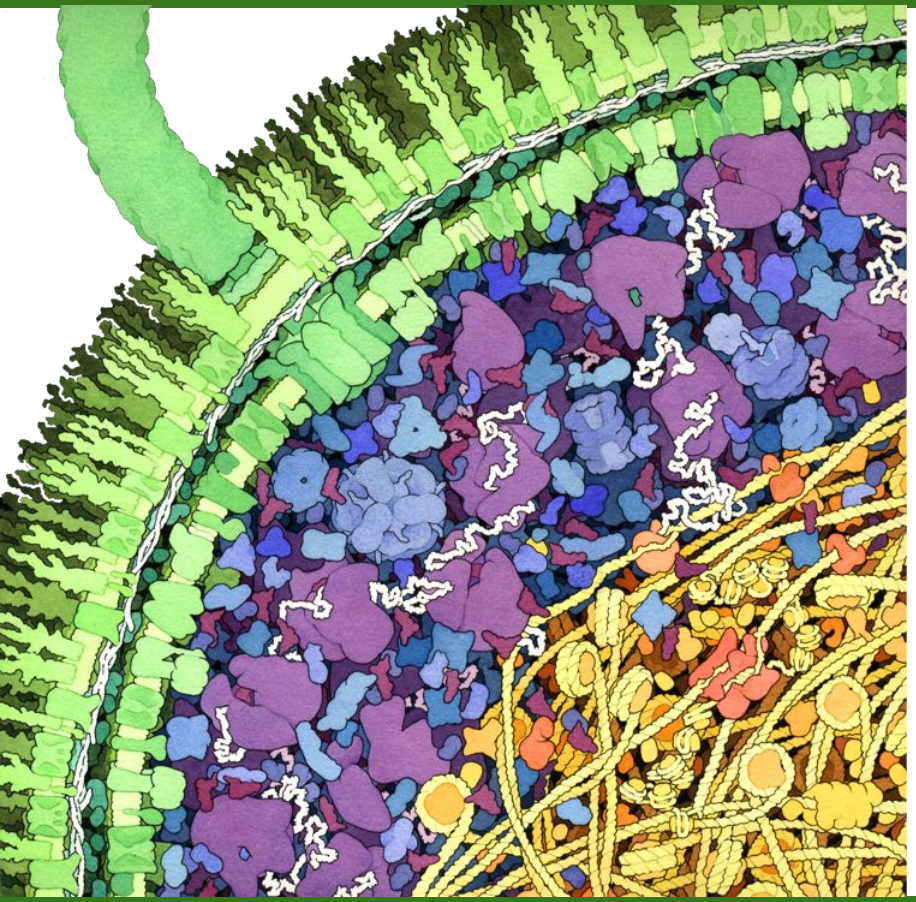
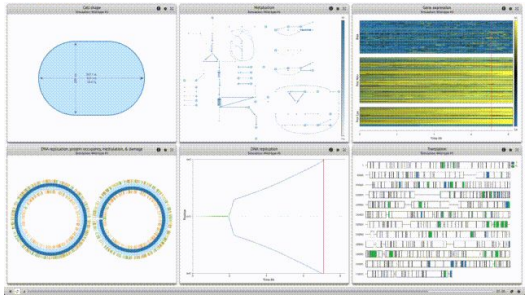


Group 31: An interactive, 3D visual reconstruction of a whole *E. coli* cell

Rohan Grover, Winnie Shi, Yuxuan Richard Xie

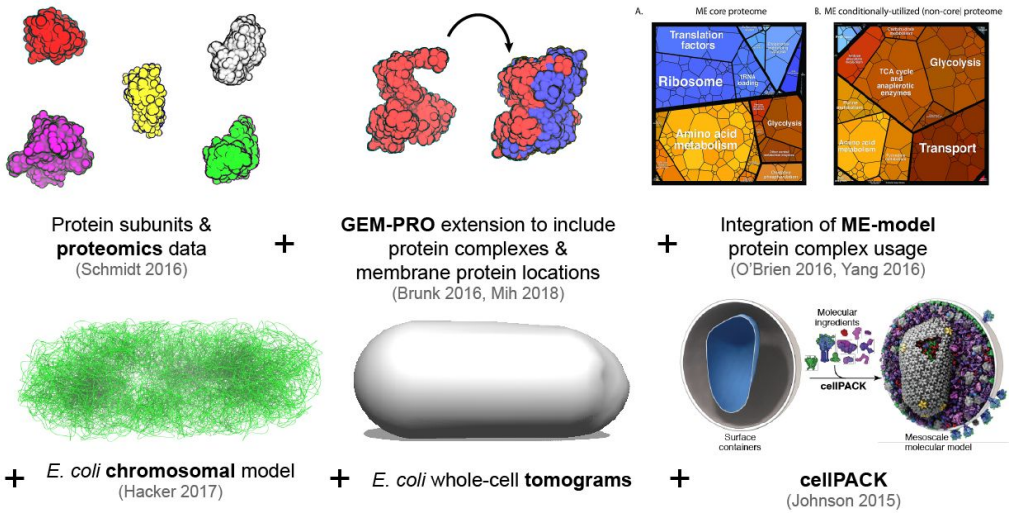
Whole Cell Models



- Help us understand:
  - Metabolic networks and expression networks
  - Relative concentrations and interactions of proteins
- Build more complex models in the future
- They are cool

Design Objectives

- Standardized pipelines for 3D reconstructions of *E. coli* and more
- Integrate high quality structural data along with:
  - Genomics
  - Proteomics
  - Metabolomics
- Interactive platform that enables easy search and manipulation

