Molecular Dynamics in the Era of Exascale Computing

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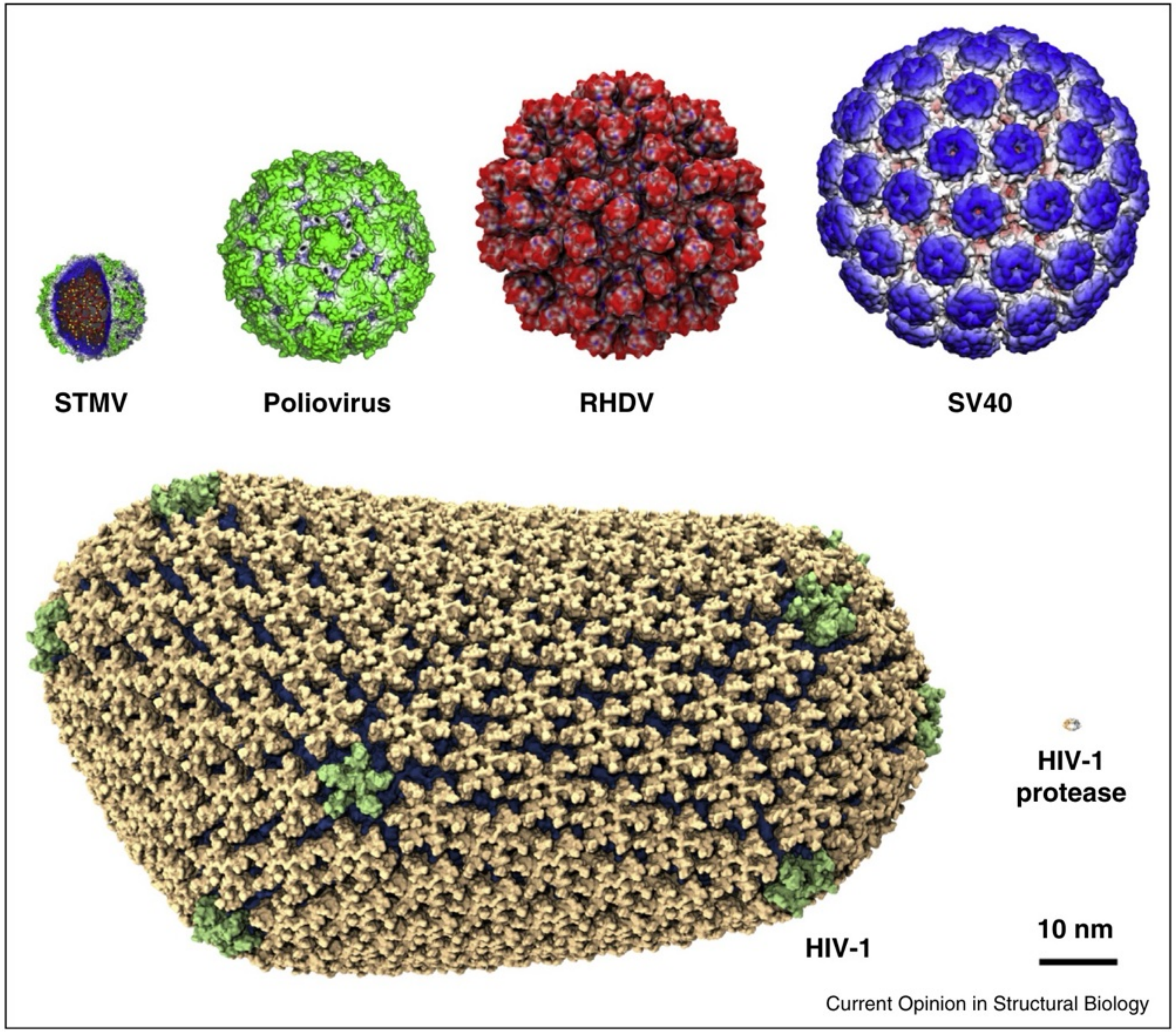
AUBURN UNIVERSITY
College of Sciences and Mathematics

Petascale revolution

Computing systems capable of calculating at least 10^{15} floating point operations per second
(1 petaFLOPS)

National Petascale Computing Facility
Blue Waters Supercomputer
x86 with GPU-acceleration
Champaign, IL



Perilla et al., Current Opinion in Structural Biology, 31 (64-74), 2015

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rcbernardi@auburn.edu

Exascale computing

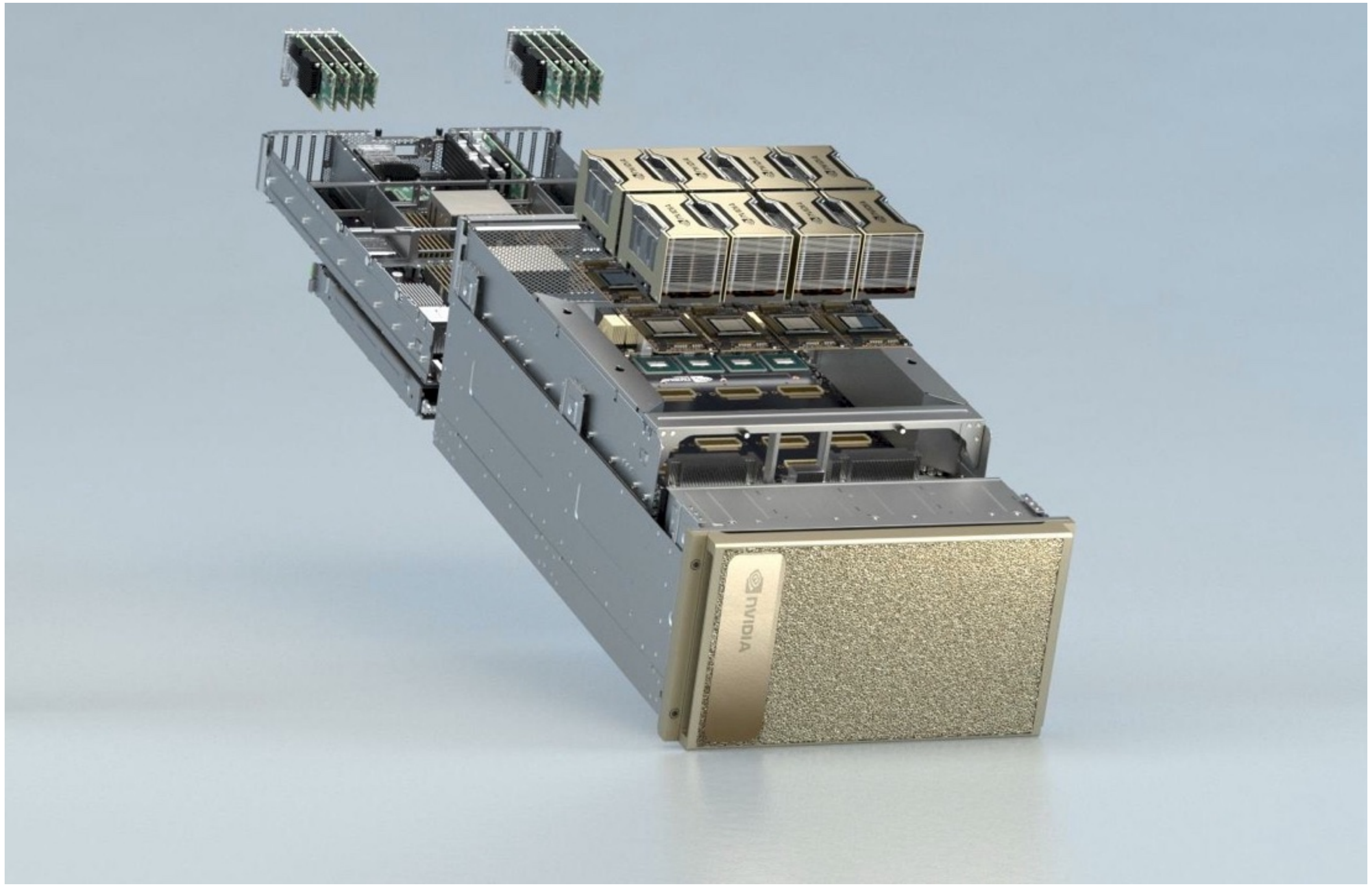
Computing systems capable of calculating at least 10^{18} floating point operations per second (1 exaFLOPS)

Japan's Fugaku Supercomputer – First exaScale Computer
ARM architecture-based.

- What the new generation of Computers look like?
- What can we do with this new generation of Computers?
- What are the challenges in the software development?
- What we are doing at the NIH Center for Macromolecular Modeling and Visualization.

Petascale Computing cost-drop

You can have your own Petascale Computer in 1 node



5 petaFLOPS of AI performance

NVIDIA DGX A-100
U$ 199,000 MSRP

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Having a Petascale Computing Lab

Computational Biophysics Group at Auburn University

960 GB of GPU memory
8x (200 Gbps) Infiniband NICs per node
15 PetaFLOPS of AI performance

Peak performance for NAMD of 1 node is equivalent to 400 Blue Waters Supercomputer Nodes

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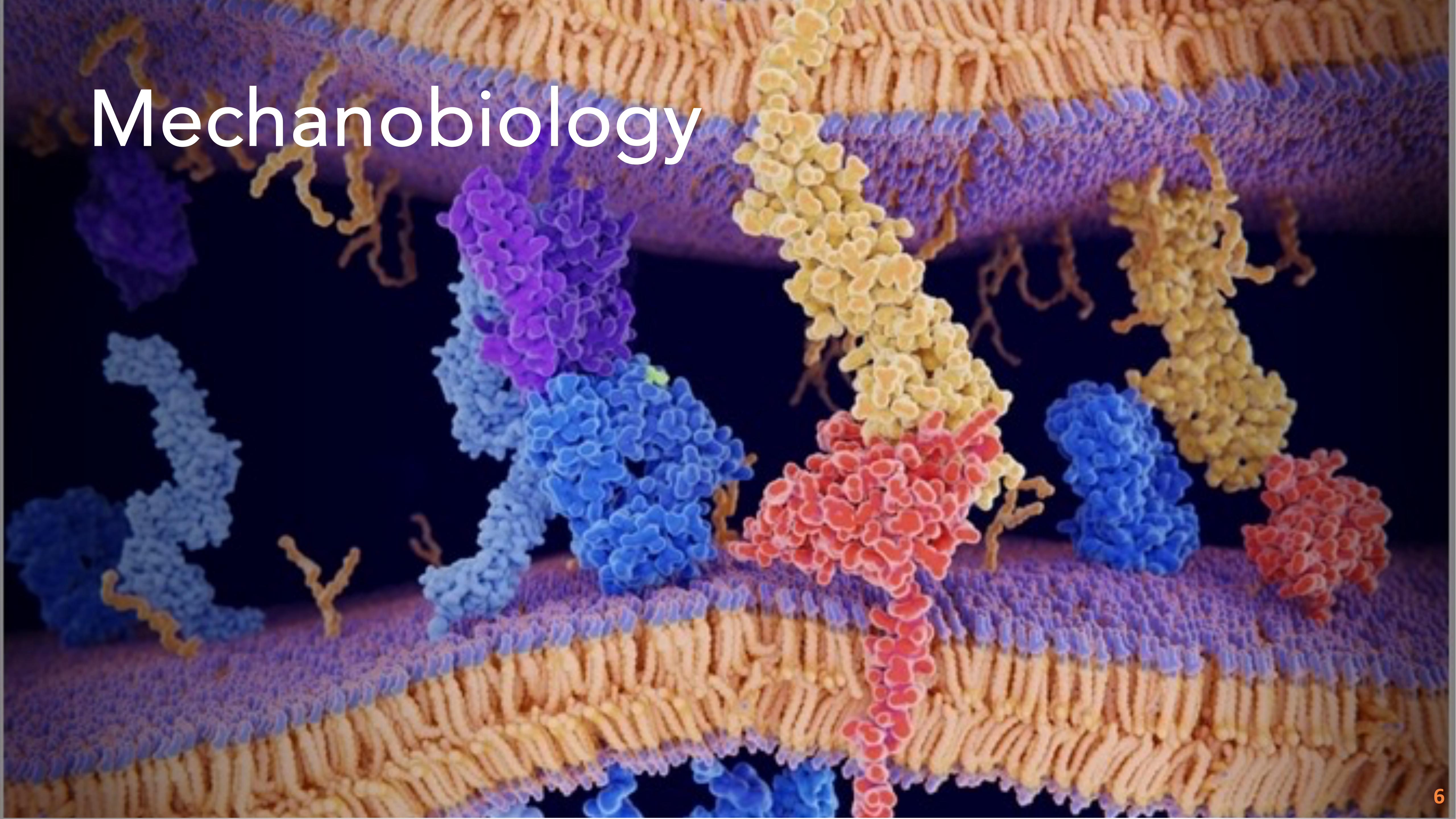
Mechanobiology



