CryoBench

Diverse and Challenging Datasets for the Heterogeneity Problem in Cryo-EM

NEURAL INFORMATION PROCESSING SYSTEMS


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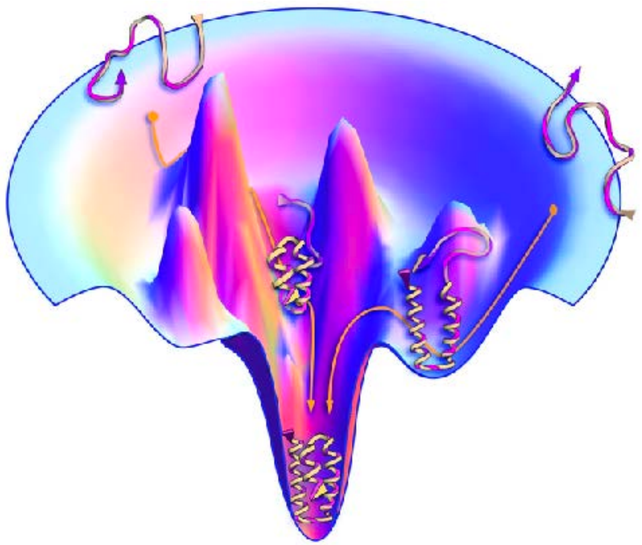
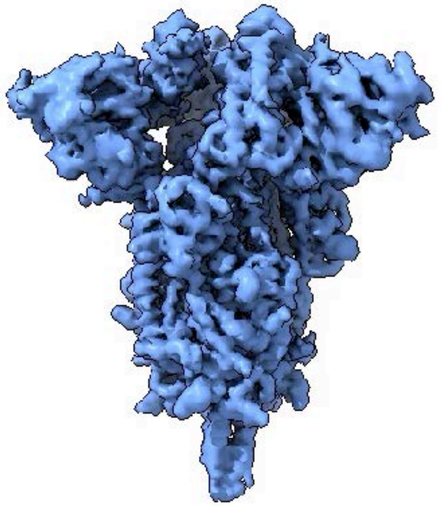
What is biomolecular heterogeneity?

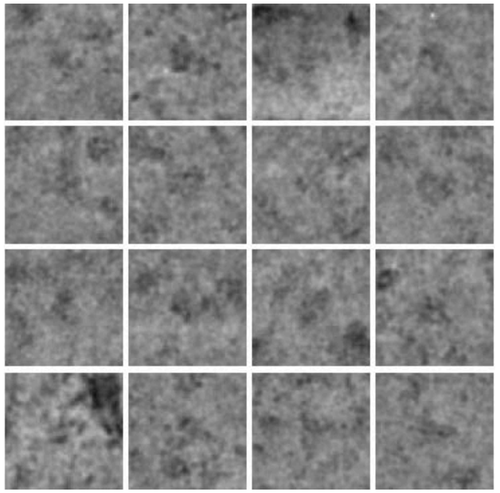
- Protein and other biomolecules form large, dynamic complexes that carry out essential biological functions.
- Existing tools for modeling structure, such as AlphaFold, are limited in their ability to predict different conformations or compositional states.
- **Cryo-electron microscopy (cryo-EM)**, in contrast, is a technique providing a unique opportunity to study biomolecules in near-native conformational states *from experimented data*.