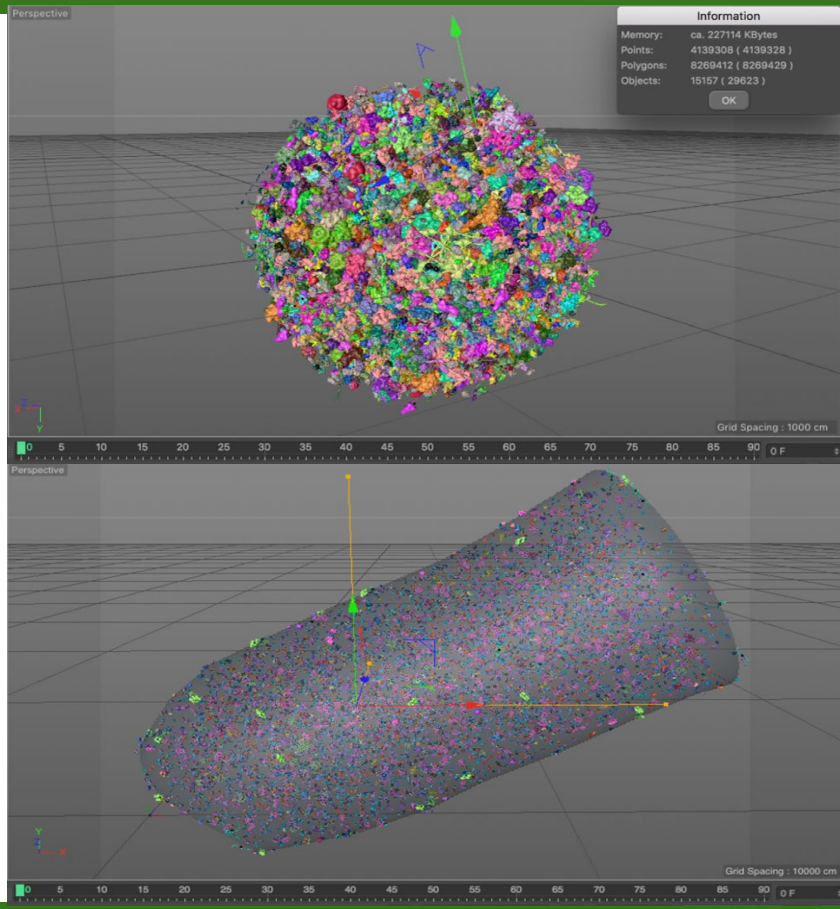


= visual reconstruction of an E. coli cell

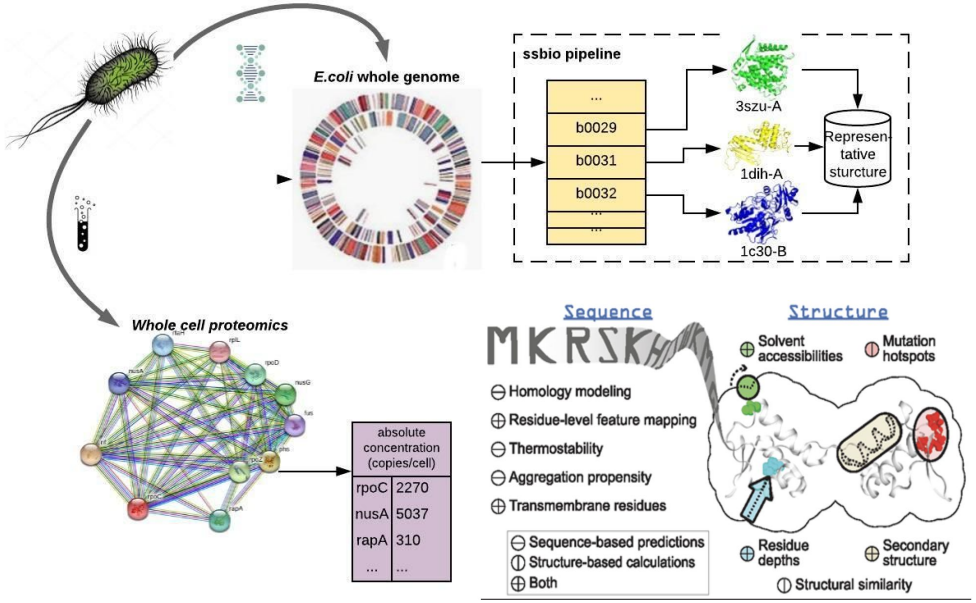
How do you curate and build a whole cell model of *E. coli*?

Prototype 1

Testing the feasibility of building a whole cell model for *E. coli*



Proteins and Structures



- ssbio (Python framework for **s**tructural **s**ystems **bio**logy): calculate and map structural information at genome scale
- Experimental protein concentrations from literature
- Building a repository of high quality protein structures with extensive annotations

What's in prototype 1?