In Silico Single-Molecule Force Spectroscopy

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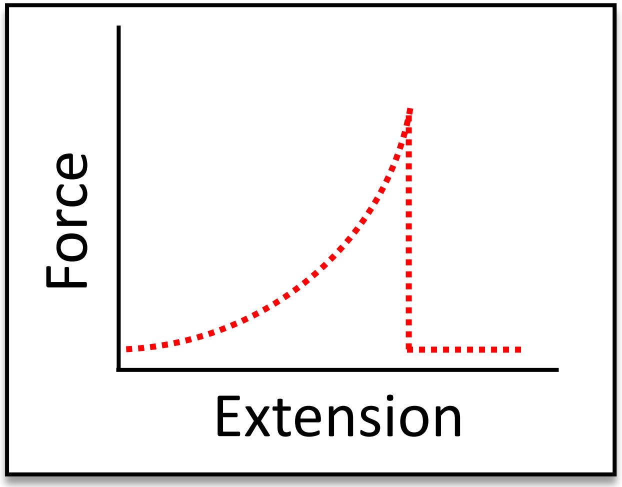
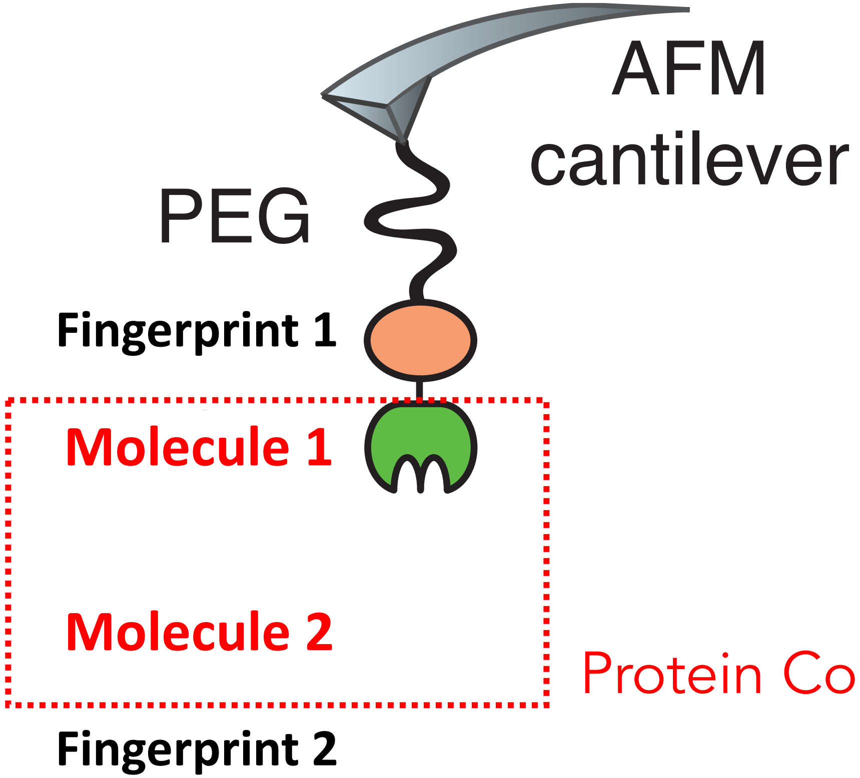
College of Sciences and Mathematics

Single-Molecule Force Spectroscopy (SMFS)

- Atomic Force Microscopy
- Centrifugal Force Microcopy
- Magnetic Tweezers
- Optical Tweezers



Atomic Force Microscope (AFM)



Protein Complex of Interest

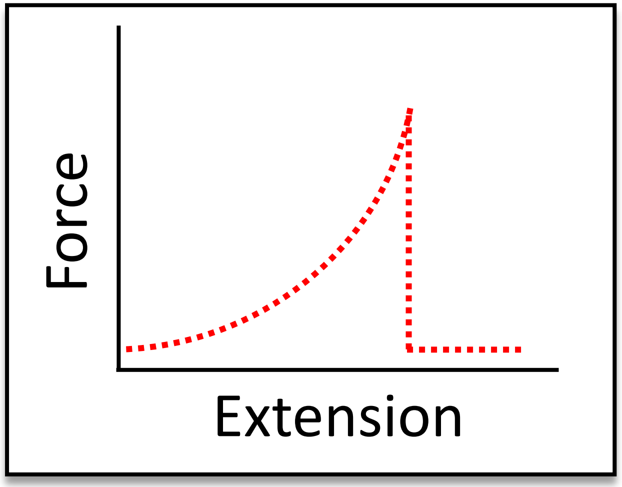
Hermann Gaub
(LMU)

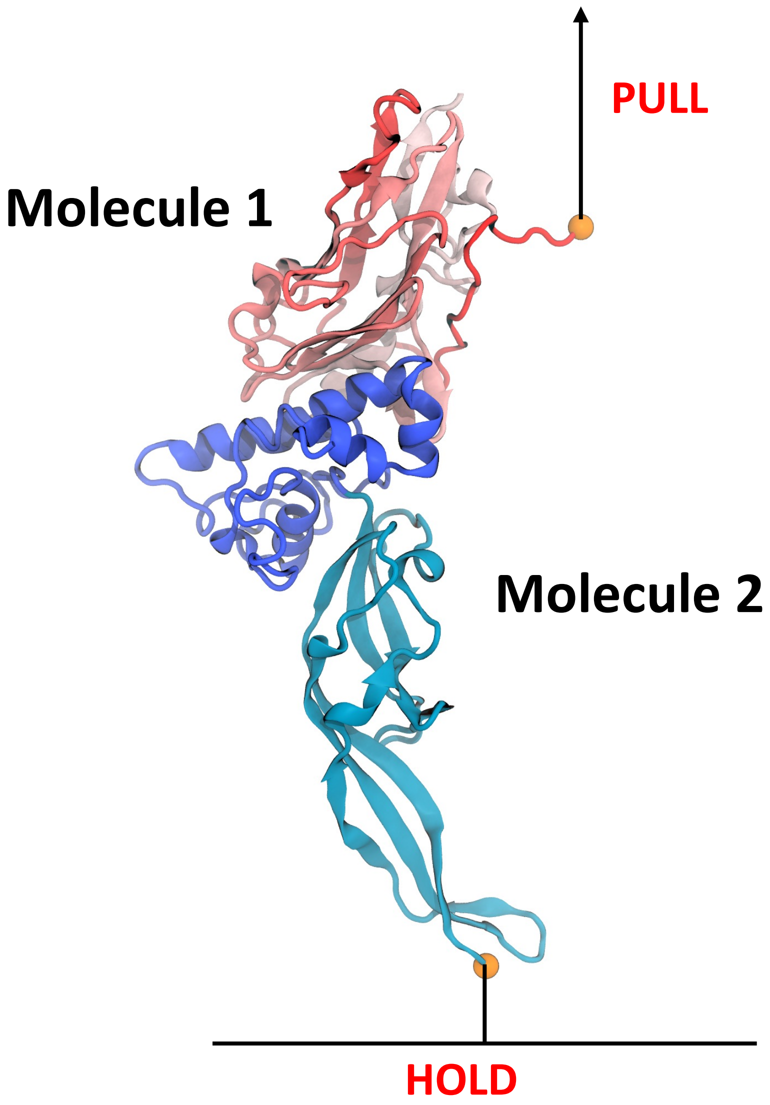
EL Florin, et. al.; Science, 1994
GU Lee, et. al.; Langmuir, 1994

Steered Molecular Dynamics

- Molecular Dynamics Simulations
- Pulling with a spring (Hooke's Law):

$$\boldsymbol{F} = -k \cdot \Delta\boldsymbol{x}$$



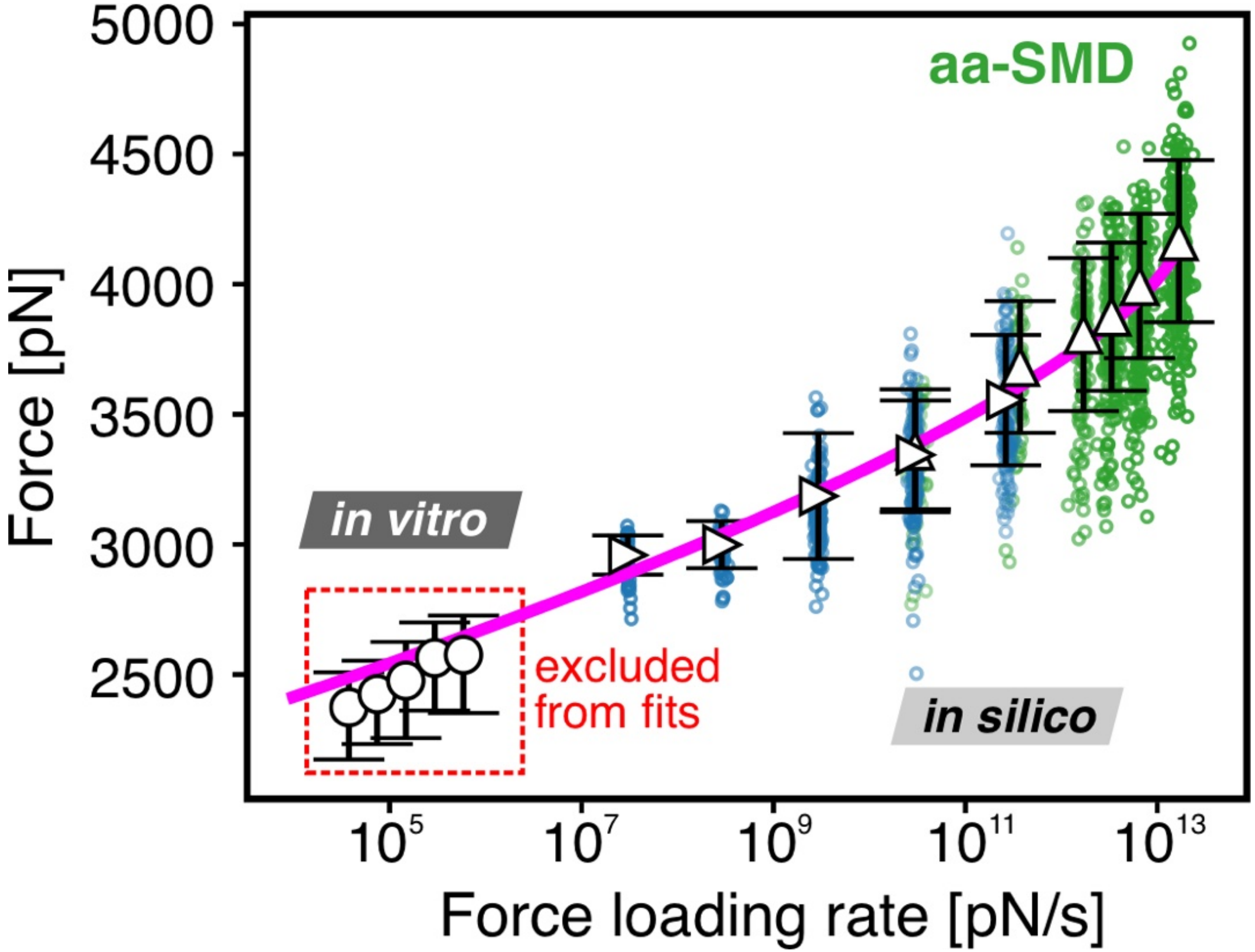


Klaus Schulten
Univ. of Illinois

H Grubmüller, et. al.; Science, 1996
S Izrailev, et. al.; Biophysical Jounal, 1997

In Silico Single Molecule Force Spectroscopy

- Pulling and anchoring points that are similar to experiments.
- Thousands of simulation replicas.
- Dozens to hundreds of microseconds of all-atom SMD.
- Dozes of Terabytes of trajectory data.
- Dynamical Network Analysis.
- Dimensionally reduction tools.
- AI tools for predicting mutations.



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So…

$$\xcancel{-\frac{\hbar^2}{2m}\nabla^2\psi + V(\boldsymbol{x})\psi = E\psi}$$

$$\boldsymbol{F} = \boldsymbol{m} \cdot \boldsymbol{a}$$

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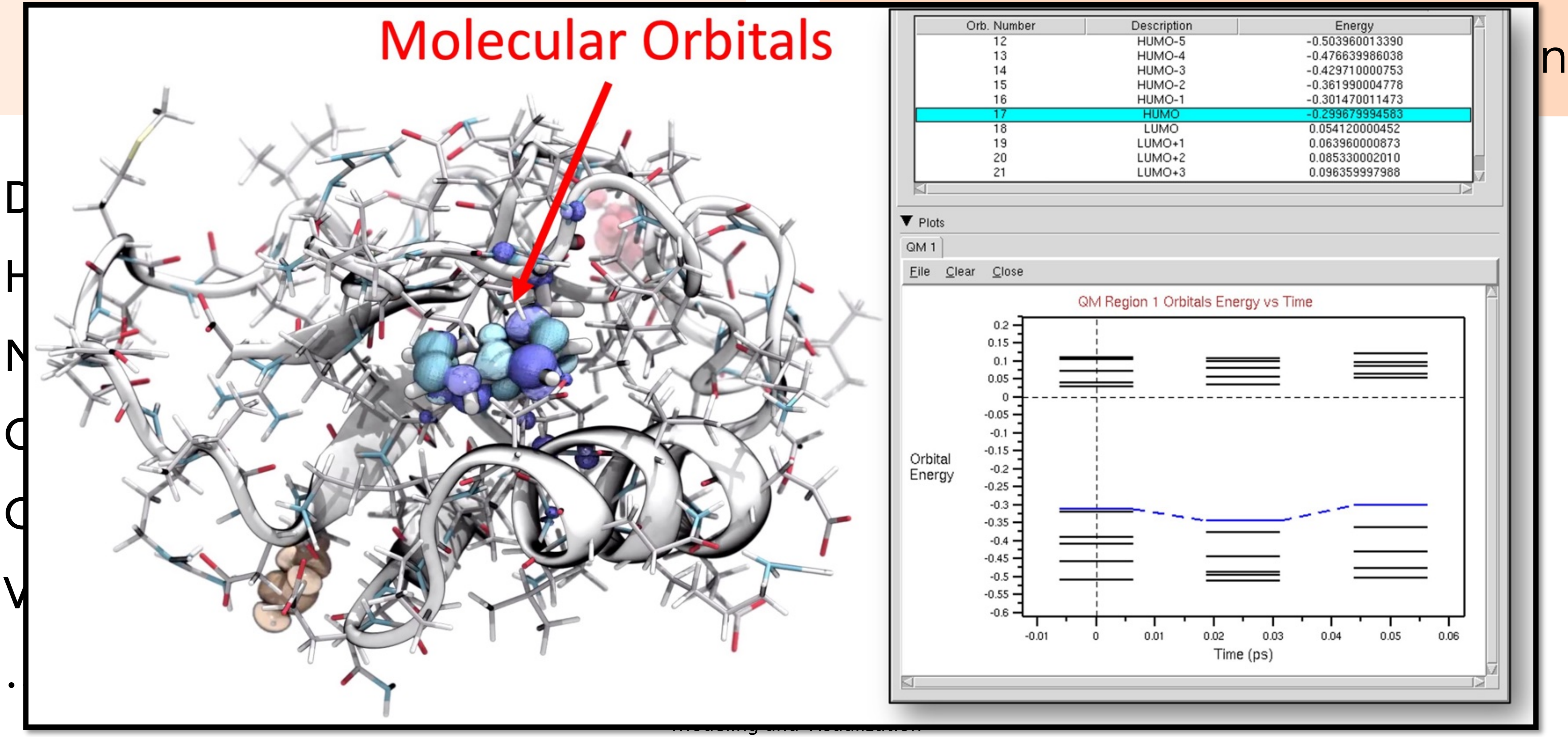
Computational Biophysics Group

