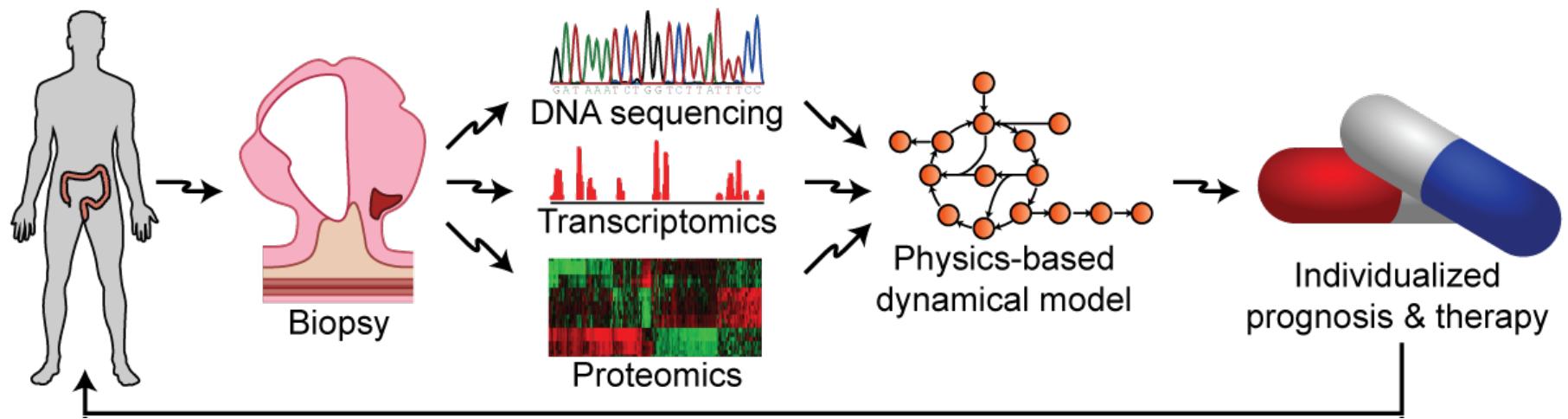


Toward methods, software, and standards for comprehensive whole-cell models

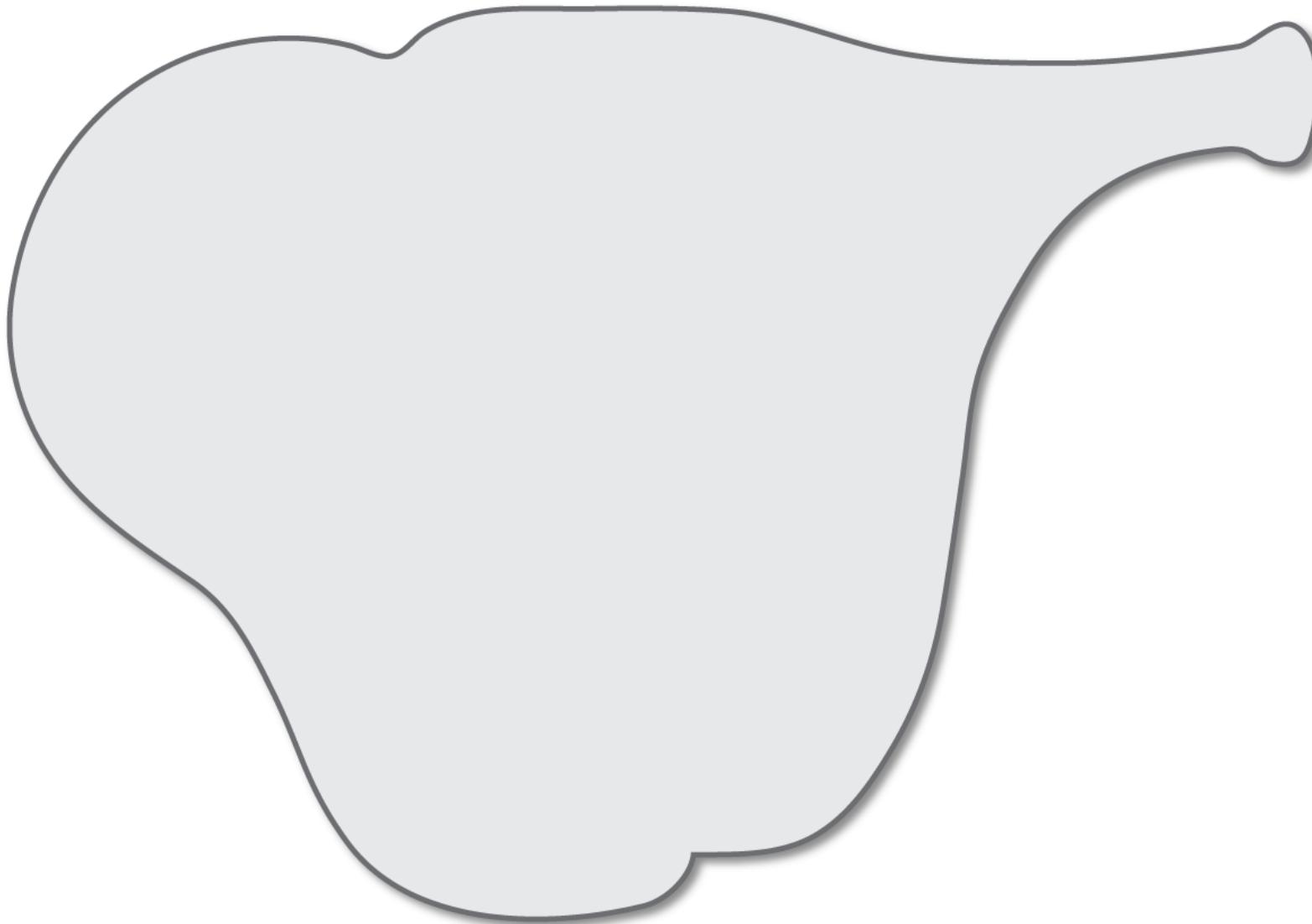


Jonathan Karr
October 12, 2015

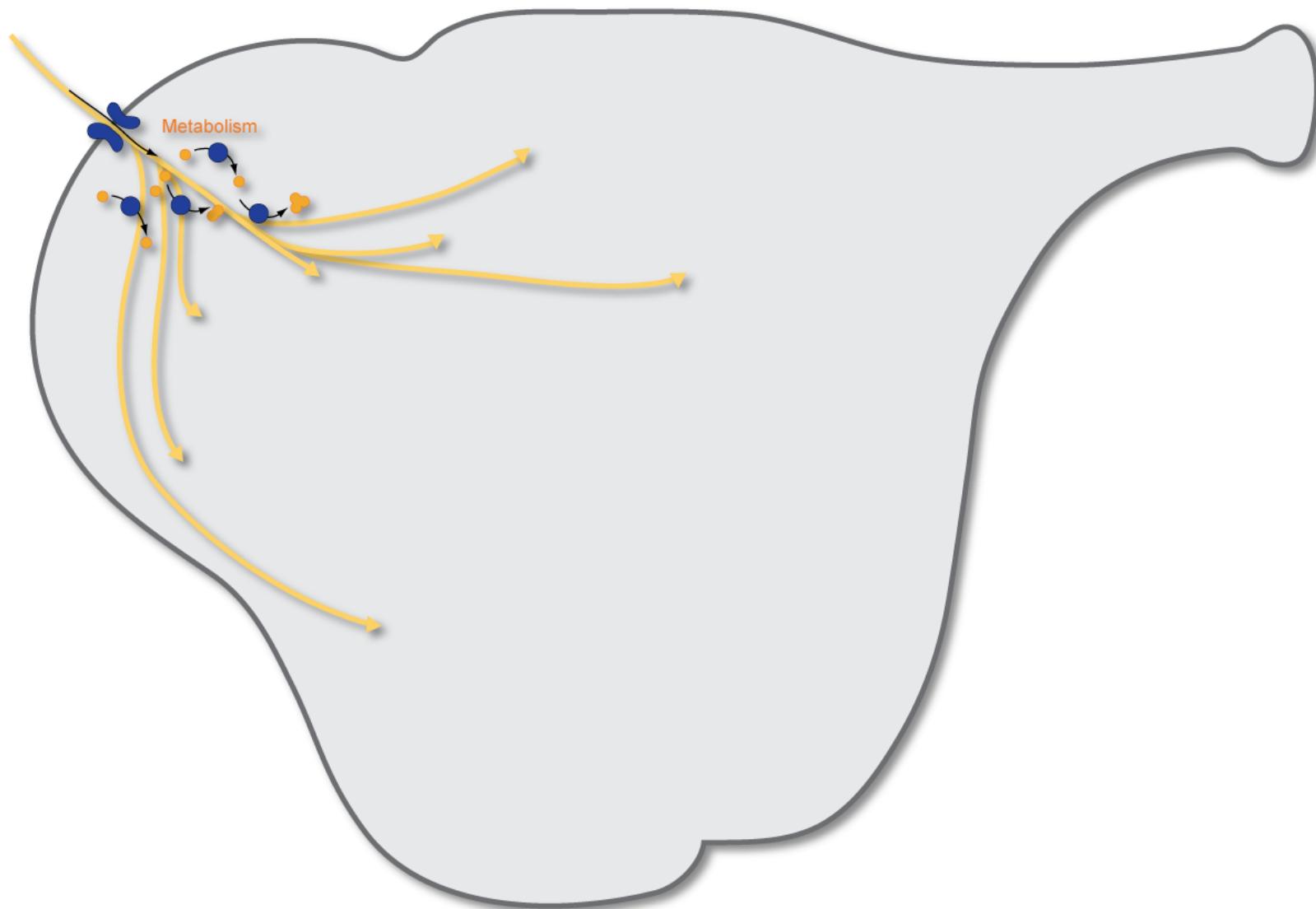


Central challenge: predict phenotype from genotype

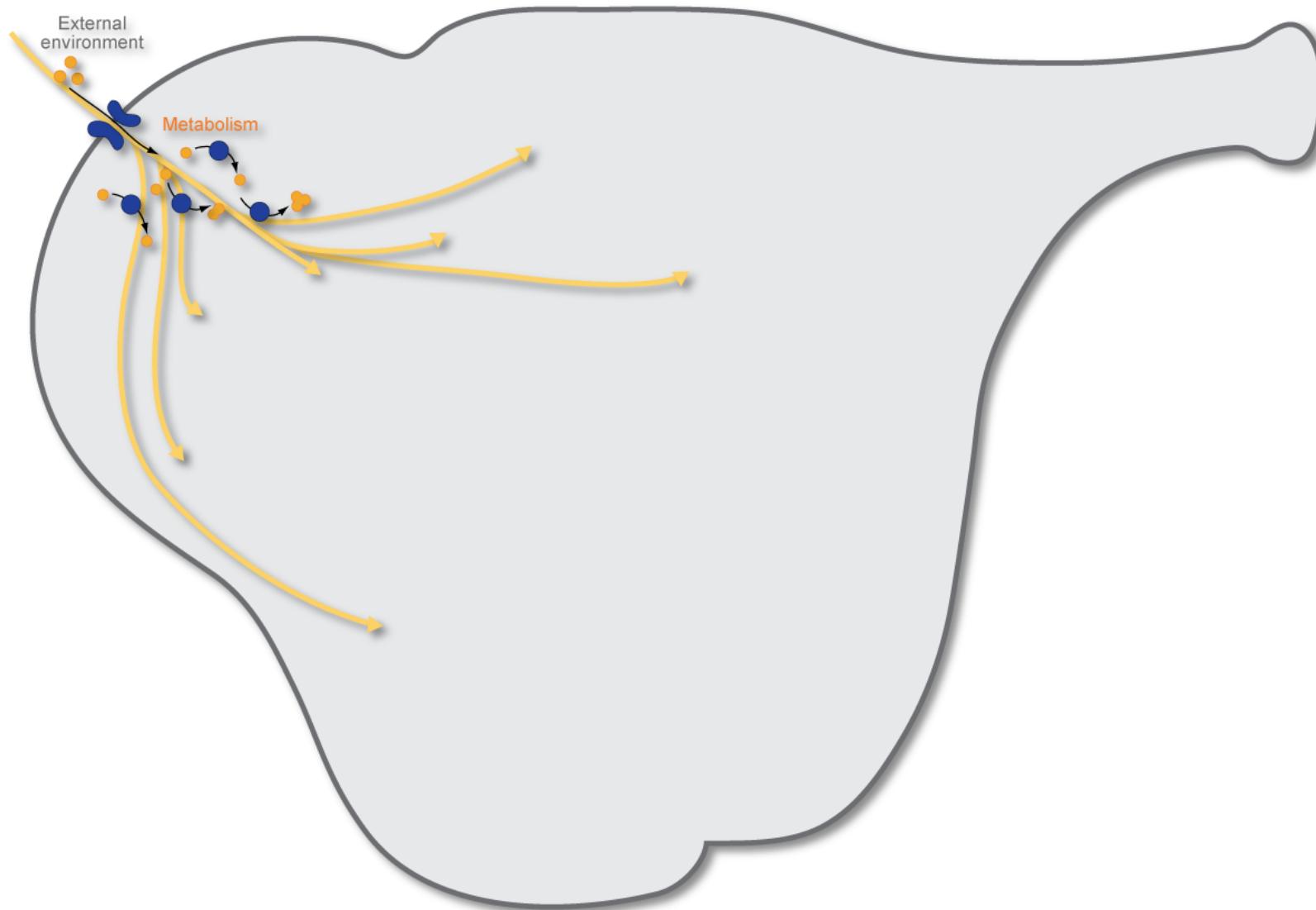
Example: drug biosynthesis



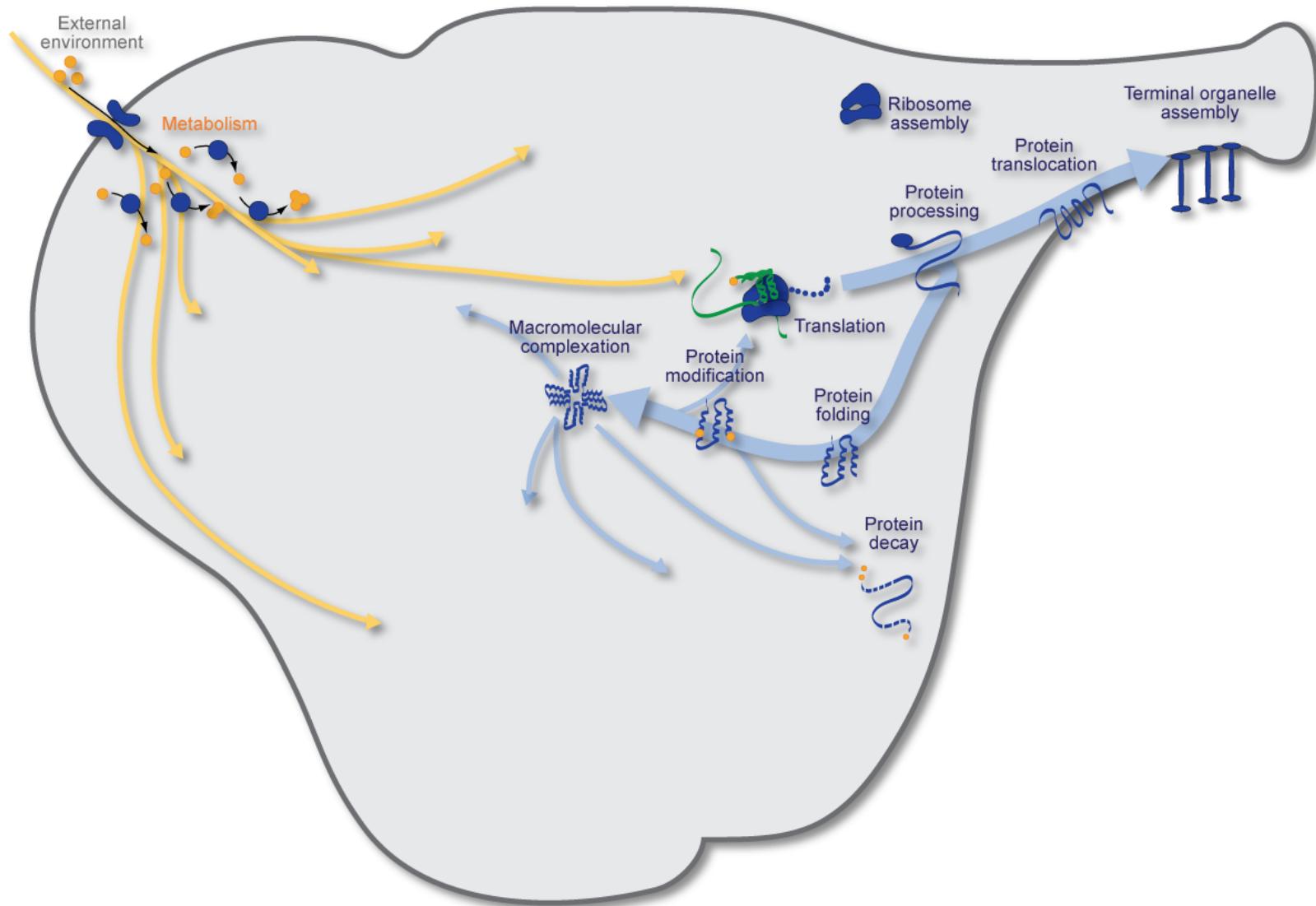
Example: drug biosynthesis



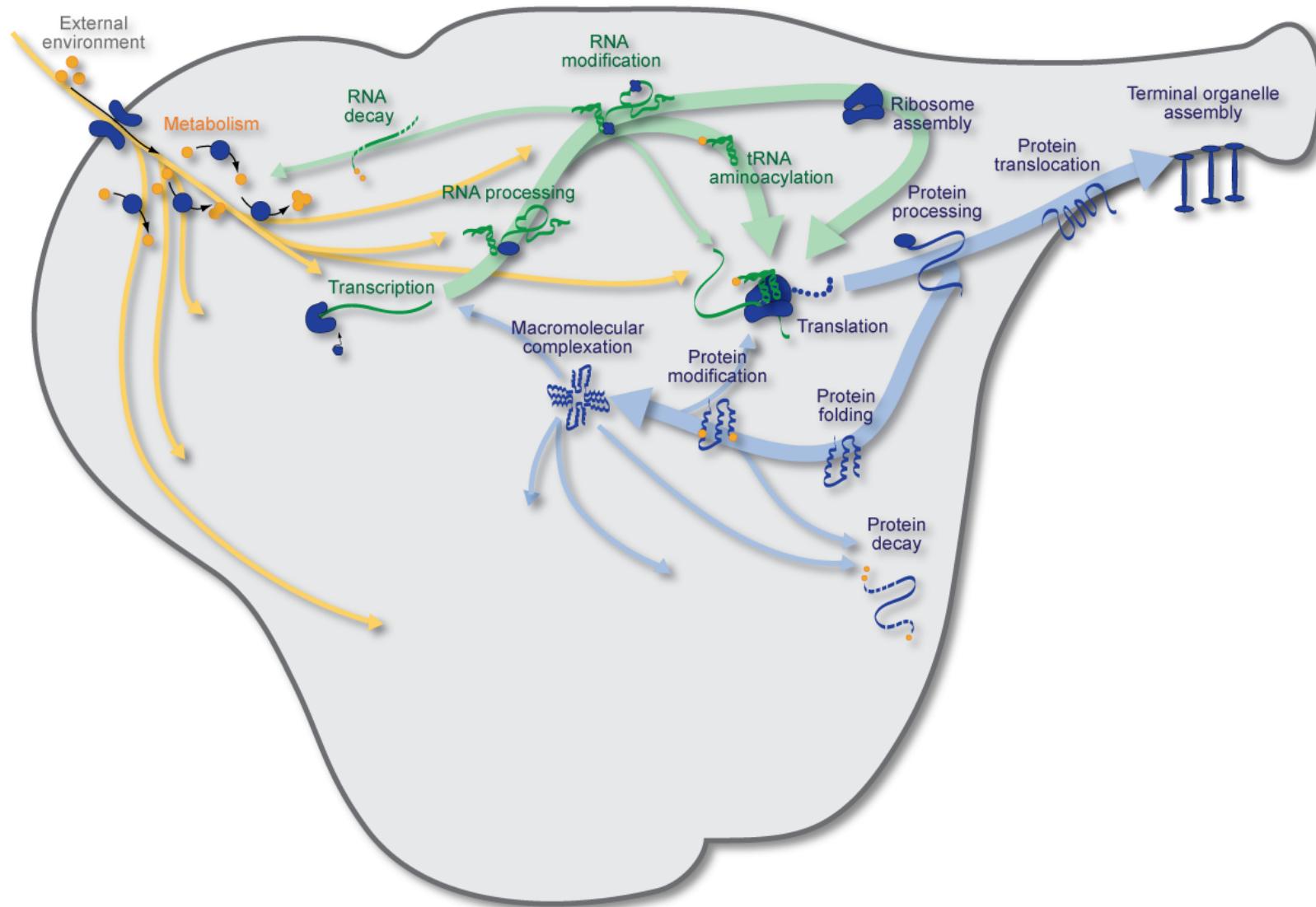
Example: drug biosynthesis



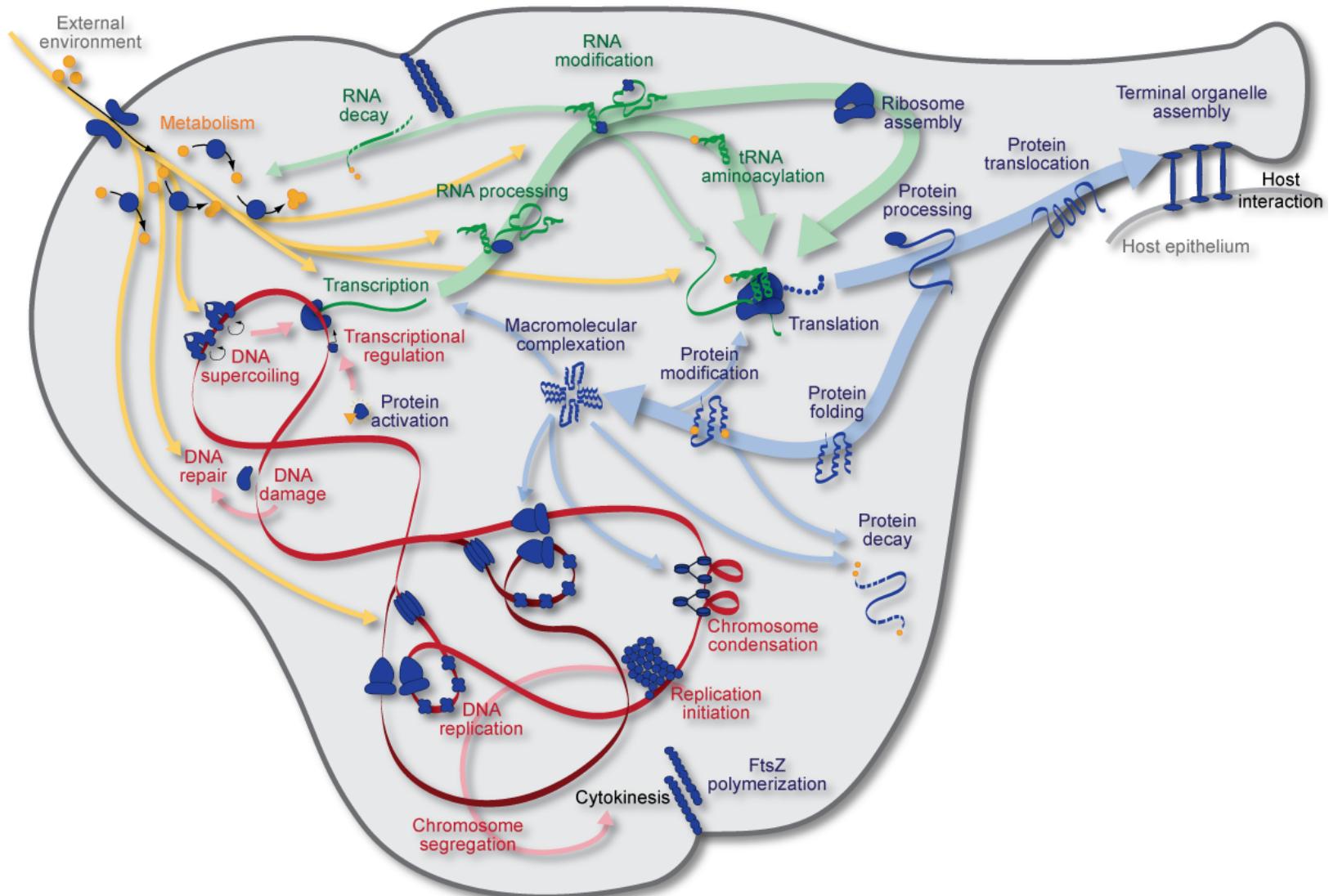
Example: drug biosynthesis



Example: drug biosynthesis

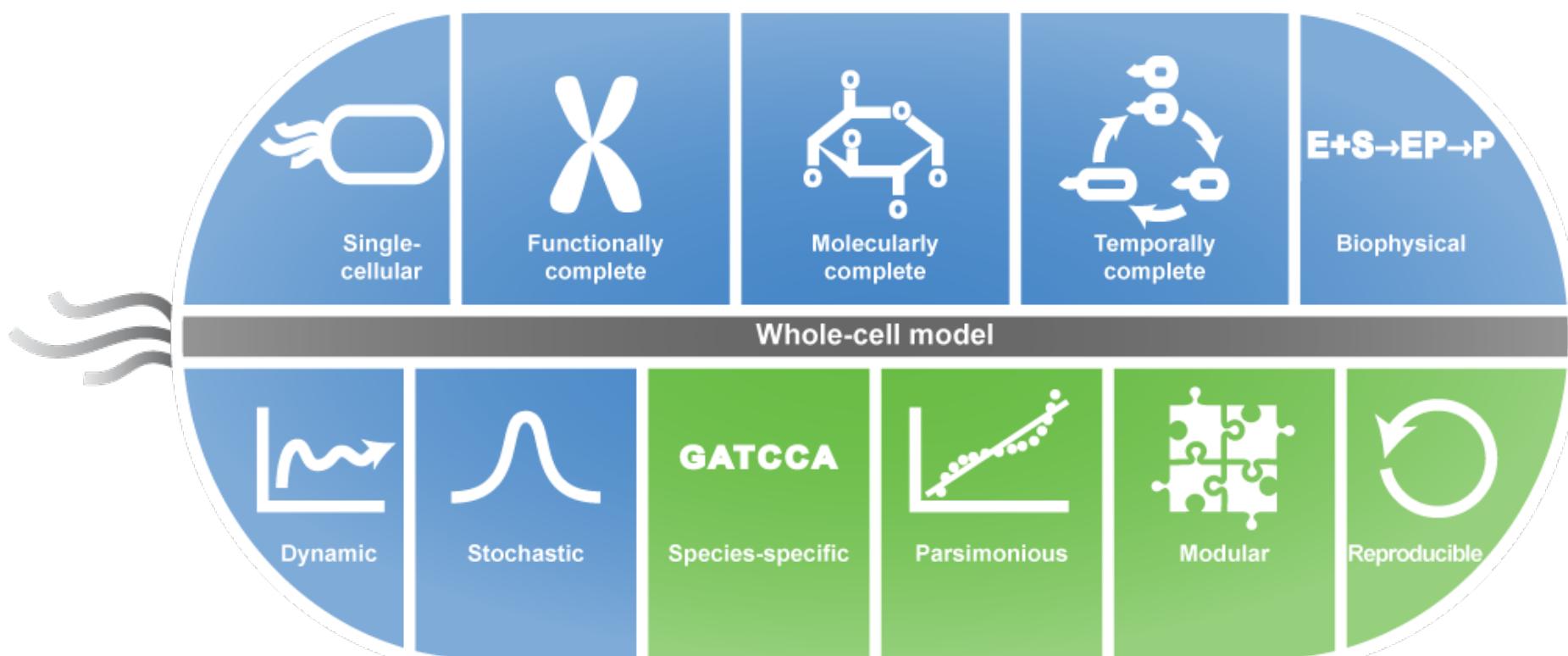


Example: drug biosynthesis

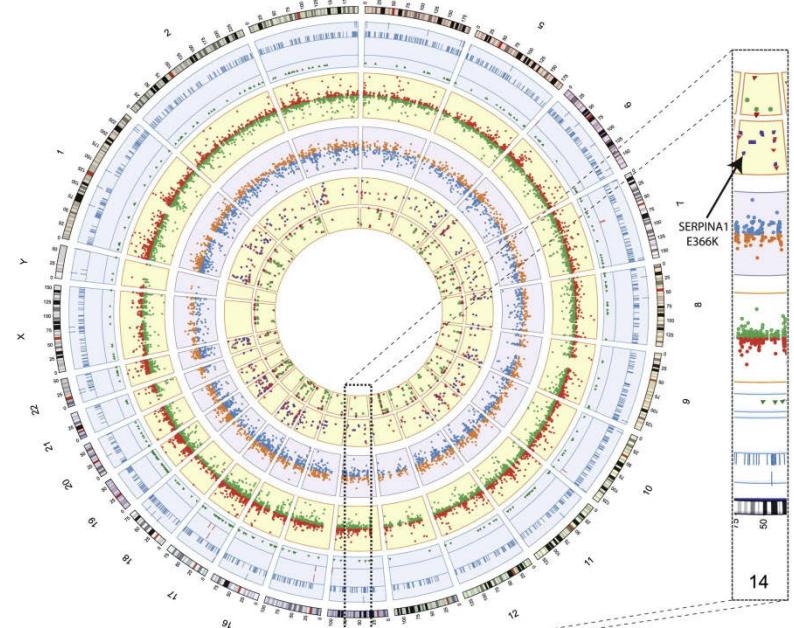


Predicting phenotype from genotype requires “whole-cell”