Not protein complexes, just the protein components themselves
(D, E in the figure, randomly distributed throughout the cell)

Protein

Structure

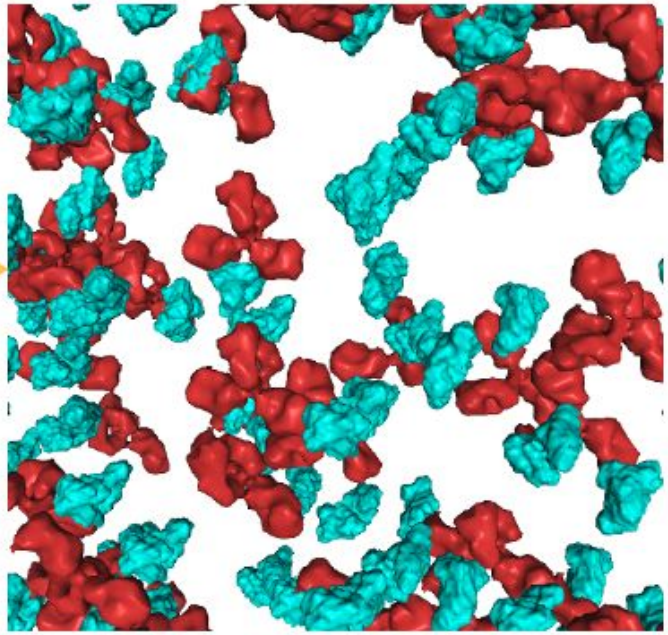
See:

- ssbio
- PDB
- Homology modeling

Proteomics measurements

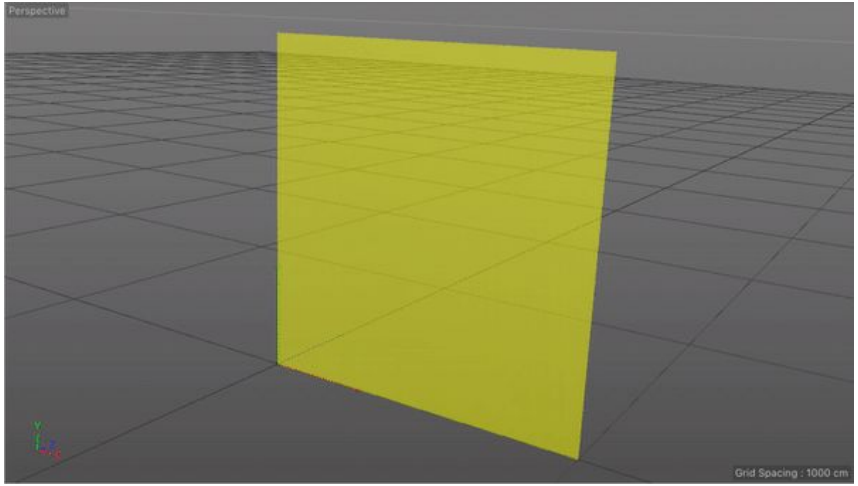
Copy number = 445

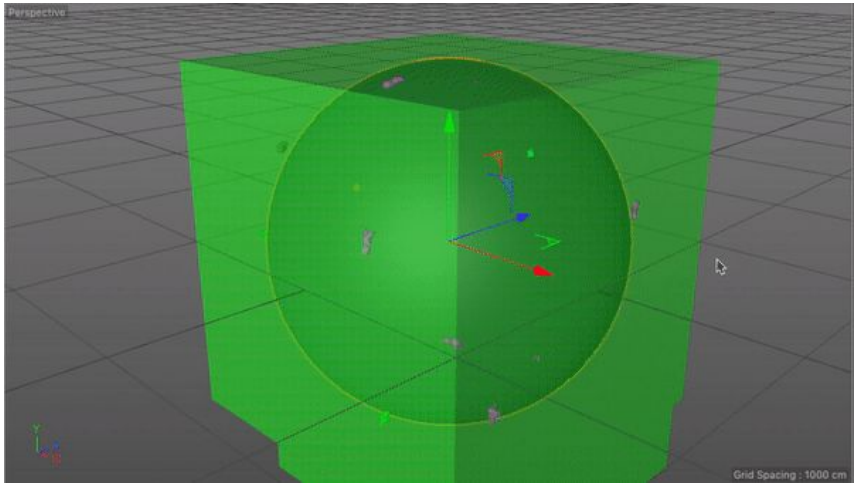
Concentration = Molarity = `(445/Avogadro's) / compartment_volume`



Gene	UniProt	NAME	Description	PDB	Structure_sourc	MOLARITY	Copy number	LOCALIZATION	INCLUDE
b0001	P0AD86	thrL	thr operon leader peptide					Cytosol	
b0002	P00561	thrA	Bifunctional aspartokinase	b0002_1_462_3c20-A_clean	SWISS-MODEL	2.59E-06	4375	Cytosol	x

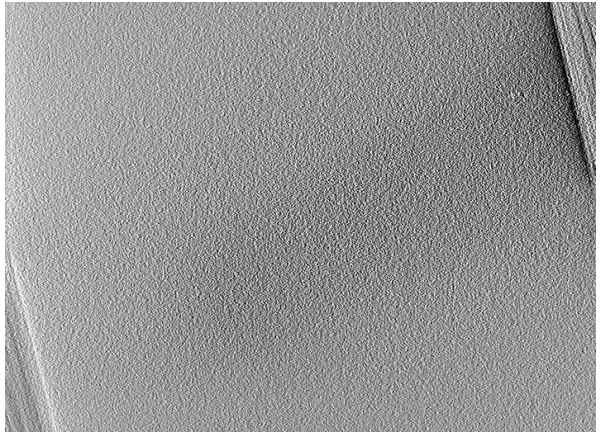
Integration Using cellPACK

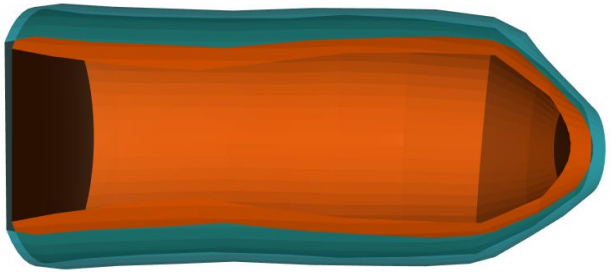




Membranes

- Built a rough object of the *E. coli* inner and outer membranes from tomograms using IMOD
- Is not a whole cell
- Volume of cell in tomograms small compared to average sizes from literature
    - Dimensions: ~1μm x ~0.4 μm



