A protein-engineering tour of Cambridge, MA

Shaun Lippow

Aileron Therapeutics

Merrimack Pharmaceuticals

Flagship Ventures

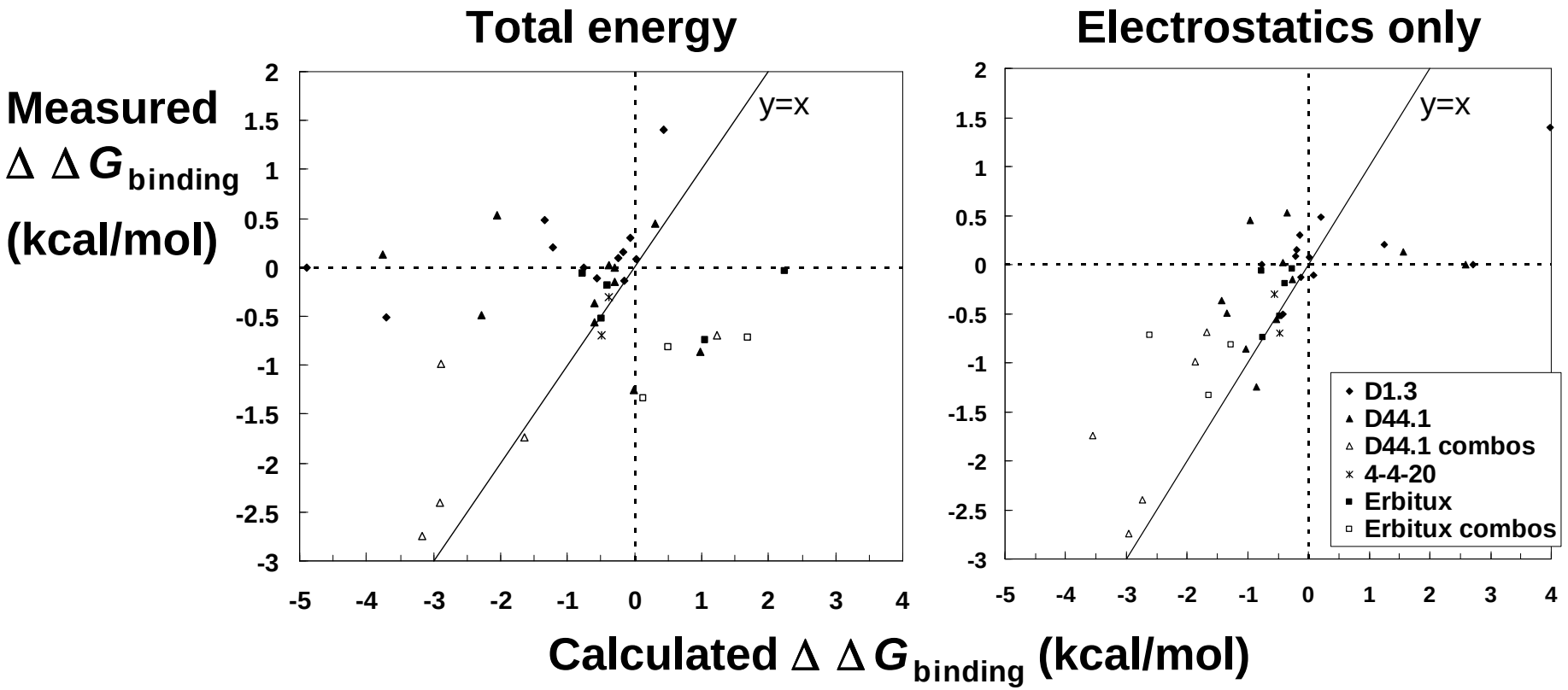
Codon Devices

Outline

- AILERON
  - Stapled helical peptides
- Electrostatics in protein–protein binding
  - A comparison of PB and Rosetta