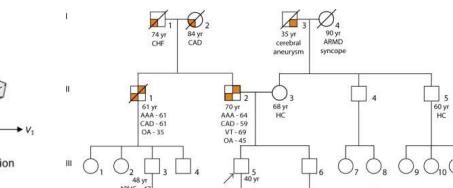
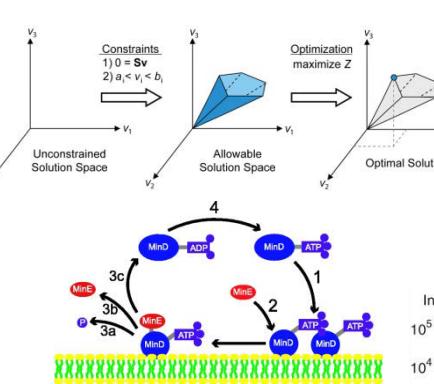
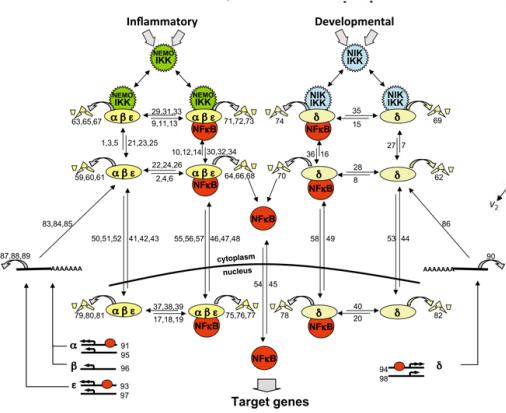
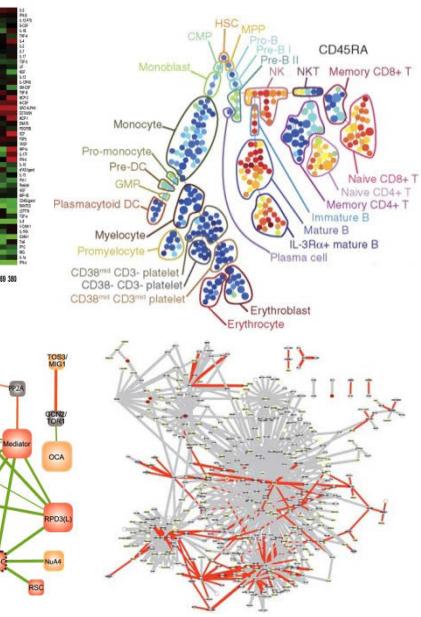
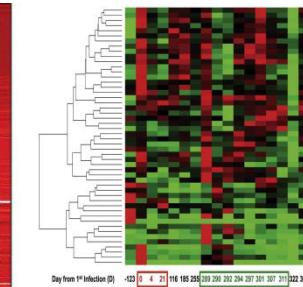
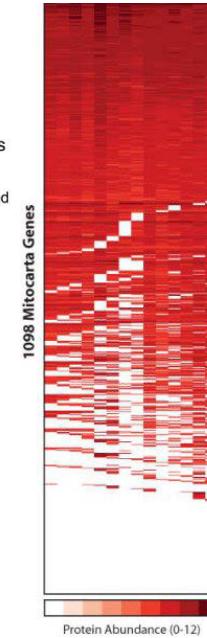
Whole-cell modeling principles



Biological data is readily available



RNA Edits
Heteroallelic SNVs
Protein-Downregulated (HRV vs Healthy)
Protein-Upregulated (HRV vs Healthy)
RNA-Downregulated (HRV vs Healthy)
RNA-Upregulated (HRV vs Healthy)
Indels
SV-Duplications
SV-Deletions
Chr. Ideogram
Chr. Number



Epic

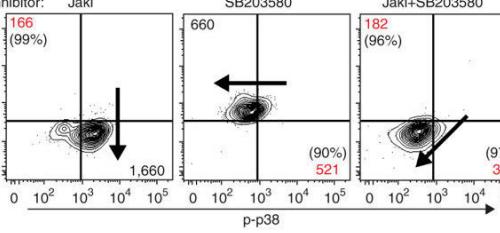
Cerner

S8ML

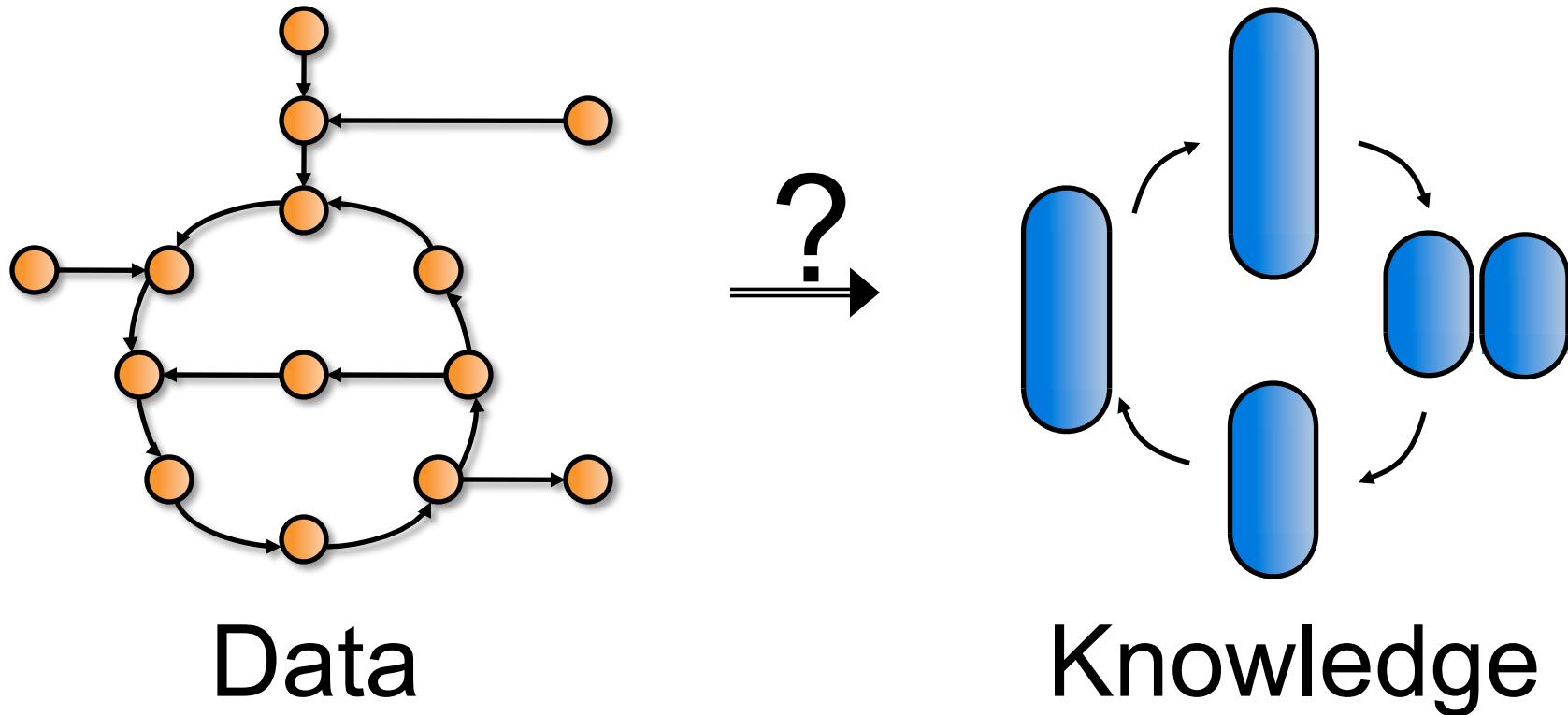
cellML

SED ML

BioModels



Whole-cell model goals



Whole-cell modeling

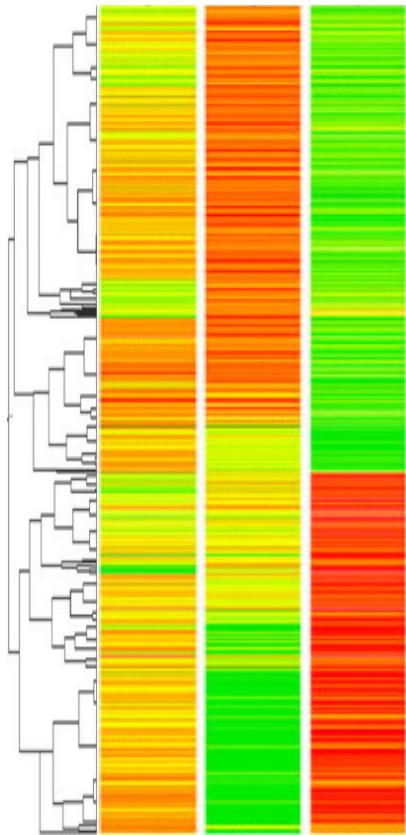
A grand challenge of the 21st century

– *Masaru Tomita*

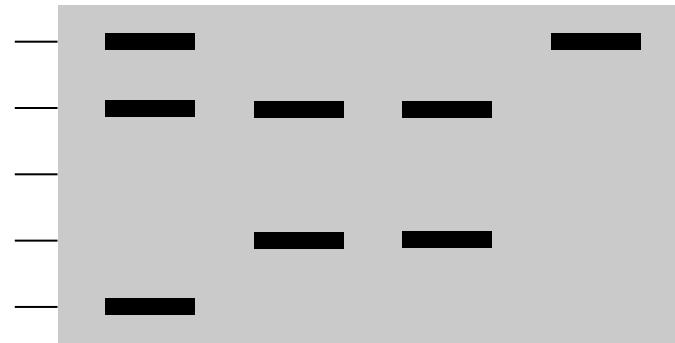
Biology urgently needs a theoretical basis to unify it

– *Sydney Brenner*

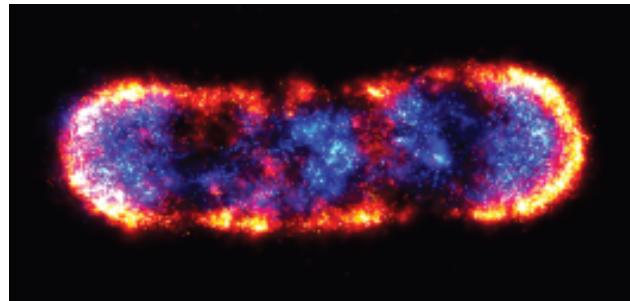
Modeling challenge: heterogeneous data



Transcription
RNA-seq

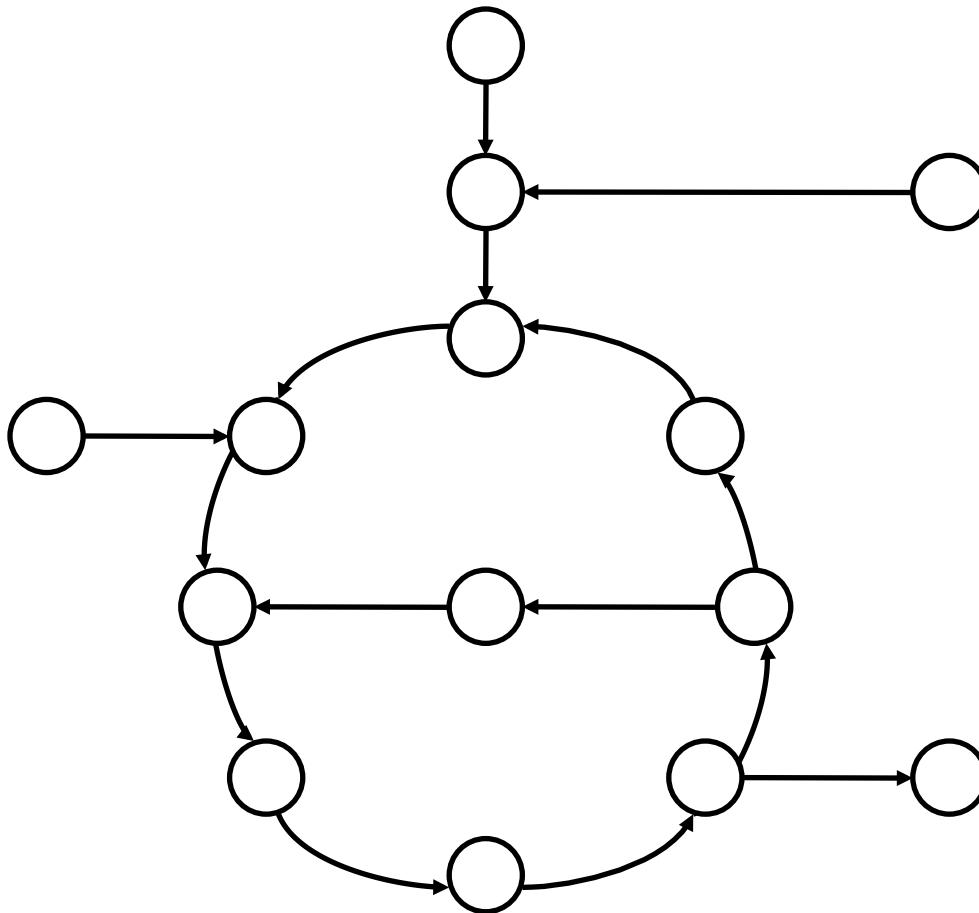


Protein expression
Mass-spec, Western blot

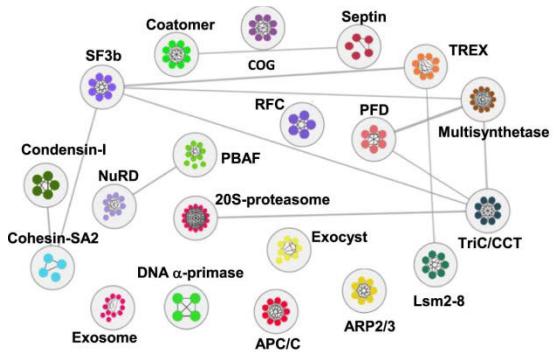
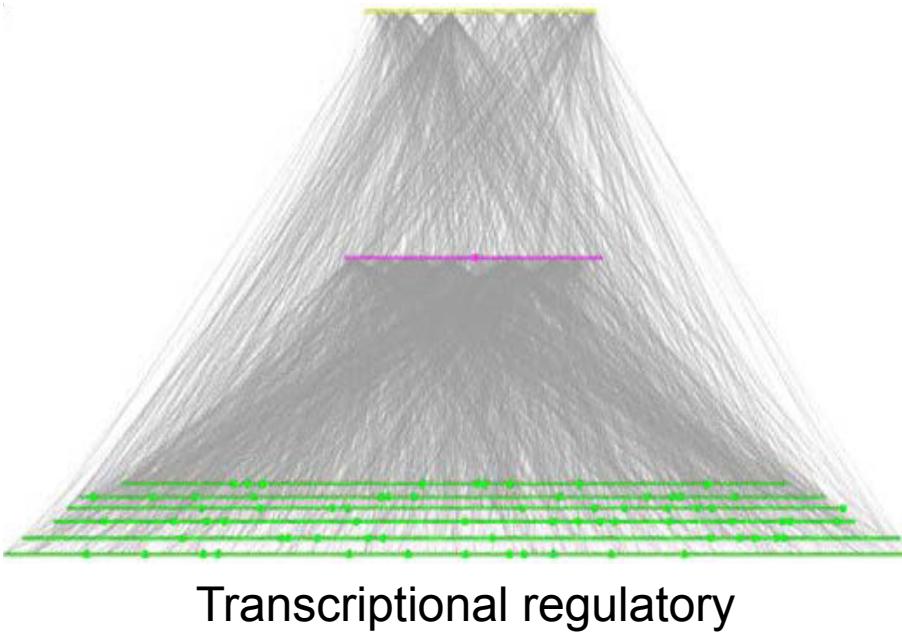


Single-cell variation
Microscopy

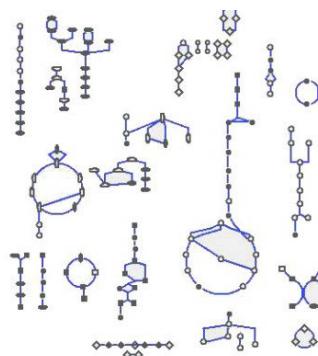
Modeling challenge: sparse data



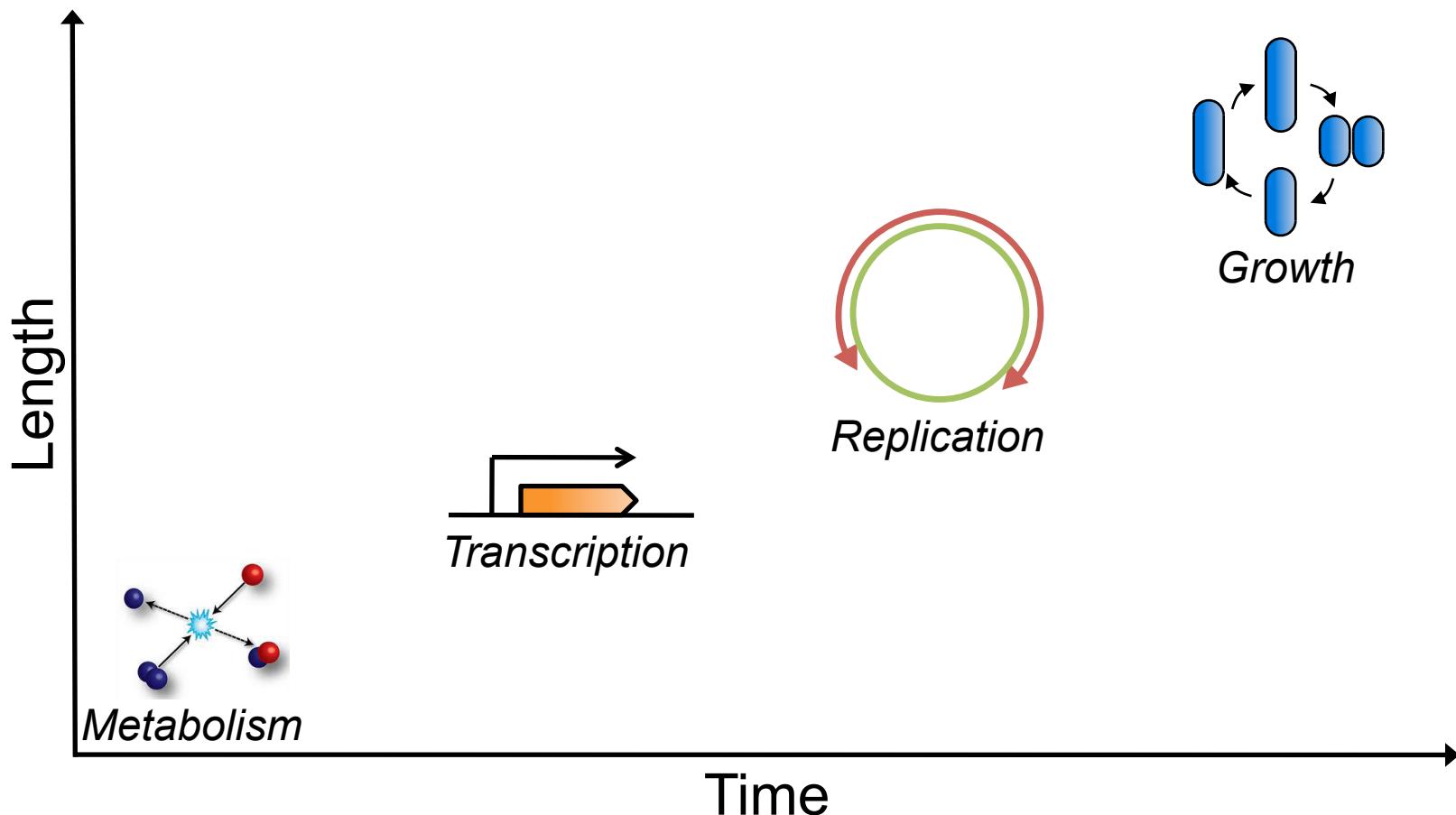
Modelling challenge: heterogenous networks



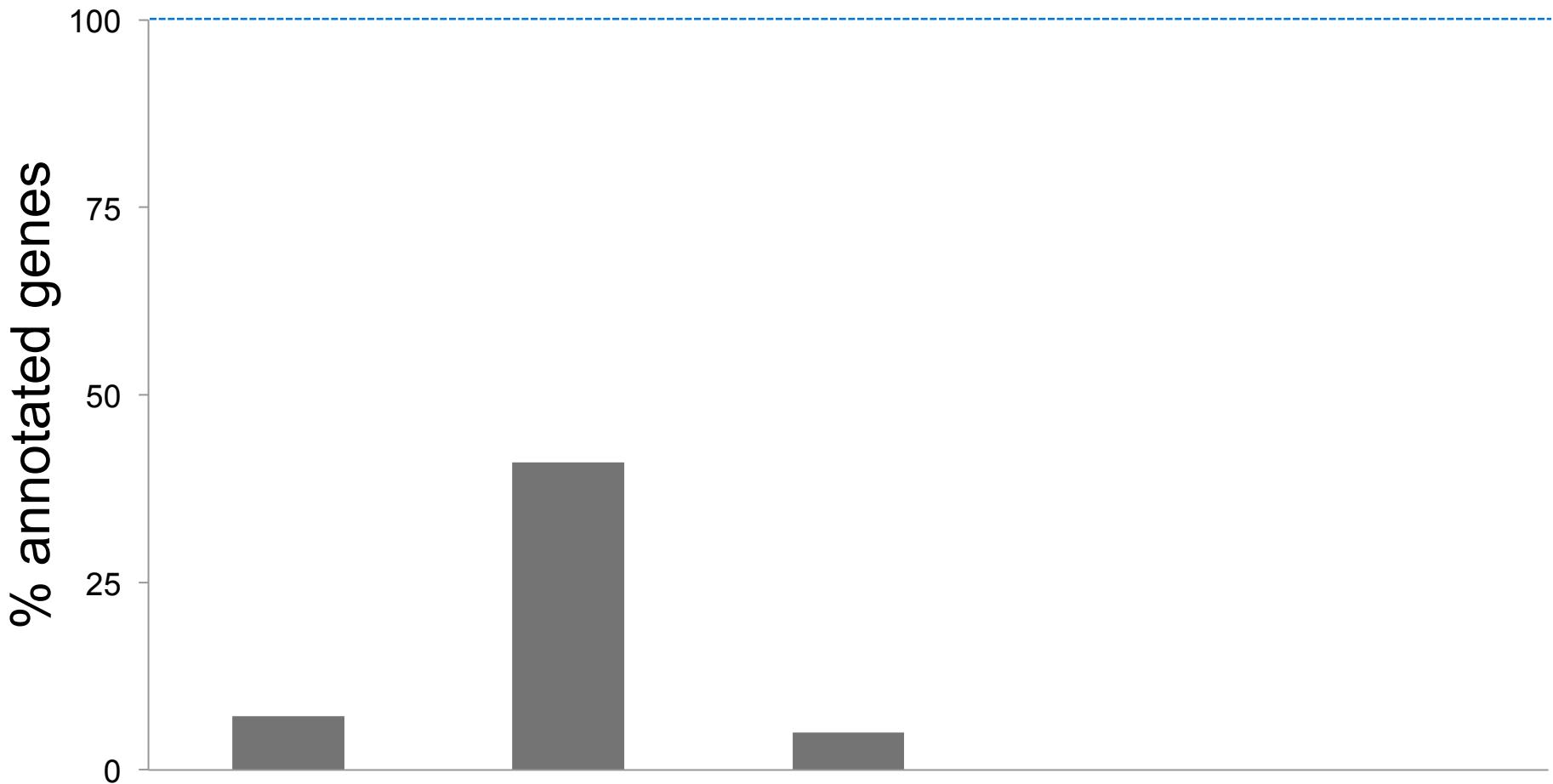
Signaling



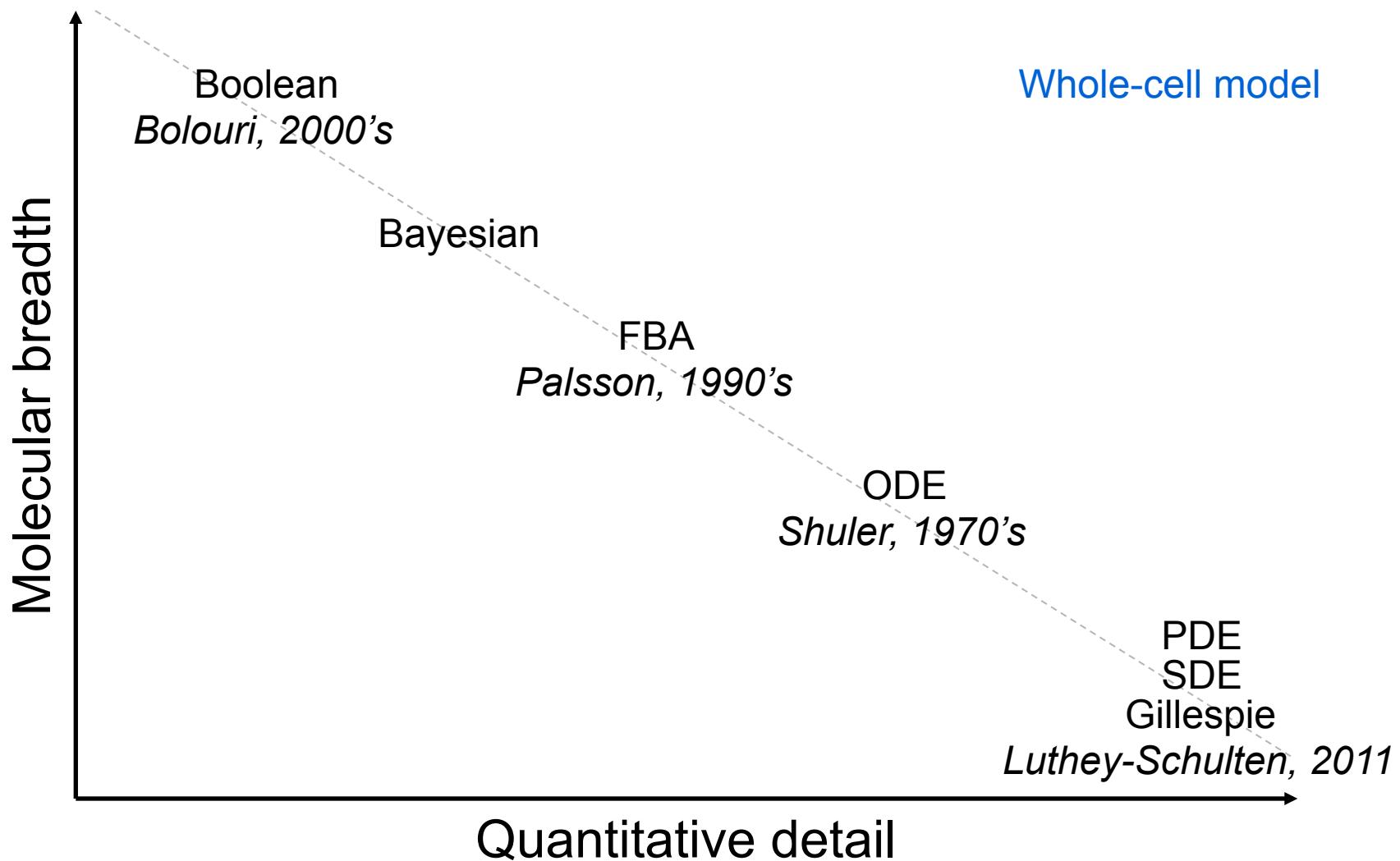
Modeling challenge: multiple time and length scales