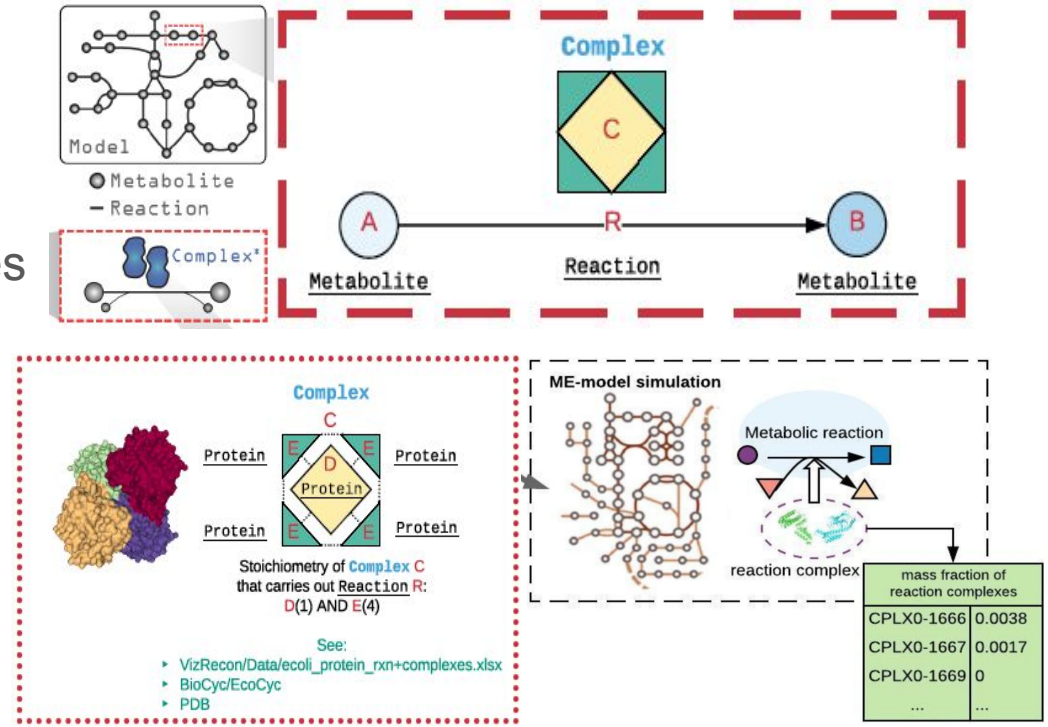


Prototype 2

A more complete representation of *E. coli* and final model for this senior design project

Addition of Complexes

- Macromolecule Expression model (ME-model) determines stoichiometry of complex structures
- Formation rate (flux) of these complexes are obtained by ME-model simulations
- Homology models used for unknown structures

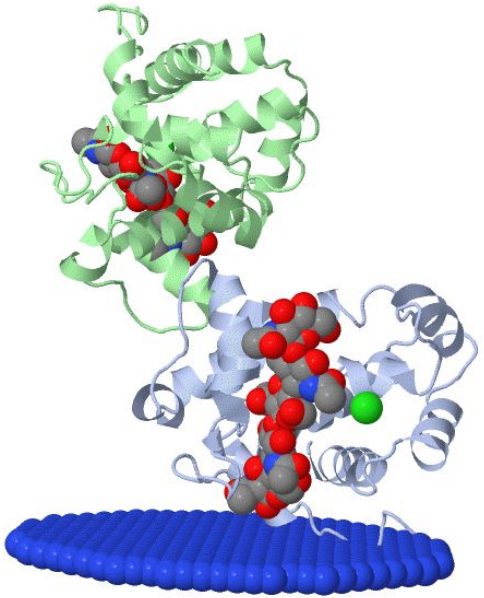


Computation of Complex Copy Numbers

- Complex copy numbers were estimated using ME-model mass fraction ratios
- Copy Numbers were determined using these assumptions:
    1. Proteins want to be in complexes
    2. There is a “limiting” protein
- The “limiting protein” is arbitrarily chosen to best maximize total number of complexes made

Membrane Proteins and Complexes

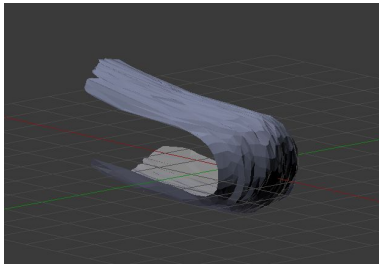
- cellPACK can pack membrane proteins by locking parameters
- Orientations of Proteins in Membranes (OPM) contains high-quality annotations for proteins
  - Alternatives: Transmembrane Helix Prediction using Hidden Markov Models (TMHMM), UniProt
- Lab member developed algorithm to confirm quality