Sorry, Aileron didn’t want their slides distributed

Electrostatic free energy as a predictor of binding improvement



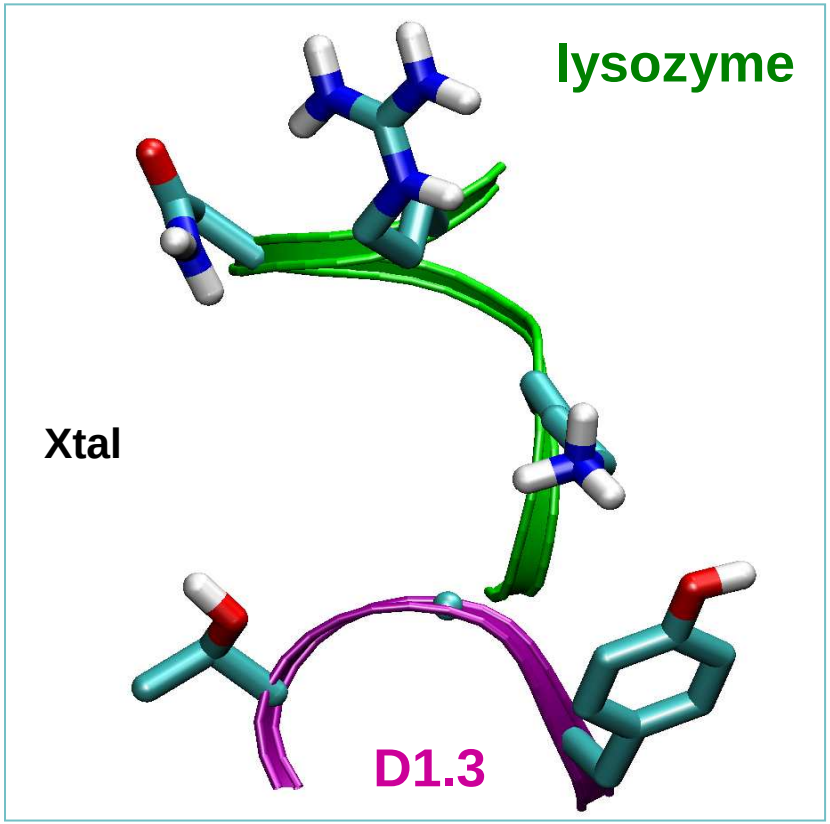
Fixbb; Rotamer library; DEE; CHARMM + PBelec
Antibody/antigen interfaces; Single mutations
Many predictions at periphery

How about Rosetta??

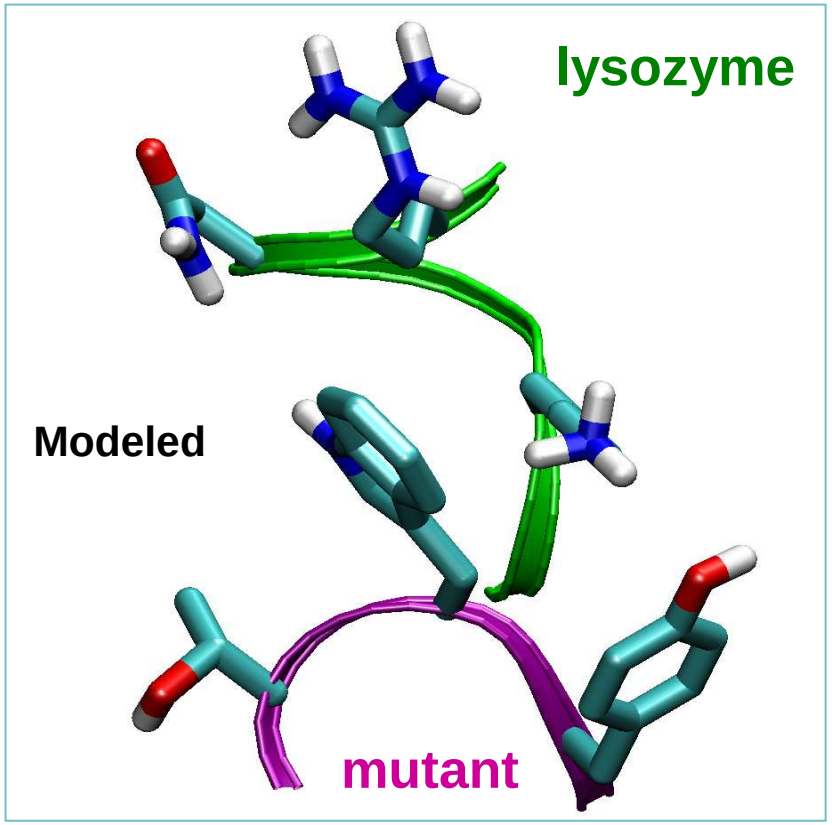
- **Periphery (surface-like)**
- **Highly polar interface**