ns

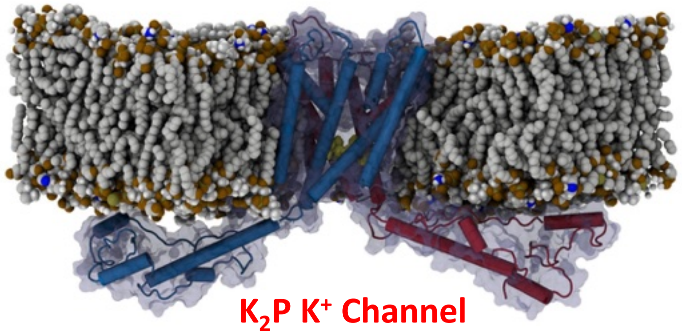
- D
- H
- N
- C
- C
- V
- .

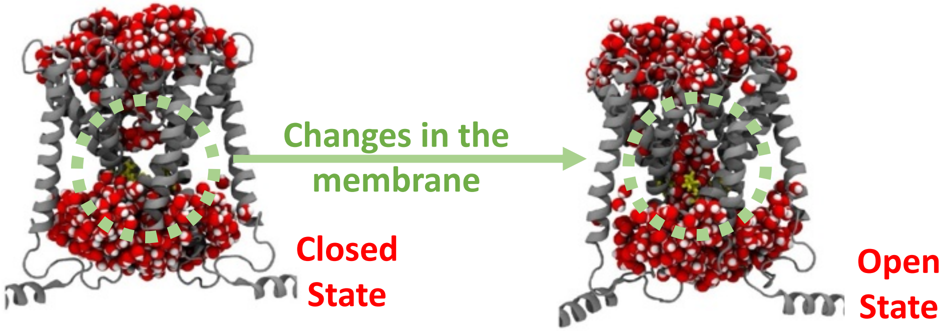


Orb. Number	Description	Energy
12	HUMO-5	-0.503960013390
13	HUMO-4	-0.476639986038
14	HUMO-3	-0.429710000753
15	HUMO-2	-0.361990004778
16	HUMO-1	-0.301470011473
17	HUMO	-0.299679994583
18	LUMO	0.054120000452
19	LUMO+1	0.063960000873
20	LUMO+2	0.085330002010
21	LUMO+3	0.096359997988

Mechanobiology: force matters in biology

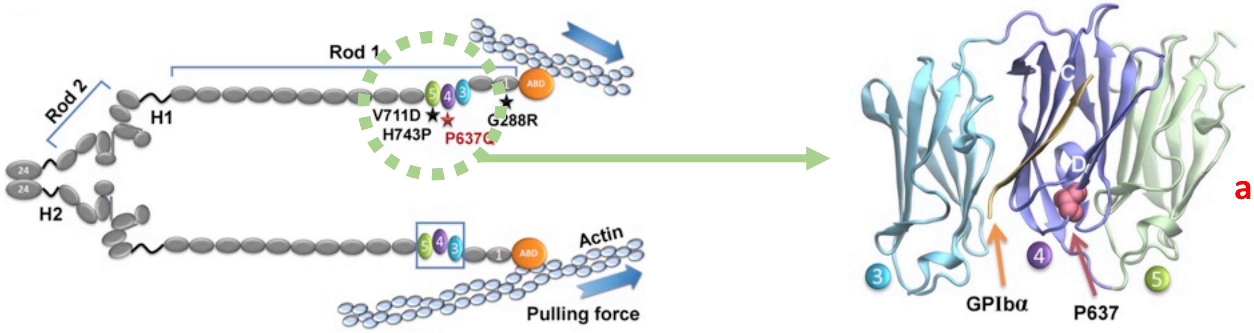
- **Mechanosensing activates signal transduction in neurons;**





PhD Work
RC Bernardi, et. al.; Molecular Physics, 2009
RC Bernardi & PG Pascutti; JCTC, 2012

- **Mechanical stress alters Filamin affinity to molecules in the cytoplasm;**

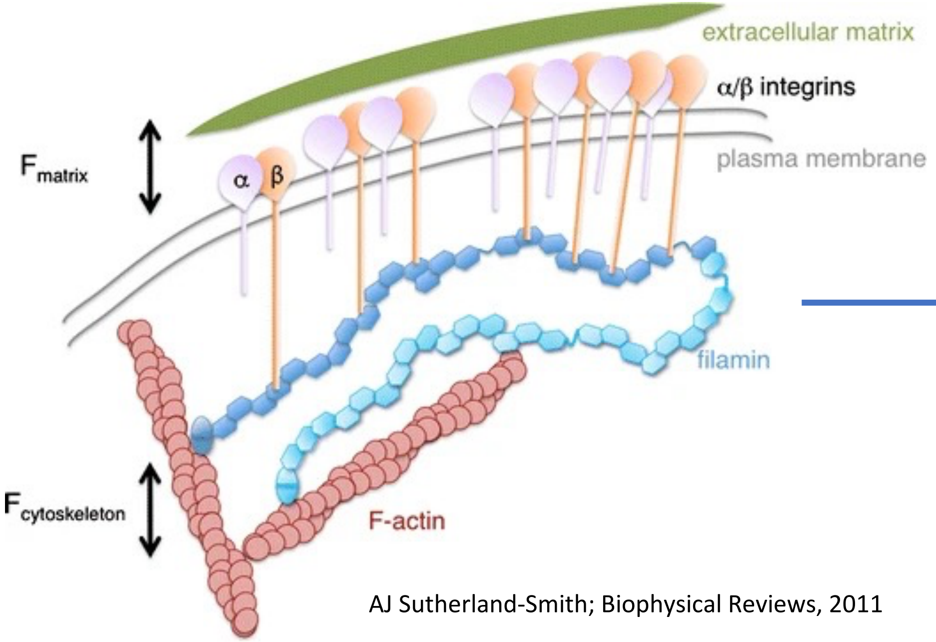


Mutations in Filamins are associated with genetic diseases

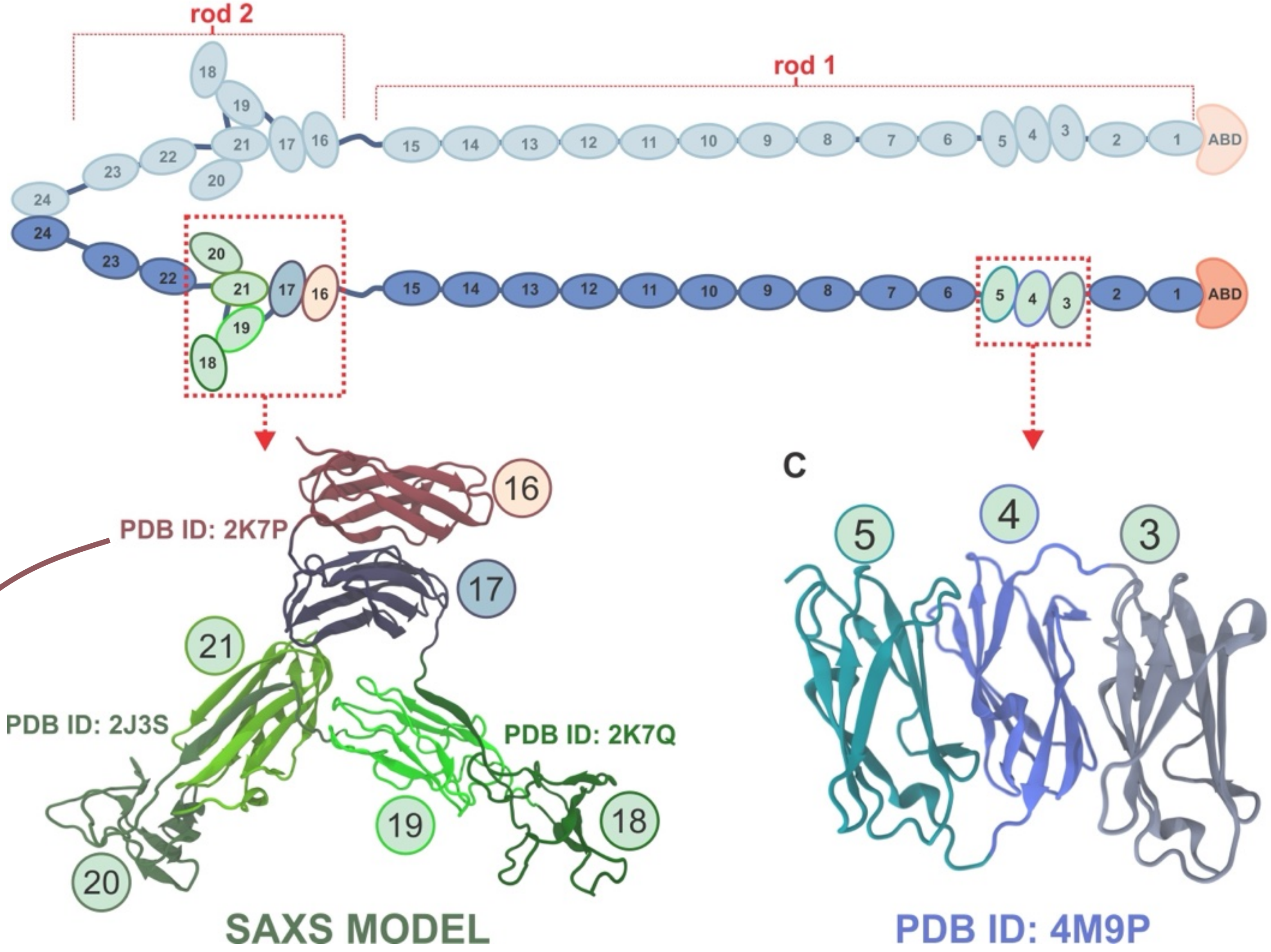
Collaboration with Ulla Pentikäinen
University of Jyväskylä, Finland
J Seppälä & RC Bernardi, et. al.; Scientific Reports, 2017
TJK Haataja & RC Bernardi, et. al.; Structure, 2019

- **Bacteria adhesion is mostly regulated by mechanoactive proteins.**

Force Resilience in Biology: Filamins and mechanosensing

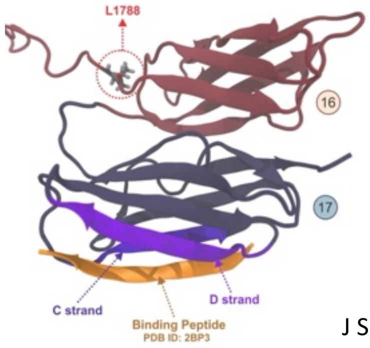


AJ Sutherland-Smith; Biophysical Reviews, 2011



Filamin cross-linking F-actin is fundamental to balance cytoskeleton and extracellular matrix tension

Mutations were found to affect mechanosensing and cause phenotype changes in humans