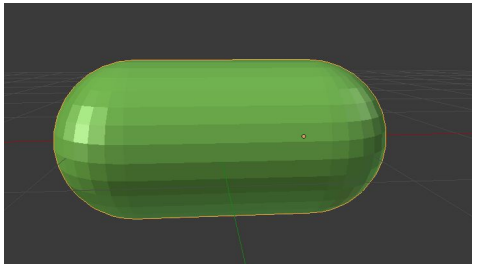


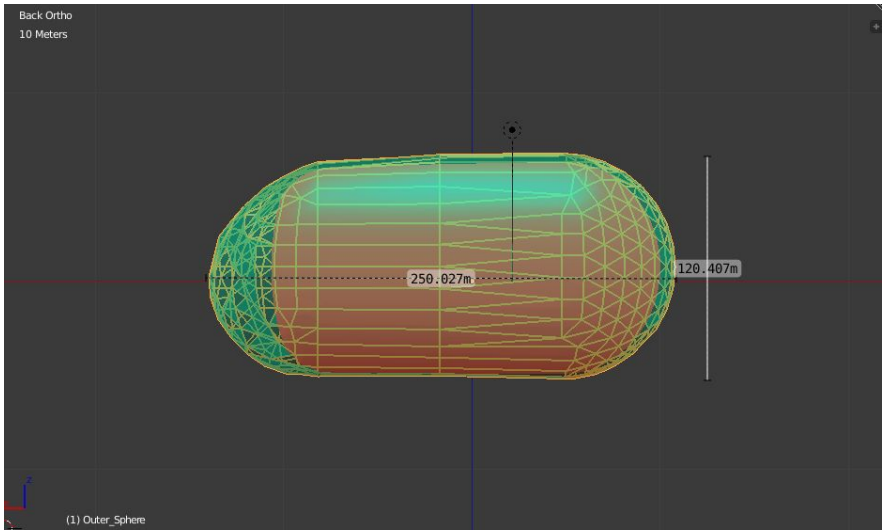
FirstGlance in Jmol

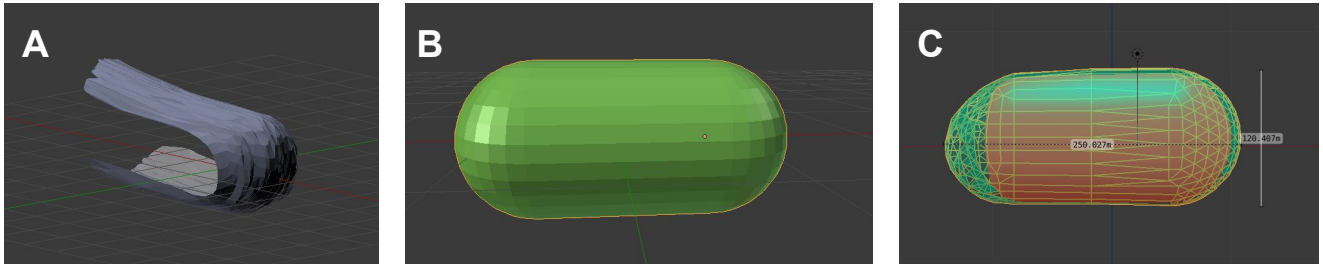
Membrane Improvements

- Made “pill” from dimensions in literature
- Total Volume: 2.2μm^3
- Drawbacks:
  - Periplasm Volume to whole cell volume: 19% (compared to 8% from experimental data)



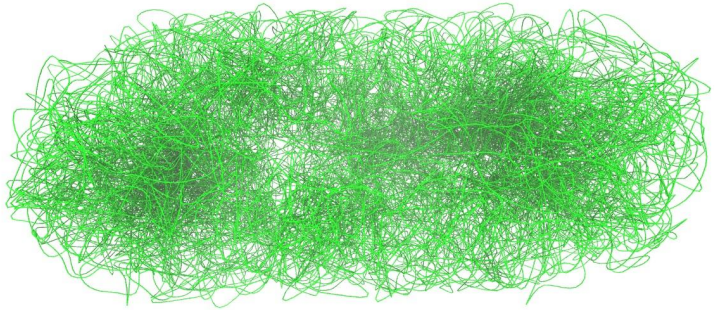


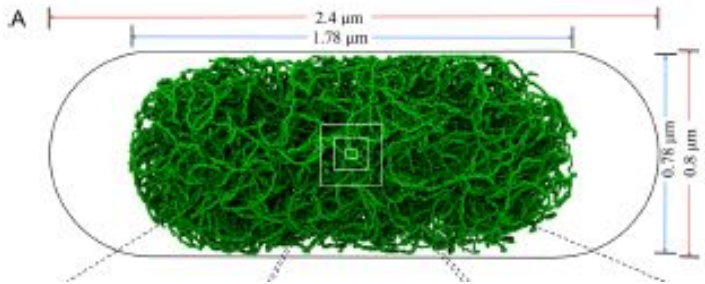



A
B
C
250.027m
120.407m

Addition of Chromosome

- 500 base pairs per bead (bpb) model of an *E. coli* chromosome
- Mid-cell origin of replication
- Shared with us by William Hacker and Professor Adrian Elcock