S.M. Lippow, K.D. Wittrup, B. Tidor (2007) *Nat. Biotechnol.* 25 (10): 1171-1176

wild-type Gly



mutation to Trp



$\Delta\Delta G_i$	vdW	nonpolar	elec.	Total	Exp.
(kcal/mol)	-7.0	-0.6	+2.7	-4.9	0.0

Rosetta: “polyTrp surface”; “just adding atoms”…

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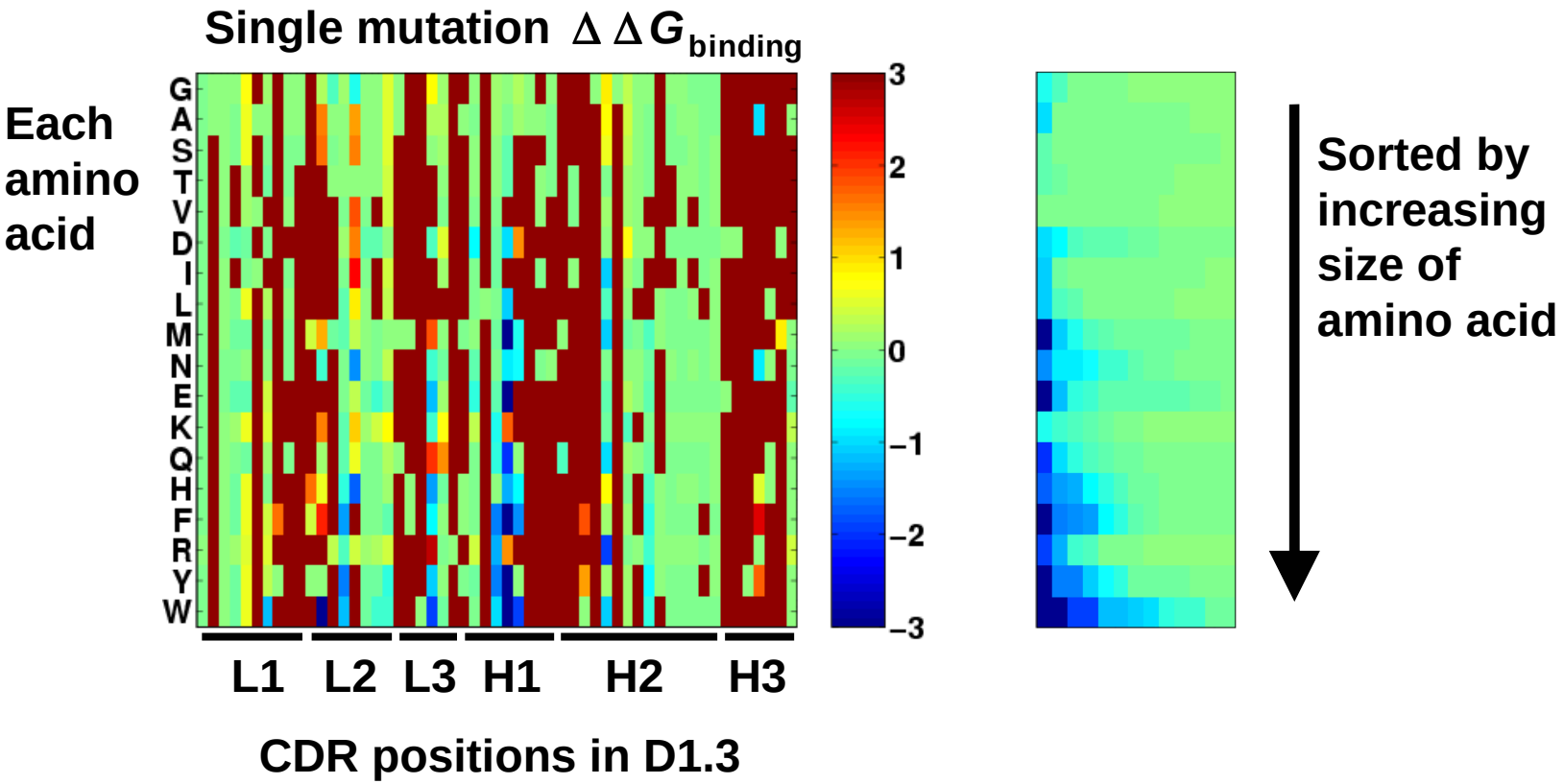
Large residues dominate predictions