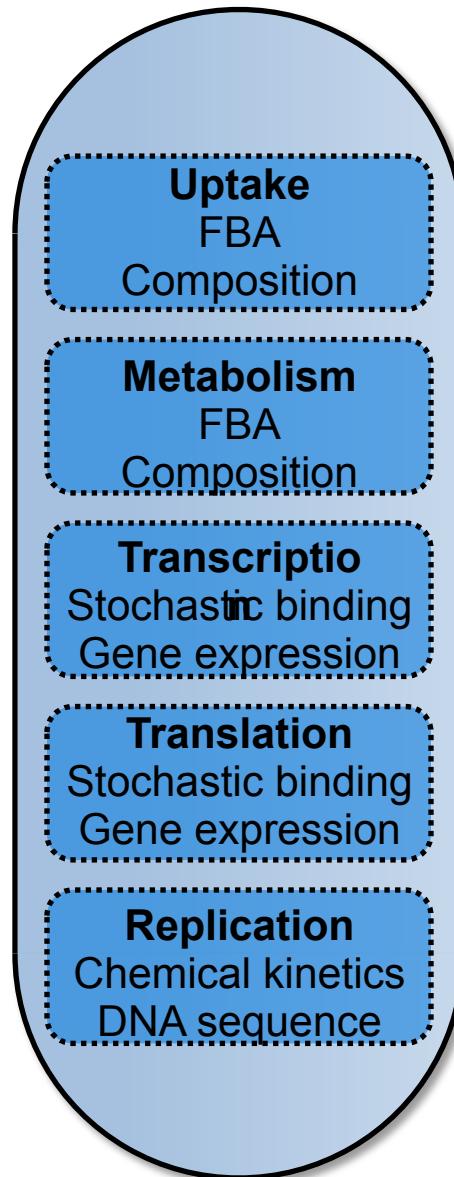
Whole-cell modeling progress



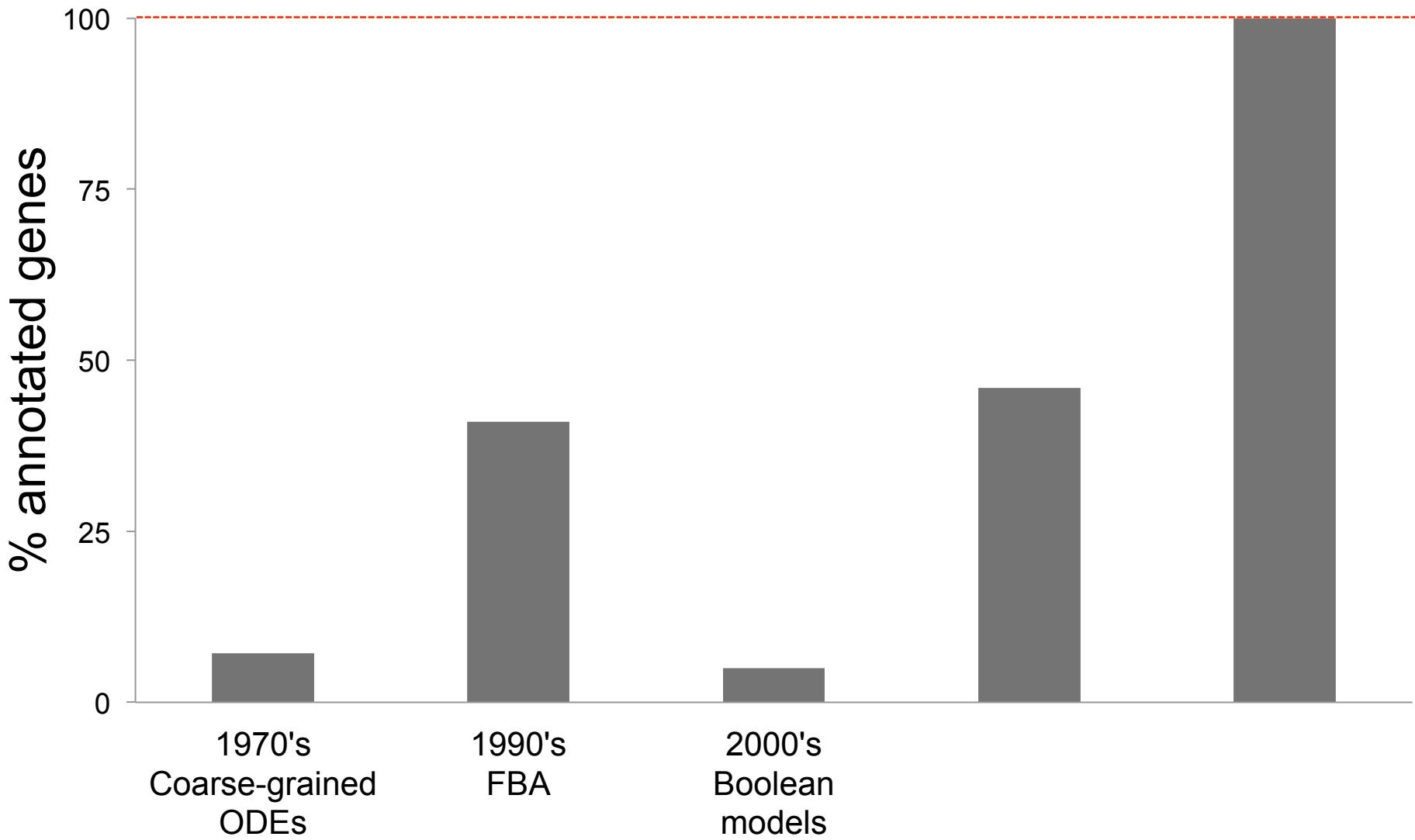
Predictive modeling methodologies



Solution: integrated models

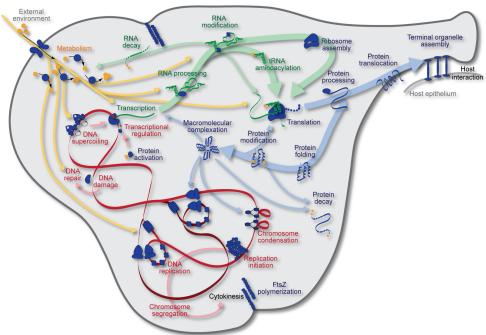


Whole-cell modeling progress

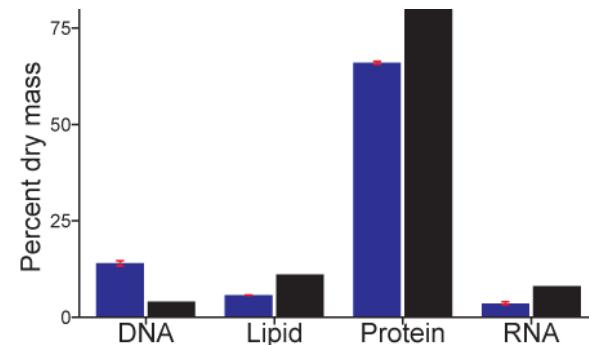


Whole-cell modeling

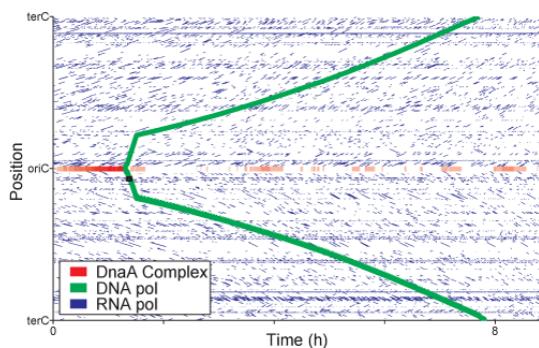
Model



Validate

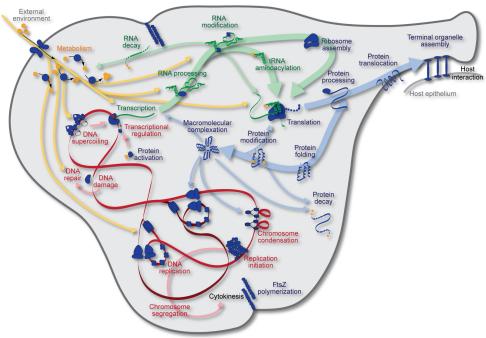


Engineer

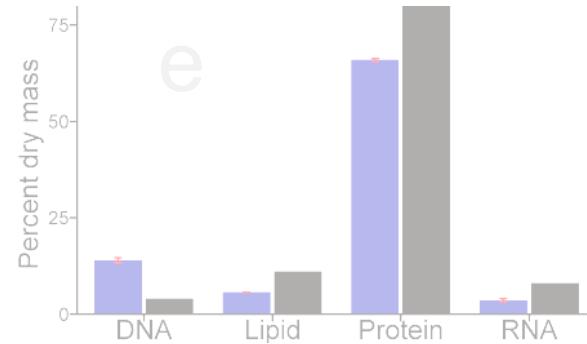


Whole-cell modeling

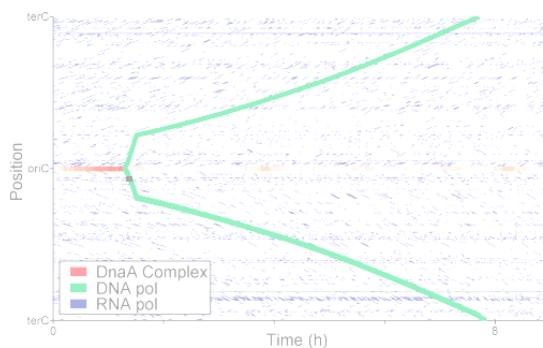
Model



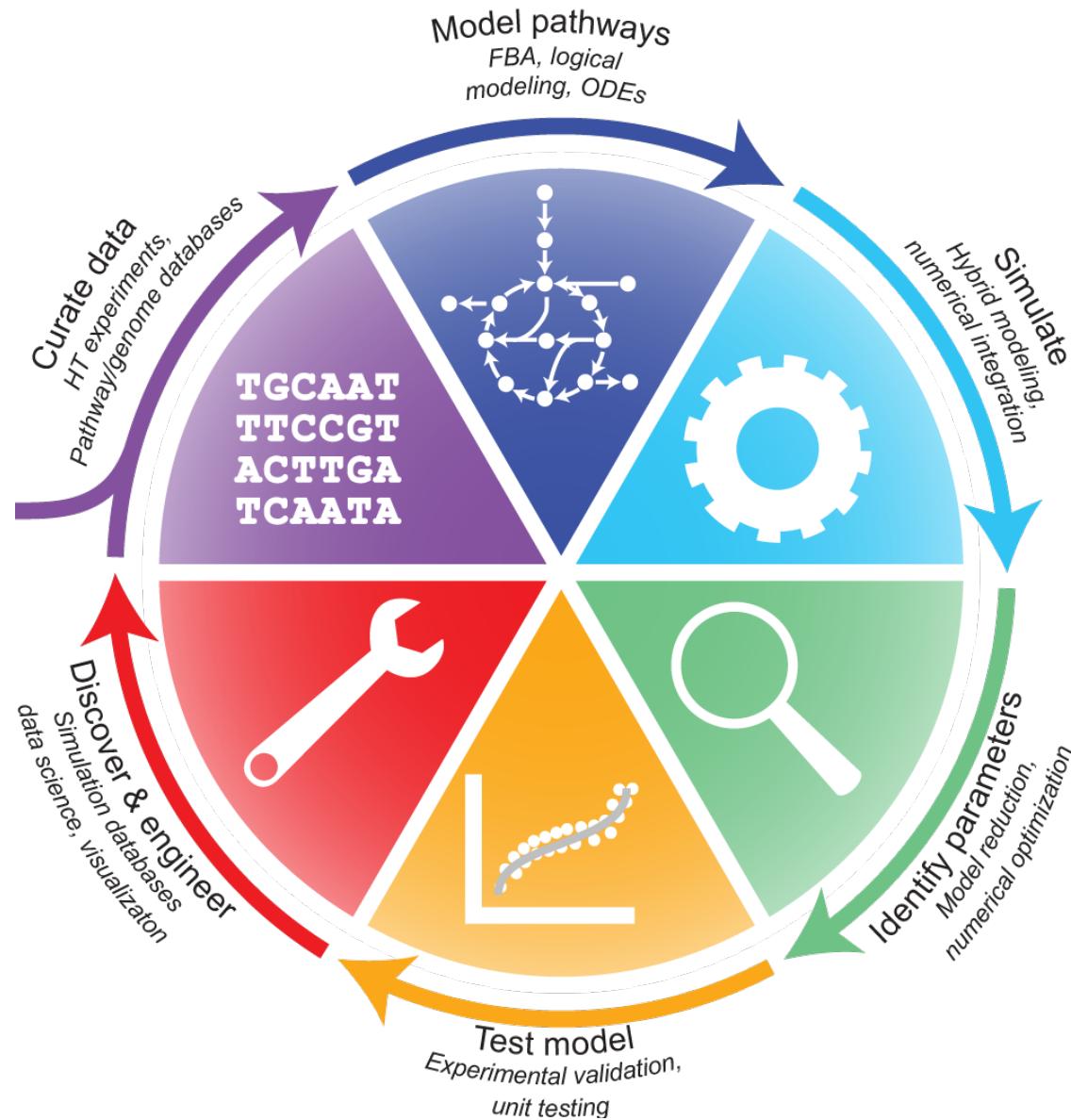
Validate



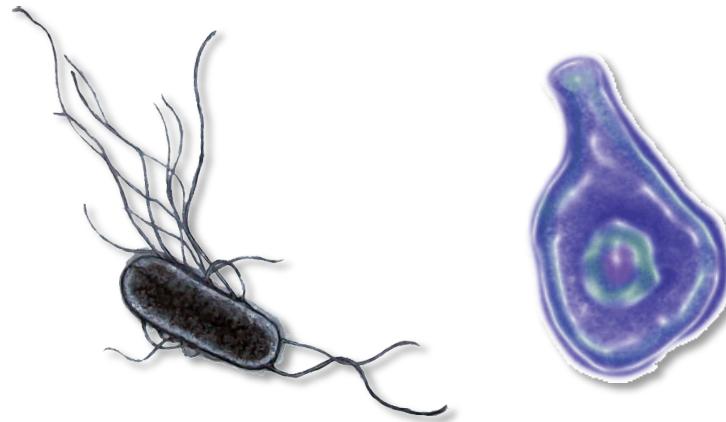
Engineer



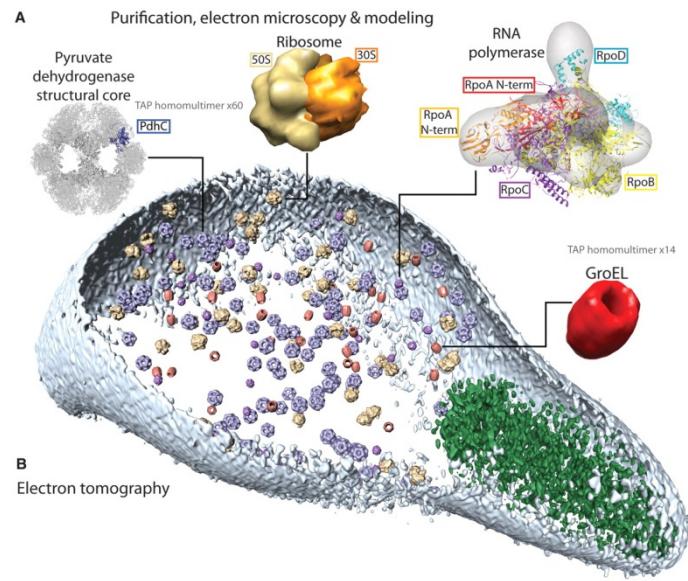
Model construction



Mycoplasma genitalium is a tractable organism

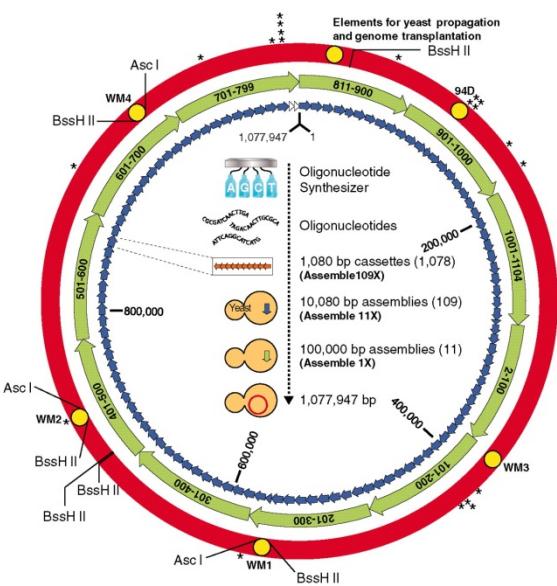


M. genitalium is well-characterized

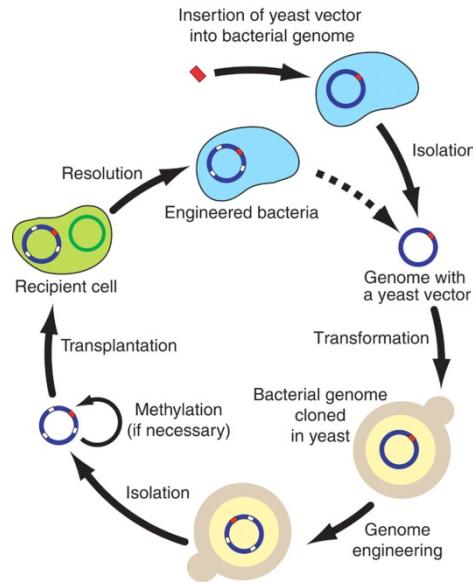


Genomic-scale data
Kühner et. al, 2009

M. genitalium has unique engineering tools

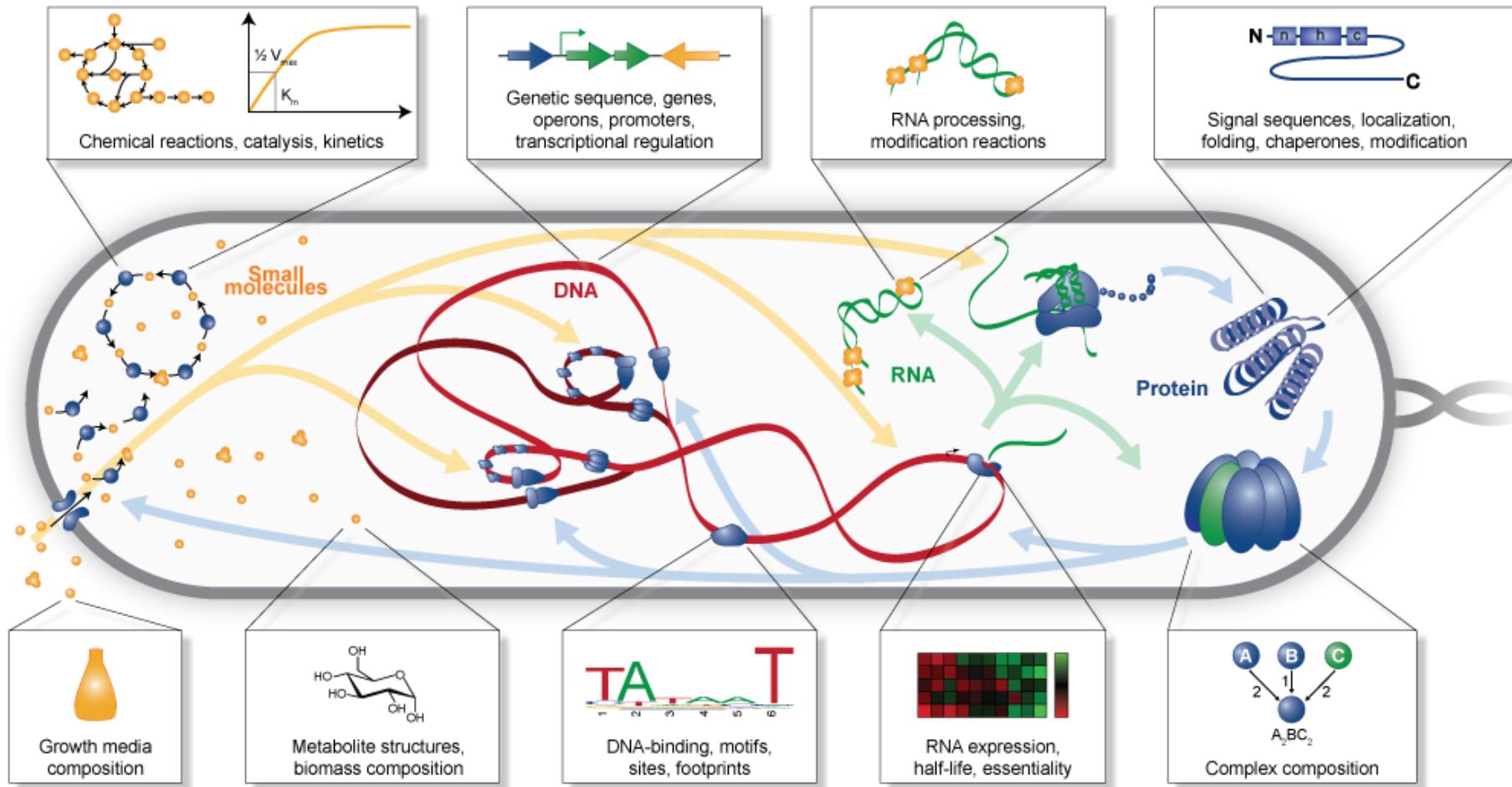


Genomic synthesis
Gibson et. al, 2009

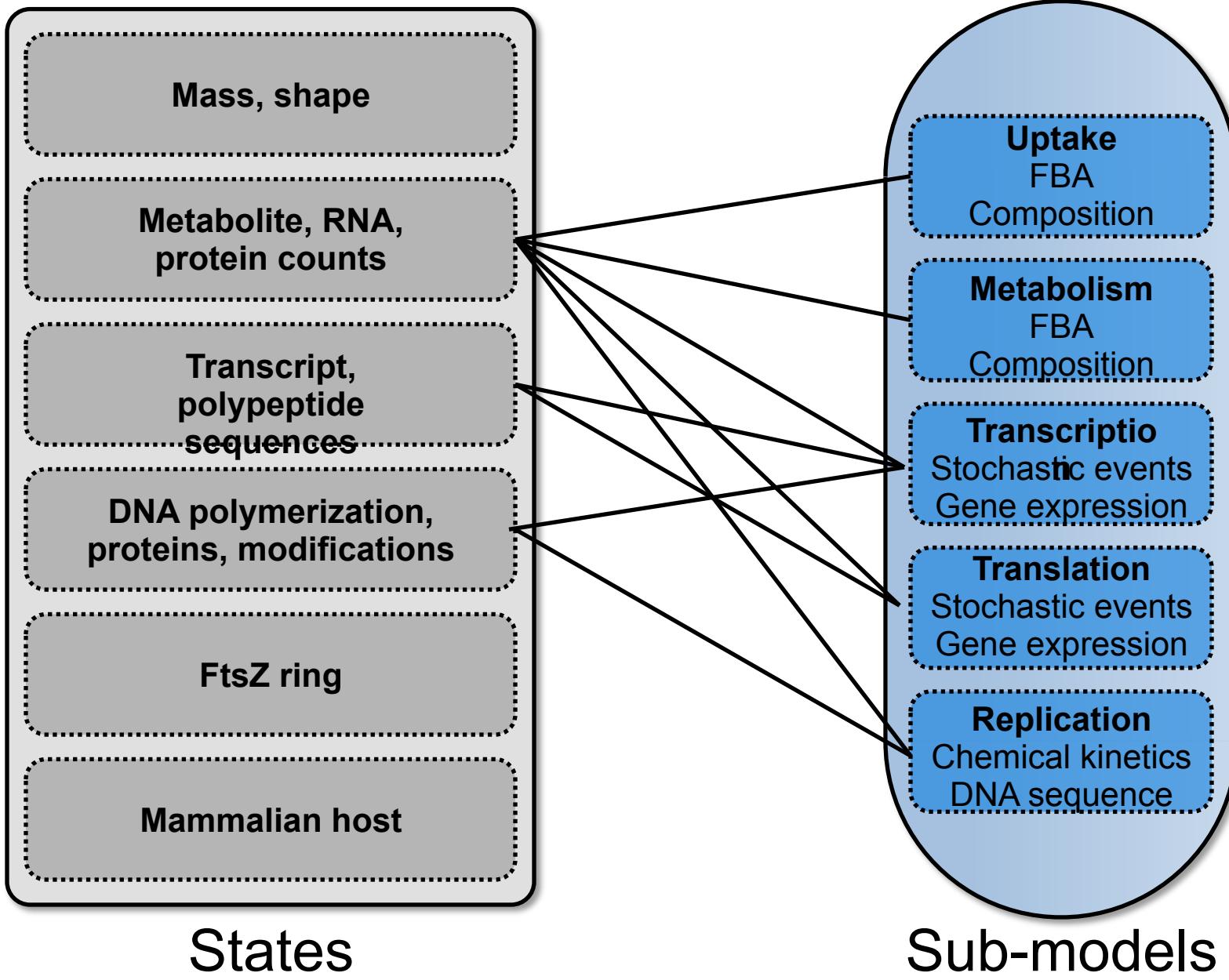


Genomic transplantation
Lartigue et. al, 2009

Broadly curate experimental data



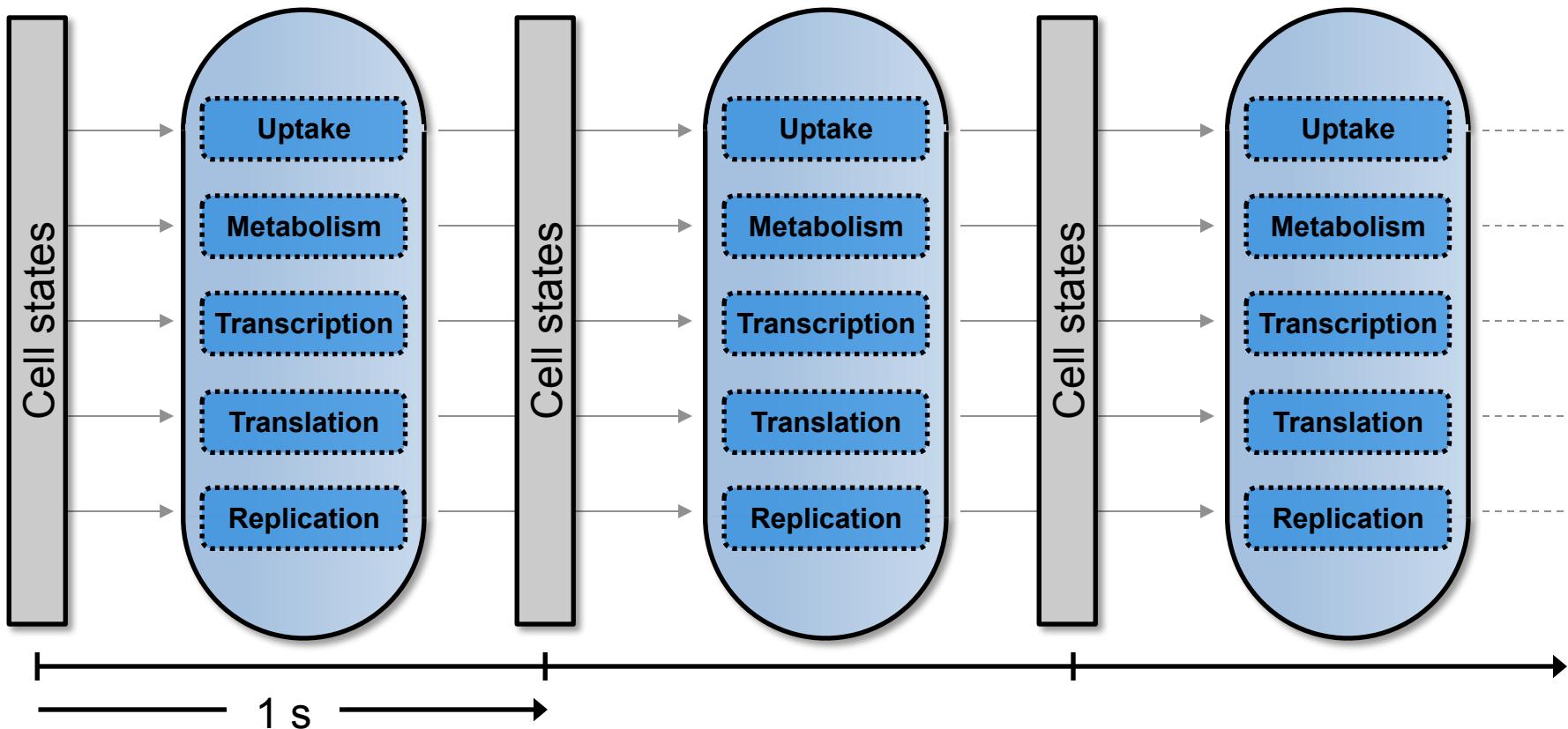
Multi-algorithm modeling



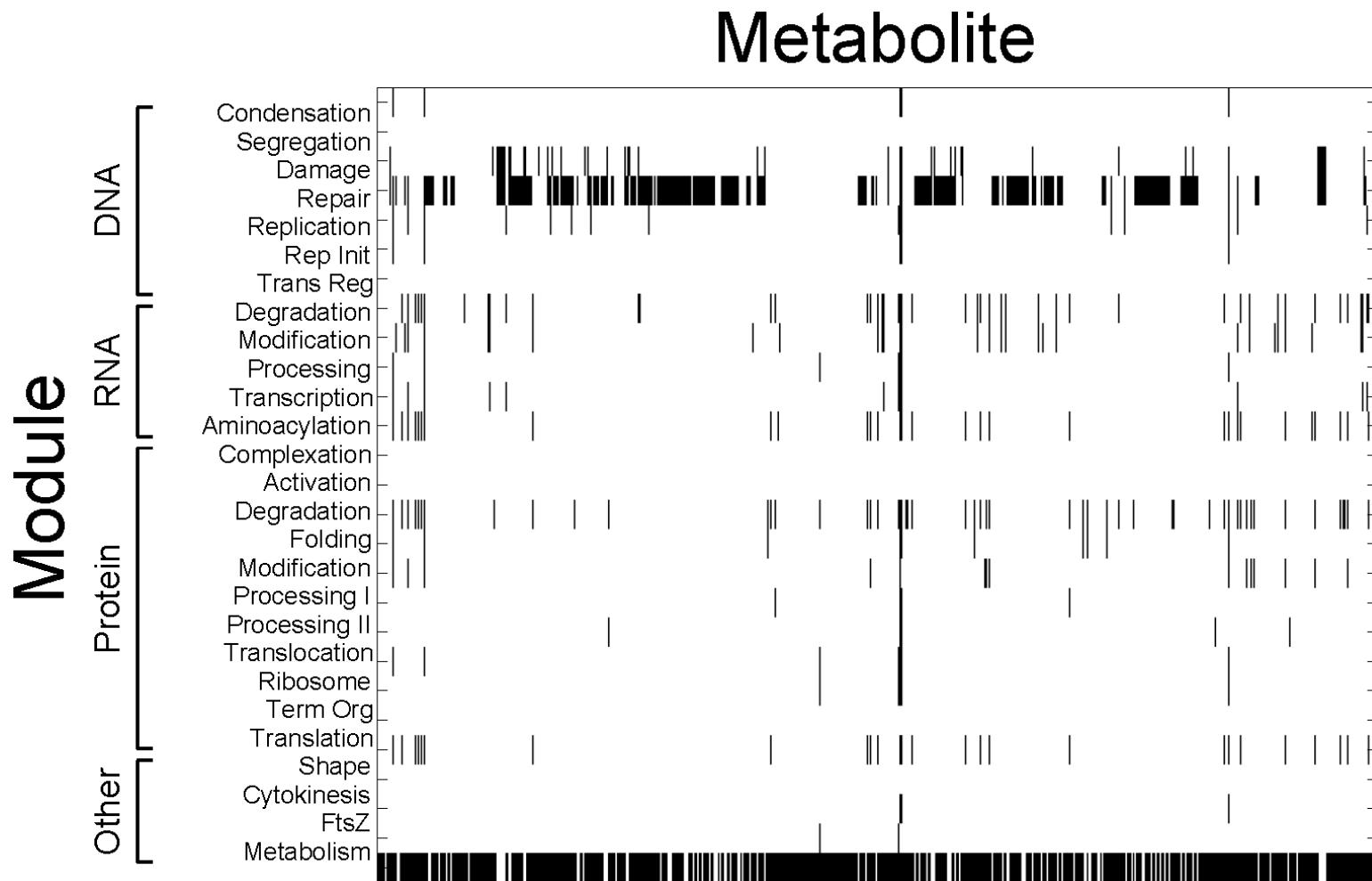
States

Sub-models

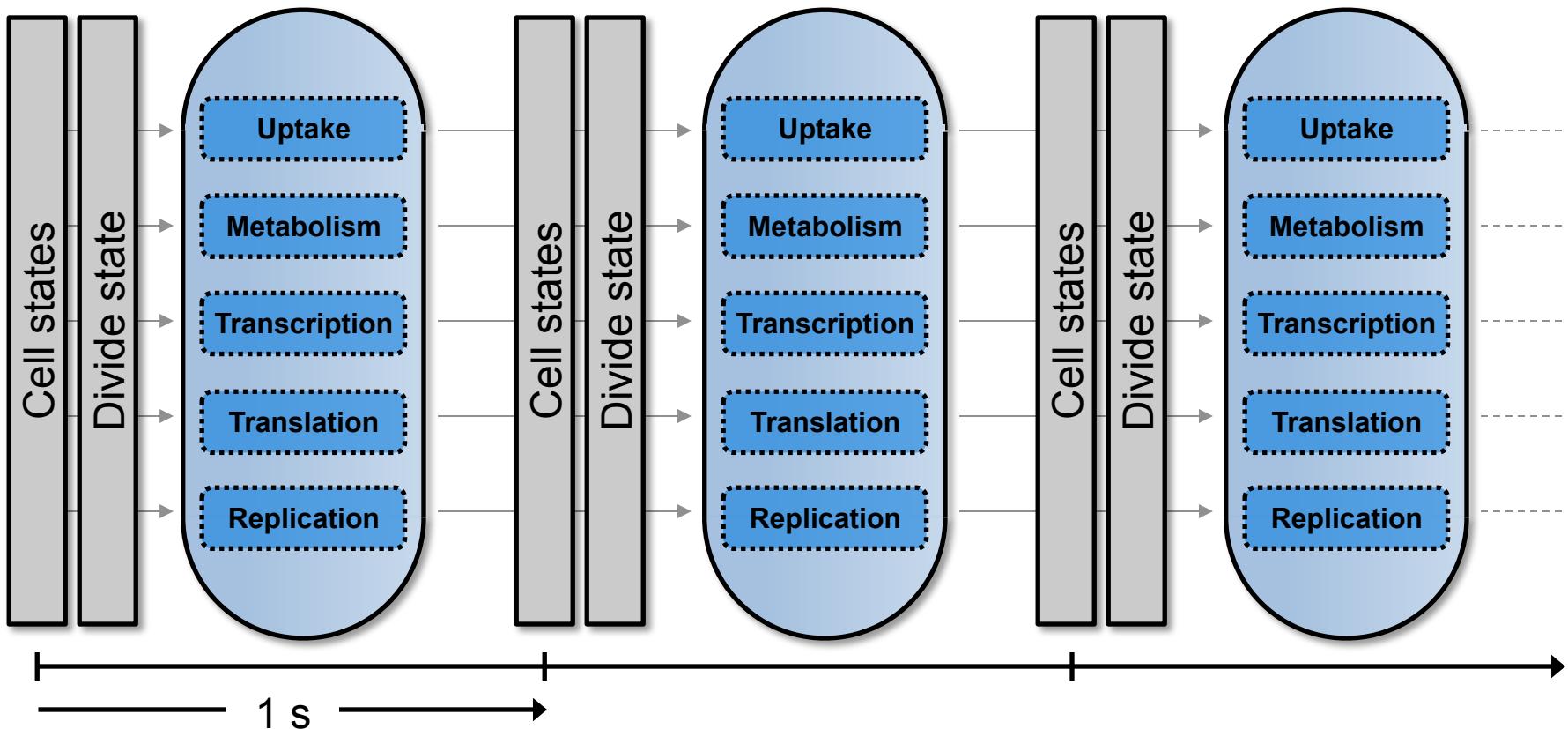
Simulation algorithm



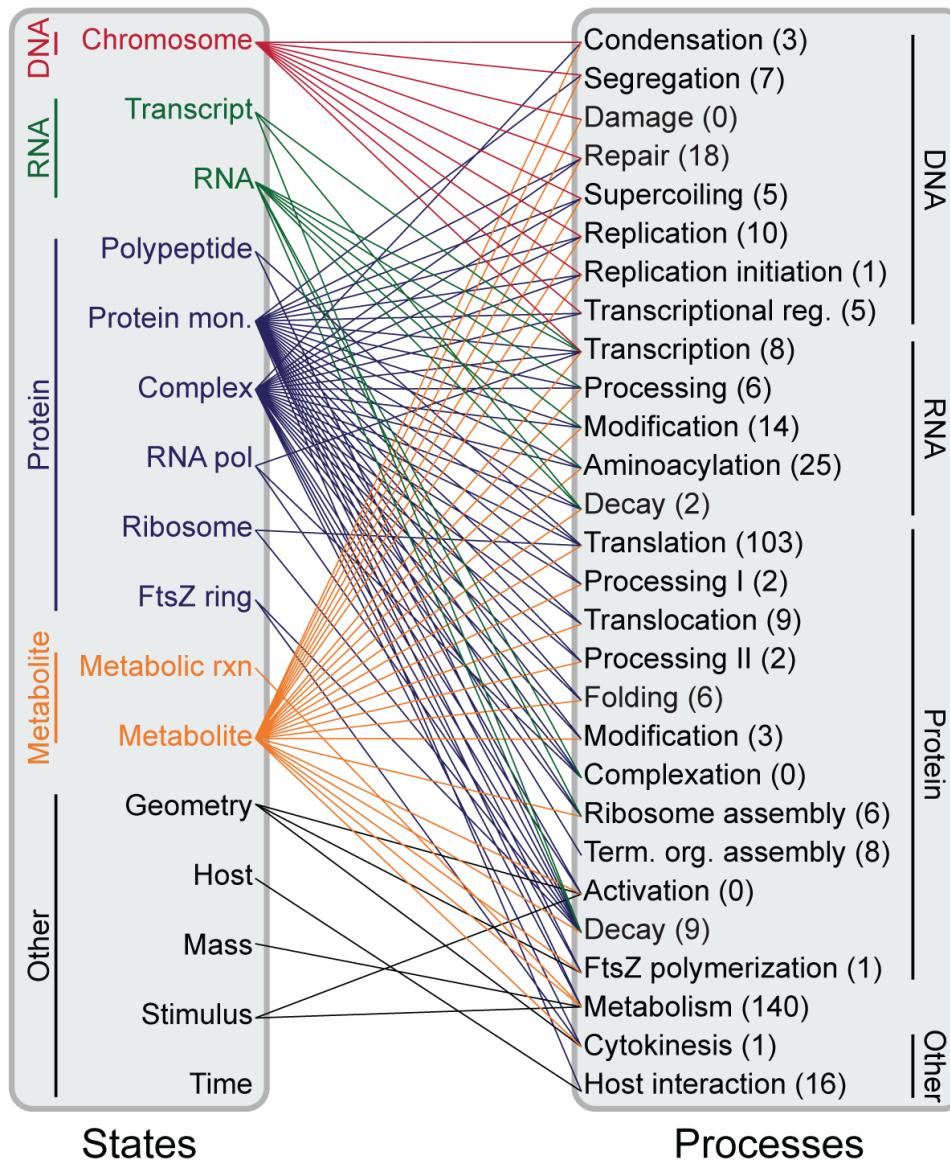
Many resources are shared



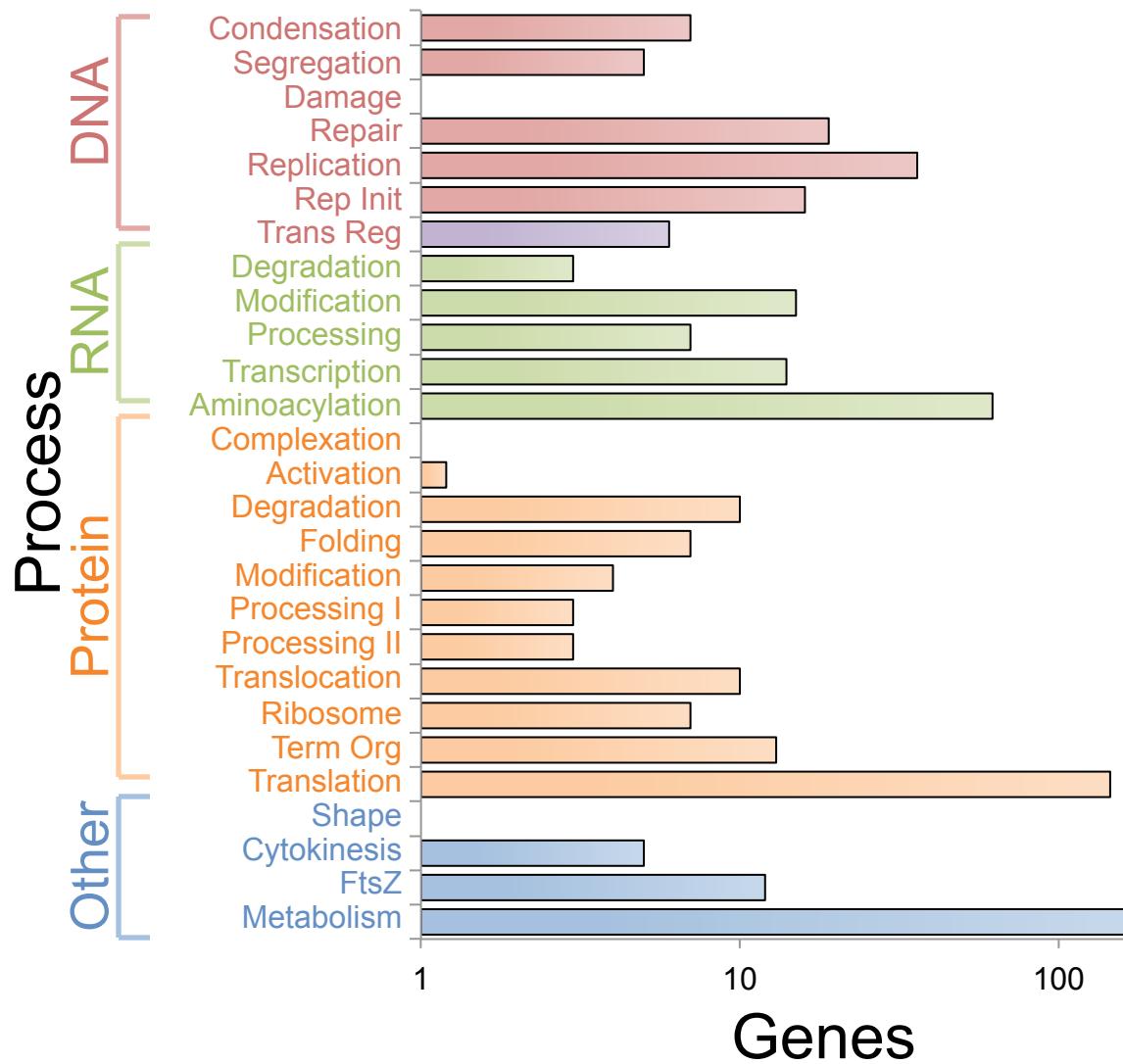
Simulation algorithm



Mycoplasma model contains 28 sub-models



Model accounts for 76% of genes



5. Identify parameters

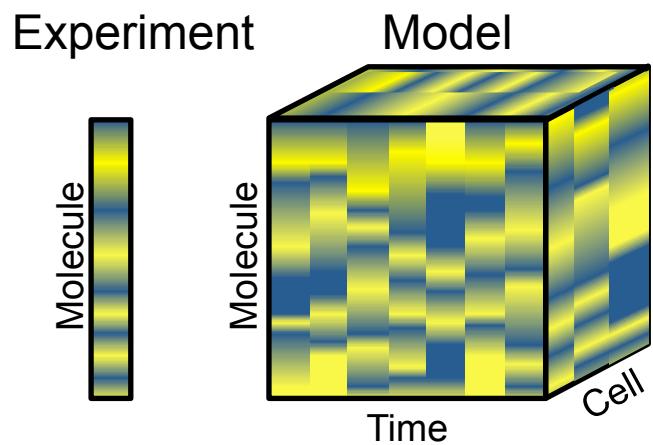