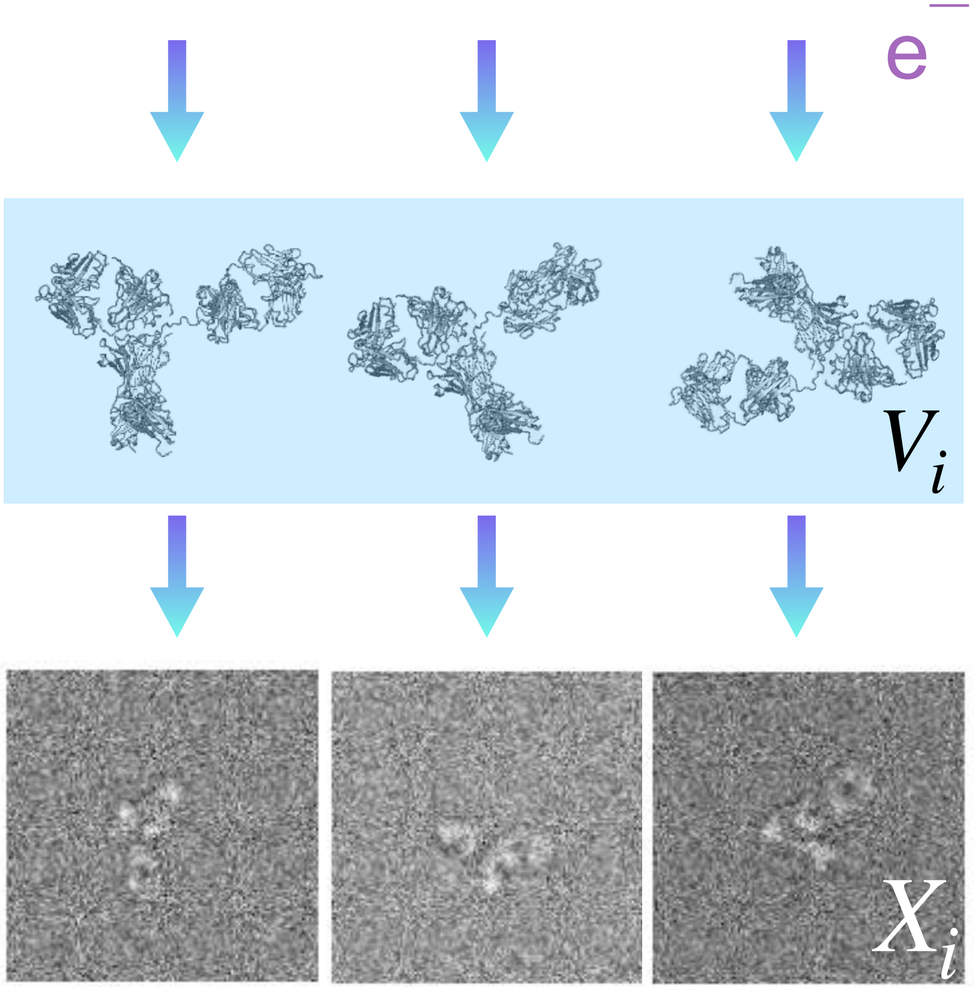
Dill & Maccallum, Science 2012



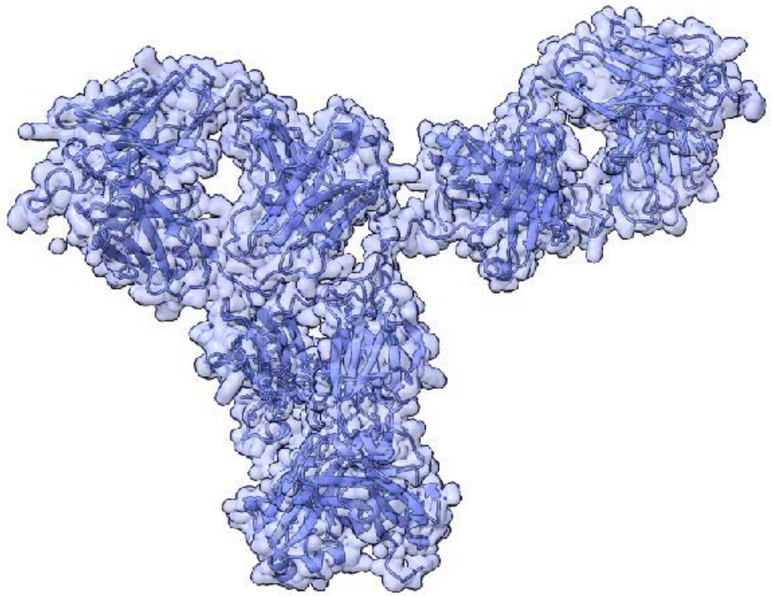
SARS-CoV-2 Spike Protein
Dataset: Walls et al 2020



Cryo-EM and 3D reconstruction



10^4 – 10^7 images
SNR ≈1 to 5%



$$V : \mathbb{R}^3 \to \mathbb{R}$$

Electron Scattering Potential

Reconstruction as an inference problem

Image Formation Model

$$Y_i = C_i * P_{\phi_i} V_i + \eta_i$$

CTF

Projection

Pose

Gaussian noise

$$\phi_i \in \mathrm{SO}(3) \times \mathbb{R}^2$$
$$\eta_i \sim \mathcal{N}$$

Goal: Estimate V and poses $\{\phi_i\}$ typically with maximum likelihood techniques