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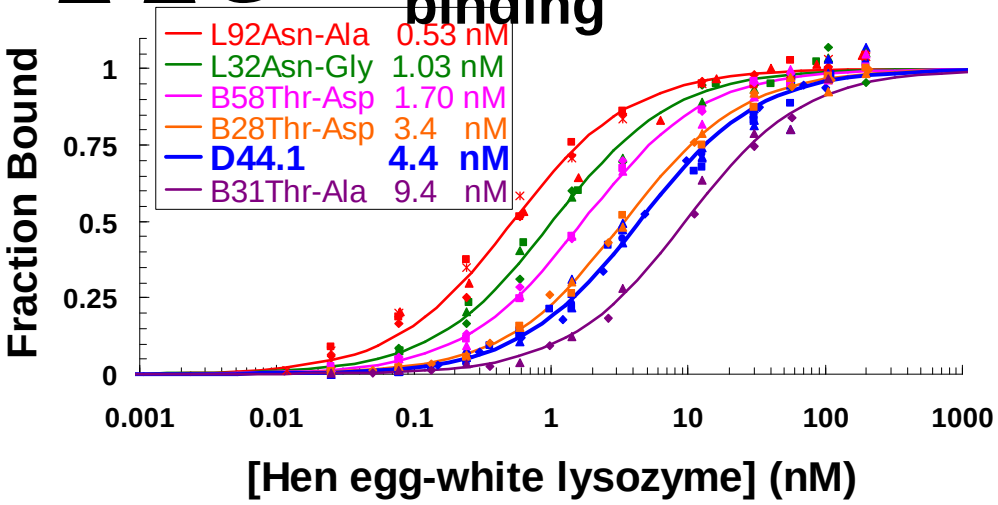
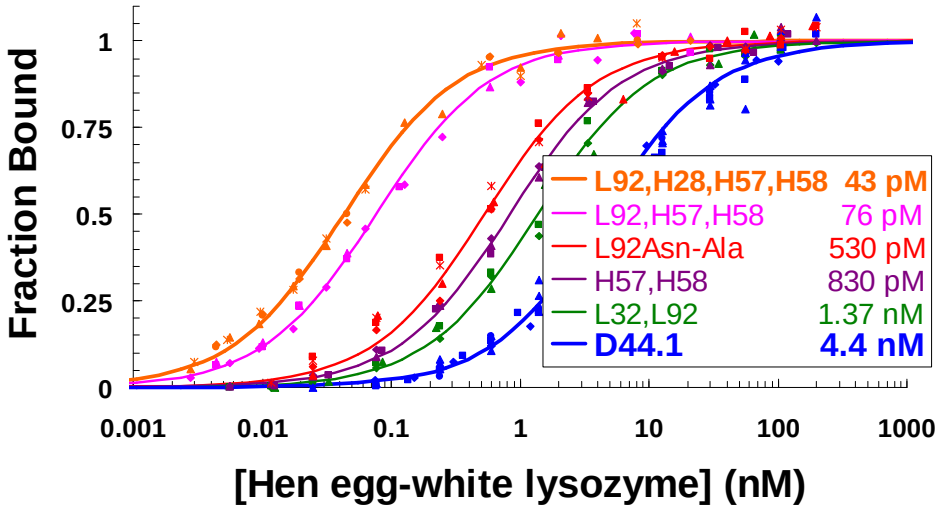
Predictions based on improved $\Delta\Delta G^{elec}$ binding