- Compare the model's predictions to data,
 $y \downarrow i$
- Define an error metric
- Numerically minimize error

5. Identify parameters

- Large parameter space
- Stochastic model
- Large computational cost
- Heterogeneous data
- Little dynamic, single cell data

Model reduction enables parameter identification

1. Reduce model



2. Identify reduced model parameters using traditional methods

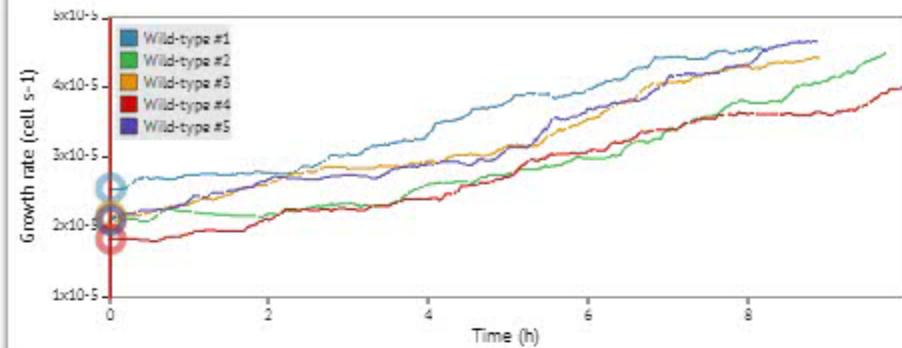
3. Manually tune parameters using full model

Model reduction

(Model selection, regularization, surrogate modeling, projection)

- Most cell biological data is time and population averaged
- Dynamics and single-cell variability is not used to calculate error metric
- Replace full model with a less computationally intensive approximate model which directly computes the time and population average
- Numerically minimize approximate error function
- Plug result into full model, simulate, and evaluate error

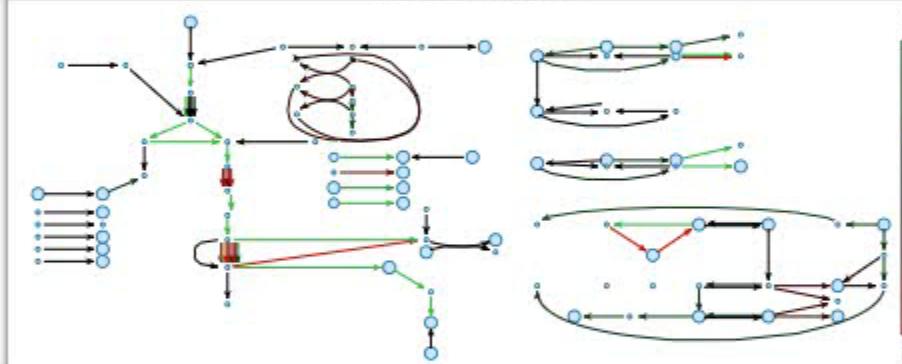
Growth rate
Simulation: multiple selected



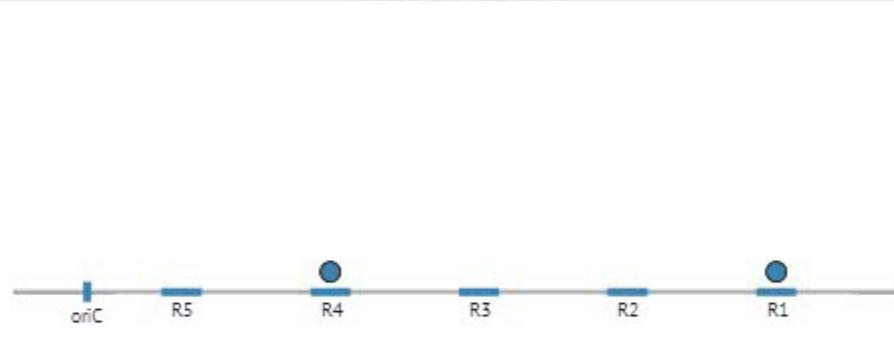
Cell shape
Simulation: Wild-type #1



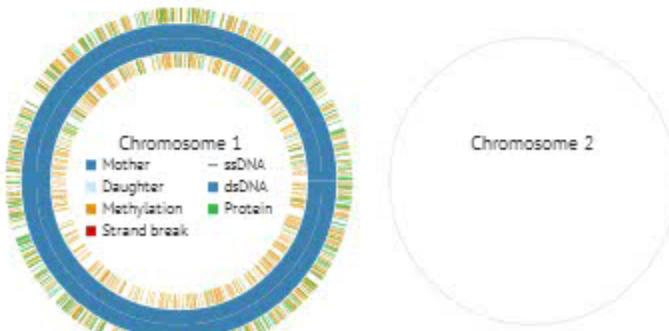
Metabolism
Simulation: Wild-type #1



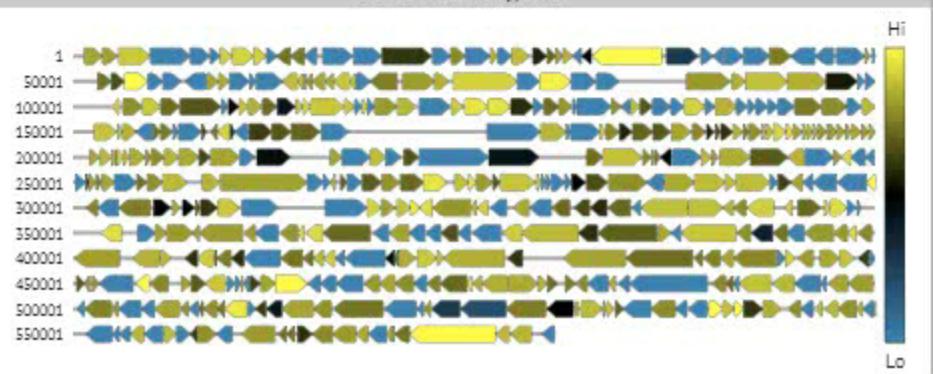
Replication initiation – oriC DnaA Boxes
Simulation: Wild-type #1



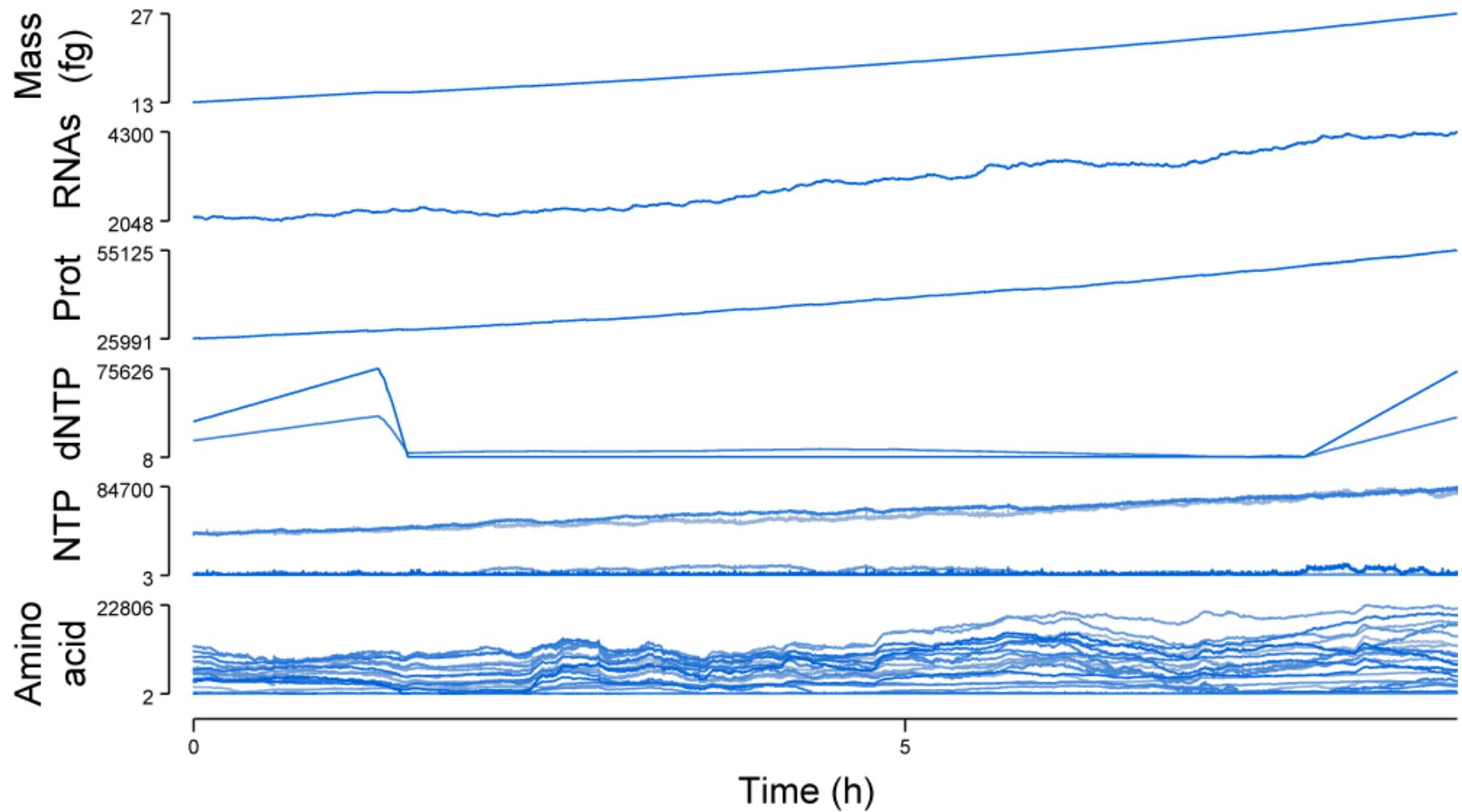
DNA replication, protein occupancy, methylation, & damage
Simulation: Wild-type #1



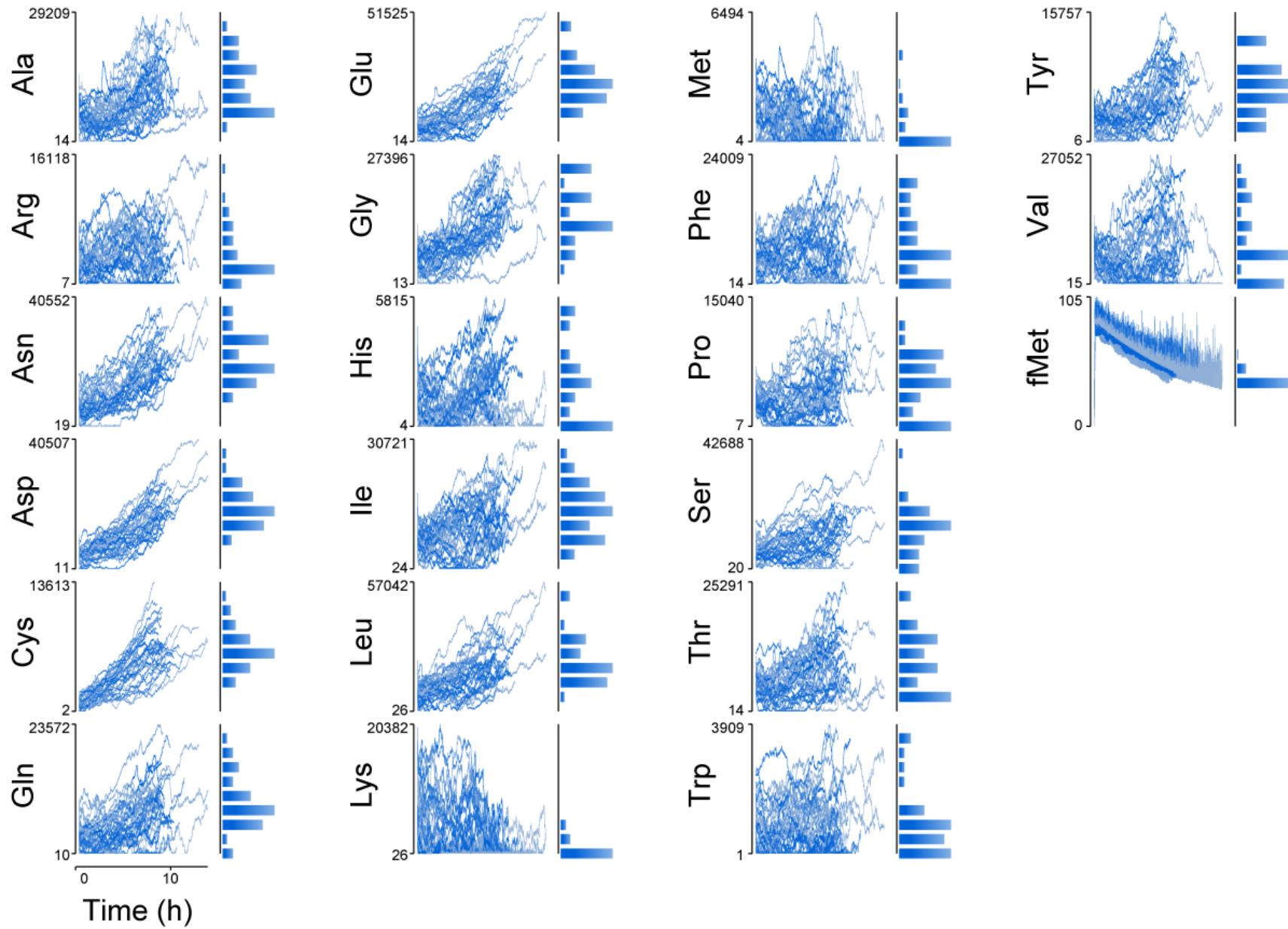
Mature protein monomer expression
Simulation: Wild-type #1



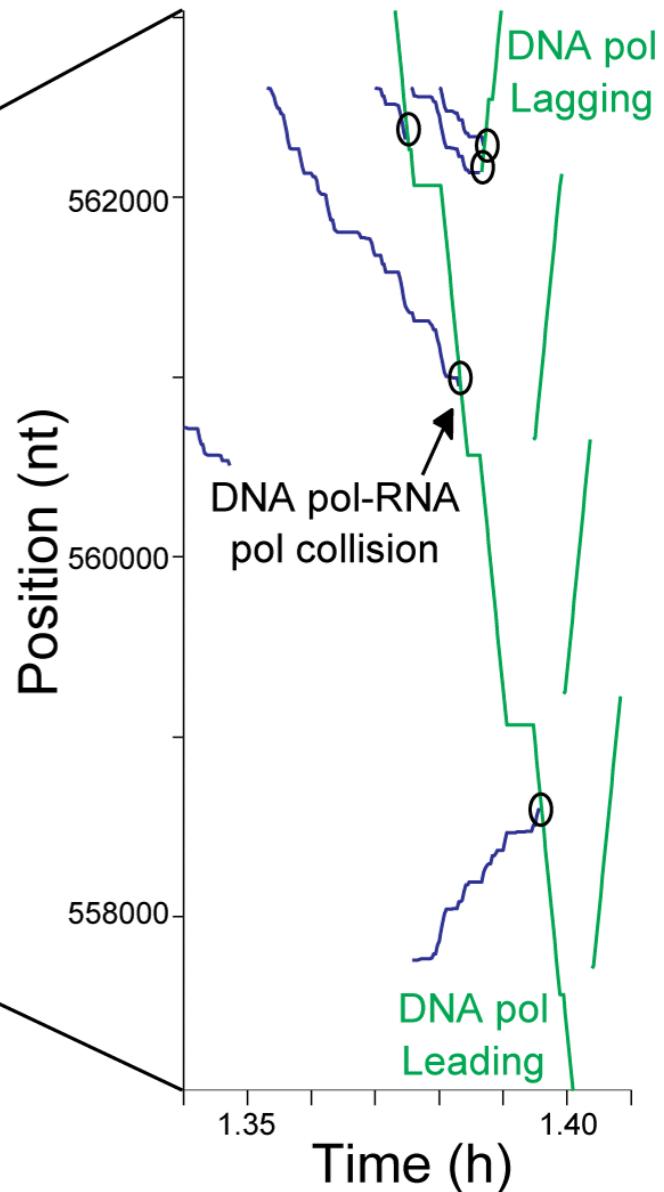
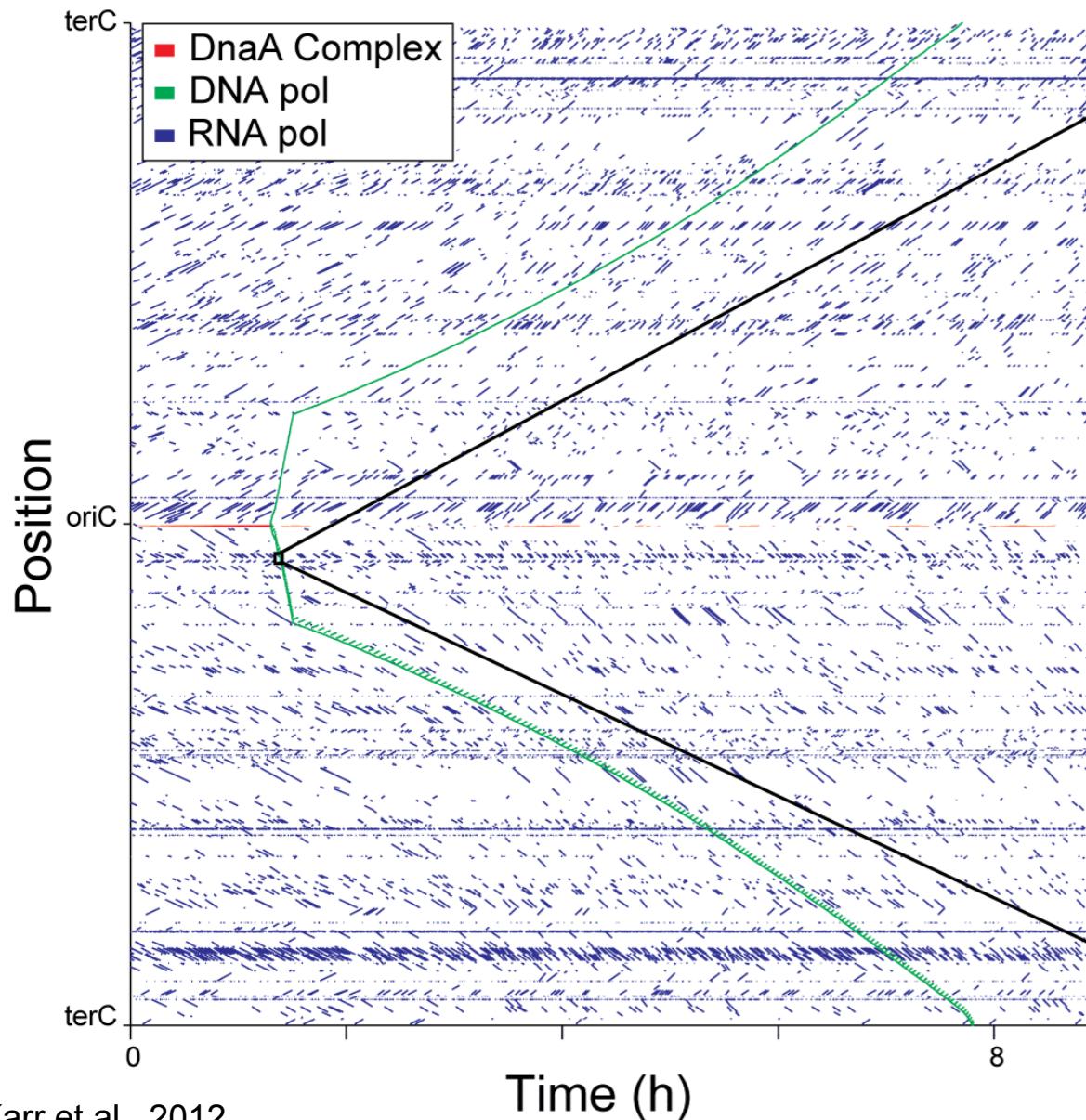
Cellular composition