Motivation

1. Reconstruction of molecular movies is now possible

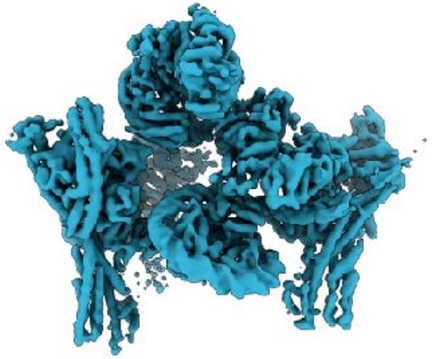
3DVA

Na_v 1.7 ion channel
[EMPIAR-10261]

3DFlex

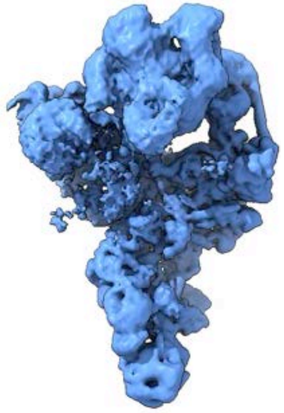
$\alpha V\beta 8$ integrin
[EMPIAR-10345]

DynaMight



Inner kinetochore
[EMPIAR-11910]

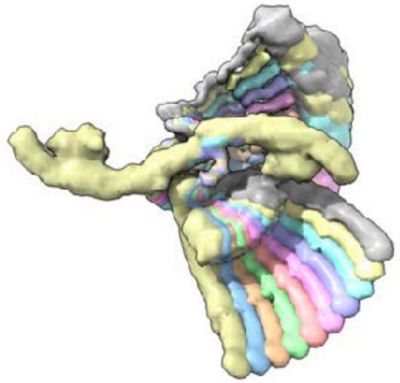
CryoDRGN



Pre-catalytic spliceosome
[EMPIAR-10180]

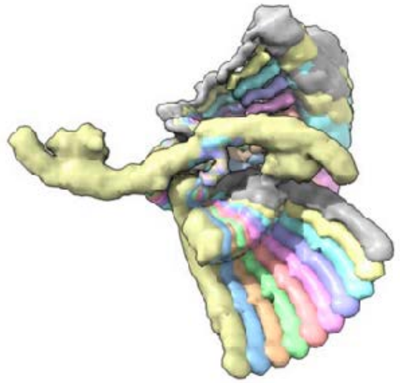
Motivation

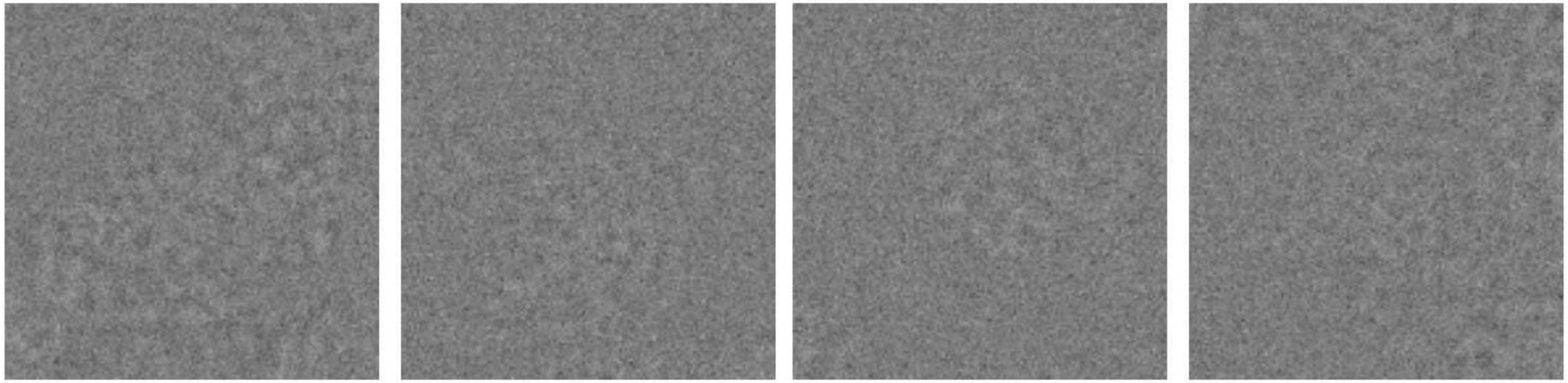
1. Reconstruction of molecular movies is now possible
2. Methods often use simple toy motions for validation and comparison with other approaches