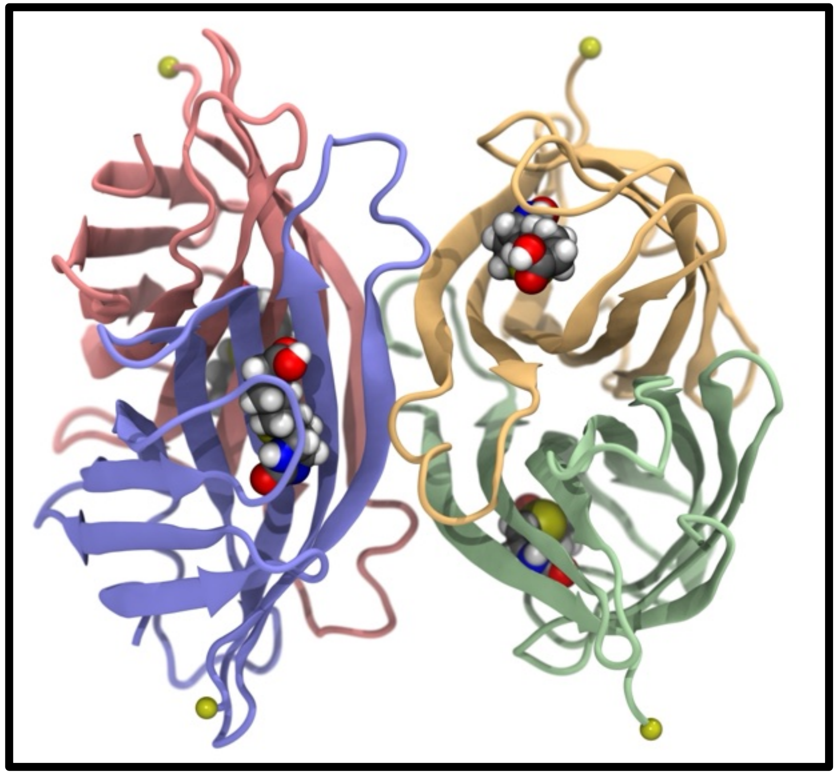


J Seppälä, RC Bernardi, et. al.; **Skeletal Dysplasia Mutations Effect on Human Filamins' Structure and Mechanosensing.** Scientific Reports, 2017

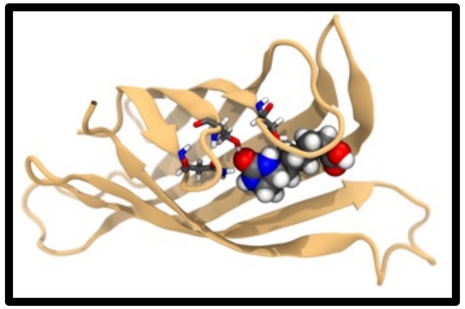
TJK Haataja, RC Bernardi, et. al.; **Non-syndromic Mitral Valve Dysplasia Mutation Changes the Force Resilience and Interaction of Human Filamin A.** Structure, 2019

Force Resilience in Biology: Pulling Geometry Matters

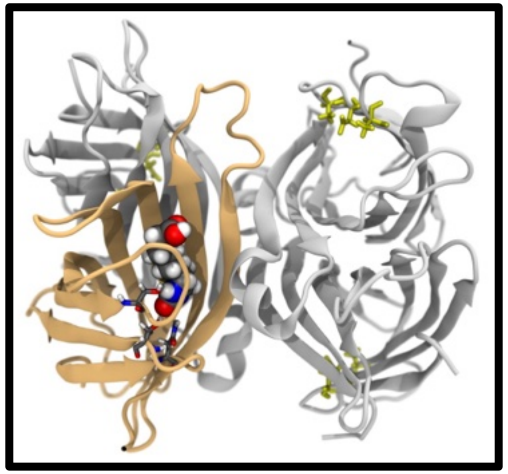
- Strongest binding affinity of any receptor:ligand complex;
- Extensivily used in biotechnology;
- Streptavidin has **femtomolar** affinity to biotin (aka vitamin B7);
- First protein system to be studied with AFM-based SMFS;
- Strongest complex under force known until 2014;



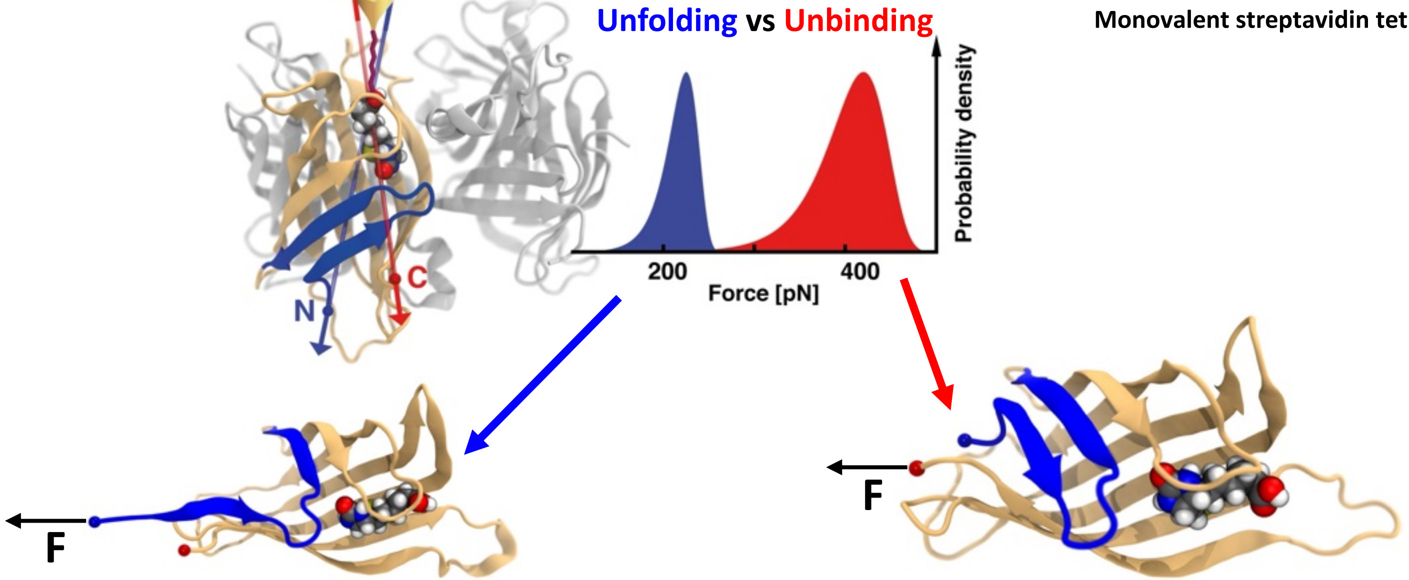
Tetravalent streptavidin tetramer bound to biotin molecules



Site-directed mutagenesis kill's affinity for biotin

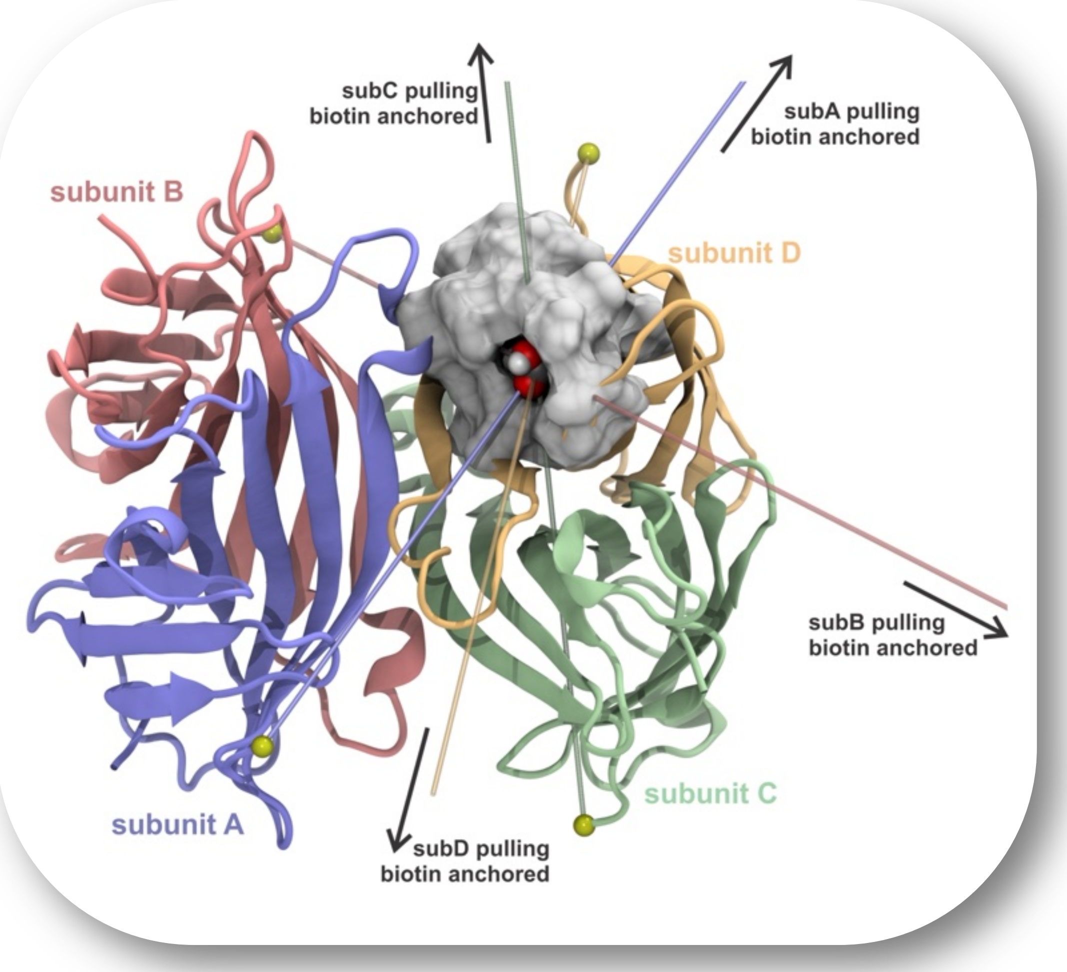


Monovalent streptavidin tetramer



SM Sedlak*, LC Schendel*, MCR Melo, DA Pippig, Z Luthey-Schulten, HE Gaub, RC Bernardi; **Direction Matters – Monovalent Streptavidin/Biotin Complex under Load.** Nano Letters, 2019

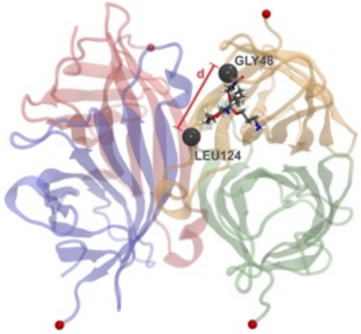
Force Resilience in Biology: The danger of unspecific tethering

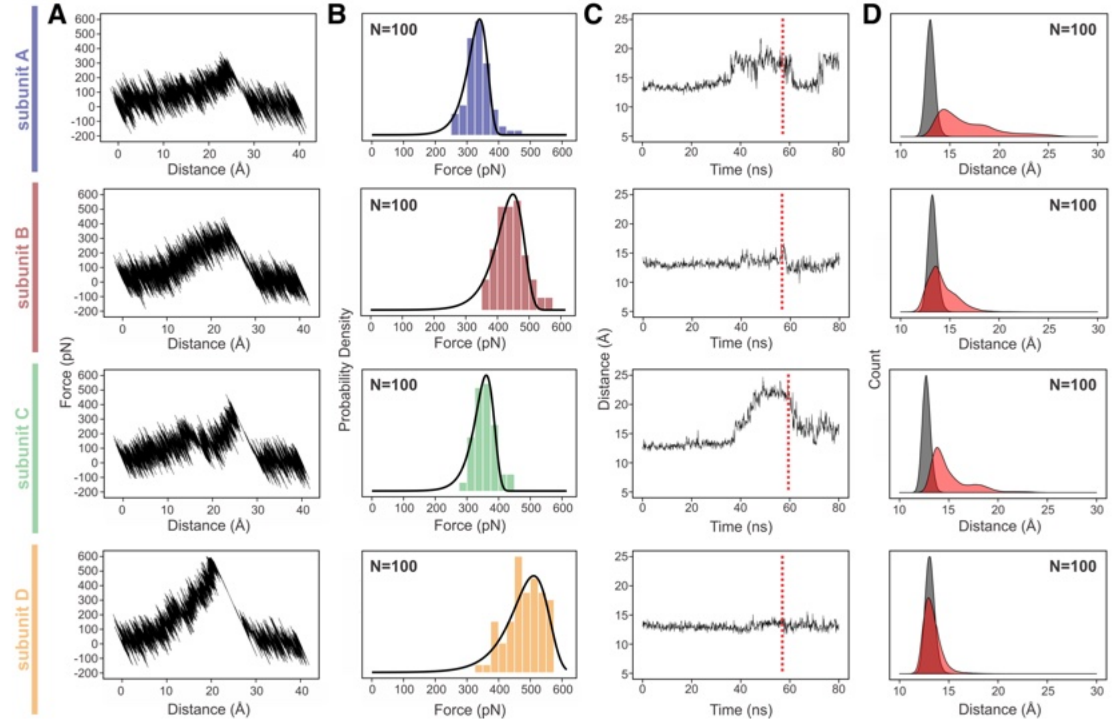


~15,000 papers published in the last 25 years using Streptavidin:Biotin biotechnology for force spectroscopy

Web of Science's search terms: "Biotin" and "Force"