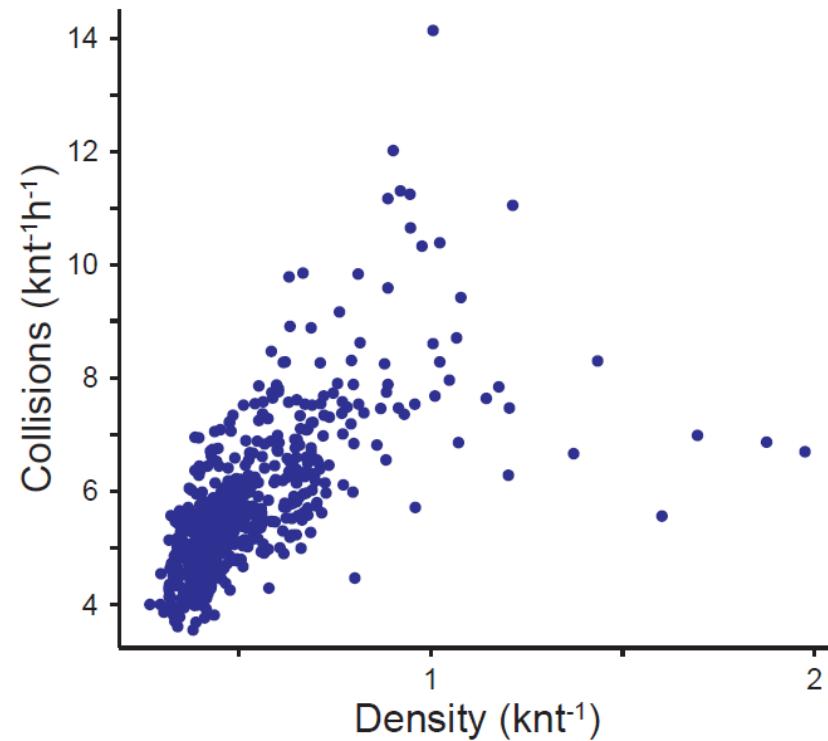
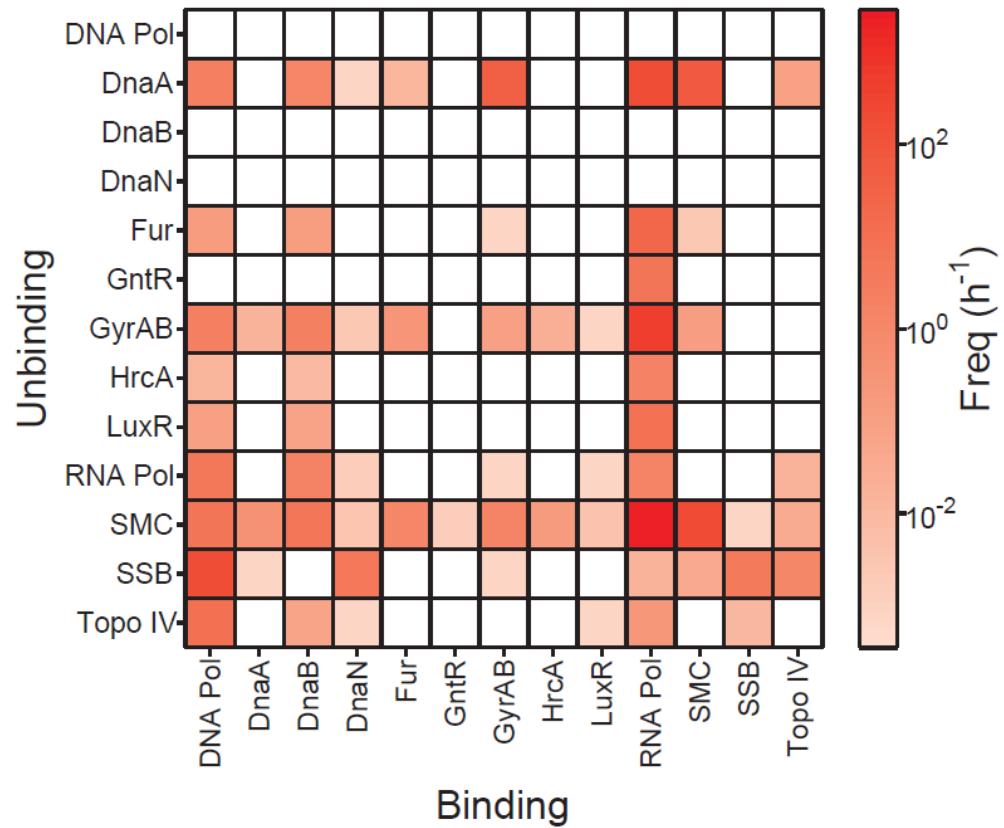
Metabolite concentrations



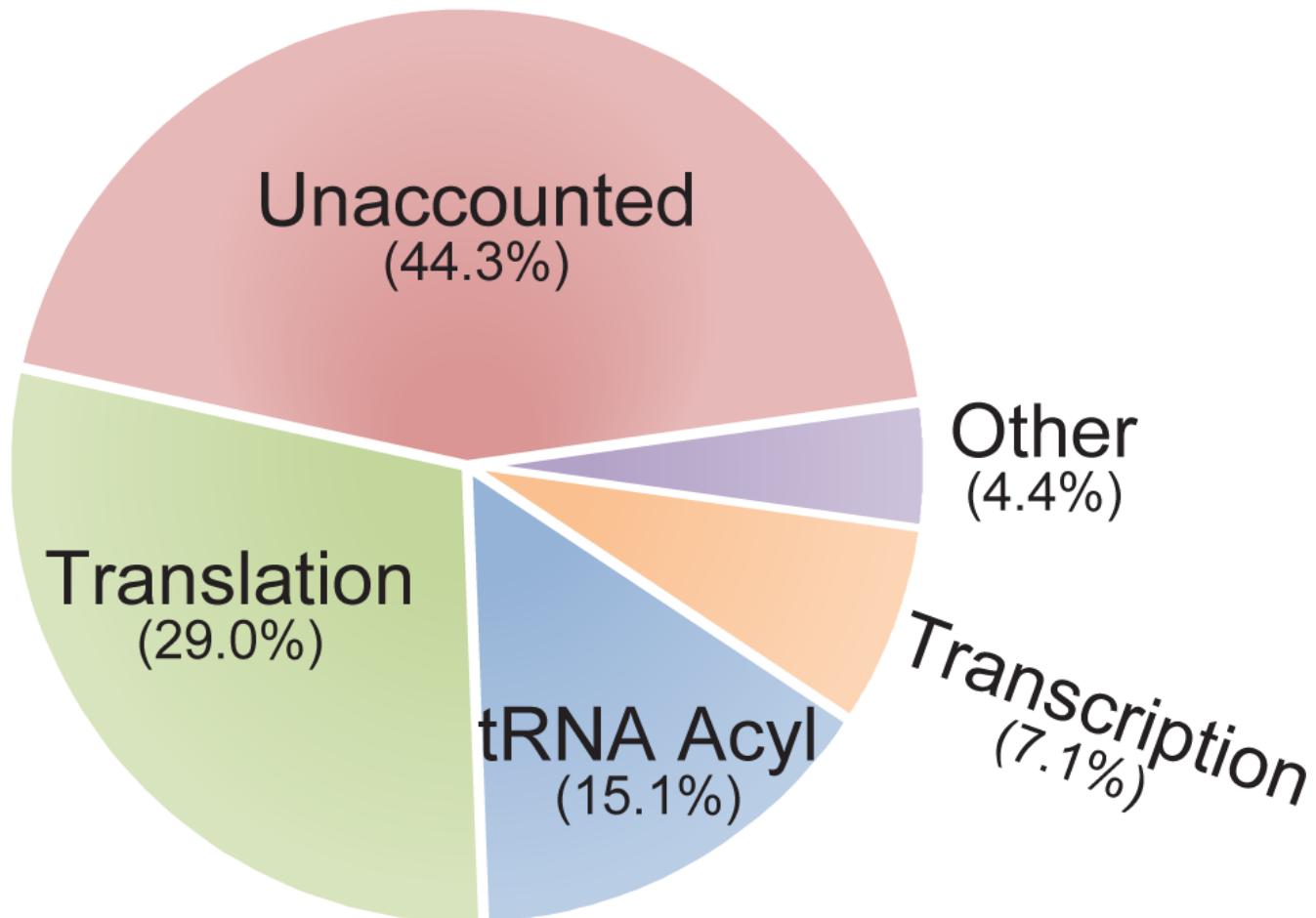
DNA binding protein collisions



DNA binding protein collisions

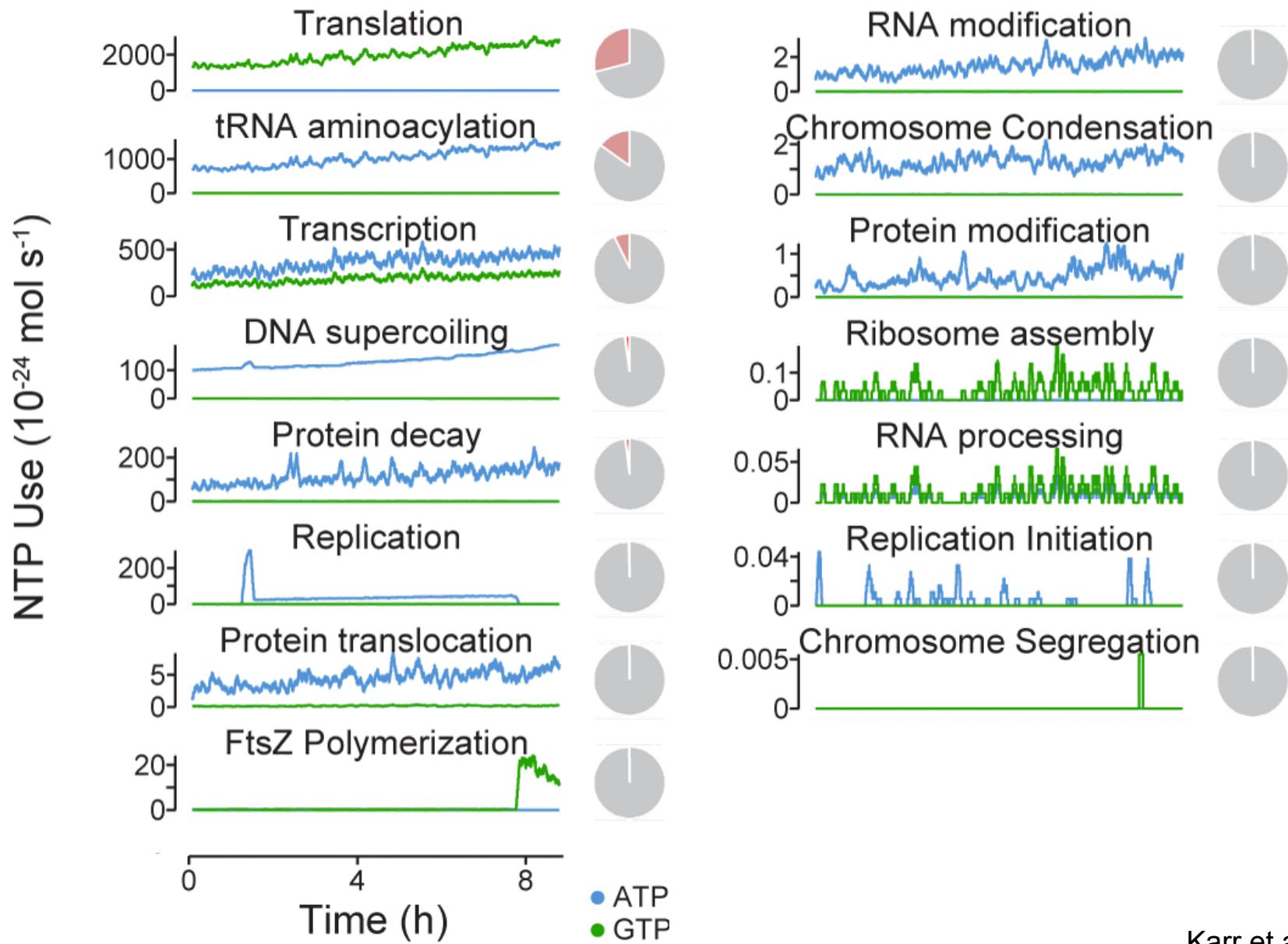


Energy consumption

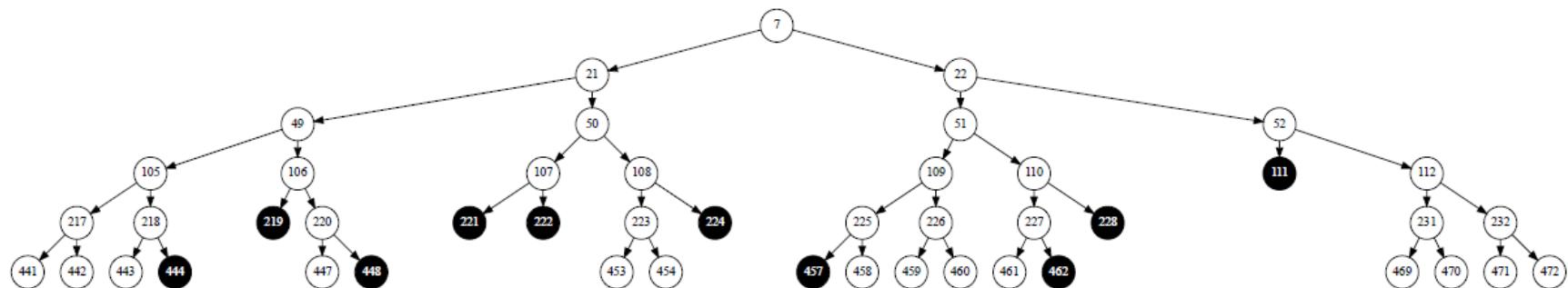


60 m mol ATP / gDCW
80 a mol ATP / cell

Energy consumption

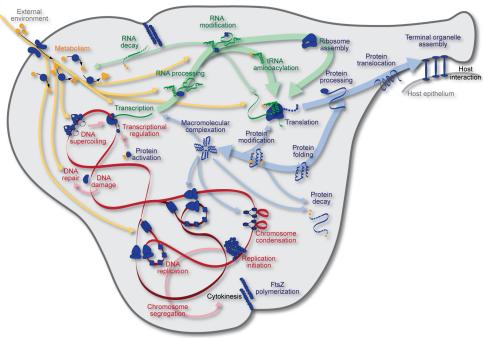


Ancestry

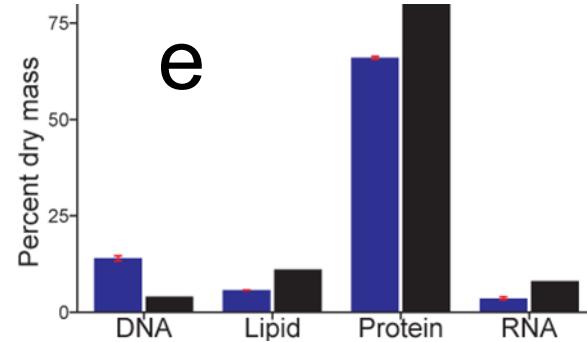


Whole-cell modeling

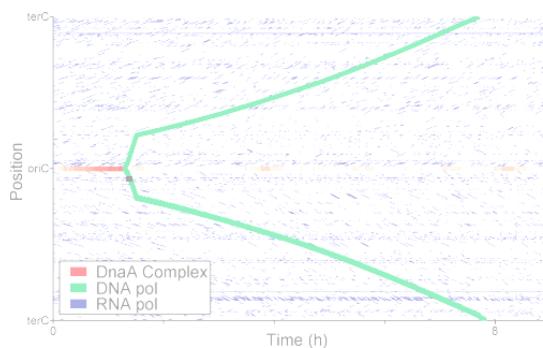
Model



Validate



Engineer



Validate model against experiments and theory

Matches training data

- Cell mass, volume
- Biomass composition
- RNA, protein expression, half-lives
- Superhelicity

Matches published data

- Metabolite concentrations
- DNA-bound protein density
- Gene essentiality

Matches new data

- Wild-type growth rate
- Disruption strain growth rates

Matches theory

- Mass conservation
- Central dogma
- Cell theory
- Evolution

No obvious errors

- Plot model predictions
- Manually inspect data
- Compare to known biology

Software stable

- Simulation code is stable
- Tests passing

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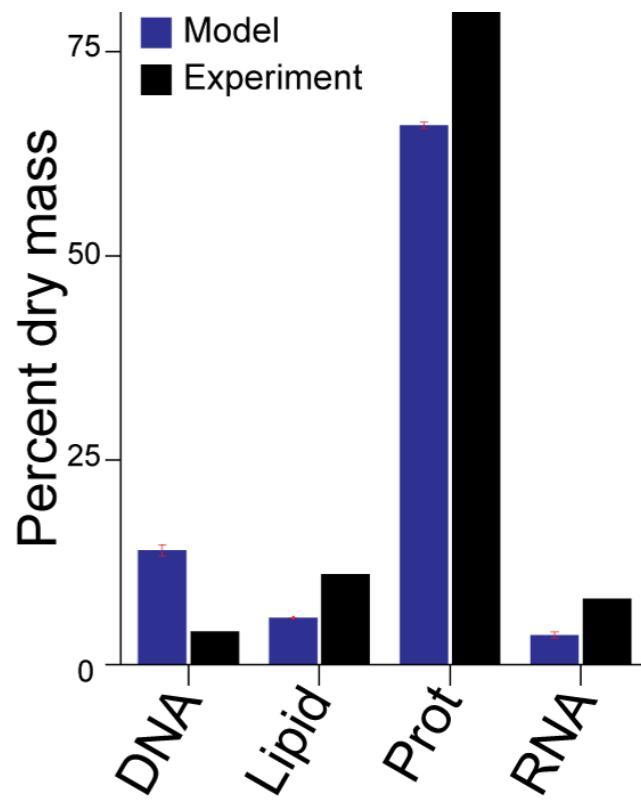
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Model reproduces observed composition



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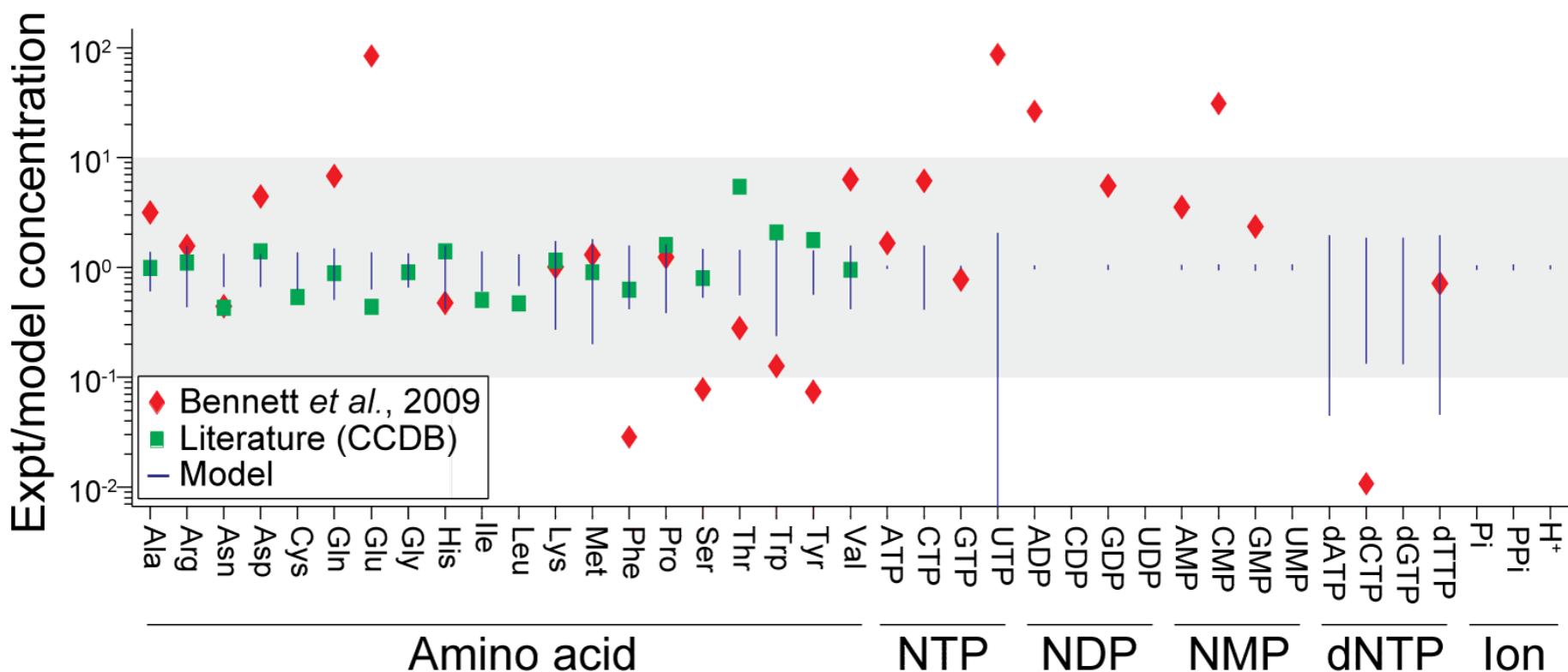
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Software stable

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- Tests passing

Model reproduces observed metabolomics



Validate model against experiments and theory

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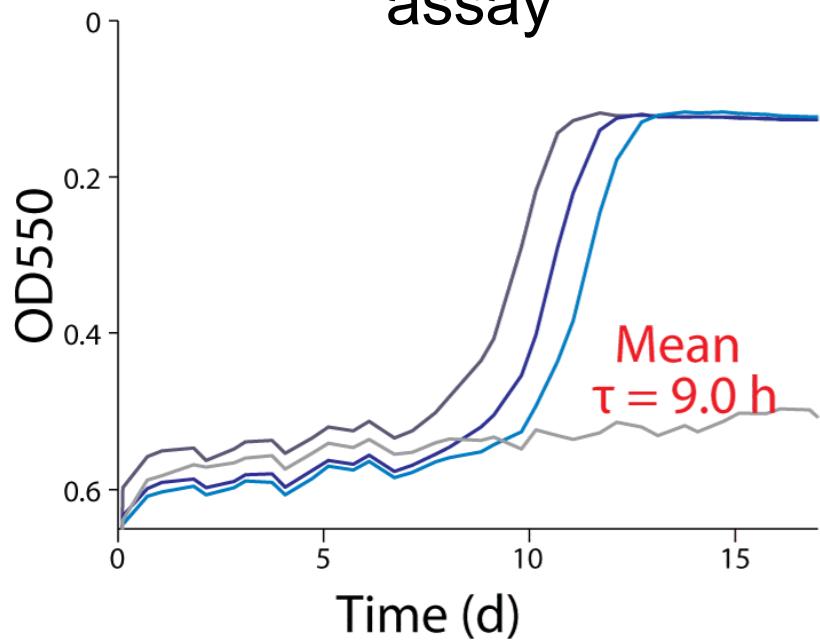
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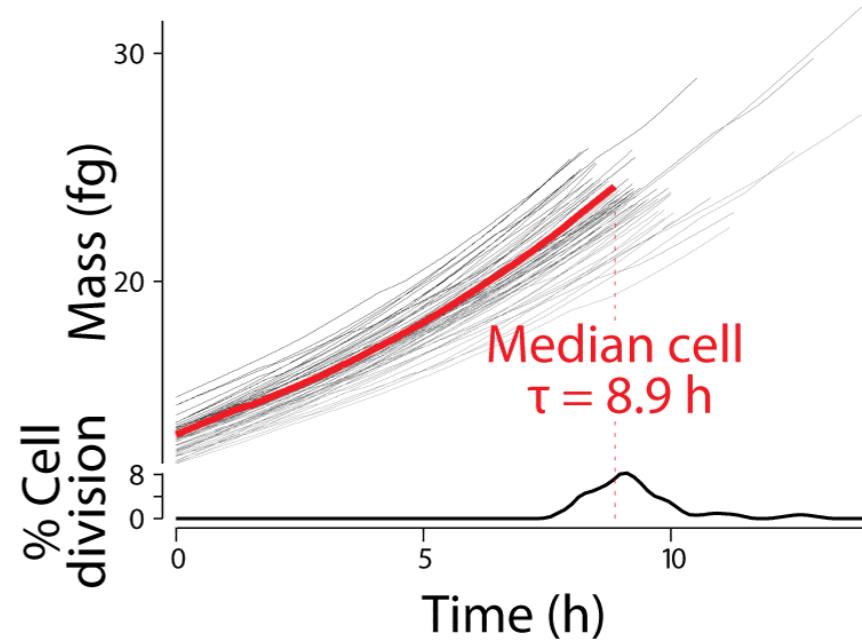
- Simulation code is stable
- Tests passing

Model reproduces measured growth rate

Colorimetric growth assay



Model predictions



Validate model against experiments and theory

Matches training data

- Cell mass, volume
- Biomass composition
- RNA, protein expression, half-lives
- Superhelicity

Matches published data

- Metabolite concentrations
- DNA-bound protein density
- Gene essentiality

Matches new data

- Wild-type growth rate
- Disruption strain growth rates

Matches theory

- Mass conservation
- Central dogma
- Cell theory
- Evolution

No obvious errors

- Plot model predictions
- Manually inspect data
- Compare to known biology

Software stable

- Simulation code is stable
- Tests passing

Validate model against experiments and theory

Matches training data

- Cell mass, volume
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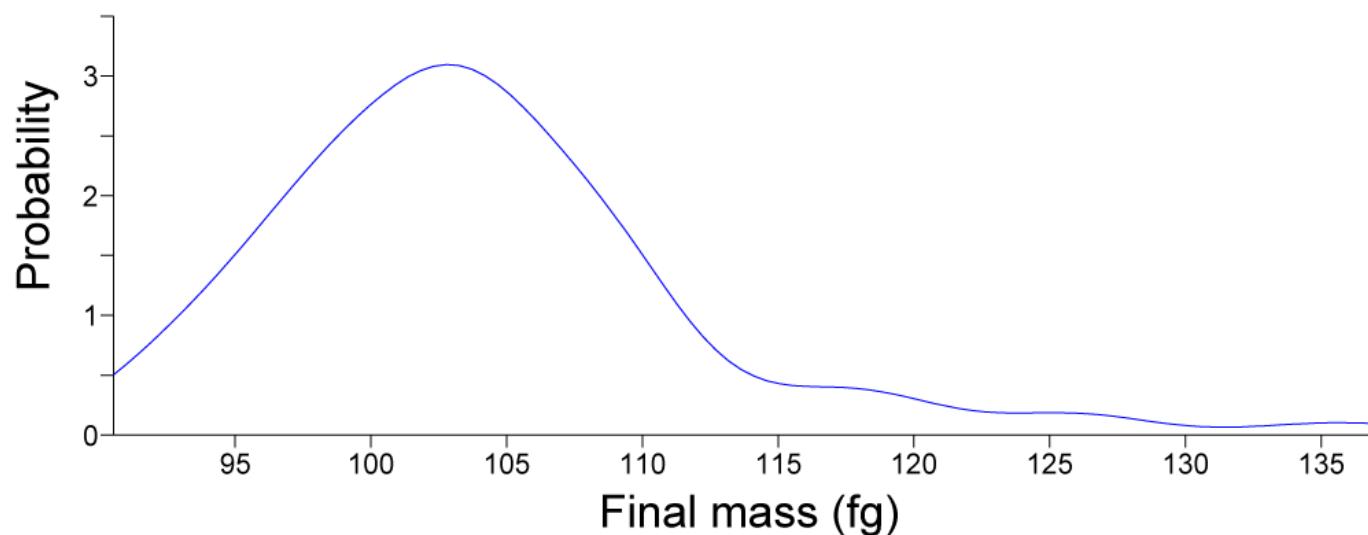
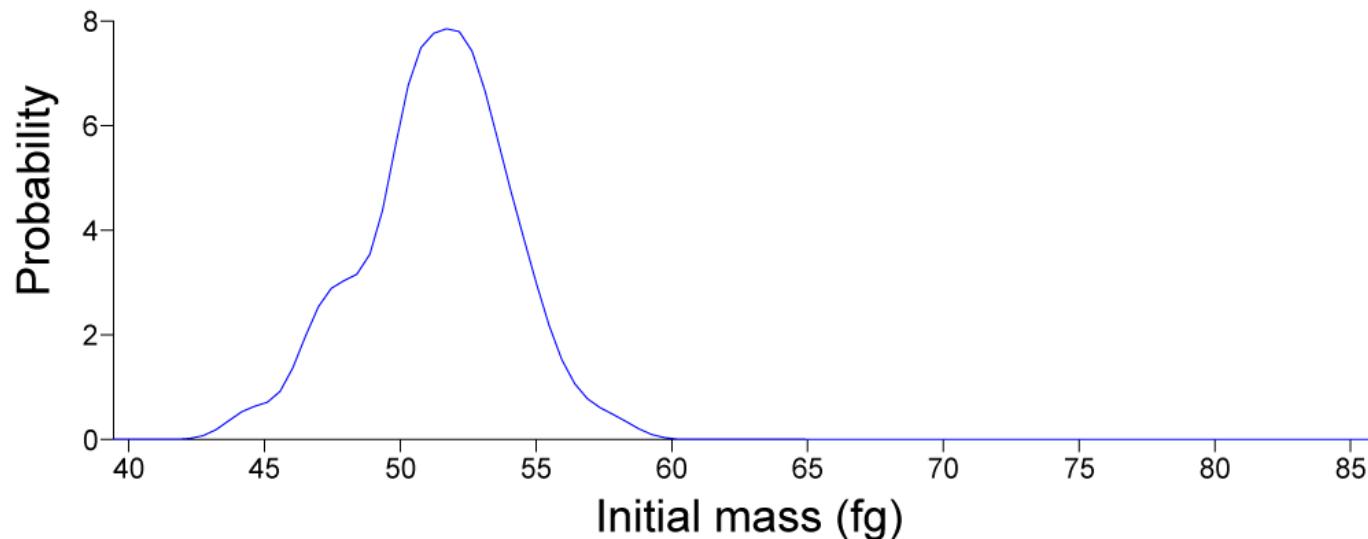
No obvious errors

- Plot model predictions
- Manually inspect data
- Compare to known biology

Software stable

- Simulation code is stable
- Tests passing

Model satisfies cell theory



Validate model against experiments and theory

Matches training data

- Cell mass, volume
- Biomass composition
- RNA, protein expression, half-lives
- Superhelicity

Matches published data

- Metabolite concentrations
- DNA-bound protein density
- Gene essentiality

Matches new data

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- Central dogma
- Cell theory
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No obvious errors

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Software stable

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Validate model against experiments and theory

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No obvious errors

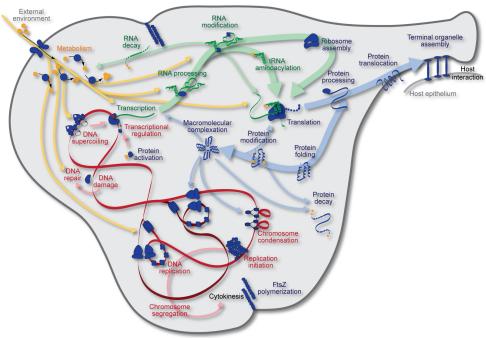
- Plot model predictions
- Manually inspect data
- Compare to known biology

Software stable

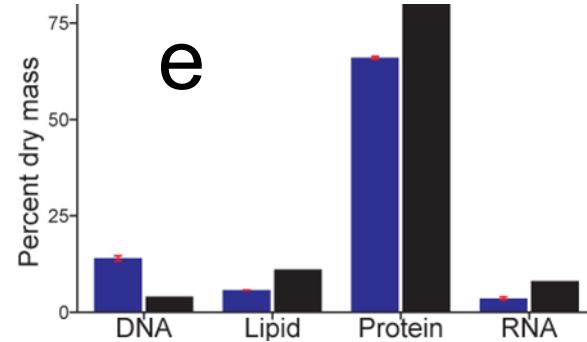
- Simulation code is stable
- Tests passing