Motivation

1. Reconstruction of molecular movies is now possible
2. Methods often use simple toy motions for validation and comparison with other approaches
3. **No ground truth** exists for real data; Evaluation currently requires **benchmarking-by-eye**





CryoBench ❄️🪑: Contributions

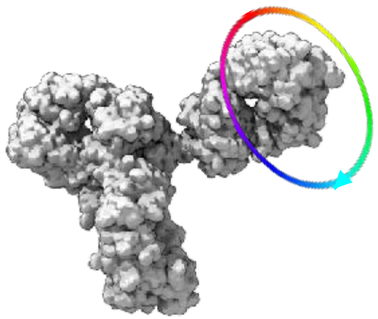
1. Design new **synthetic datasets** with challenging forms of heterogeneity to motivate new tasks and methods development
2. Introduce **metrics** for quantitative comparison of methods for heterogeneity reconstruction
3. **Benchmark** existing state-of-the-art methods

CryoBench ❄️🪑: Datasets

Conformational Heterogeneity

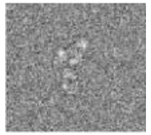
Compositional Heterogeneity

Diagnostic

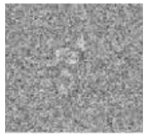


IgG-1D

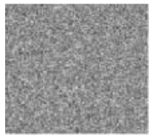
IgG-1D

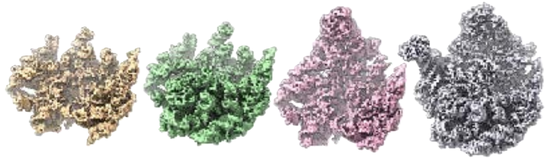


IgG-1D-noisier



IgG-1D-noisiest

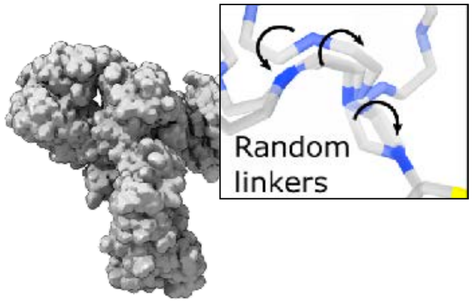




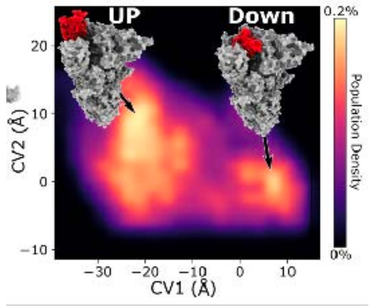
16 structures

Ribosembly

Challenging

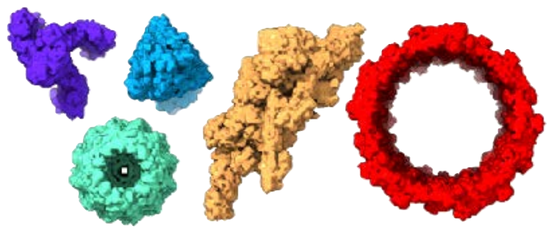


IgG-RL



~46k structures

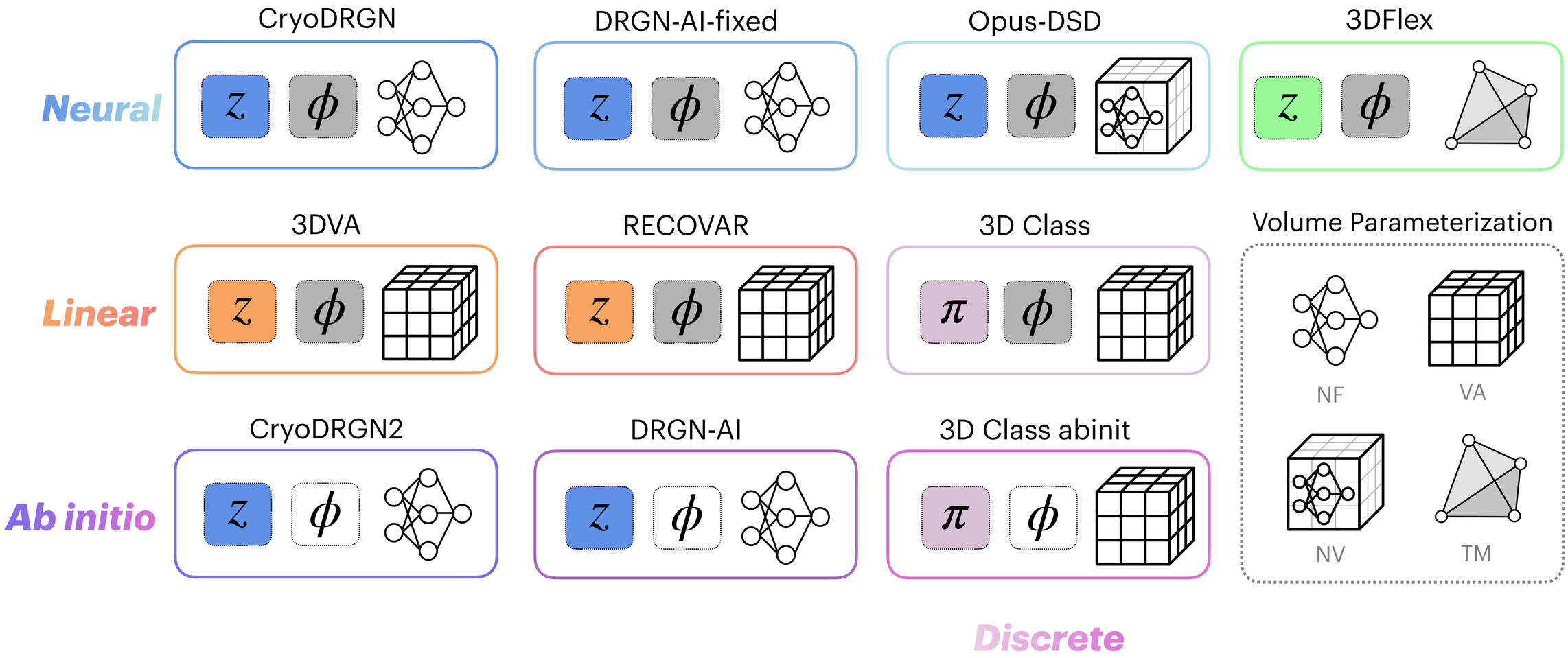
Spike-MD



100 structures

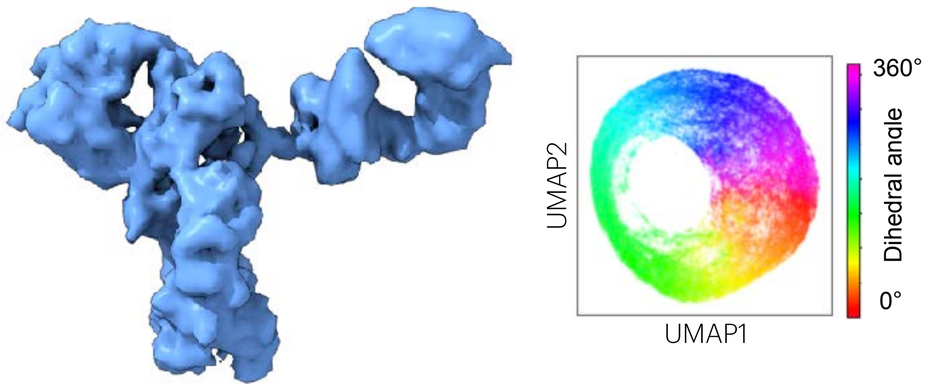
Tomotwin-100

Methods

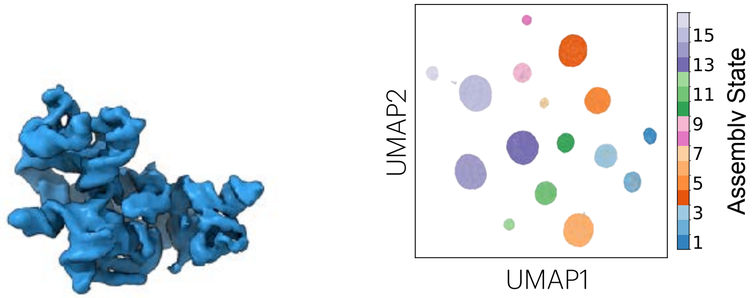

Neural
CryoDRGN
DRGN-AI-fixed
Opus-DSD
3DFlex
Linear
3DVA
RECOVAR
3D Class
Volume Parameterization
NF
VA
NV
TM
Ab initio
CryoDRGN2
DRGN-AI
3D Class abinit
Discrete