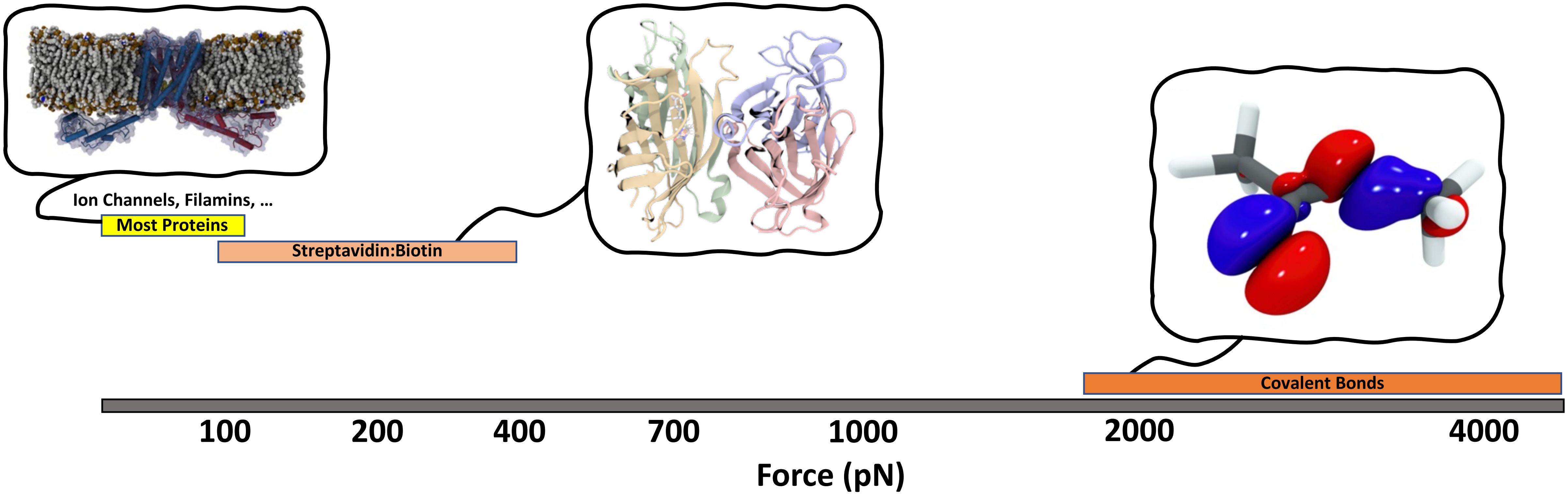
Depending on tethering geometry mechanical resilience can be completely different





SM Sedlak*, LC Schendel*, HE Gaub, RC Bernardi; **Streptavidin/Biotin: Tethering Geometry Defines Unbinding Mechanics.** Science Advances, 2020

Force Resilience in Biology



Ion Channels, Filamins, …

Most Proteins

Streptavidin:Biotin

Covalent Bonds

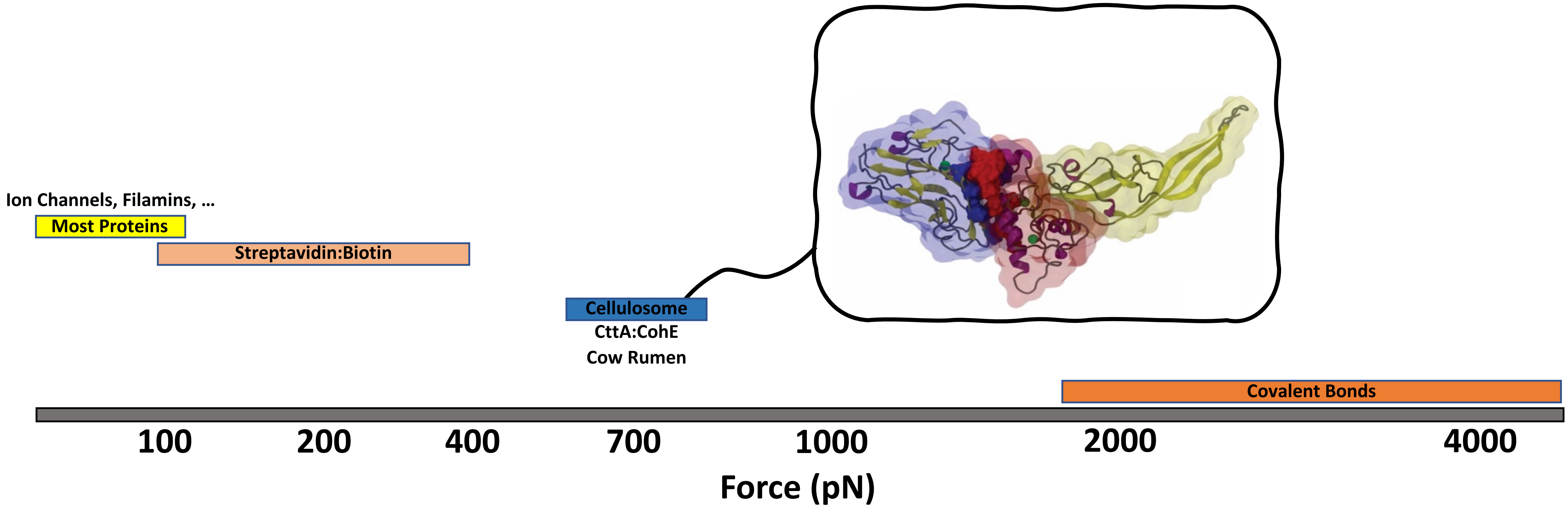
100 200 400 700 1000 2000 4000

Force (pN)

BIOLOGY

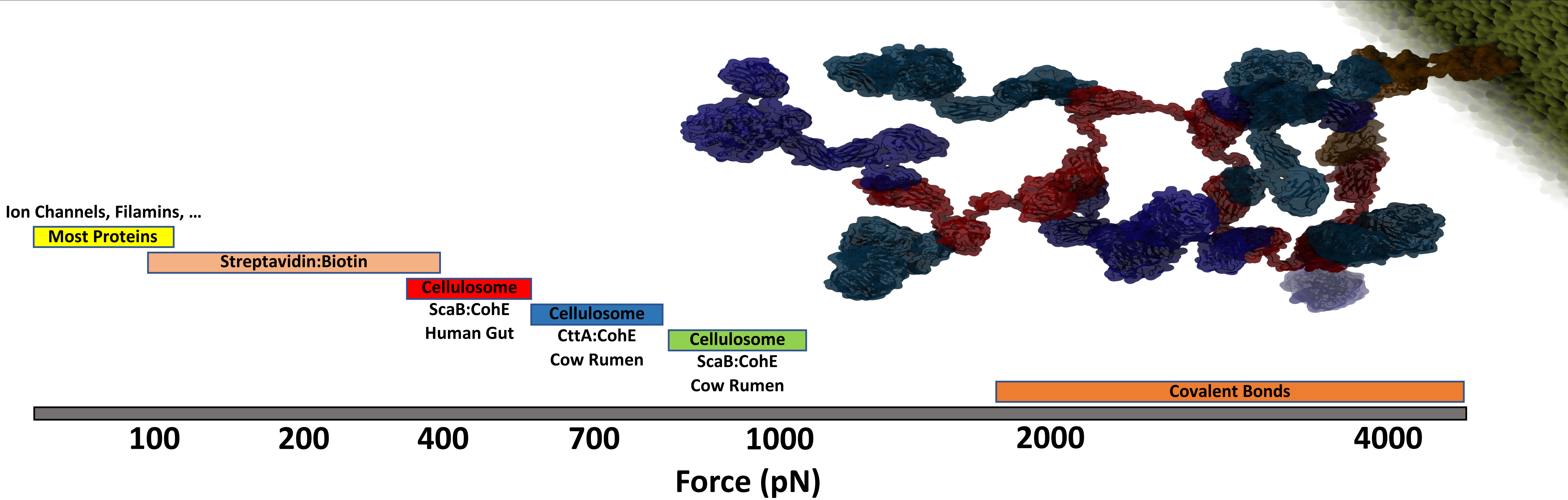
CHEMISTRY

Force Resilience in Biology



C Schoeler*, KH Malinowska*, RC Bernardi, et. al.; **Ultrastable cellulosome-adhesion complex tightens under load.** Nature Communications, 2014

Force Resilience in Biology



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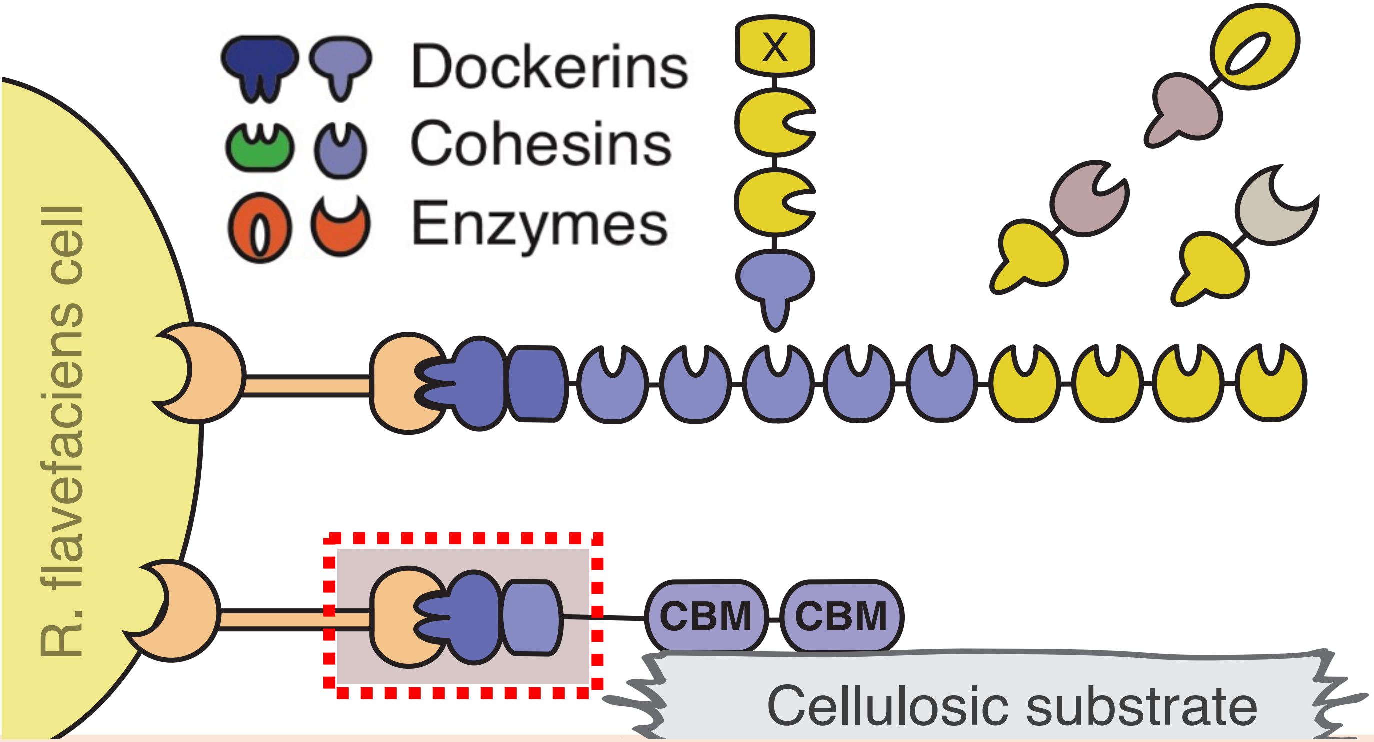
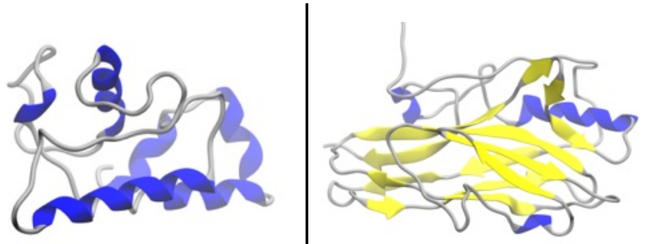
Z Liu, H Liu, AM Vera, RC Bernardi, et. al.; **High Force Catch Bond Mechanism of Bacterial Adhesion in the Human Gut.** Nature Communications, 2020

Mechanoactive bonds: Cellulosomes



Main Building Blocks:



Dockerins *Cohesins*

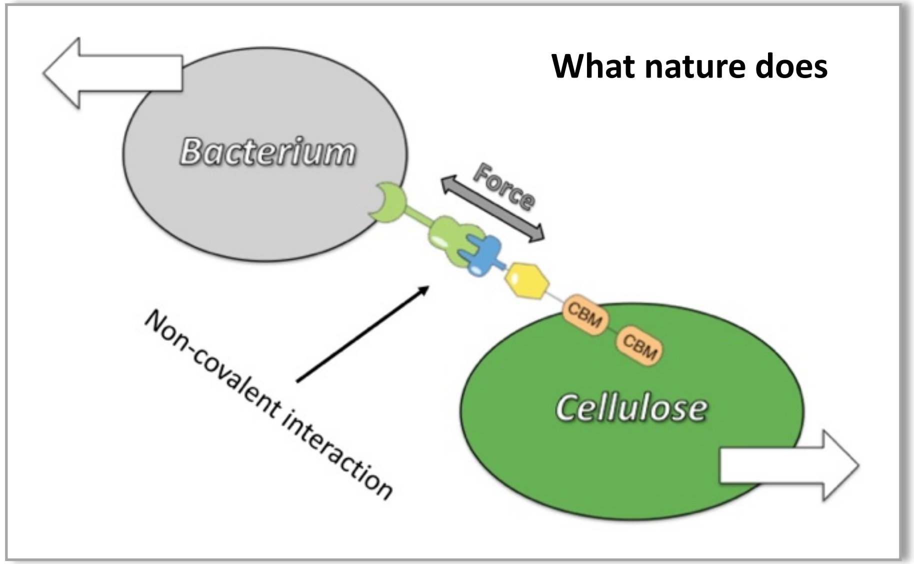
Very efficient in digesting plant-biomass – Ideal for 2nd generation biofuel production