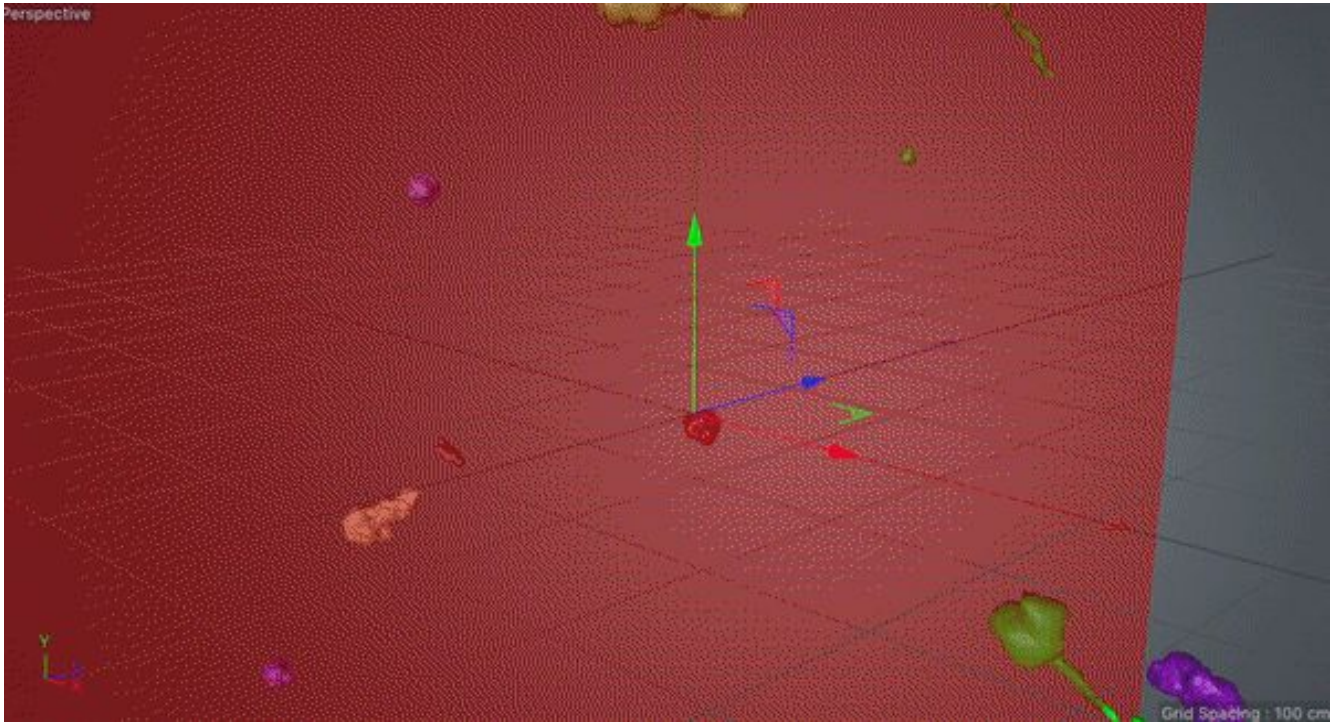




Final “Menu” and Recipe

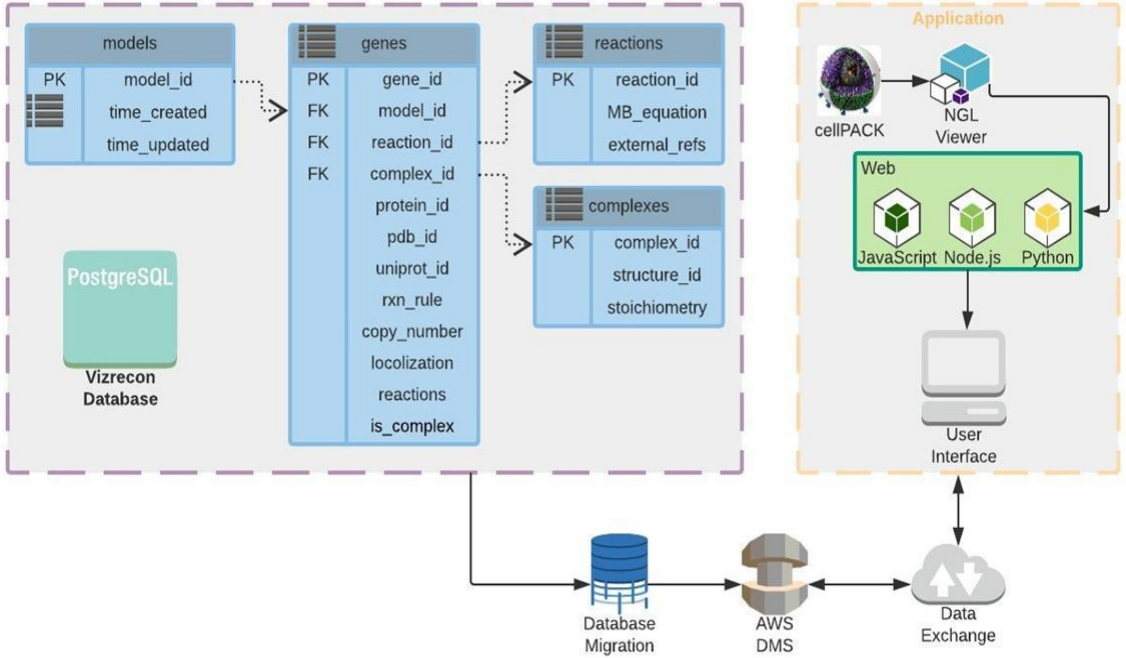
	B	C	D	E	F	G	H	I	J	K	L	M	N	O	
1	UniProt	NAME	Description	Structure_ID	PDB	Structure_sc	MW	MOLARITY	Copy_numb	Localization	Localization_	INCLUDE	COLOR	NOTES	model_
2	P0AD86	thrL	thr operon leader	REP-E00001	E00001_clean-X	metaTASSER	2138.4426			Cytosol	EchoLOCATION	predicted			iML151
3	P00561	thrA	Bifunctional aspa	REP-E00002	E00002_clean-X	metaTASSER	89119.2202	4.11E-06	4375	Cytosol	iJL1678	x			iML151
4	P00547	thrB	Homoserine kina	REP-1_308_4rpf	b0003_1_308_4r	SWISS-MODEL	33623.2905	0	844	Cytosol	iJL1678				iML151
5	P00934	thrC	Threonine syntha	REP-1vb3	1vb3-A_clean.pd	Protein Data Ban	47113.2536	1.28E-05	13676	Cytosol	iJL1678	x			iML151
6	P75616	yaaX	Uncharacterized	REP-E00005	E00005_clean-X	metaTASSER	11354.7994			Periplasm	EchoLOCATION	predicted			iML151
7	P0A8I3	yaaA	UPF0246 protein	REP-5caj	5caj-A_clean.pdb	Protein Data Ban	29585.4538	5.75E-07	613	Cytosol	EchoLOCATION	x			iML151
8	P30143	yaaJ	Uncharacterized	REP-E00007	E00007_clean-X	metaTASSER	51662.291			Inner_Membrane	iJL1678				iML151
9	P0A870	talB	Transaldolase B	REP-4s2b	4s2b-A_clean.pd	Protein Data Ban	35218.8107	8.68E-06	9247	Cytosol	iJL1678	x			iML151
10	P0AF03	mog	Molybdopterin ad	REP-1di6	1di6-A_clean.pdb	Protein Data Ban	21222.105	1.45E-07	155	Cytosol	iJL1678	x			iML151
11	P0AC98	satP	Succinate-acetat	REP-E00010	E00010_clean-X	metaTASSER	20070.6303			Cytosol	Arbitrary assignment				iML151