Qualitative Results

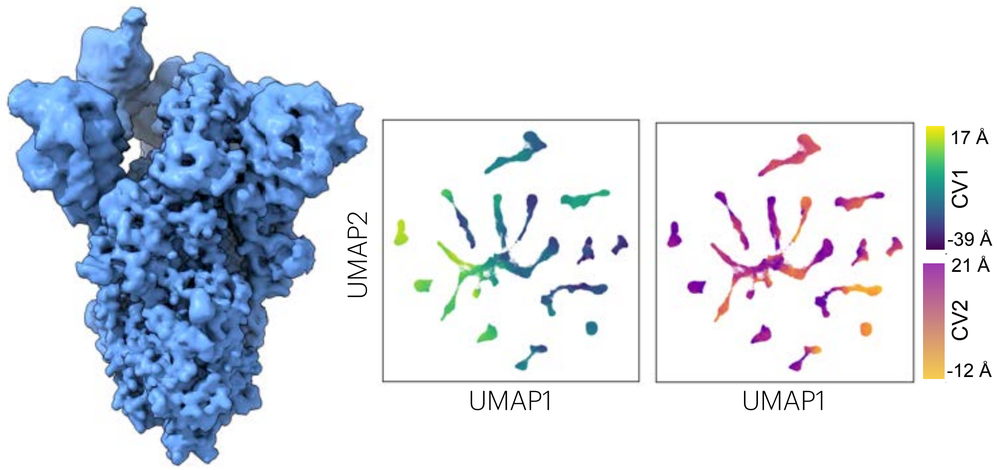
IgG-1D



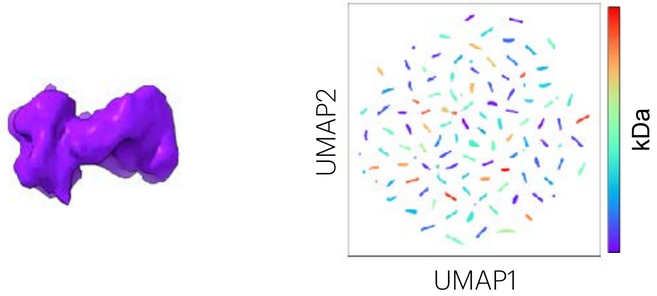
Ribosembly



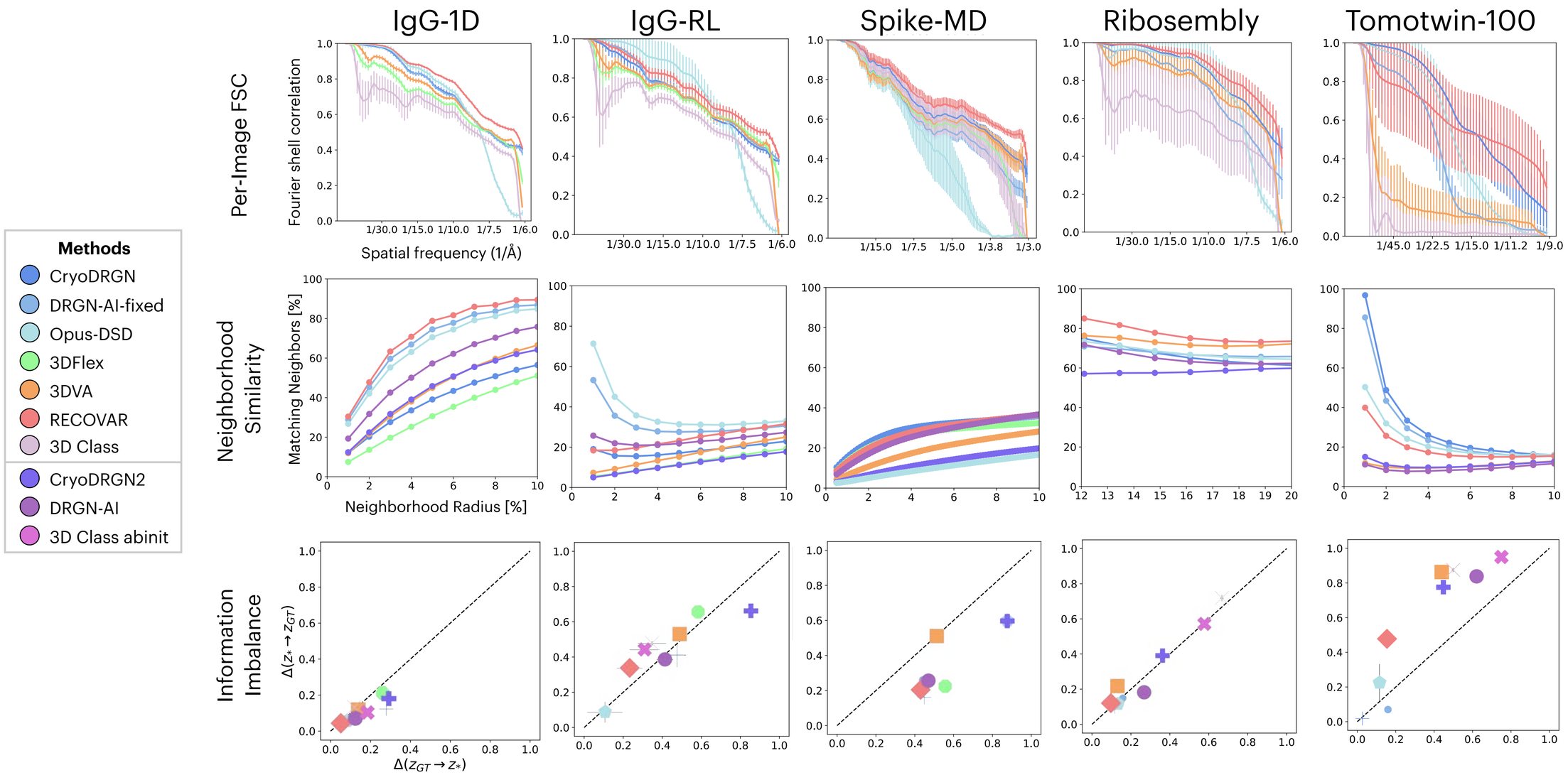
Spike-MD



Tomotwin-100



Quantitative Results

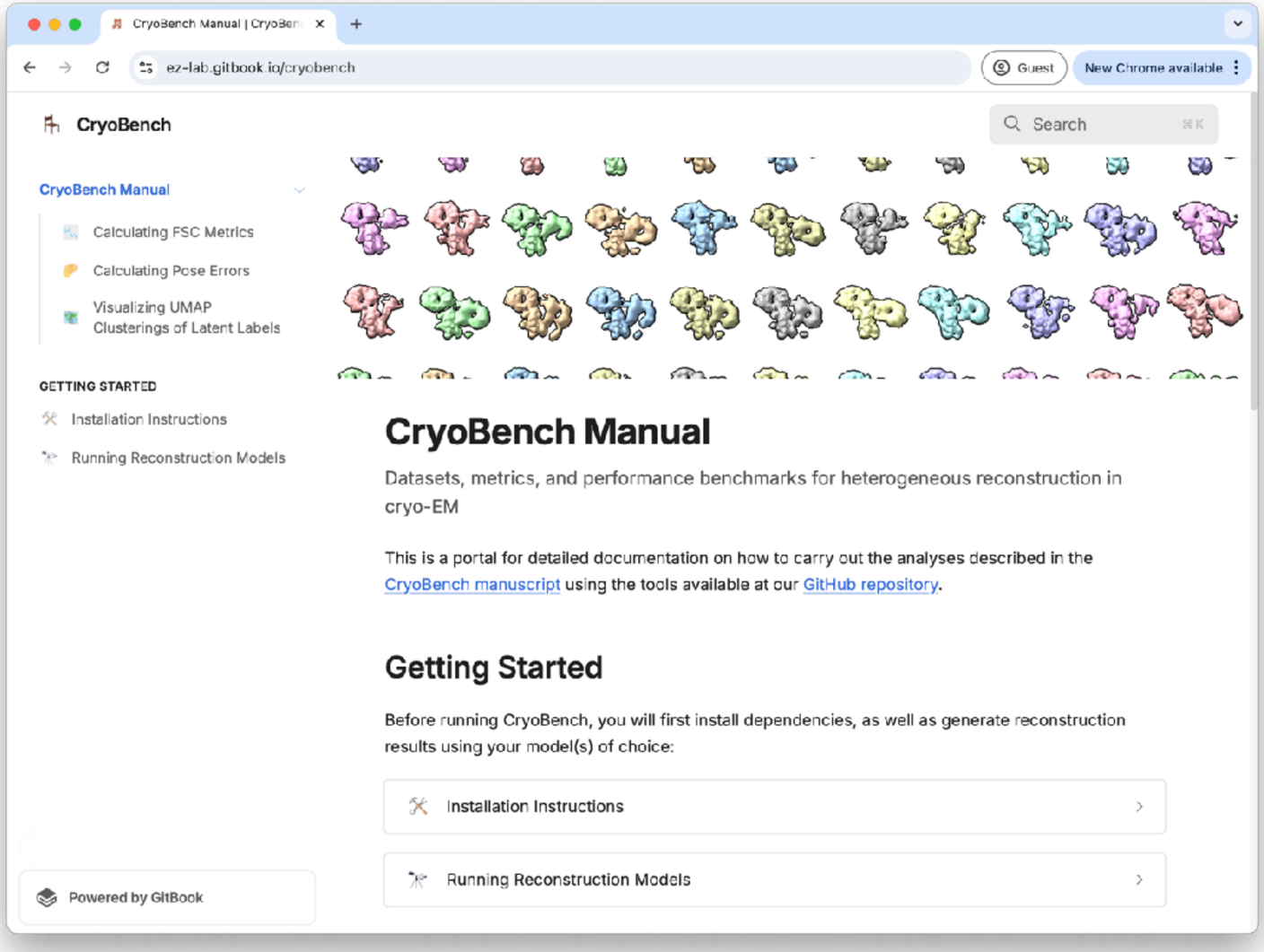


Quantitative Results

Method	IgG-1D		IgG-RL		Ribosembly		Tomotwin-100		Spike-MD	
	Mean (std)	Median	Mean (std)	Med	Mean (std)	Med	Mean (std)	Med	Mean (std)	Med
CryoDRGN	0.351 (0.028)	0.356	0.331 (0.016)	0.333	0.412 (0.023)	0.415	**0.316 (0.046)**	**0.321**	0.340 (0.009)	0.340
DRGN-AI-fixed	0.364 (0.002)	0.364	0.348 (0.012)	0.350	0.372 (0.032)	0.375	0.202 (0.044)	0.207	0.301 (0.012)	0.303