Ed Bayer (Weizmann)

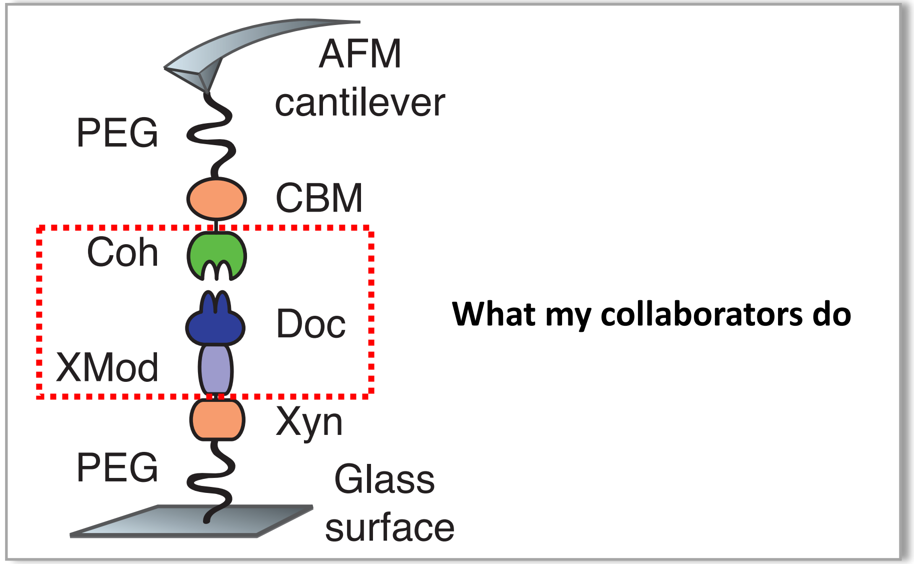
Cellulosomes are a large consortium of enzymes arranged as highly efficient nanomachines.

C Schoeler*, KH Malinowska*, RC Bernardi, et. al.; **Ultrastable cellulosome-adhesion complex tightens under load.** Nature Communications, 2014
C Schoeler*, RC Bernardi*, et. al.; **Mapping mechanical force propagation through biomolecular complexes.** Nano Letters, 2015
I Cann, RC Bernardi, RI Mackie; **Cellulose degradation in the human gut: *Ruminococcus champanellensis* expands the cellulosome paradigm.** Environmental Microbiology, 2016
T Verdorfer, RC Bernardi, et. al.; **Combining in Vitro and in Silico Single-Molecule Force Spectroscopy to Characterize and Tune Cellulosomal Scaffoldin Mechanics.** JACS, 2017
RC Bernardi, et al.; **Mechanisms of Nanonewton Mechanostability in a Protein Complex Revealed by Molecular Dynamics Simulations and Single-Molecule Force Spectroscopy.** JACS, 2019
I Cann, …, RC Bernardi, et. al.; **Thermophilic degradation of hemicellulose, a critical feedstock in the production of bioenergy and other value-added products.** Applied Environmental Microbiology, 2020
Z Liu, H Liu, AM Vera, RC Bernardi, et. al.; **High Force Catch Bond Mechanism of Bacterial Adhesion in the Human Gut.** Nature Communications, 2020

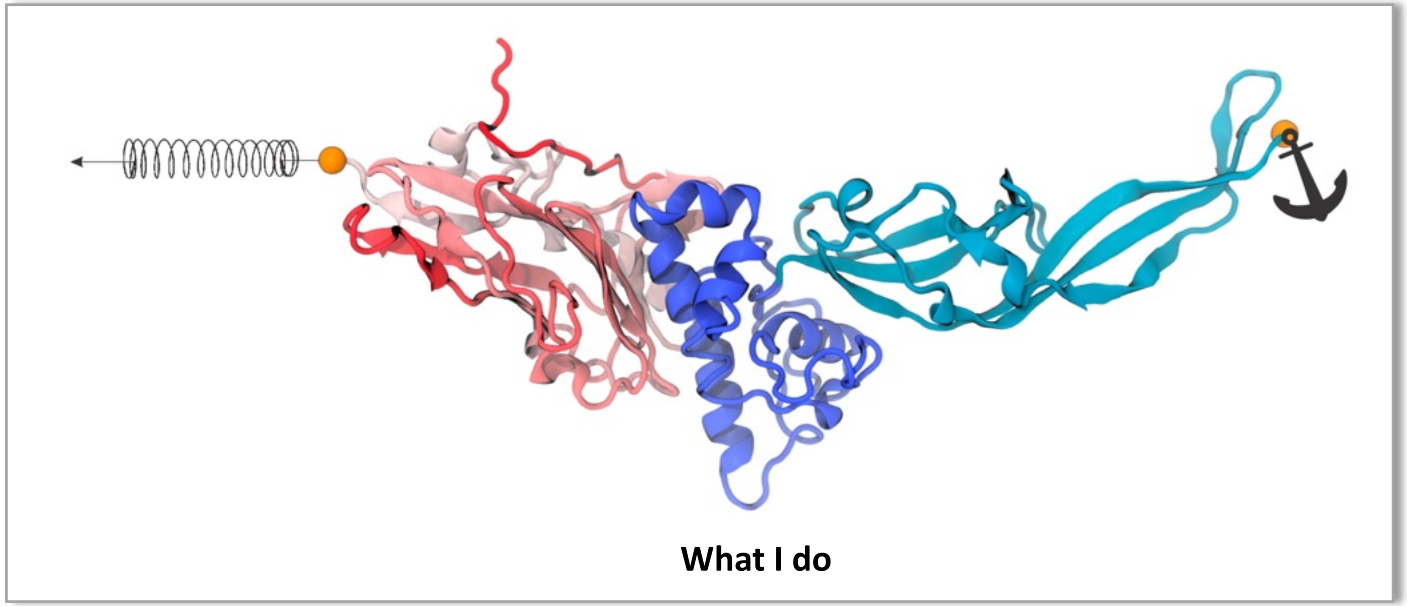
Mechanoactive bonds: Cellulosomes



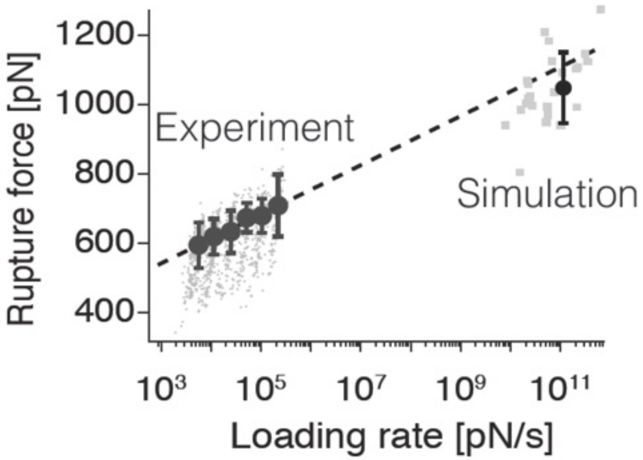
What nature does



What my collaborators do



What I do

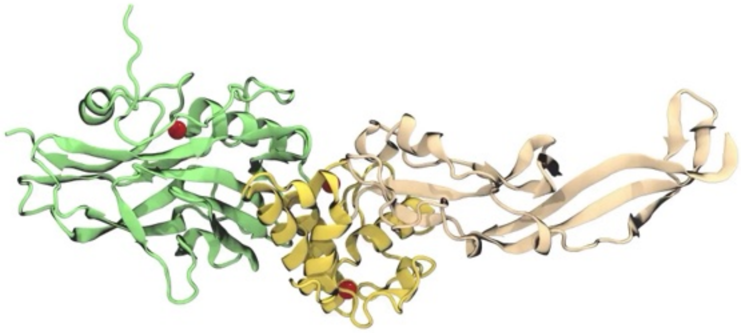


Strongest Protein Complex (2014)

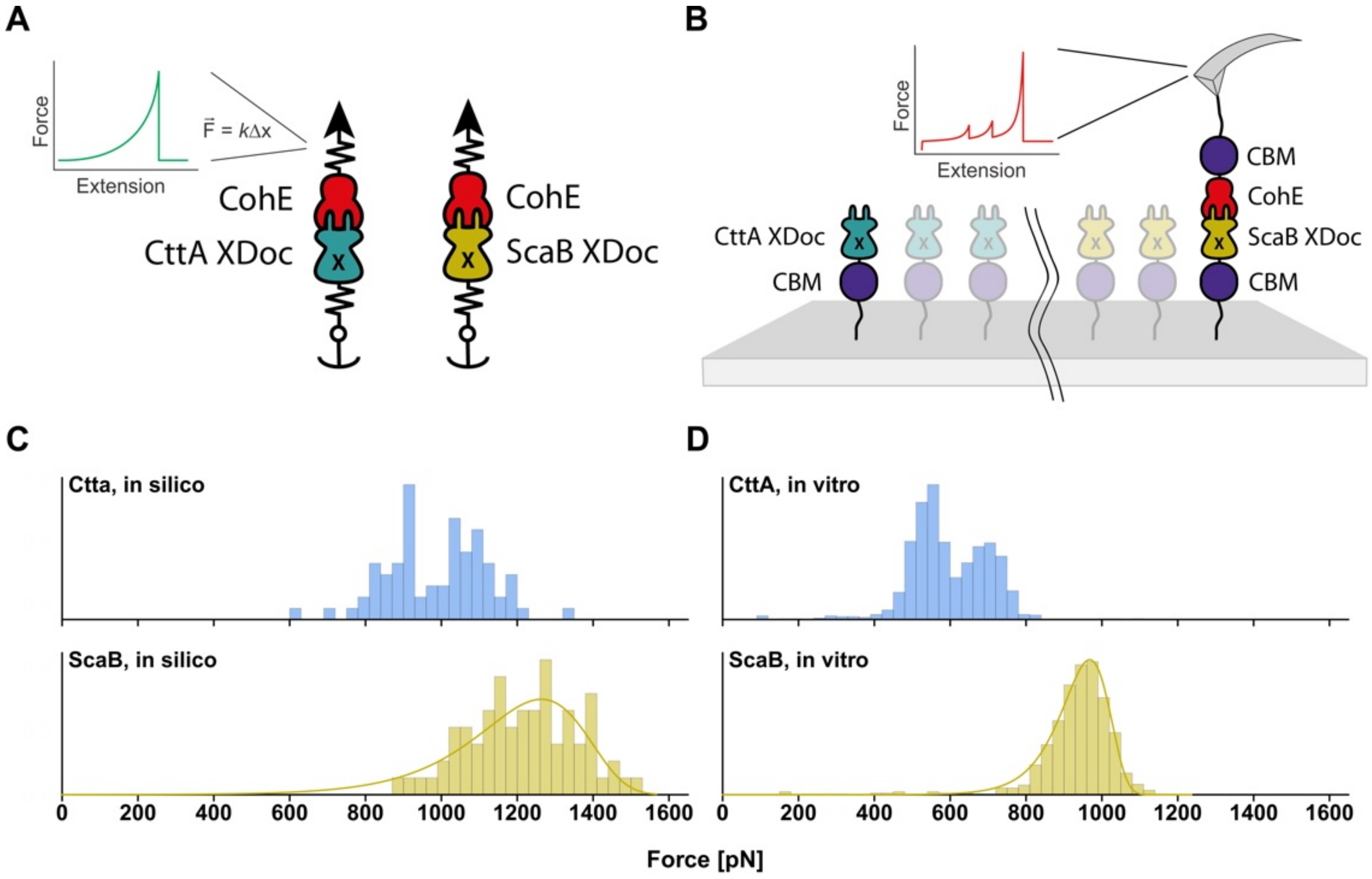
C Schoeler*, KH Malinowska*, RC Bernardi, et. al.; **Ultrastable cellulosome-adhesion complex tightens under load.** Nature Communications, 2014

In silico Single-Molecule Force Spectroscopy: Simulations Predicting the Experiments