12	P75617	yaaW	UPF0174 protein	REP-E00011	E00011_clean-X	metaTASSER	26664.5763			Cytosol	EchoLOCATION	predicted			iML151
13	P28697	mbiA	Uncharacterized protein MbiA				17637.1282			Cytosol	Arbitrary assignment				iML151
14	P28696	yaaI	UPF0412 protein	REP-E00012	E00012_clean-X	metaTASSER	14481.418			Periplasm	EchoLOCATION	predicted			iML151
15	P0A6Y8	dnaK	Chaperone prote	REP-4jne	4jne-A_clean.pdb	Protein Data Ban	69114.1284	1.70E-05	18133	Cytosol	iJL1678	x			iML151
16	P08622	dnaJ	Chaperone prote	REP-4_348_4j80	b0015_4_348_4j	SWISS-MODEL	41099.8758	7.02E-07	748	Cytosol	iJL1678	x			iML151
17	P0CF91	insL1	Putative transpos	REP-INSL2_ECC	INSL2_ECOLI_m	I-TASSER	40908.3273			Cytosol	Arbitrary assignment				iML151
18	P33236	mokC	Regulatory protei	REP-E00016	E00016_clean-X	metaTASSER	7742.206			Periplasm	EchoLOCATION	predicted			iML151
19	P13738	nhaA	Na(+)/H(+) antipo	REP-1zcd	1zcd-A_clean.pd	Protein Data Ban	41355.1815			Inner_Membrane	iJL1678				iML151
20	P0A9G2	nhaR	Transcriptional a	REP-6_298_3k1	b0020_6_298_3k	SWISS-MODEL	34284.0399			Cytosol	EchoLOCATION	predicted			iML151
21	P0CF25	insB1	Insertion element	IS1 1 protein InsB			19564.3535			Cytosol	Arbitrary assignment				iML151
22	P0CF07	insA1	Insertion element	IS1 1 protein InsA			9868.2797			Cytosol	Arbitrary assignment				iML151
23	P0A7U7	rpsT	30S ribosomal pr	REP-5afi	5afi-t_clean.pdb	Protein Data Ban	9684.2814	2.19E-05	31222	Cytosol	iJL1678	x			iML151
24	P75620	yaaY	Uncharacterized	REP-E00023	E00023_clean-X	metaTASSER	7890.2841			Inner_Membrane	EchoLOCATION	predicted			iML151