Single mutations validated

Rosetta "elec" terms: sol + hbnds + pair

Combinations of single mutations yield 100-fold improvement to 43 pM



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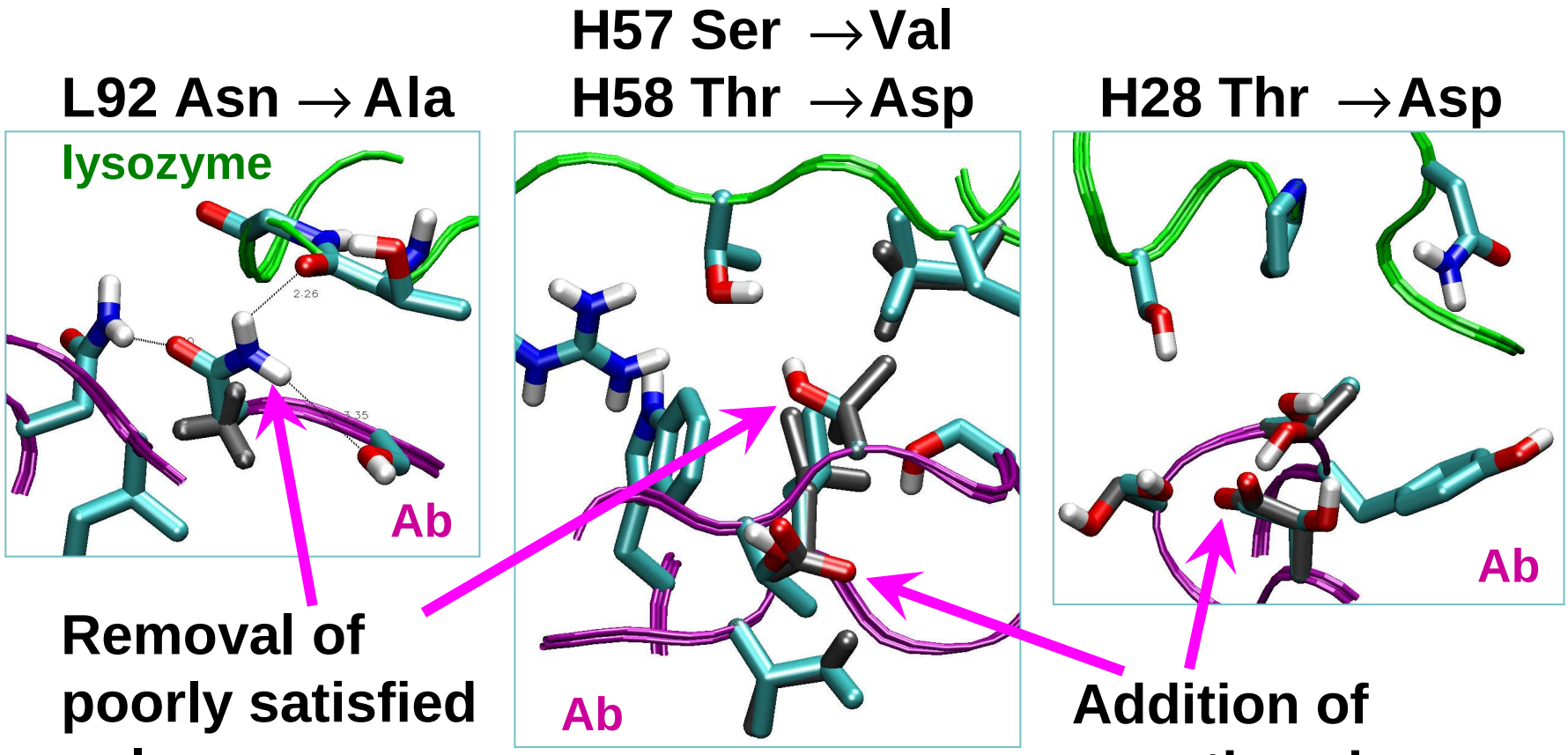
Two mechanisms lead to 100-fold improved quadruple mutant