Simulations were used to predict stronger complexes

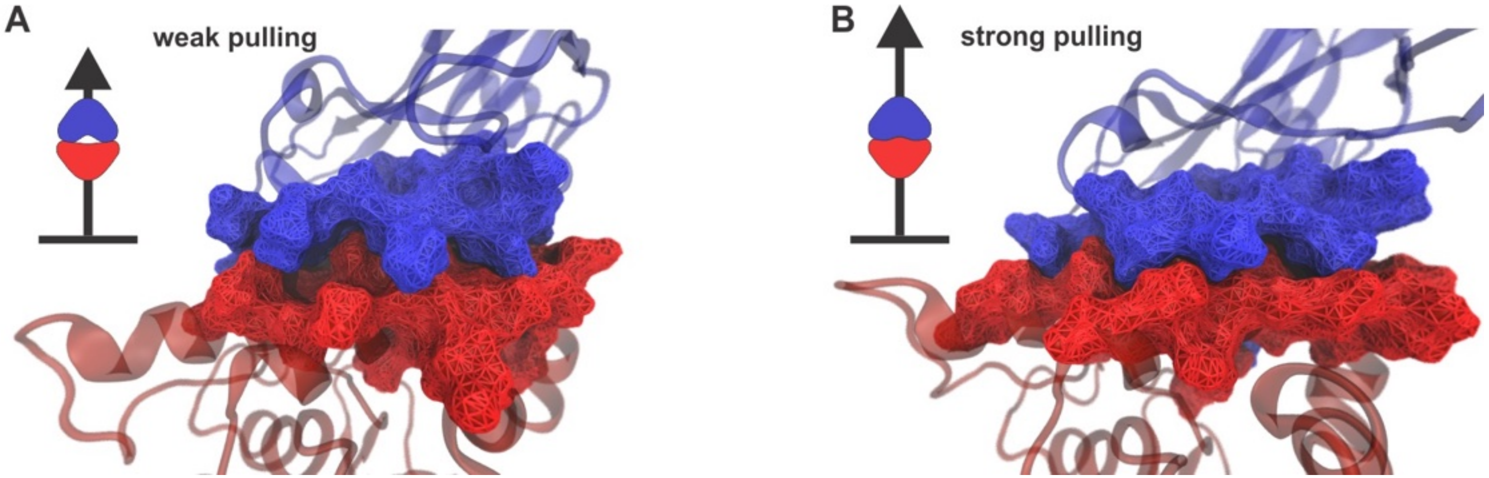


Simulations were used to predict function of the X-module, a domain with previously unknown function

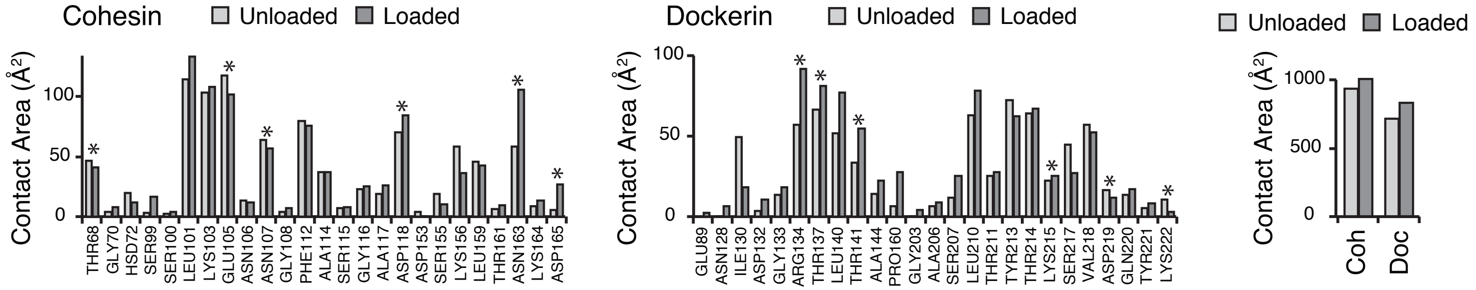


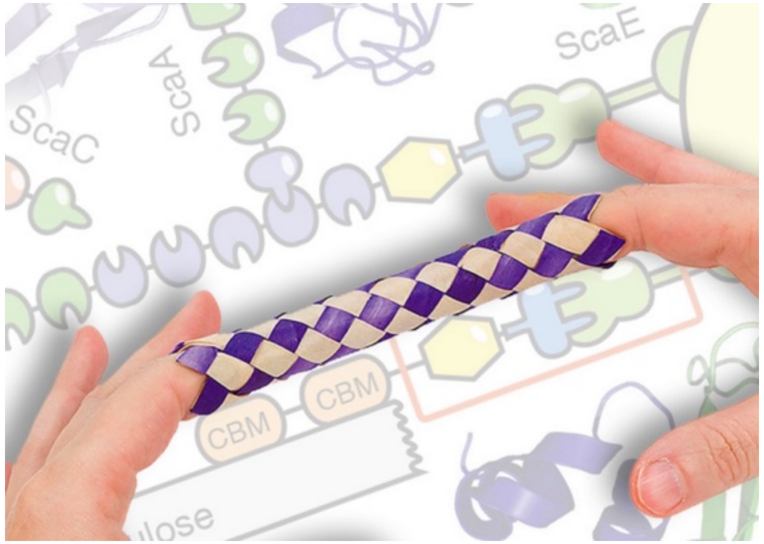
RC Bernardi, et al.; **Mechanisms of Nanonewton Mechanostability in a Protein Complex Revealed by Molecular Dynamics Simulations and Single-Molecule Force Spectroscopy.** JACS, 2019

Cellulosomes: the bond puzzle



Pulling Cohesin-Dockerin apart bring them closer together!





Molecular Finger Trap Puzzle

K_D = 20 nM

Rupture Under Force = 500-1100 pN

Antibody-antigen rupture at only ~60 pN

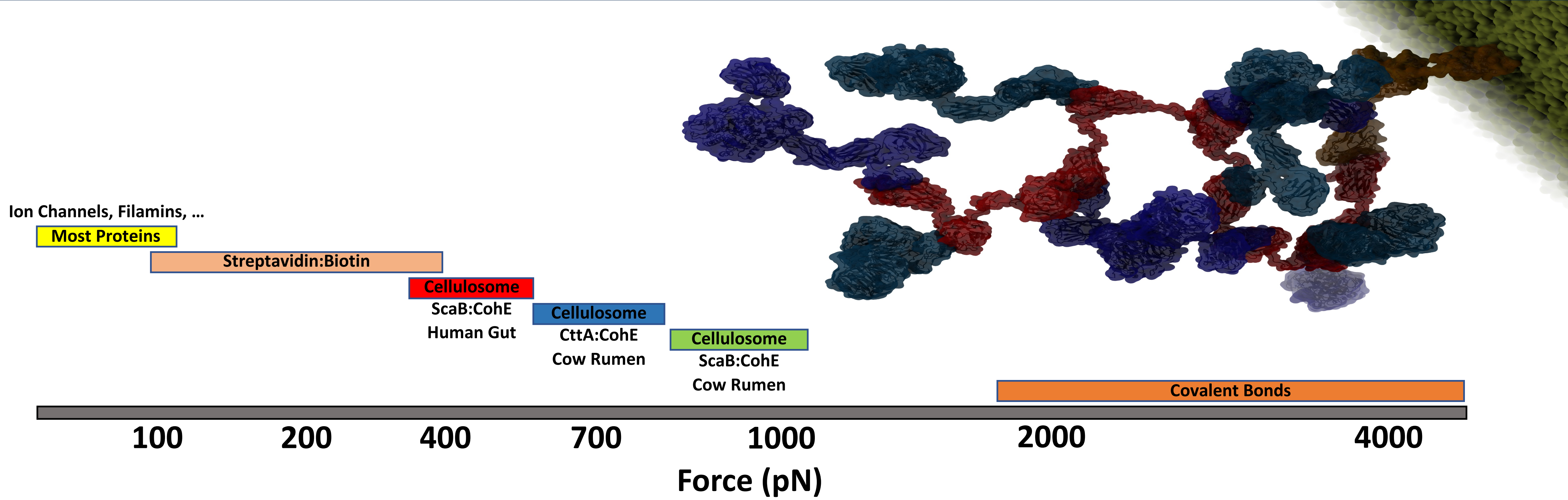
About half the rupture force of a covalent gold-thiol bond

C Schoeler*, KH Malinowska*, RC Bernardi, et. al.; **Ultrastable cellulosome-adhesion complex tightens under load.** Nature Communications, 2014

M Scheurer, P Rodenkirch, M Siggel, RC Bernardi, et. al.; **PyContact: Rapid, customizable, and visual analysis of noncovalent interactions in MD simulations.** Biophysical Journal, 2018

RC Bernardi, et al.; **Mechanisms of Nanonewton Mechanostability in a Protein Complex Revealed by Molecular Dynamics Simulations and Single-Molecule Force Spectroscopy.** JACS, 2019

Force Resilience in Biology



Ion Channels, Filamins, …

Most Proteins

Streptavidin:Biotin

Cellulosome
ScaB:CohE
Human Gut

Cellulosome
CttA:CohE
Cow Rumen

Cellulosome
ScaB:CohE
Cow Rumen

Covalent Bonds

C Schoeler*, KH Malinowska*, RC Bernardi, et. al.; **Ultrastable cellulosome-adhesion complex tightens under load.** Nature Communications, 2014

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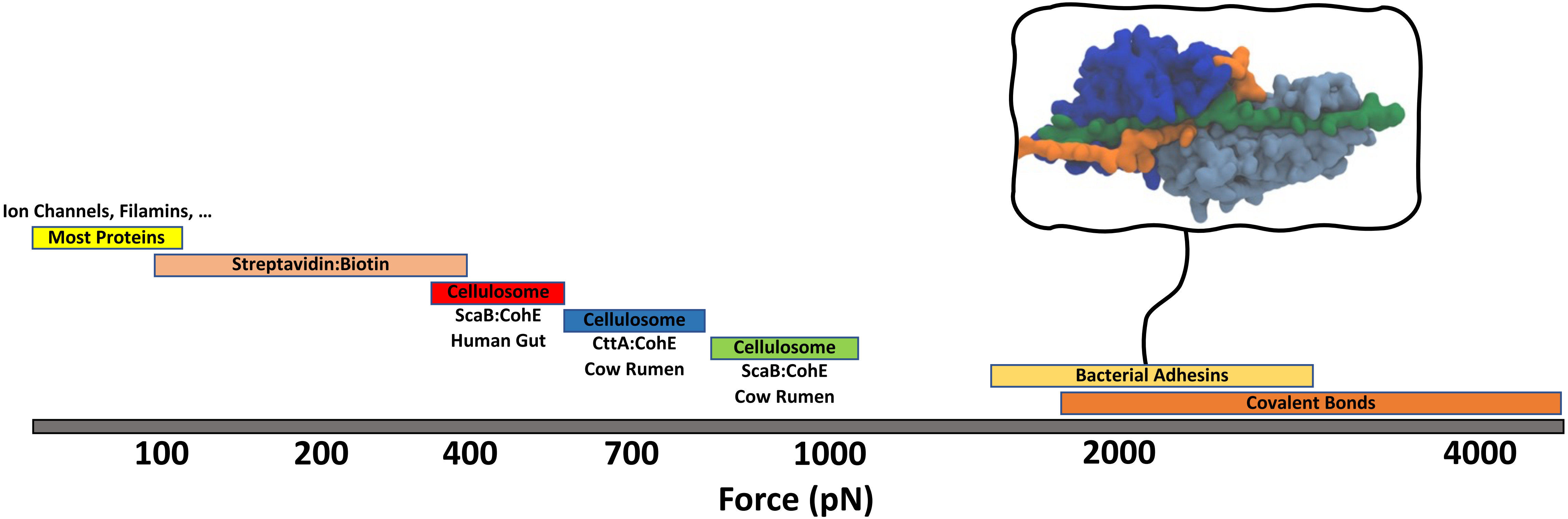

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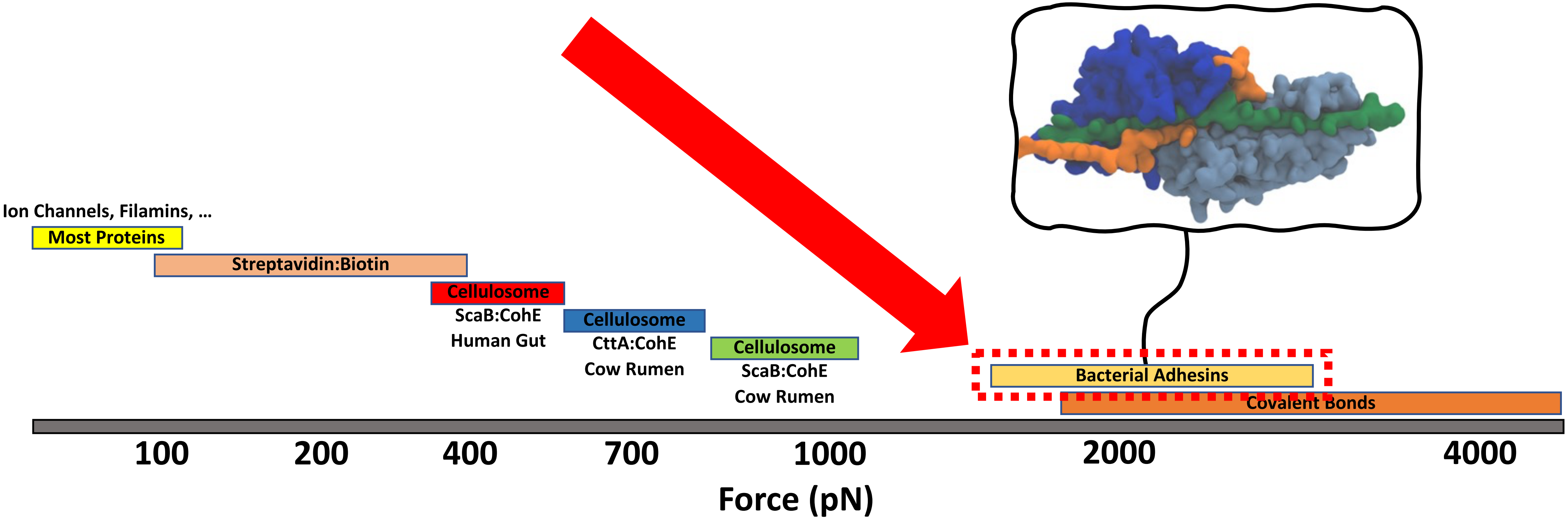
Force Resilience in Biology