Whole-cell modeling

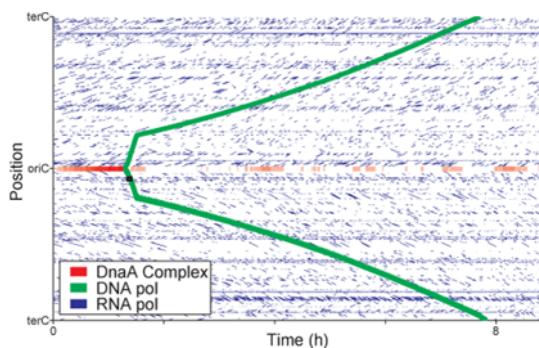
Model



Validate

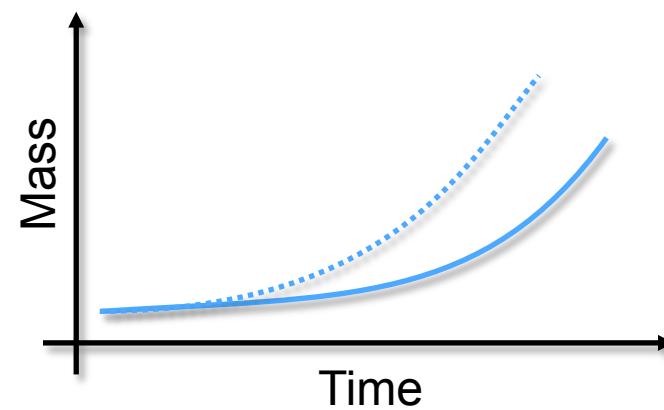
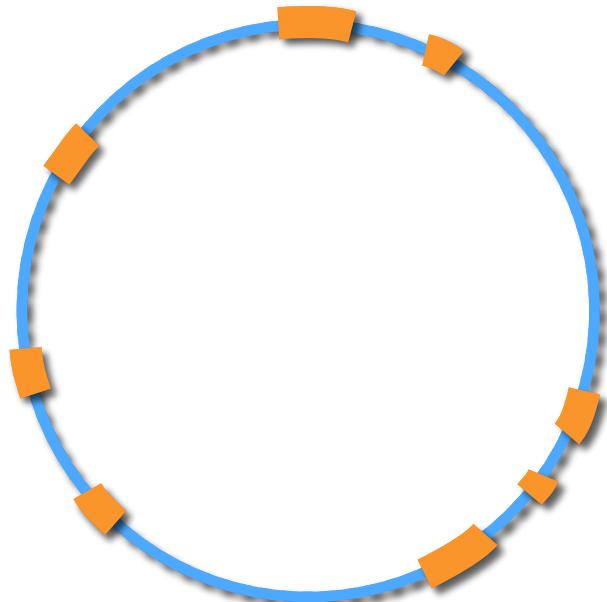


Engineer

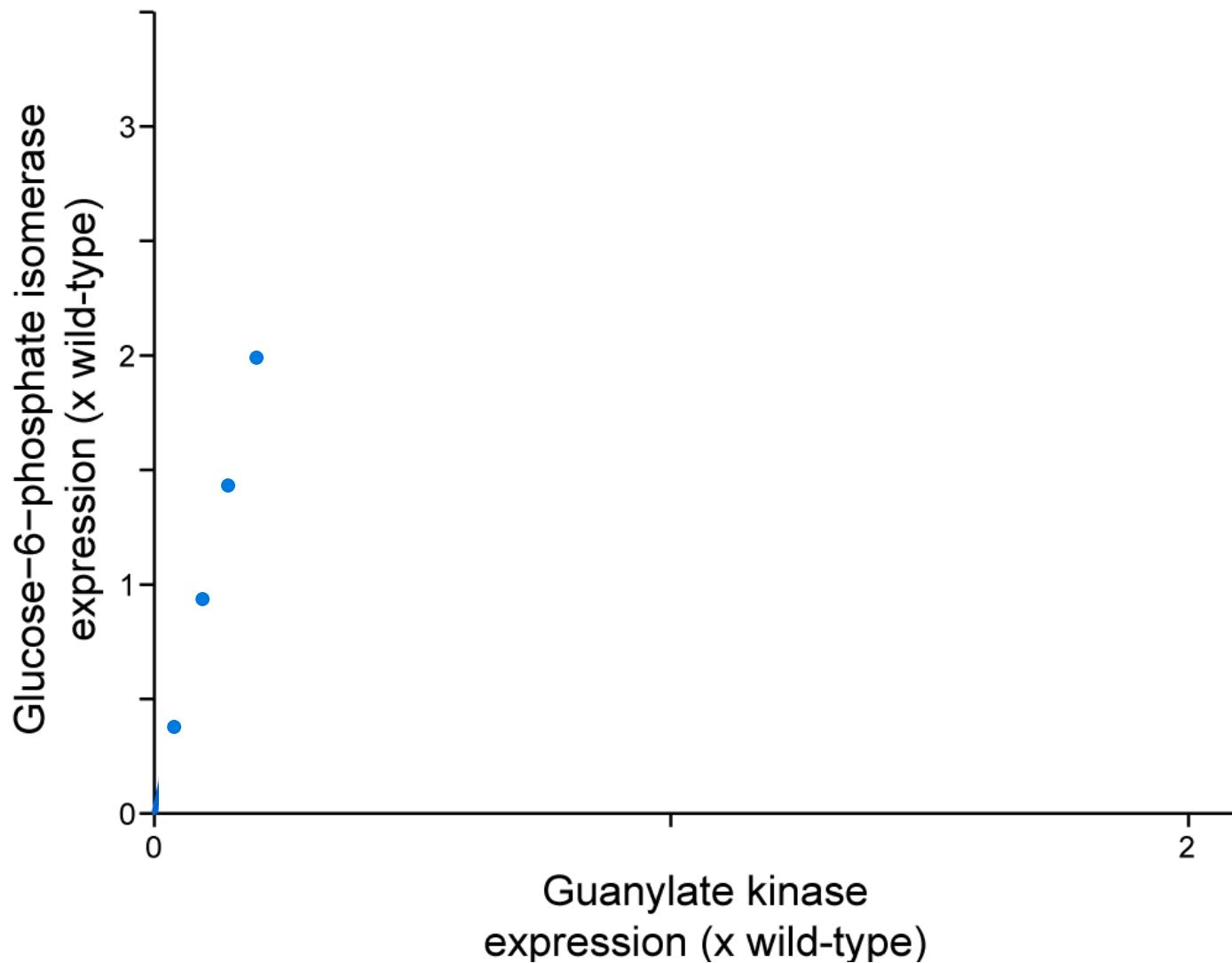


Example: growth optimization

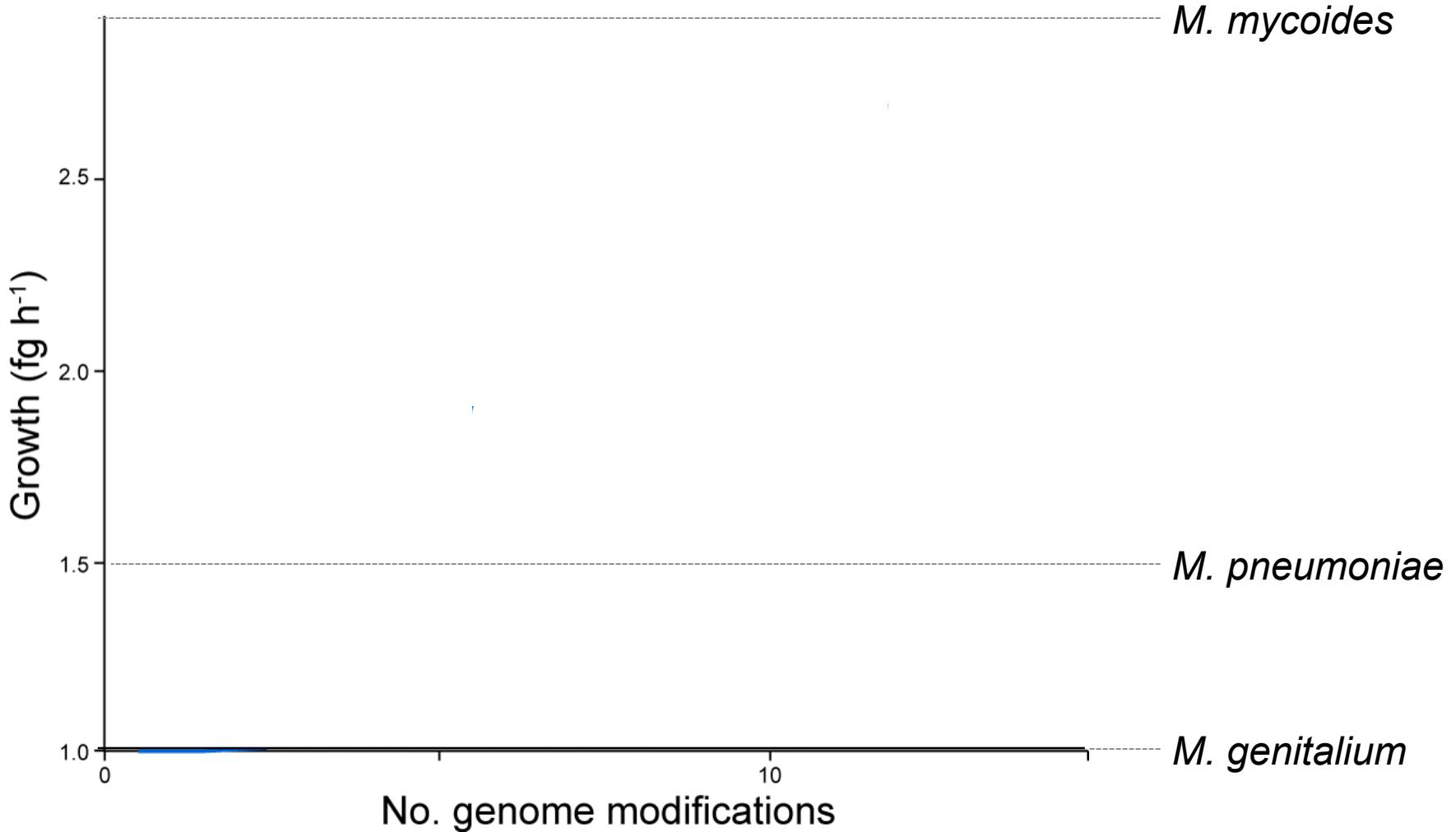
What genomic modifications maximize growth?



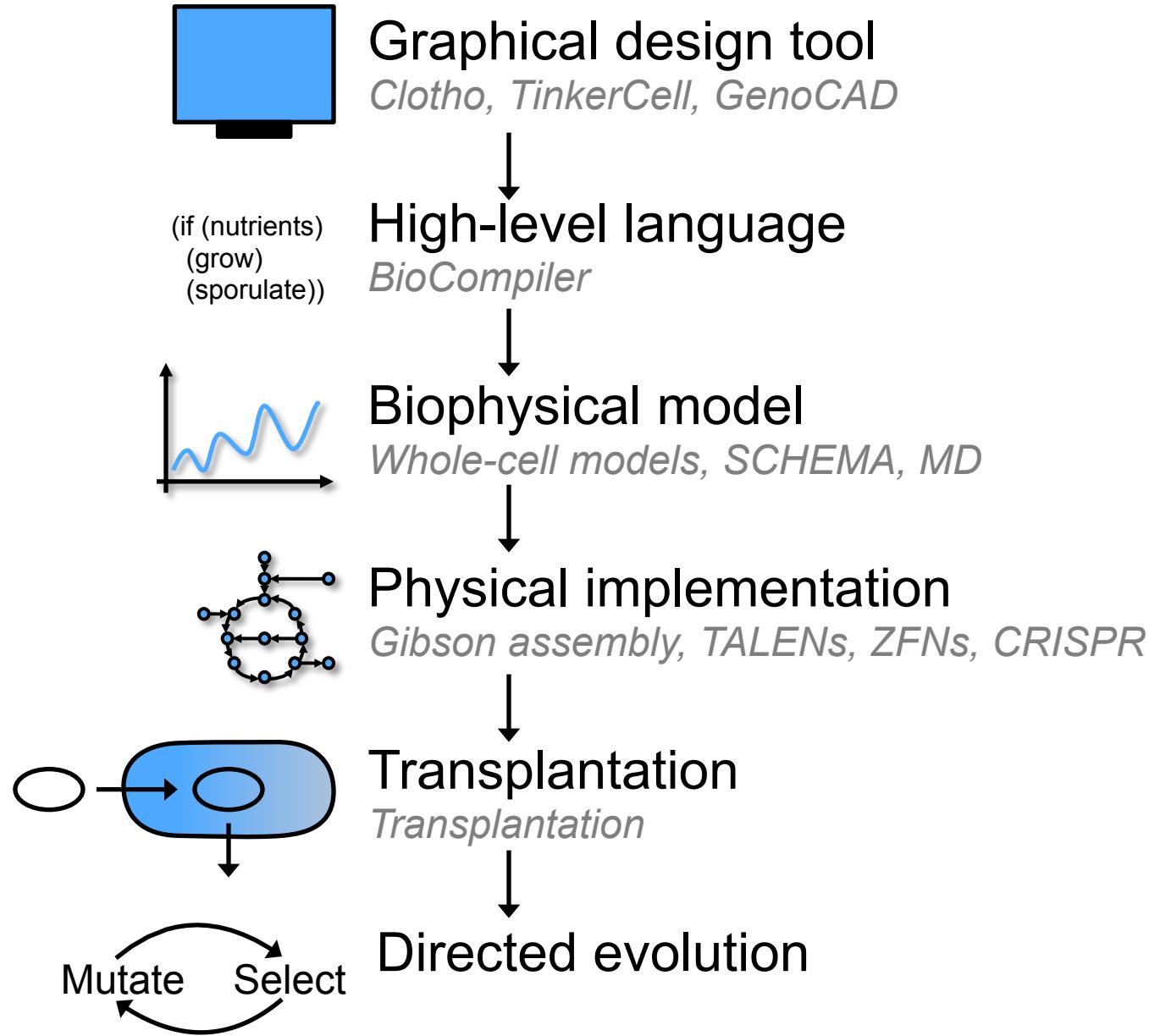
Optimal gene expression



Optimal gene expression



Synthetic design landscape



Open challenges

- How can we model more complex physiology?
 - Regulation
 - Aging
 - Cell death
 - Evolution
- How can we model more complex organisms?
 - Larger bacteria
 - Eukaryotes
 - Multicellularity
 - Humans
- How can we use models to direct engineering?

Karr lab: expanding whole-cell models



M. pneumoniae

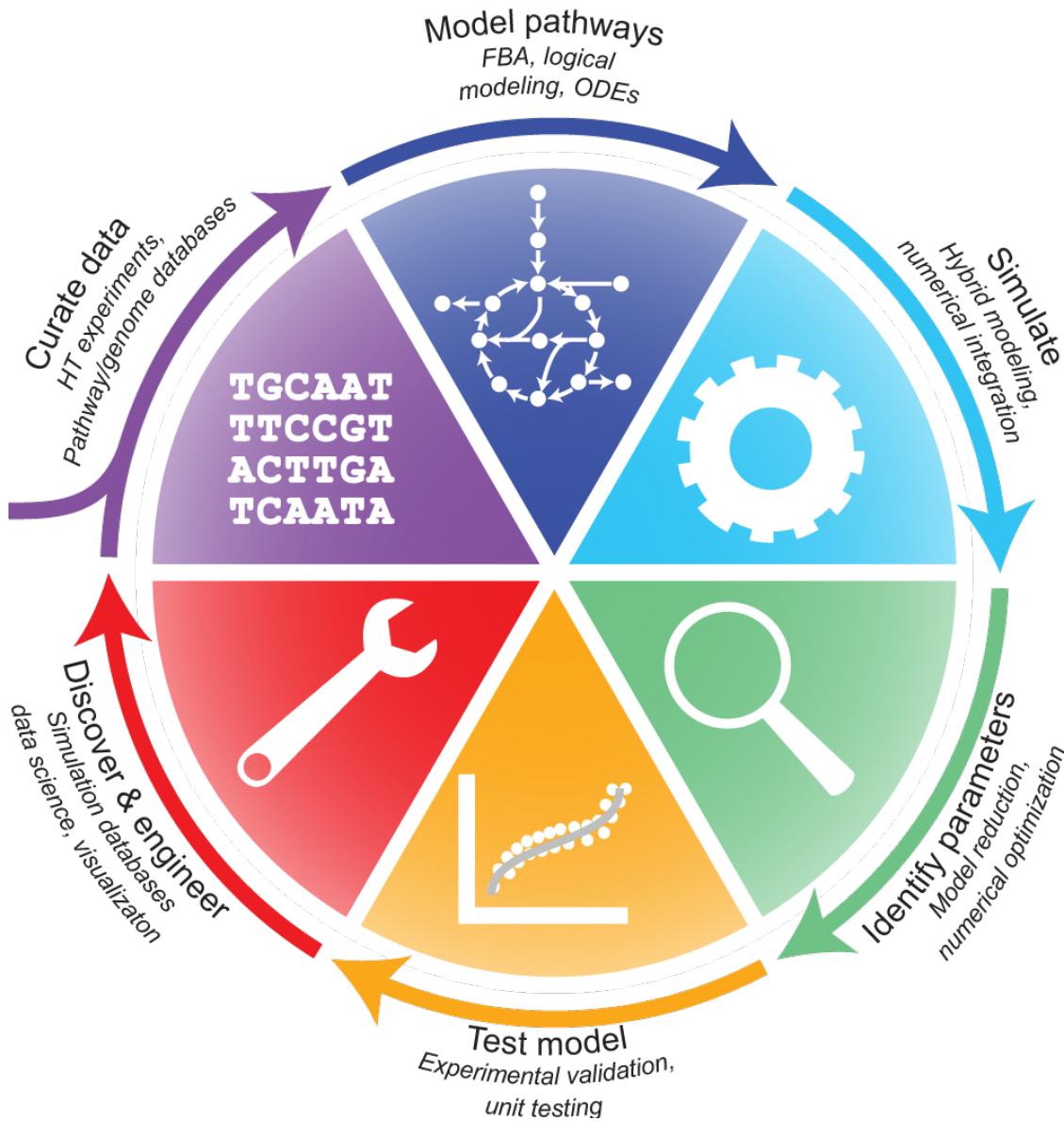
- Expand scope: regulation
- Improve accuracy: species-specific data
- Enable rational genome engineering
- Cell-based drug therapy