cellPACK

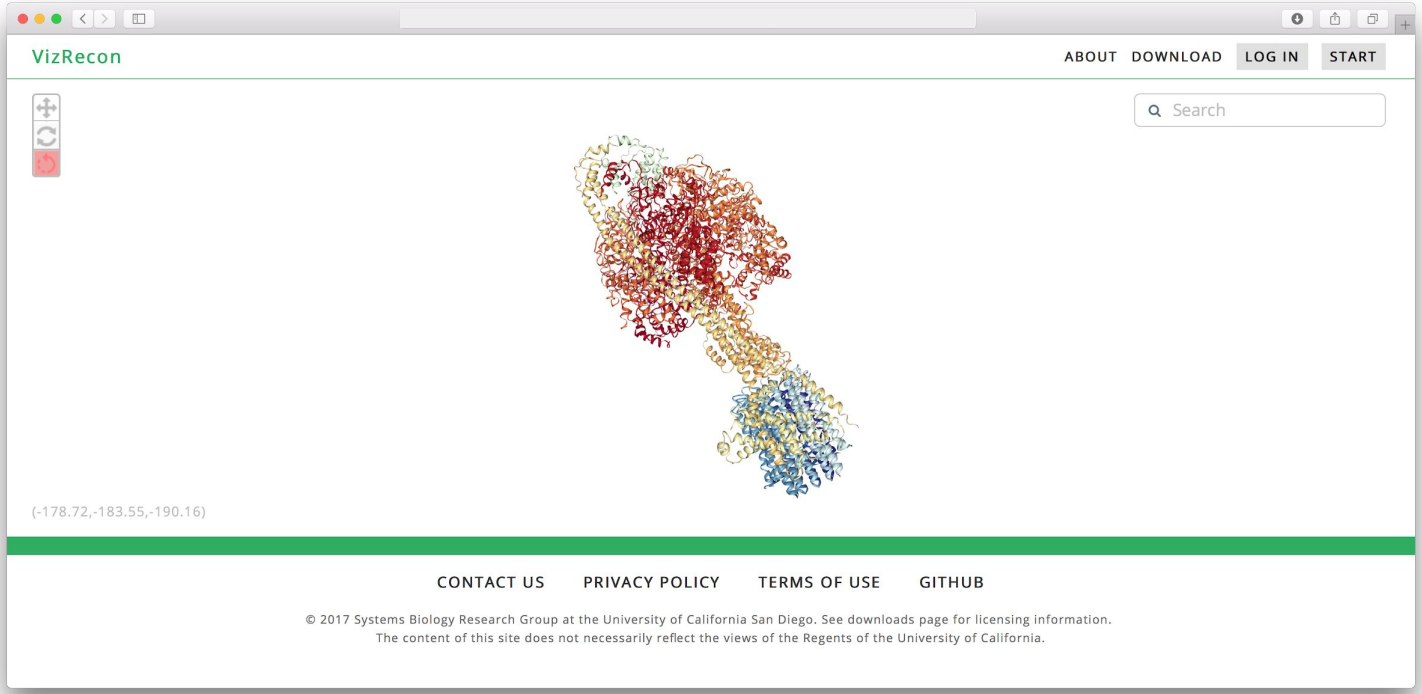


How do we display the pipeline and model?

Integration of the components



Building an interactive platform



Conclusions and Future Goals

- Created pipeline to link structural and spatial info with “omics” data
- Able to generate appropriate complexes and membrane structures
- Still in progress of packing final model
- Working on finding an efficient way of displaying the model using NGL
- Improve the detail of visualization (i.e. interactions between components, more constraints for localizations, thermodynamics, kinetics, etc.)
- Update model to include more experimentally determined components
- Apply the pipeline and create reconstructions to other organisms
- Integrate metabolic networks to simulate directly on the 3D model with the aid of molecular dynamics (MD)

Acknowledgements

- Dr. Nathan Mih
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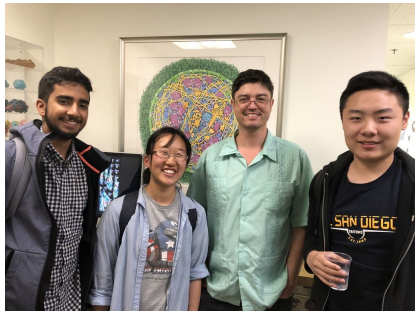
- Anjulie Agrusa
- Marissa Keller

- Dr. Brett Barbaro
- Dr. Alex Rose
- Dr. Bernhard Ø. Palsson



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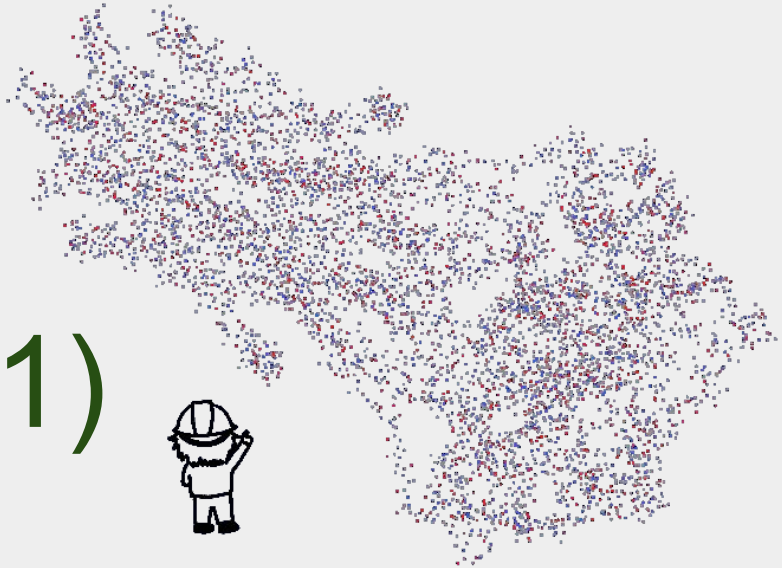
Questions?
(we are Group 31)

((for those who are wondering))



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- **The quantitative and condition-dependent *Escherichia coli* proteome 2016**, Alexander Schmidt, Karl Kochanowski, Silke Vedelaar, Erik Ahrné, Benjamin Volkmer, Luciano Callipo, Kèvin Knoops, Manuel Bauer, Ruedi Aebersold & Matthias Heinemann
- **cellPACK: a virtual mesoscope to model and visualize structural systems biology 2015**, Graham T Johnson, Ludovic Autin, Mostafa Al-Alusi, David S Goodsell, Michel F Sanner& Arthur J Olson