Opus-DSD	0.335 (0.026)	0.339	0.343 (0.016)	0.346	0.362 (0.083)	0.382	0.237 (0.049)	0.251	0.229 (0.027)	0.242
3DFlex	0.335 (0.003)	0.335	0.337 (0.007)	0.337	-	-	-	-	0.304 (0.011)	0.306
3DVA	0.349 (0.004)	0.350	0.333 (0.014)	0.335	0.375 (0.038)	0.375	0.088 (0.04)	0.077	0.324 (0.010)	0.323
RECOVAR	**0.386 (0.005)**	**0.388**	**0.363 (0.011)**	**0.363**	**0.429 (0.018)**	**0.432**	0.258 (0.109)	0.254	**0.362 (0.011)**	**0.365**
3D Class	0.297 (0.019)	0.291	0.309 (0.01)	0.307	0.289 (0.081)	0.288	0.046 (0.026)	0.037	0.307 (0.023)	0.308
CryoDRGN2	0.32 (0.062)	0.342	0.301 (0.03)	0.306	**0.341 (0.059)**	0.356	**0.076 (0.016)**	**0.072**	0.245 (0.042)	0.260
DRGN-AI	**0.351 (0.01)**	**0.352**	**0.329 (0.028)**	**0.333**	**0.341 (0.083)**	**0.367**	0.072 (0.015)	**0.072**	**0.279 (0.017)**	**0.281**
3D Class abinit	0.13 (0.046)	0.119	0.184 (0.022)	0.188	0.144 (0.036)	0.138	0.032 (0.012)	0.031	0.206 (0.009)	0.208

- Per-image Fourier Shell Correlation (Per-image FSC) is a distributional metric measuring volume reconstruction quality (higher better, best 0.5)
- No method dominates across different forms of heterogeneity

Datasets and code: cryobench.cs.princeton.edu



Thank you