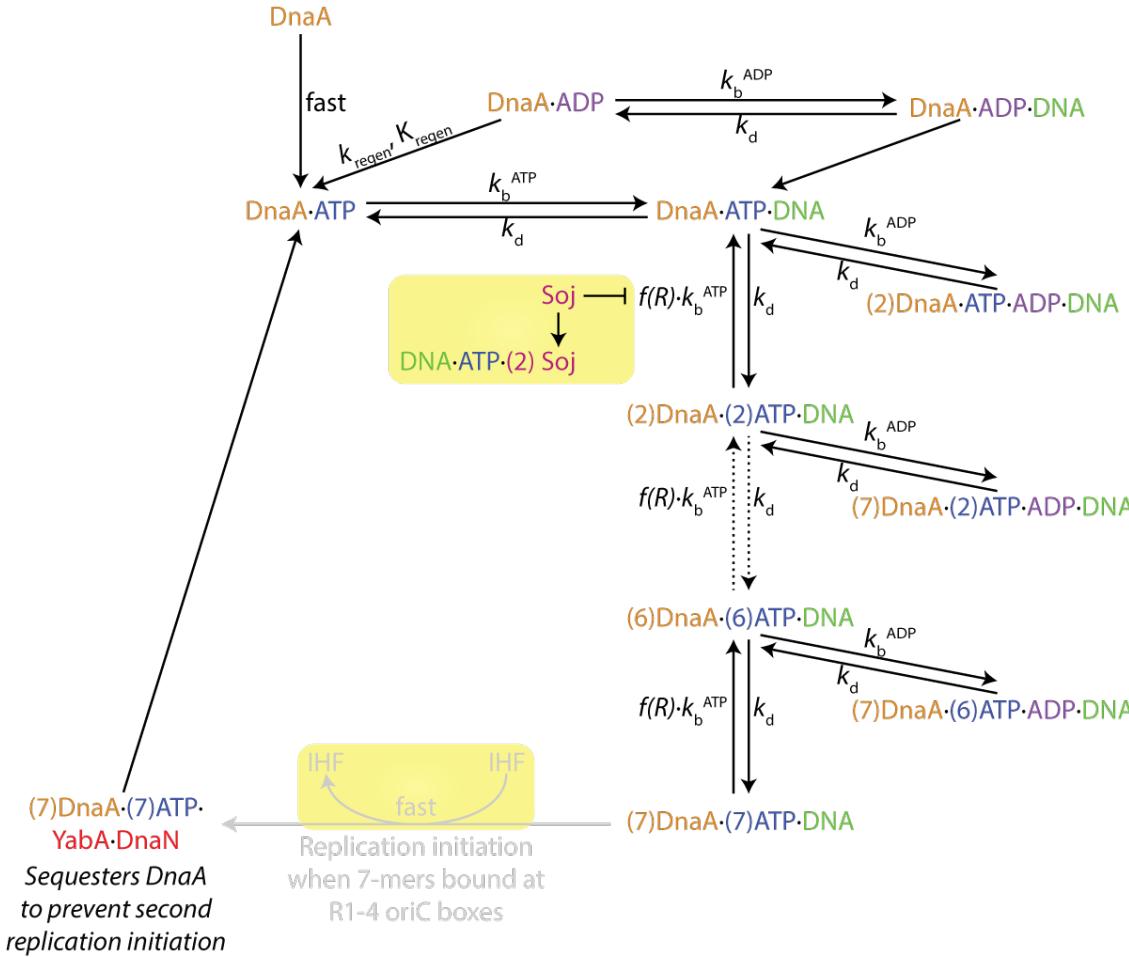
Human cancer

- Colorectal cancer
- Personalized models
- Precision medicine

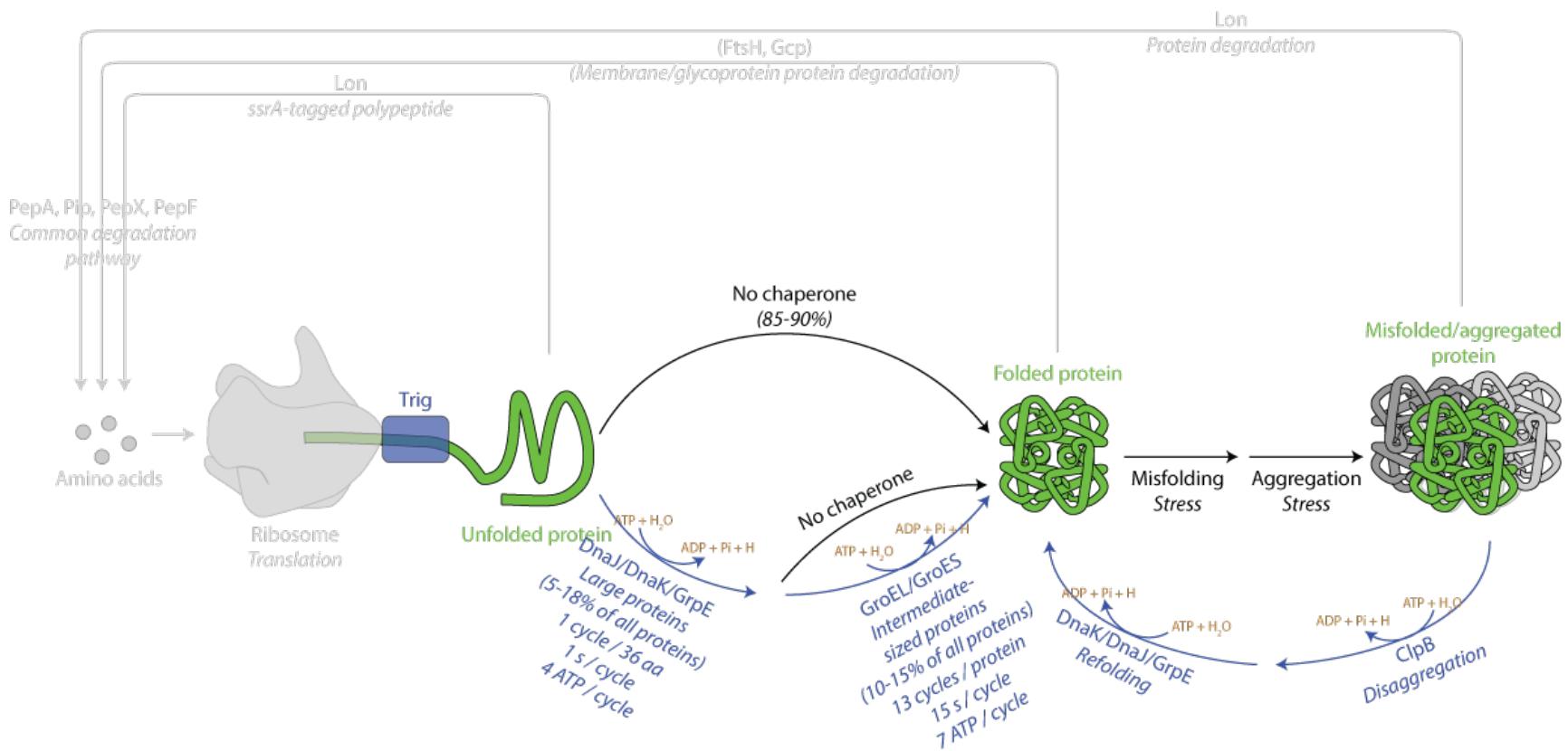
Toward more comprehensive models



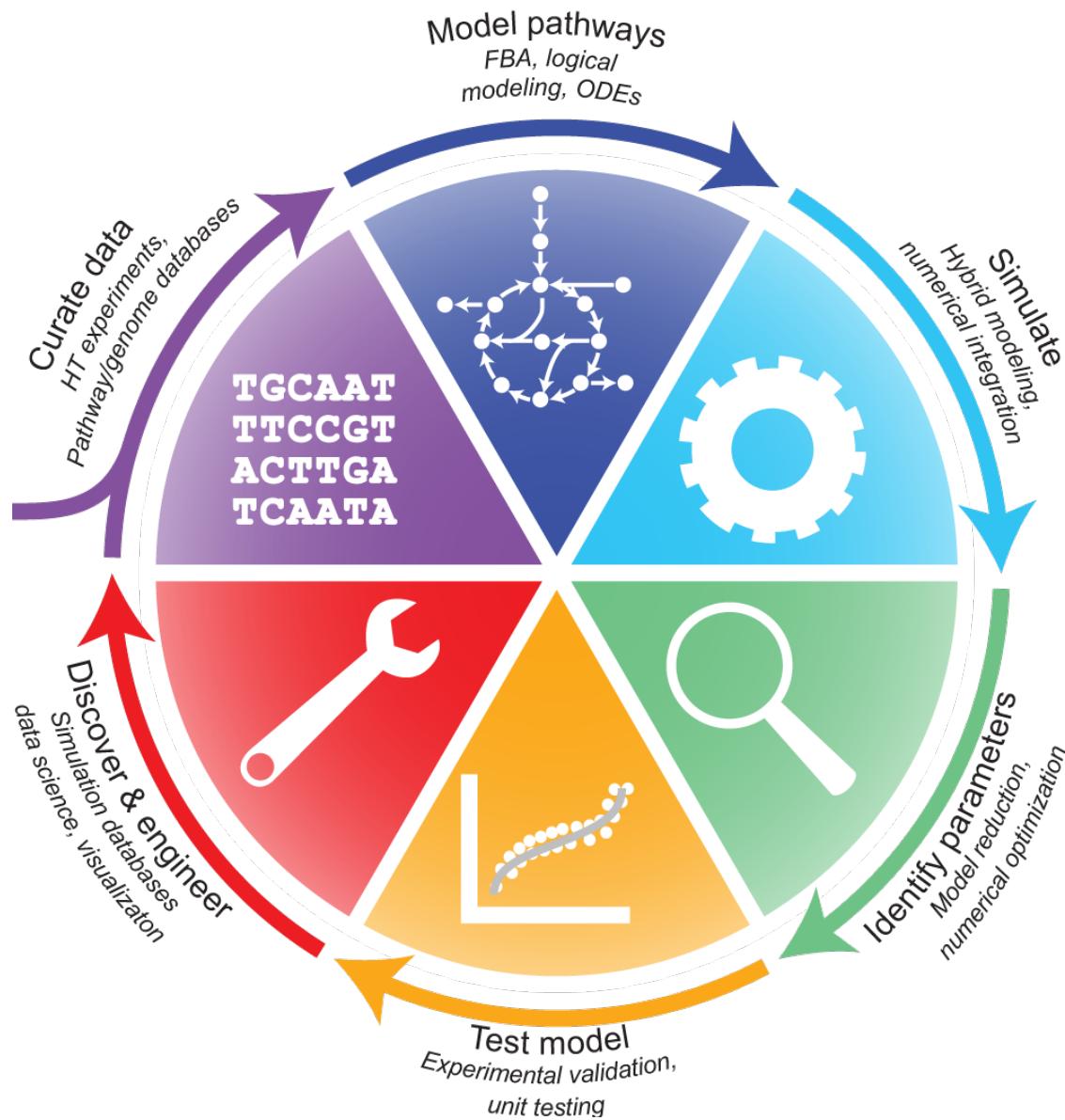
Toward more comprehensive models



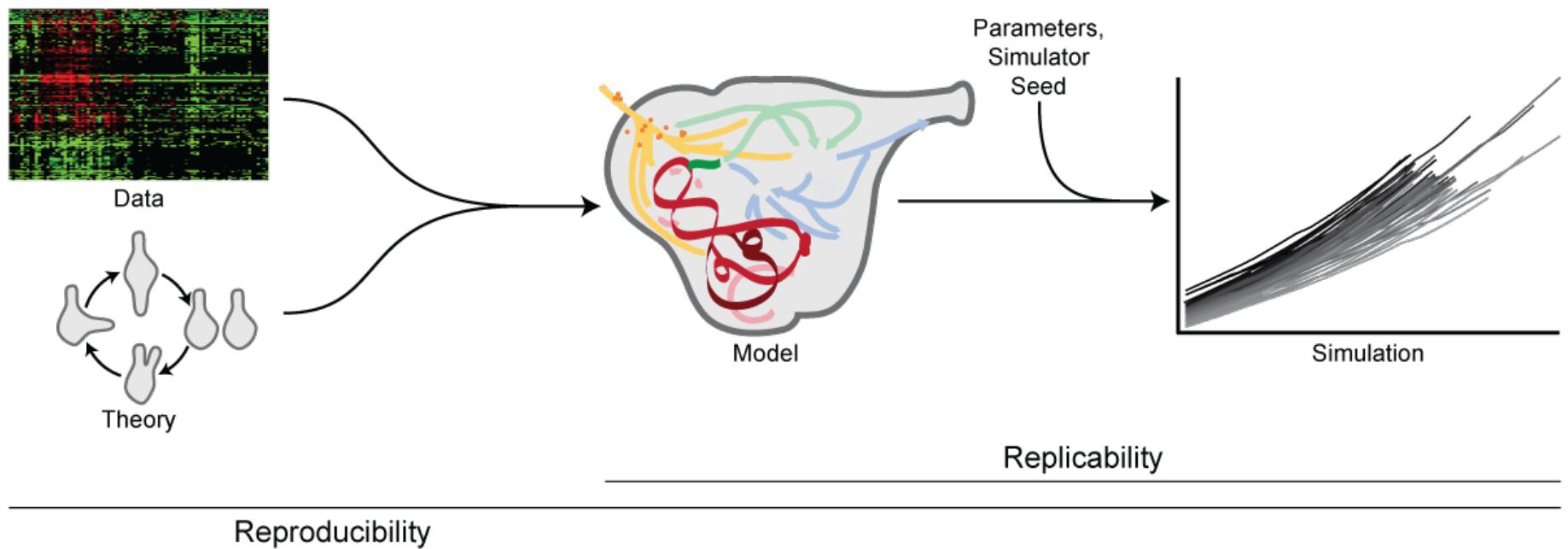
Toward more comprehensive models



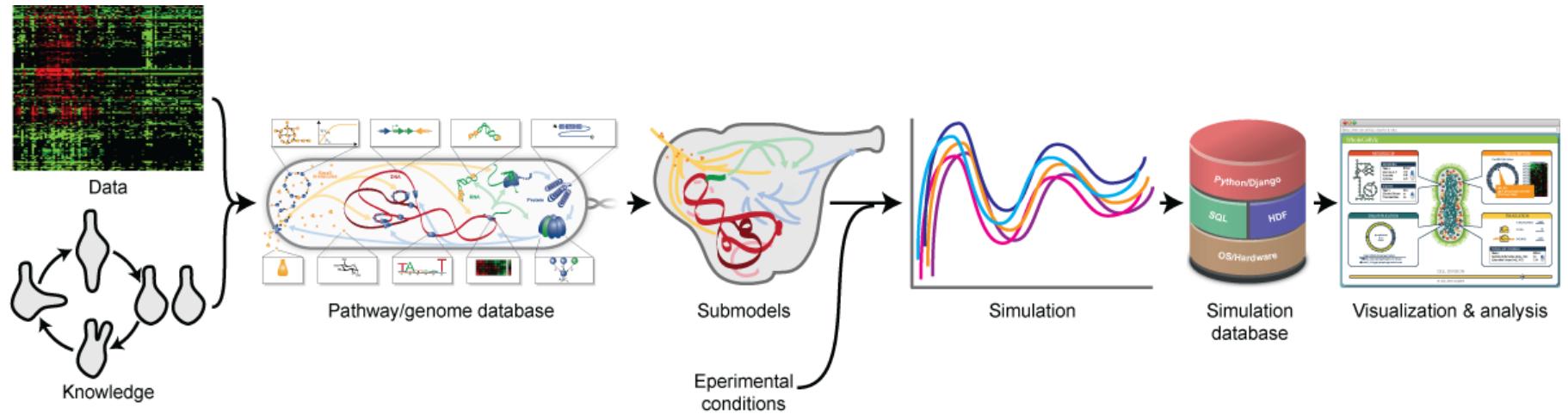
Toward more comprehensive models



Toward more comprehensive models

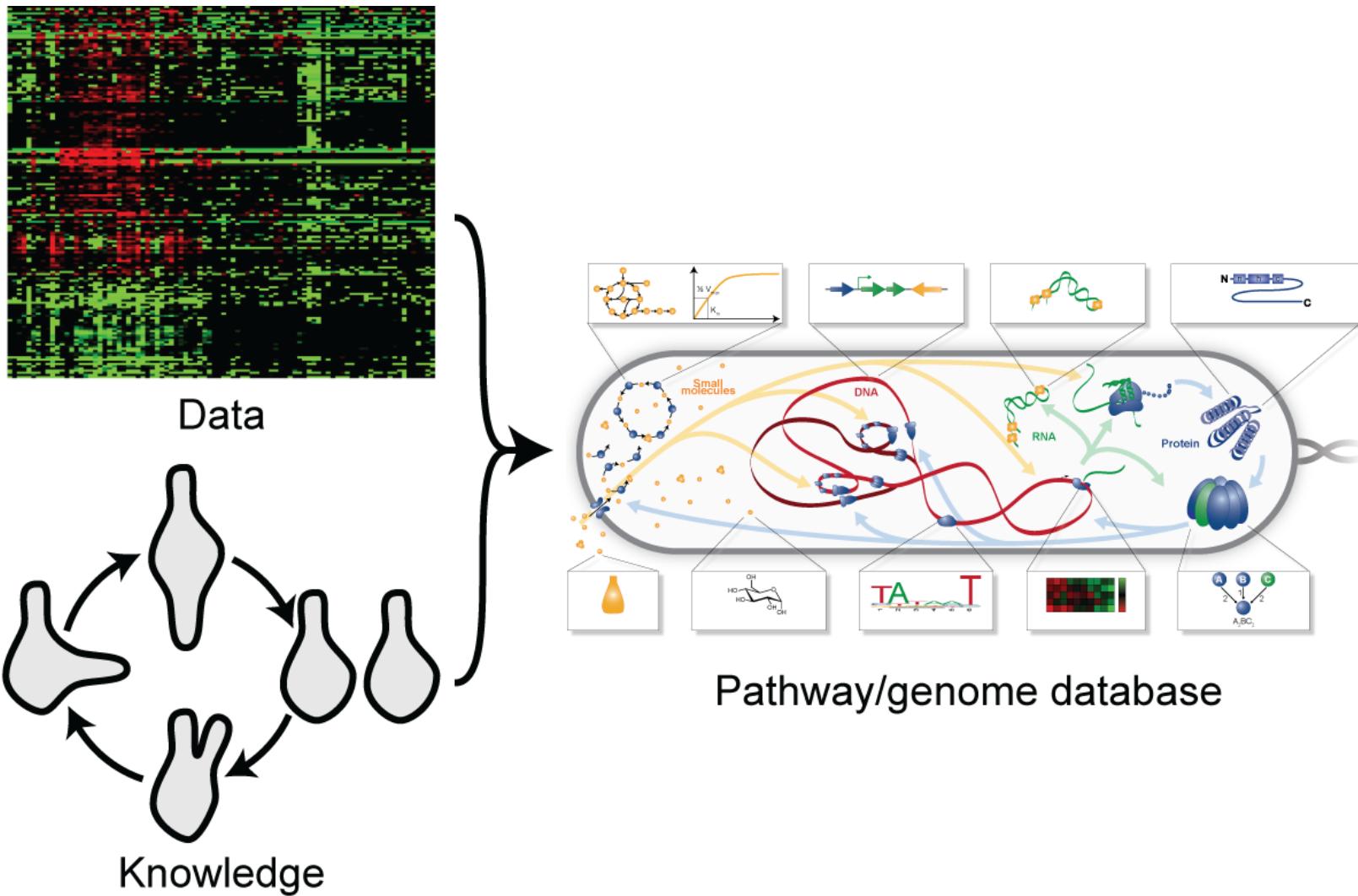


Toward more comprehensive models



Data → Knowledge,
Models applications

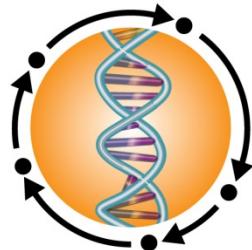
Data curation



Data curation

	Data type	Source
Baseline	Chaperones	Literature
Disease	Complex composition	Human Soluble Protein Complexes DB
	DNA binding sites	TRANSFAC, AnimalTFDB
	DNA footprints	Literature
	DNA methylation	MethBase
	DNA sequence	Genome Reference Consortium
	Gene-drug interactions	DrugBank, PharmaGKB
	Genome annotation	Ensembl
	Growth rates	Hapmap, NCI-60
	Metabolite concentrations	Human Metabolome Database
	Protein cofactors	UniProt
	Protein expression	Human Protein Atlas
	Protein half-lives	Literature
	Protein localization	Human Protein Atlas
	Protein modification	Human Protein Reference DB
	RNA editing	RADAR, DARNED
	RNA expression	GEO, Human Protein Atlas
	RNA half-lives	Literature
	RNA modification	RNA Modification DB, MODOMICS
	RNA maturation	RNApathwaysDB
	Reaction fluxes	Literature
	Reaction kinetics	SABIO-RK, BRENDA
	Reaction stoichiometries	Recon X, UniProt, HumanCyc
	Signaling pathways	Literature
	DNA mutations	CCLE, COSMIC
	DNA methylation	TCGA
	Gene-drug interactions	CCLE
	Growth rates	NCI-60
	Metabolite concentrations	Literature
	Protein expression	TCGA
	RNA expression	CCLE

Data curation



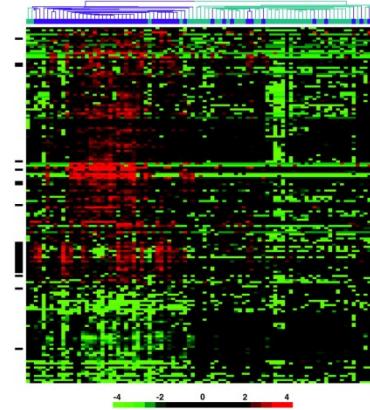
BioCyc



Databases

Low throughput annotation,
biochemical data

Semi-automated curation



Articles

High-throughput,
genomic measurements

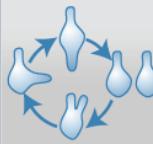
Assisted curation

Pathway/genome database

Data organization

WholeCellKB x

www.wholecellkb.org



WholeCellKB

Tmk – thiamine kinase

Name

WID	Tmk	
Name	thiamine kinase	
Cross references	EC: 2.7.1.89, BioCyc: DTMPKI-RXN, KEGG: R02094, BIGG: TMK	

Classification

Type	chemical	
------	----------	--

Reaction

Stoichiometry	[c]: ATP + DTMP ⇌ ADP + DTDP	
---------------	------------------------------	--

Catalysis

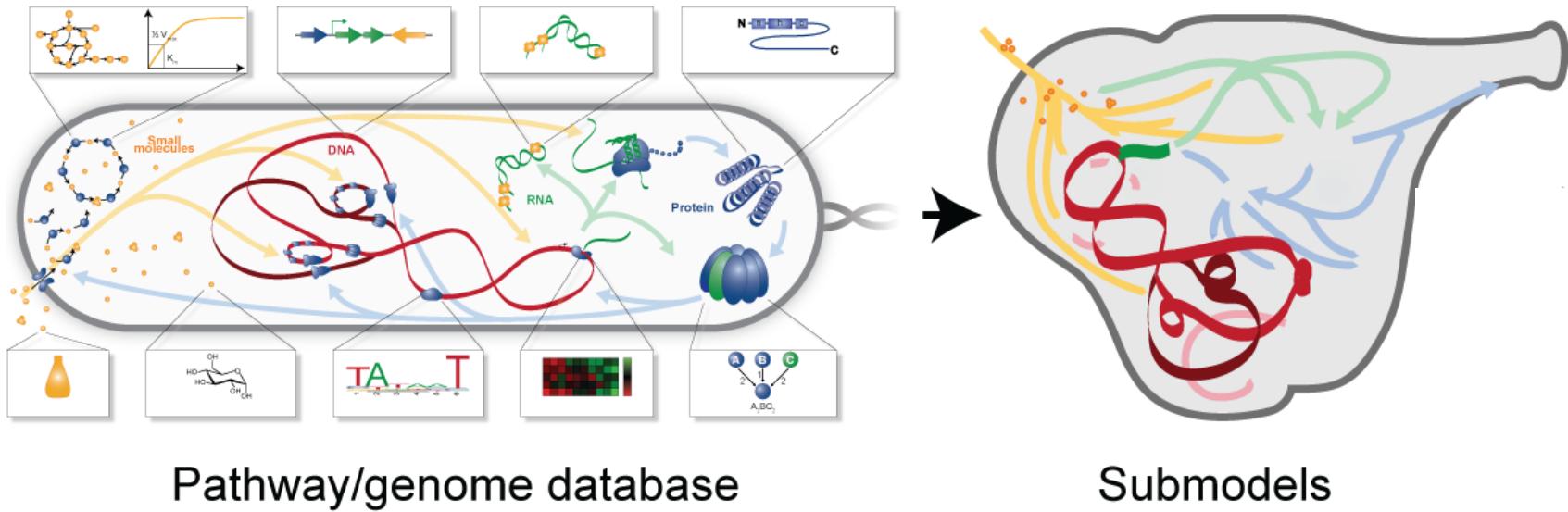
Enzyme	MG_006_DIMER [c]	
Optimal pH	7.9 (dimensionless)	
Optimal temperature	45.0 (C)	

Energetics

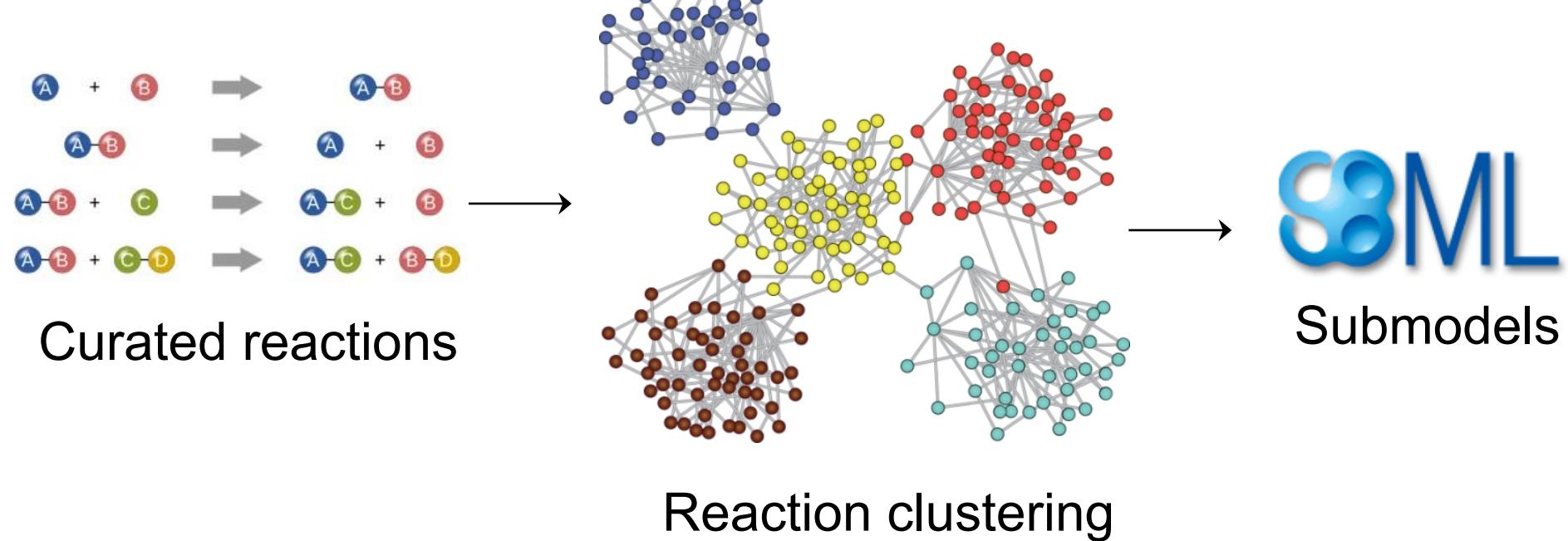
Is spontaneous (pH 7.5, 25C, $J = 0$)	False	
ΔG (pH 7.5, 25C, $J = 0$; kJ mol ⁻¹)	-5.80	

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Model design



Model design



Model encoding

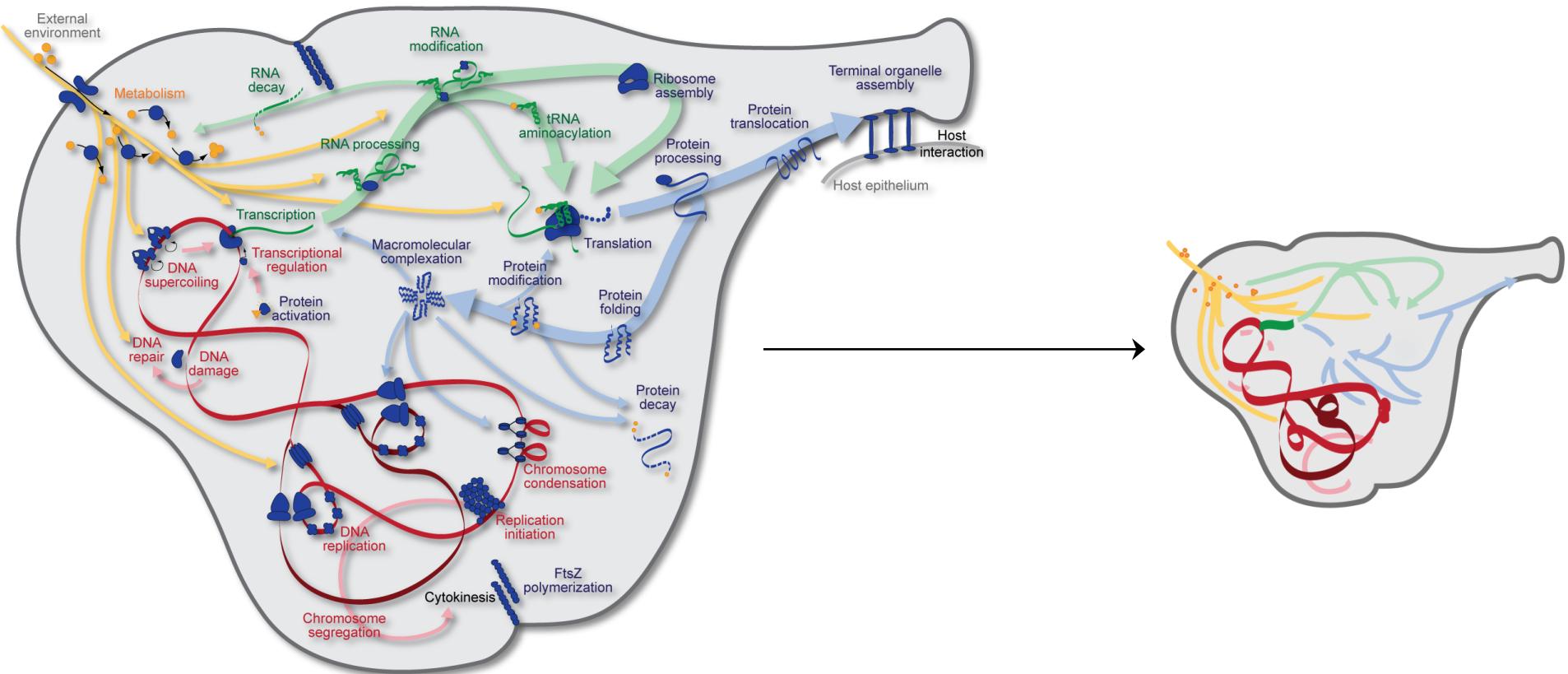
Goals

- Represent the entire model
- Edit, expand models
- Transparency
- Exchange
- Reproducibility

SBML

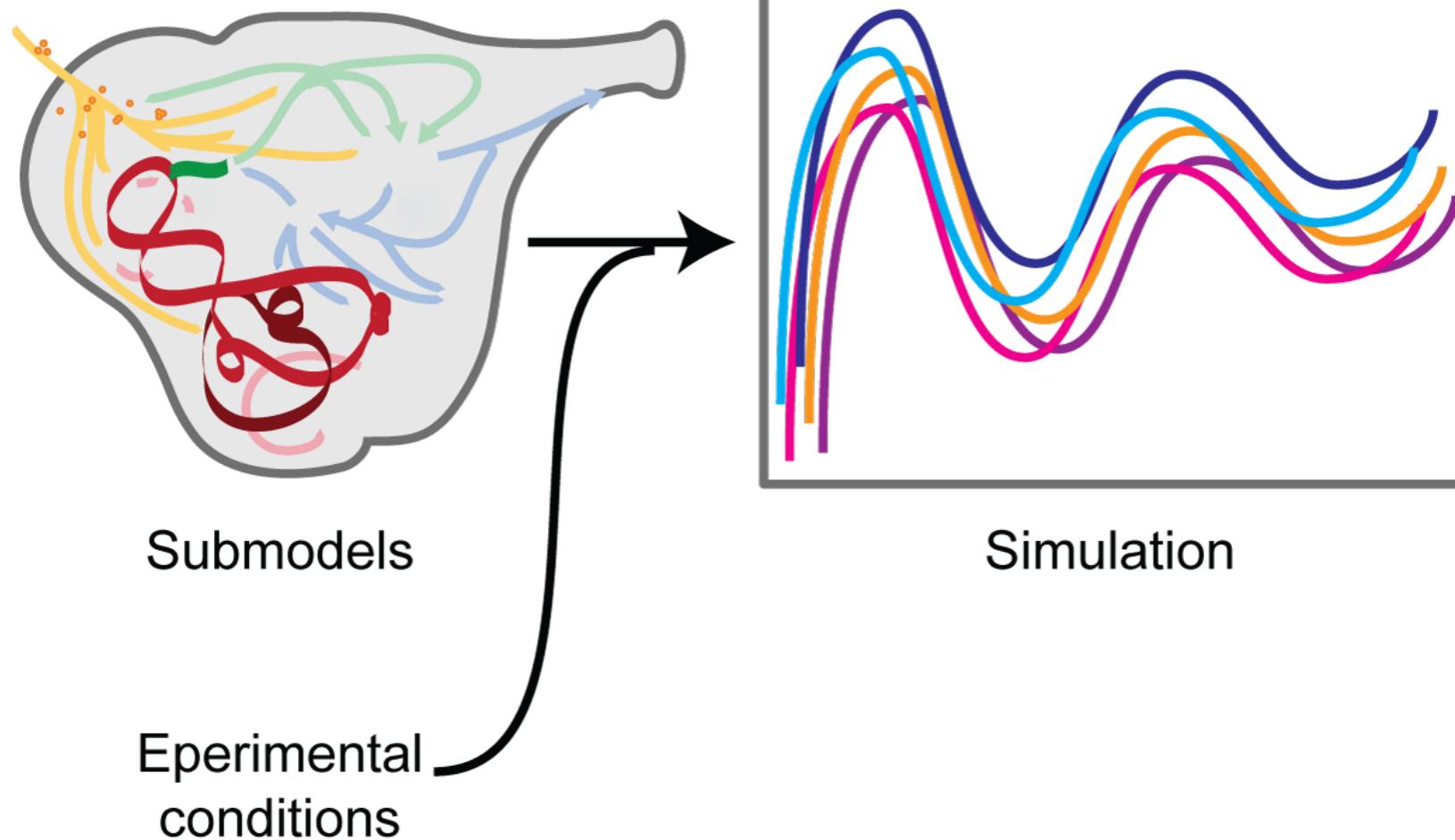
- Requires numerous features
 - Multistate and multicomponent species
 - Hierarchical model composition
 - Distributions
 - Annotations
- How to represent bioinformatic portions of the models?
- How to represent initial conditions model?