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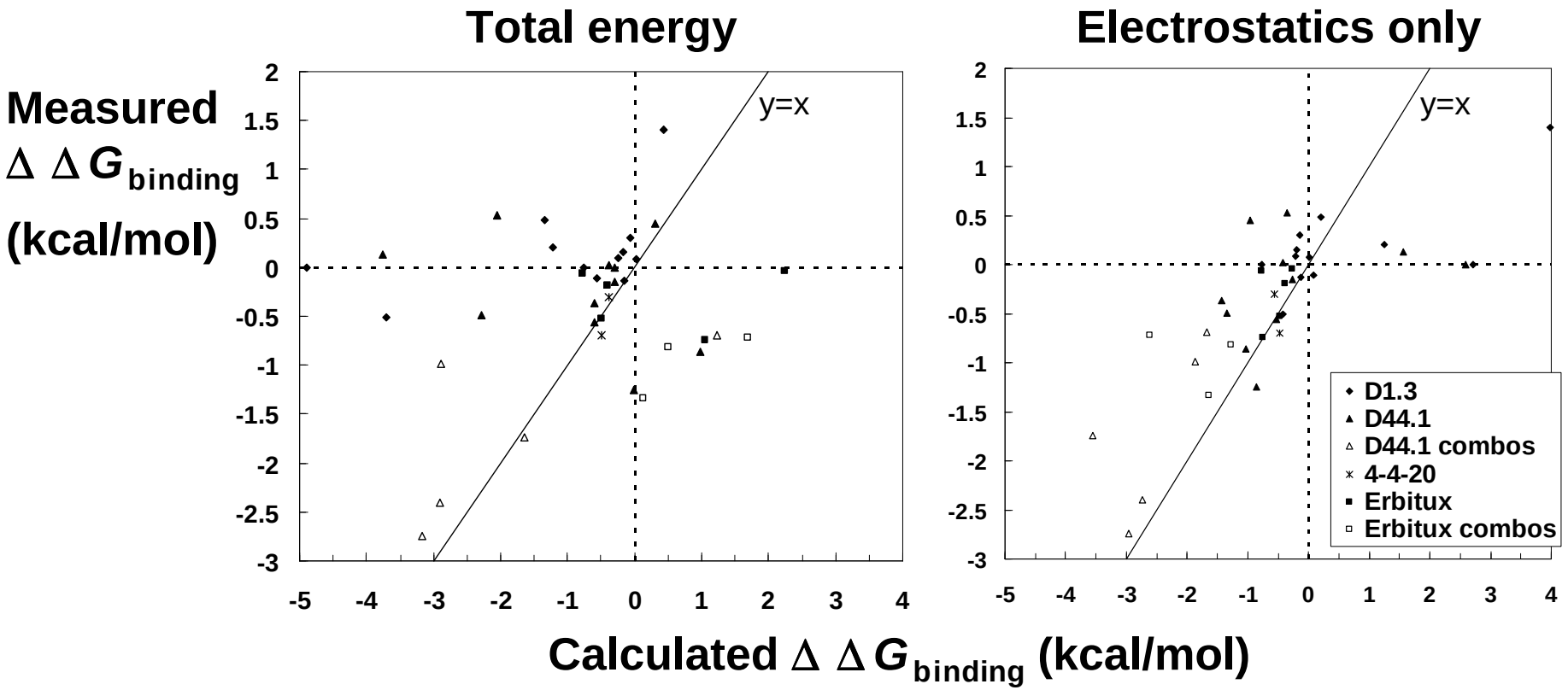

Rosetta: similar failure for periphery mutations

similar success with subset of electrostatic terms

(except mutations utilizing medium-range electrostatics)

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  - Tidor Lab
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Future work

- It depends!
  - Reduce my 4-body “problem” to a 2-body problem