Parameter identification

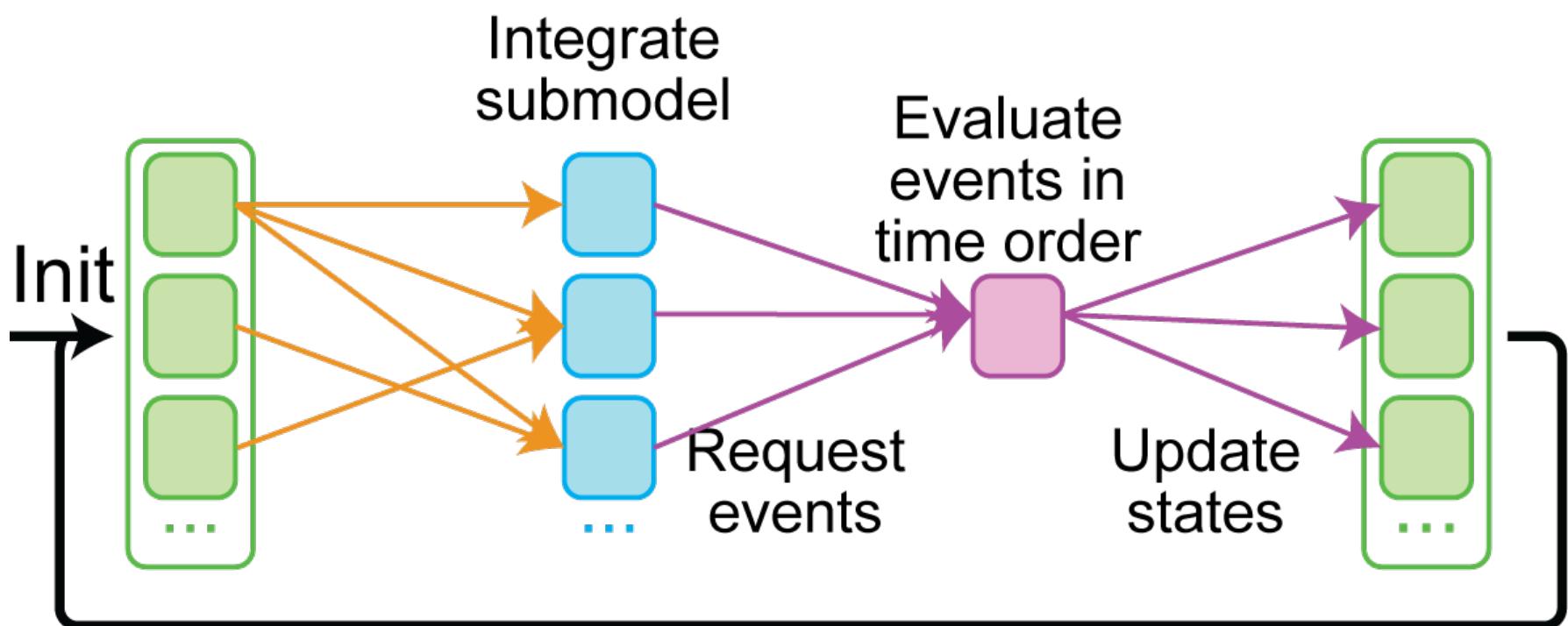


Model reduction, distributed optimization

Simulation

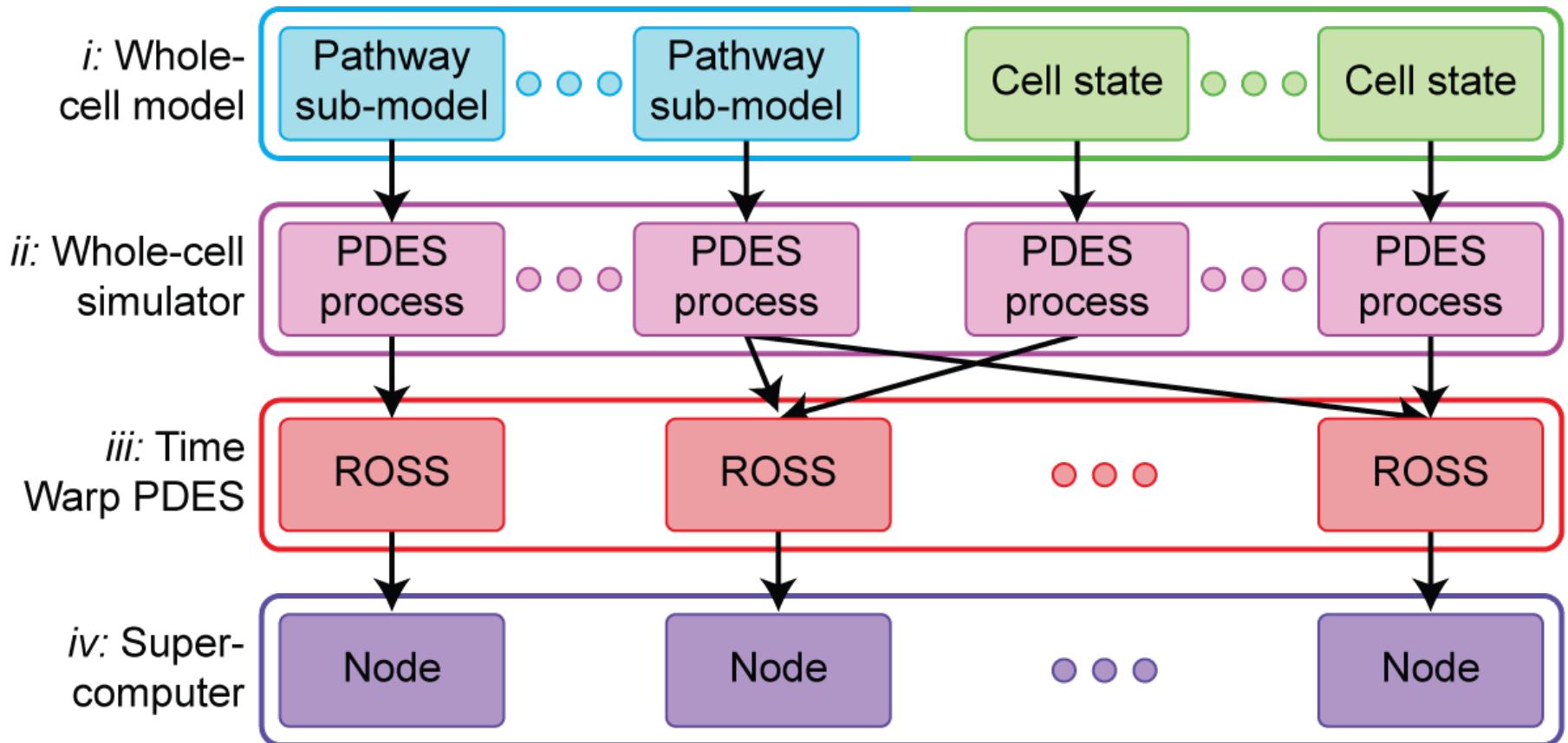


Multi-algorithm simulation

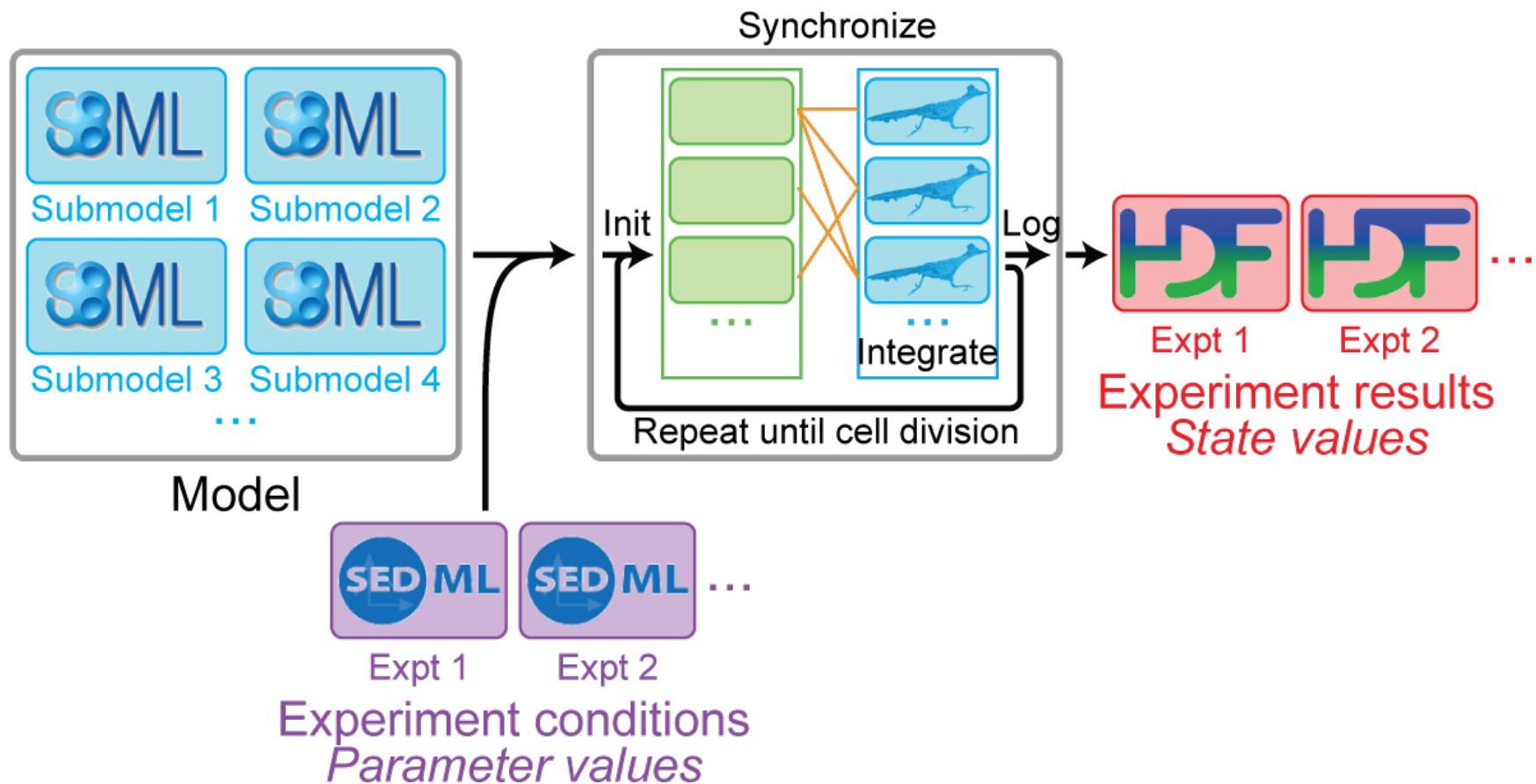


$t \leftarrow t + \Delta t$, Repeat until cell division

Multi-algorithm simulation



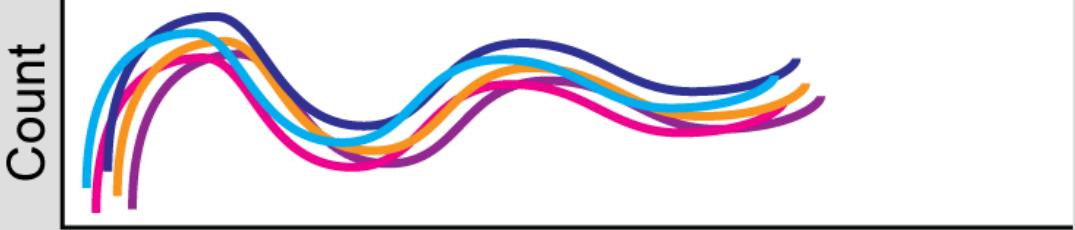
High-performance simulator



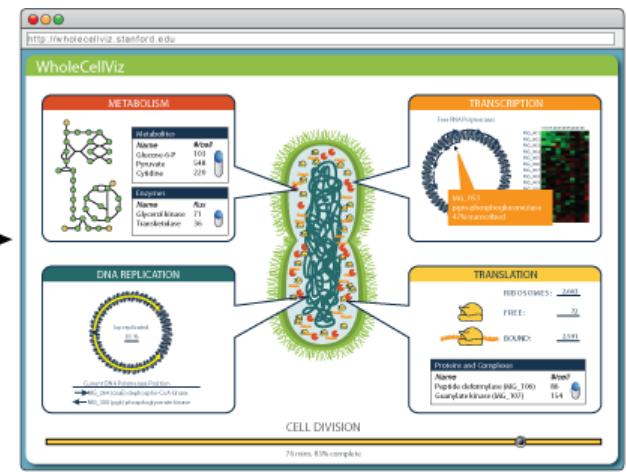
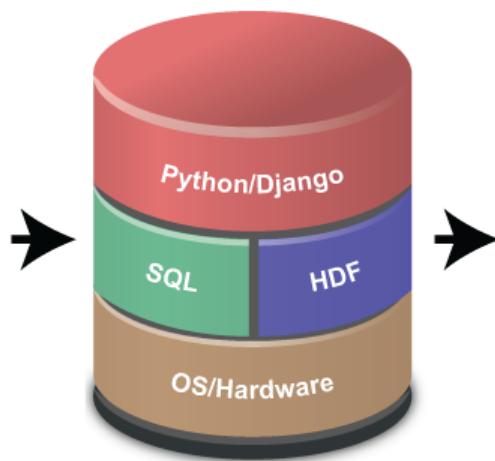
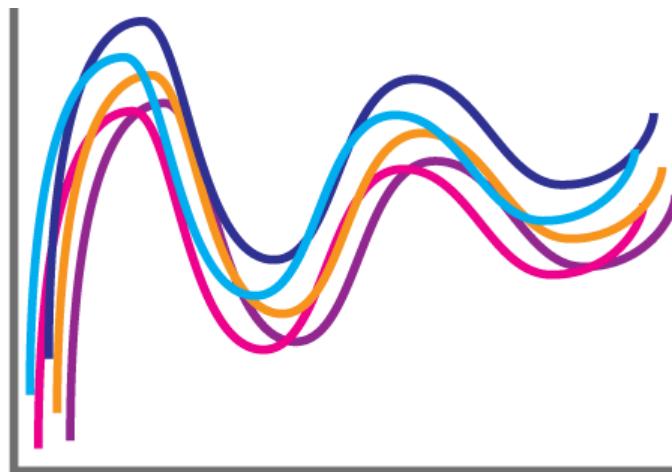
High-performance simulator

Whole-cell simulator

-

Config simulations	Results
(1) Select model Select <u>Mge.sbml</u>	(1) Select simulations <input checked="" type="checkbox"/> 1 <input type="checkbox"/> 2 <input checked="" type="checkbox"/> 3 <input type="checkbox"/> 4
(2) Parameter vals [ATP] _i <u>1,000</u> k _{cat} <u>10</u> T <u>10</u>	(2) Select states <input checked="" type="checkbox"/> RNA-1 <input type="checkbox"/> RNA-3 <input type="checkbox"/> RNA-2 <input type="checkbox"/> Protein-1
(3) Stat sampling Simulations <u>1,000</u> Length (h) <u>10</u>	(3) View results  <p>The graph displays the count of various proteins over a 10-hour period. The y-axis is labeled 'Count' and the x-axis is labeled 'Time (h)'. Multiple colored lines (blue, orange, yellow, green, red, purple) show oscillatory behavior, with some lines peaking earlier than others.</p>
(4) Cluster config Nodes <u>100</u>	

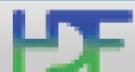
Simulation analysis



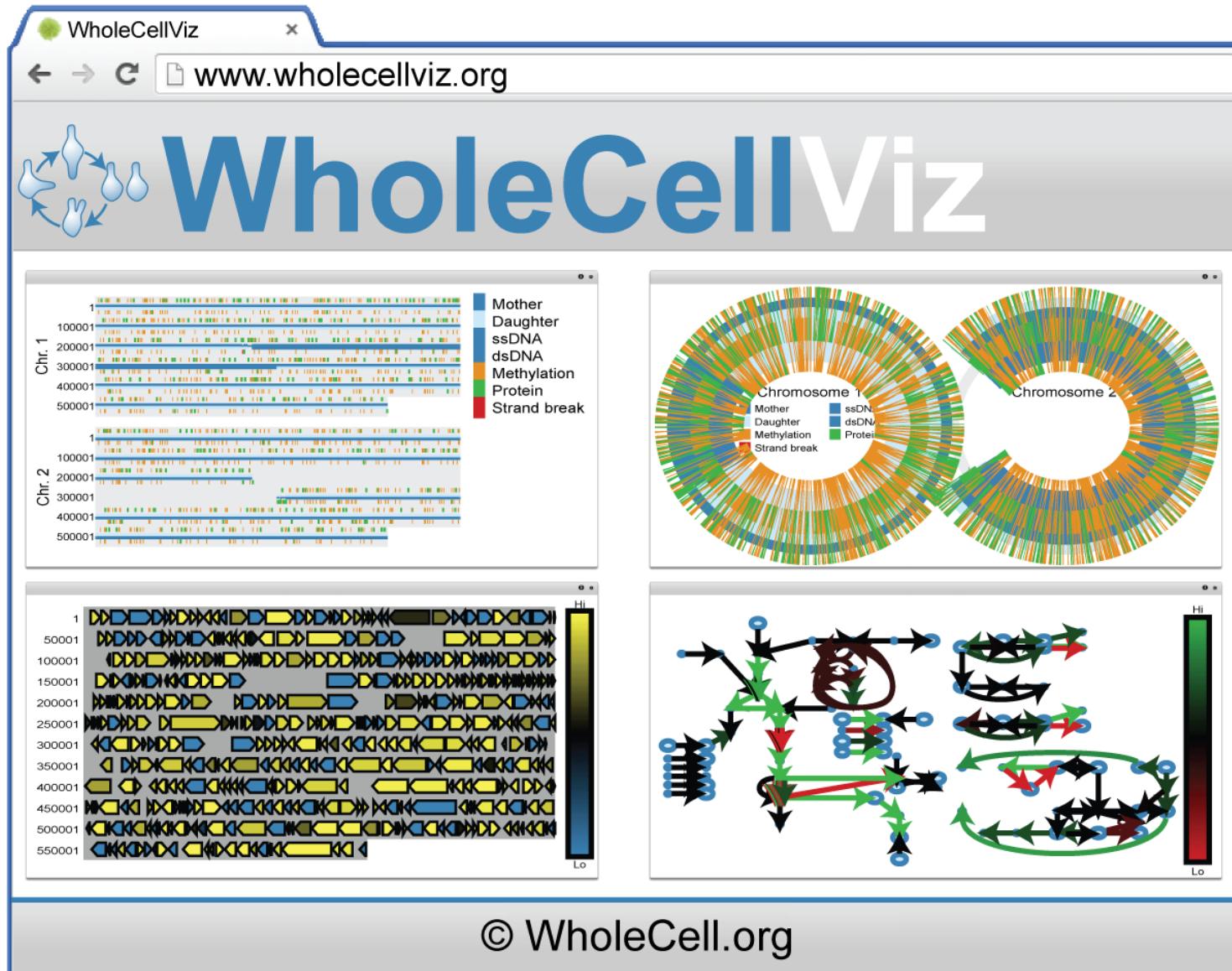
Simulation database

The screenshot shows a web browser window for 'WholeCellSimDB' at the URL www.wholecellsimdb.org/list/simulation. The page title is 'Simulations'. The table lists 12 entries, all for 'Mycoplasma genitalium' at version 2125, named 'Wild-type set #1', assigned to user 'Jonathan Karr' with ID 1.

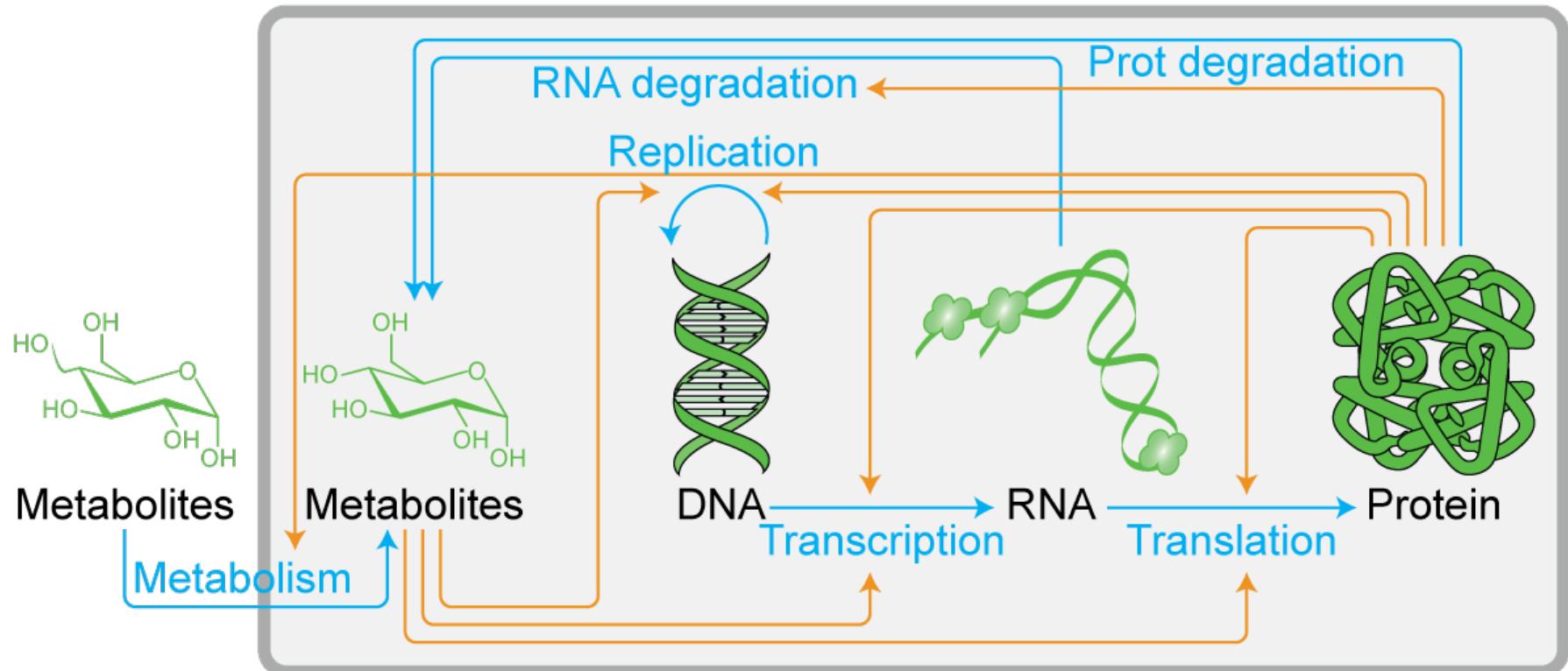
Organism Name	Ver	Simulation Name	No.	User
Mycoplasma genitalium	2125	Wild-type set #1	1	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	2	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	3	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	4	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	5	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	6	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	7	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	8	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	9	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	10	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	11	Jonathan Karr
Mycoplasma genitalium	2125	Wild-type set #1	12	Jonathan Karr

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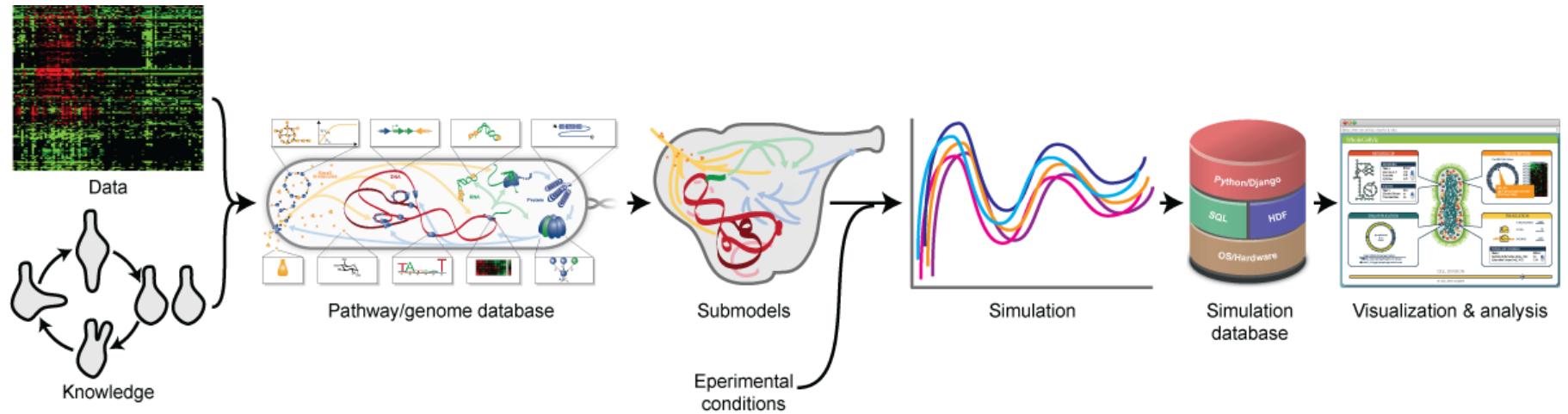
Simulation visualization



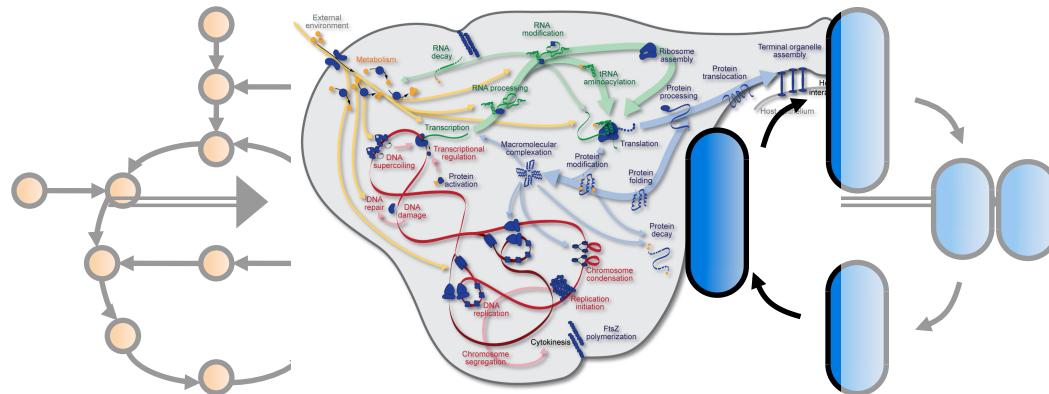
Benchmark model



Toward more comprehensive models



Data → Knowledge,
Models applications



Data ~~Whole-cell~~ knowledge models

Broadly predicts cell physiology

Integrates heterogeneous data and models

Guides bioengineering and medicine

Substantial effort needed to develop improved methods and tools

2016 whole-cell modeling school

April 3-8, 2016, Barcelona
wholecell.org/school-2016



Javier Carrera, Jonathan Karr, Maria Lluch-Senar, Luis Serrano
James Faeder, Tim Gardner, Anne-Claude Gavin, Peter Karp, Edda Klipp,
Julio Saez-Rodriguez, Ian Stansfield, Jorg Stelling, Bas Teusink, Barbara Di Ventura

Acknowledgements

Stanford
University

Stanford University

Markus Covert
Jayodita Sanghvi
Derek Macklin
Jared Jacobs



**Center for Regulatory
Genomics**

Luis Serrano
Maria Lluch-Senar
Veronica Llorens

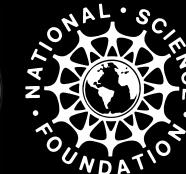


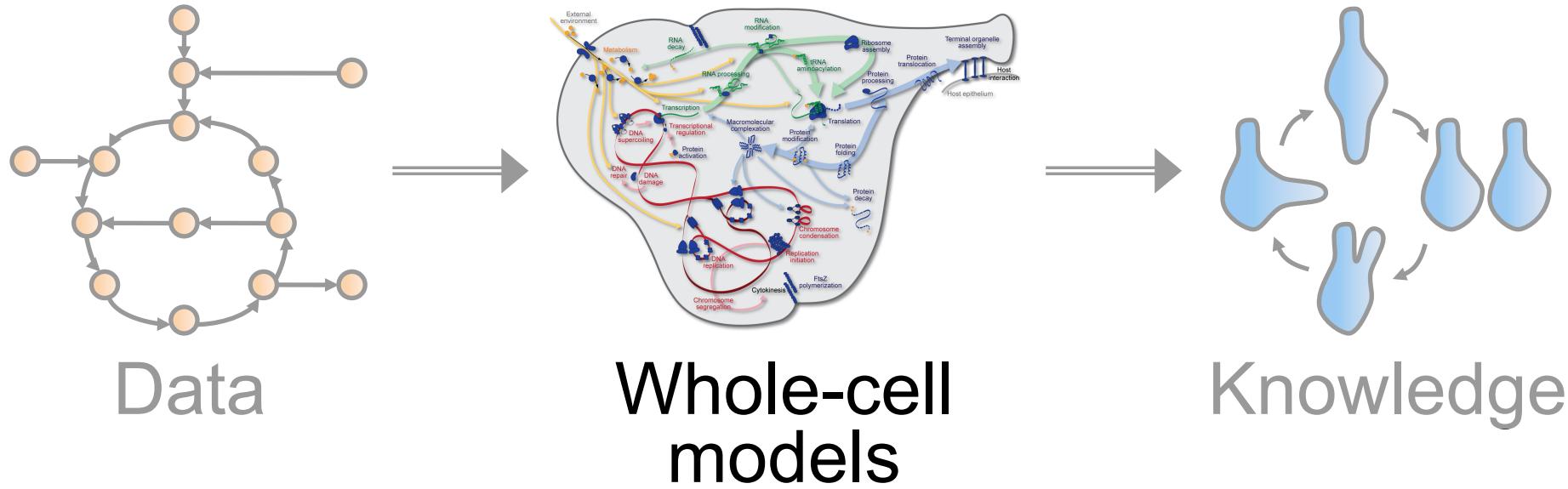
Mount Sinai
Arthur Goldberg

JAMES S.
MCDONNELL
FOUNDATION



THE PAUL G. ALLEN
FAMILY FOUNDATION





Broadly **predicts** cell physiology

Integrates heterogeneous data and
models

Guides bioengineering and medicine

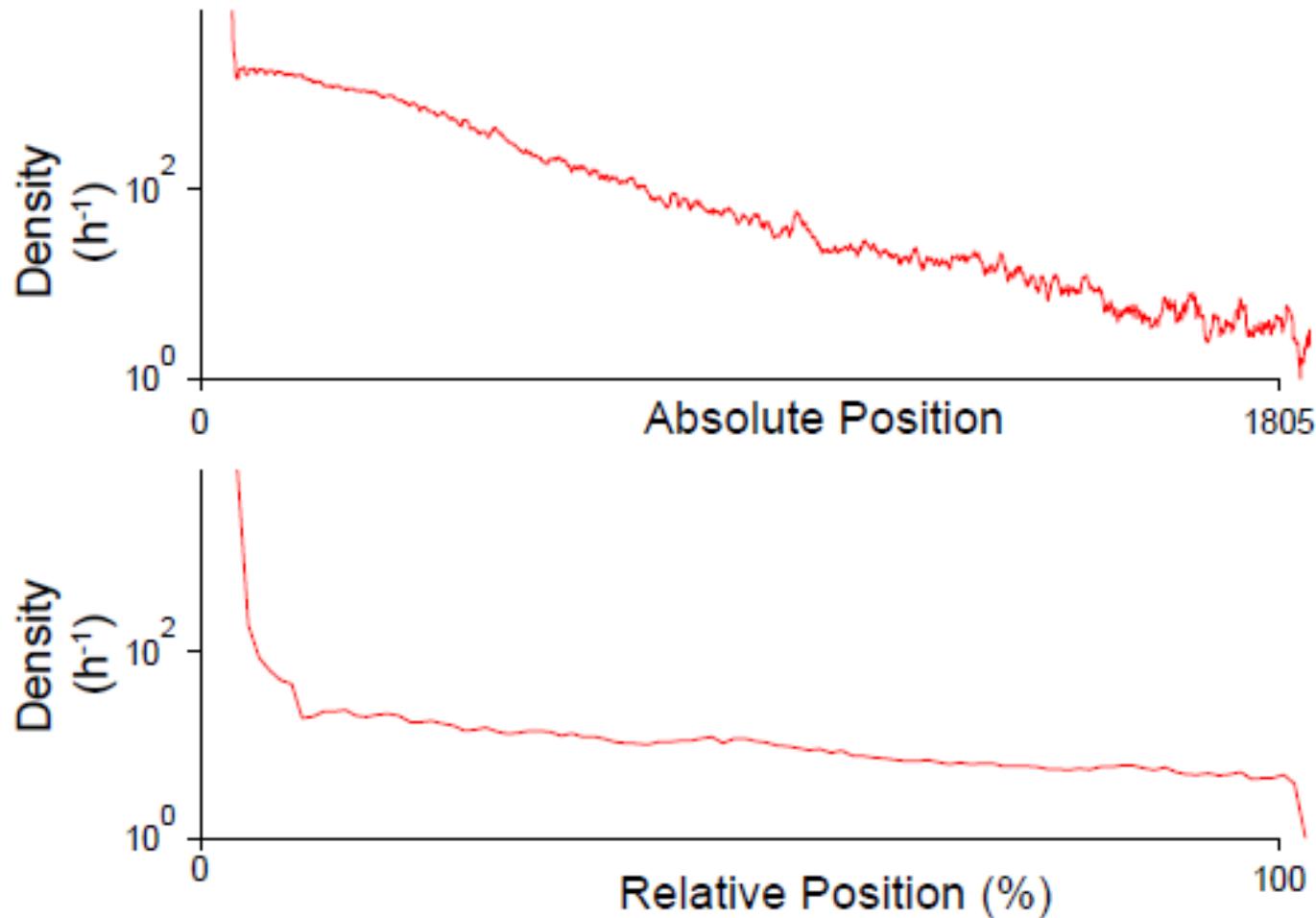
Recommended reading

- Karr JR et al. (2012) A Whole-Cell Computational Model Predicts Phenotype from Genotype. *Cell*, **150**, 389-401.
- Macklin DN, Ruggero NA, Covert MW (2014) The future of whole-cell modeling. *Curr Opin Biotechnol*, **28C**, 111-115.
- Shuler ML, Foley P, Atlas J (2012). Modeling a minimal cell. *Methods Mol Biol*, **881**, 573-610.
- Joyce AR, Palsson BØ (2007). Toward whole cell modeling and simulation: comprehensive functional genomics through the constraint-based approach. *Prog Drug Res* **64**, 267-309.
- Tomita M (2001). Whole-cell simulation: a grand challenge of the 21st century. *Trends Biotechnol* **6**, 205-10.
- Surovtsev IV et al. (2009) Mathematical modeling of a minimal protocell with coordinated growth and division. *J Theor Biol*, **260**, 422-9.

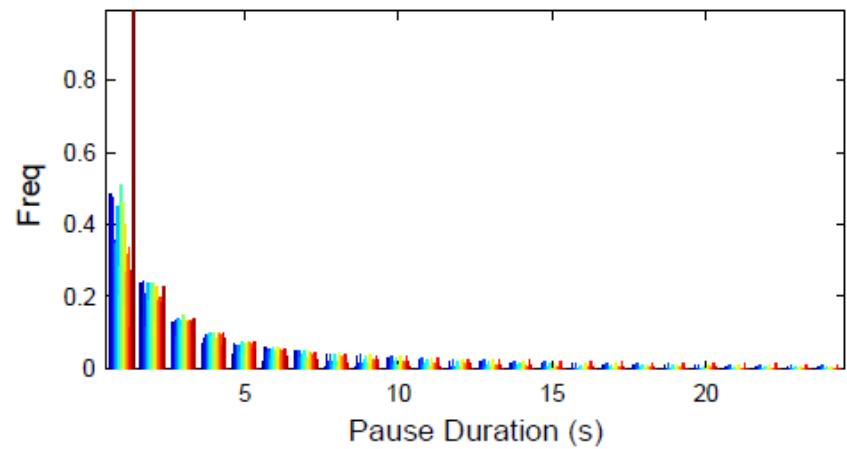
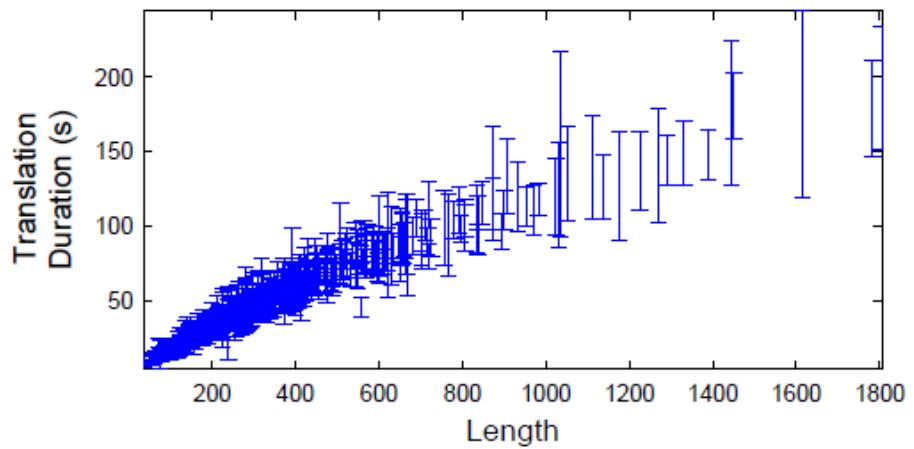
Recommended reading: FBA

- Thiele I et al. (2009). Genome-scale reconstruction of *Escherichia coli*'s transcriptional and translational machinery: a knowledge base, its mathematical formulation, and its functional characterization. *PLoS Comput Biol.* **5**, e1000312.
- Orth JD, Thiele I, Palsson BØ (2010). What is flux balance analysis? *Nat Biotechnol*, **28**, 245-8.
- Covert MW et al (2008). Integrated Flux Balance Analysis Model of *Escherichia coli*. *Bioinformatics* **24**, 2044–50.
- Covert MW et al (2004). Integrating high-throughput and computational data elucidates bacterial networks. *Nature*, **429**, 92-6.

Transcription



Translation



Growth is sensitive to enzyme expression