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Most Proteins

Streptavidin:Biotin

Cellulosome
ScaB:CohE
Human Gut

Cellulosome
CttA:CohE
Cow Rumen

Cellulosome
ScaB:CohE
Cow Rumen

Bacterial Adhesins

Covalent Bonds

100 200 400 700 1000 2000 4000

Force (pN)

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Z Liu, H Liu, AM Vera, RC Bernardi, et. al.; **High Force Catch Bond Mechanism of Bacterial Adhesion in the Human Gut.** Nature Communications, 2020

MS Bauer, …, RC Bernardi, et. al.; **A Tethered Ligand Assay to Probe SARS-CoV-2:ACE2 Interactions.** PNAS, 2022

MCR Melo, DEB Gomes, RC Bernardi; **Molecular Origins of Force-Dependent Protein Complex Stabilization during Bacterial Infections.** JACS, 2023

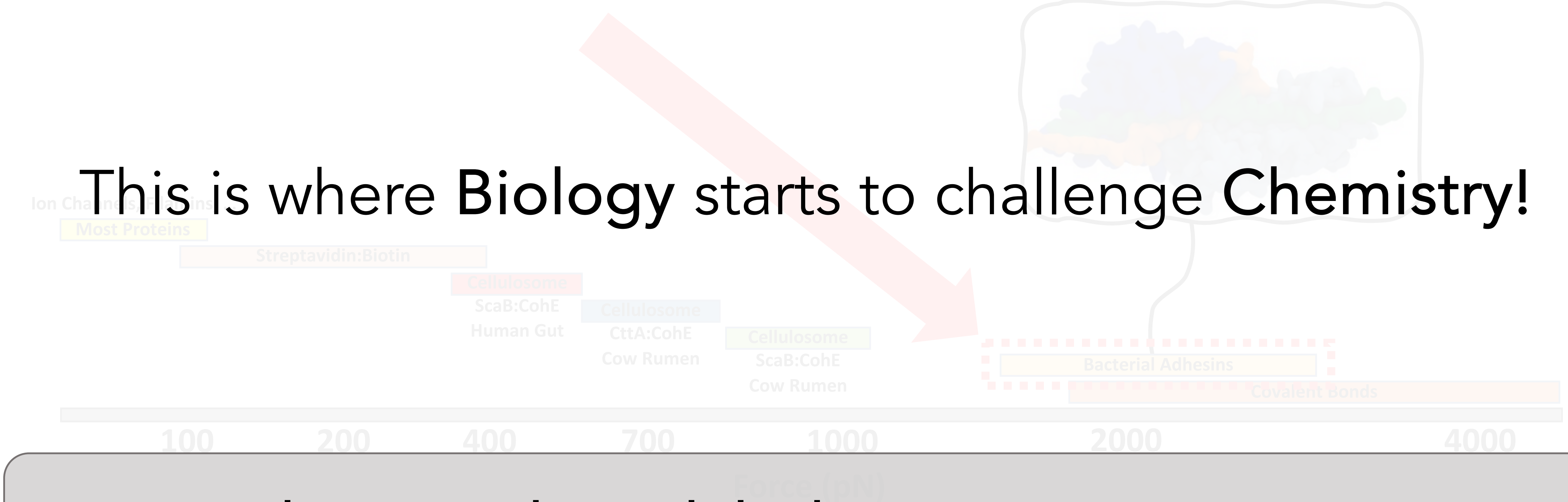
PSFC Gomes, …, RC Bernardi ; **May the force be with you: the role of hyper-mechanostability of the bone sialoprotein binding protein during early stages of Staphylococci infections.** Frontiers in Chemistry, 2023

RS Natividade, …, RC Bernardi, et. al.; **Deciphering molecular mechanisms stabilizing the reovirus-binding complex.** PNAS, 2023

S Gruber, …, RC Bernardi‡, J Lipfert‡; **Mechanical properties of SARS-CoV-2:ACE2 interaction and the role of the variants of concern.** Nature Nanotechnology, 2024

Force Resilience in Biology

This is where **Biology** starts to challenge **Chemistry!**



> Nature does not have labels!
>
> *Klaus Schulten*

T Verdorfer, RC Bernardi, et. al.; **Combining in Vitro and in Silico Single-Molecule Force Spectroscopy to Characterize and Tune Cellulosomal Scaffoldin Mechanics.** JACS, 2017

LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

TJK Haataja, RC Bernardi, et. al.; **Non-syndromic Mitral Valve Dysplasia Mutation Changes the Force Resilience and Interaction of Human Filamin A.** Structure, 2019

SM Sedlak*, LC Schendel*, MCR Melo, DA Pippig, Z Luthey-Schulten, HE Gaub, RC Bernardi; **Direction Matters – Monovalent Streptavidin/Biotin Complex under Load.** Nano Letters, 2019

RC Bernardi, et al.; **Mechanisms of Nanonewton Mechanostability in a Protein Complex Revealed by Molecular Dynamics Simulations and Single-Molecule Force Spectroscopy.** JACS, 2019

SM Sedlak*, LC Schendel*, HE Gaub, RC Bernardi; **Streptavidin/Biotin: Tethering Geometry Defines Unbinding Mechanics.** Science Advances, 2020

Z Liu, H Liu, AM Vera, RC Bernardi, et. al.; **High Force Catch Bond Mechanism of Bacterial Adhesion in the Human Gut.** Nature Communications, 2020

MS Bauer, ..., RC Bernardi, et. al.; **A Tethered Ligand Assay to Probe SARS-CoV-2:ACE2 Interactions.** PNAS, 2022

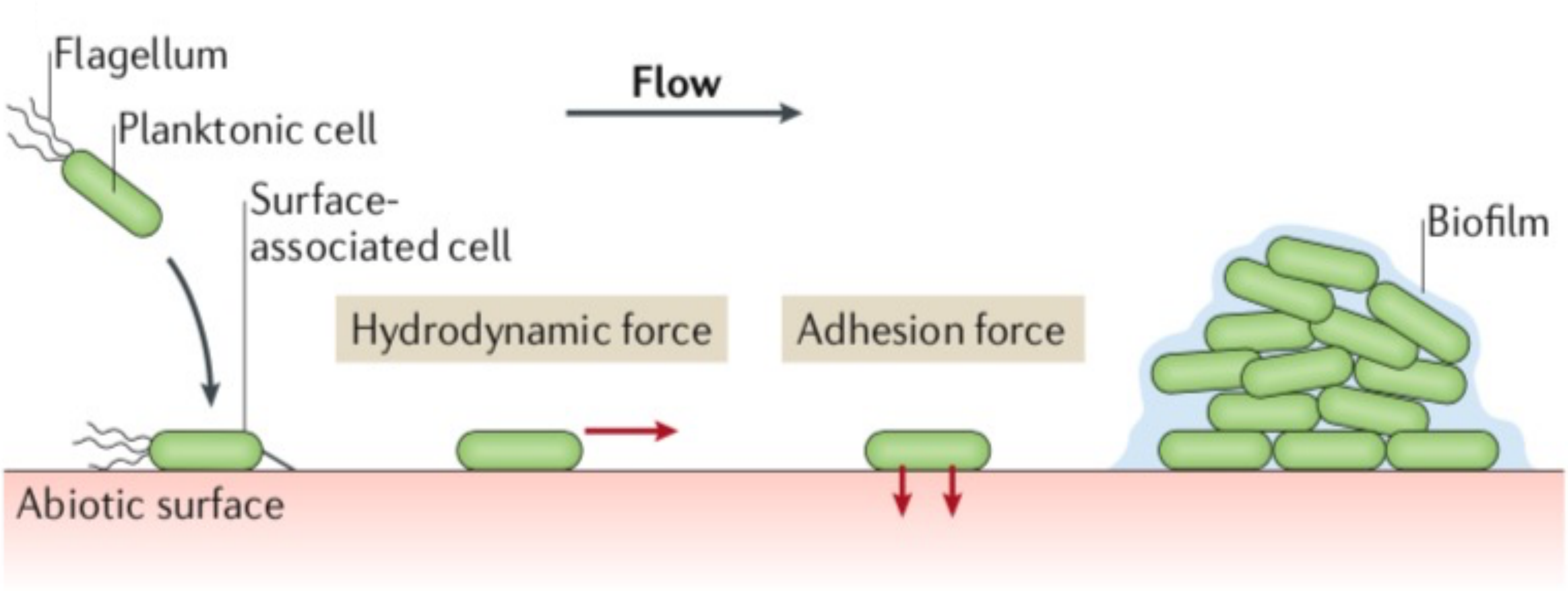
MCR Melo, DEB Gomes, RC Bernardi; **Molecular Origins of Force-Dependent Protein Complex Stabilization during Bacterial Infections.** JACS, 2023

PSFC Gomes, ..., RC Bernardi ; **May the force be with you: the role of hyper-mechanostability of the bone sialoprotein binding protein during early stages of Staphylococci infections.** Frontiers in Chemistry, 2023

RS Natividade, ..., RC Bernardi, et. al.; **Deciphering molecular mechanisms stabilizing the reovirus-binding complex.** PNAS, 2023

S Gruber, ..., RC Bernardi‡, J Lipfert‡; **Mechanical properties of SARS-CoV-2:ACE2 interaction and the role of the variants of concern.** Nature Nanotechnology, 2024

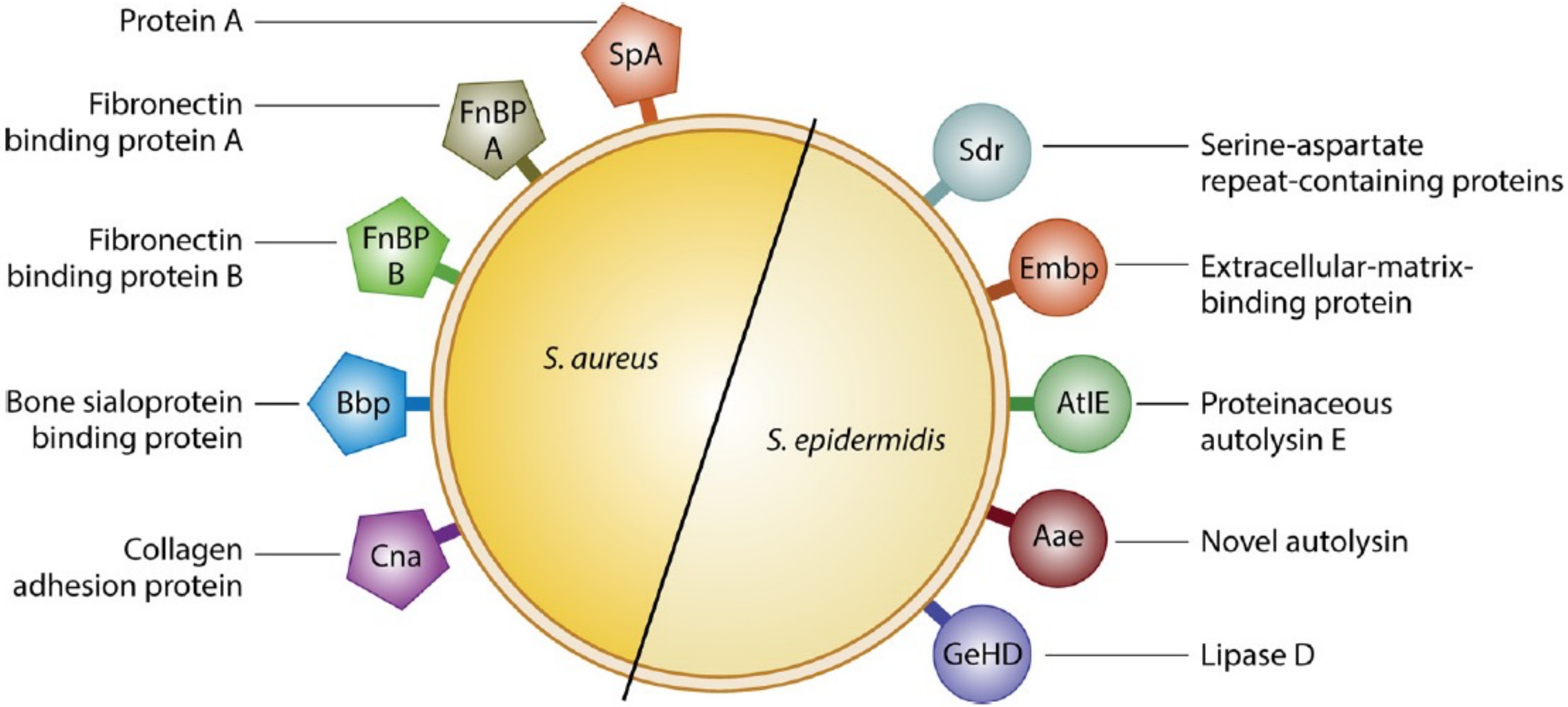
Mechanics of bacteria infection



YF Dufrêne & A Persat; Nature Reviews Microbiology, 2020

Adhesion by Pathogenic Bacteria: Staphylococcus' Cell Surface Proteins

Microbial Surface Components Recognizing Adhesive Matrix Molecules (MSCRAMMs)



Human targets include Fibrinogen (Fg, all chains), Fibronectin (Fn), Keratin, Collagen, Elastin, Complement Factor H, …

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Adapted from: Kavanagh, et. al.; Clinical Microbiology Reviews, 2018

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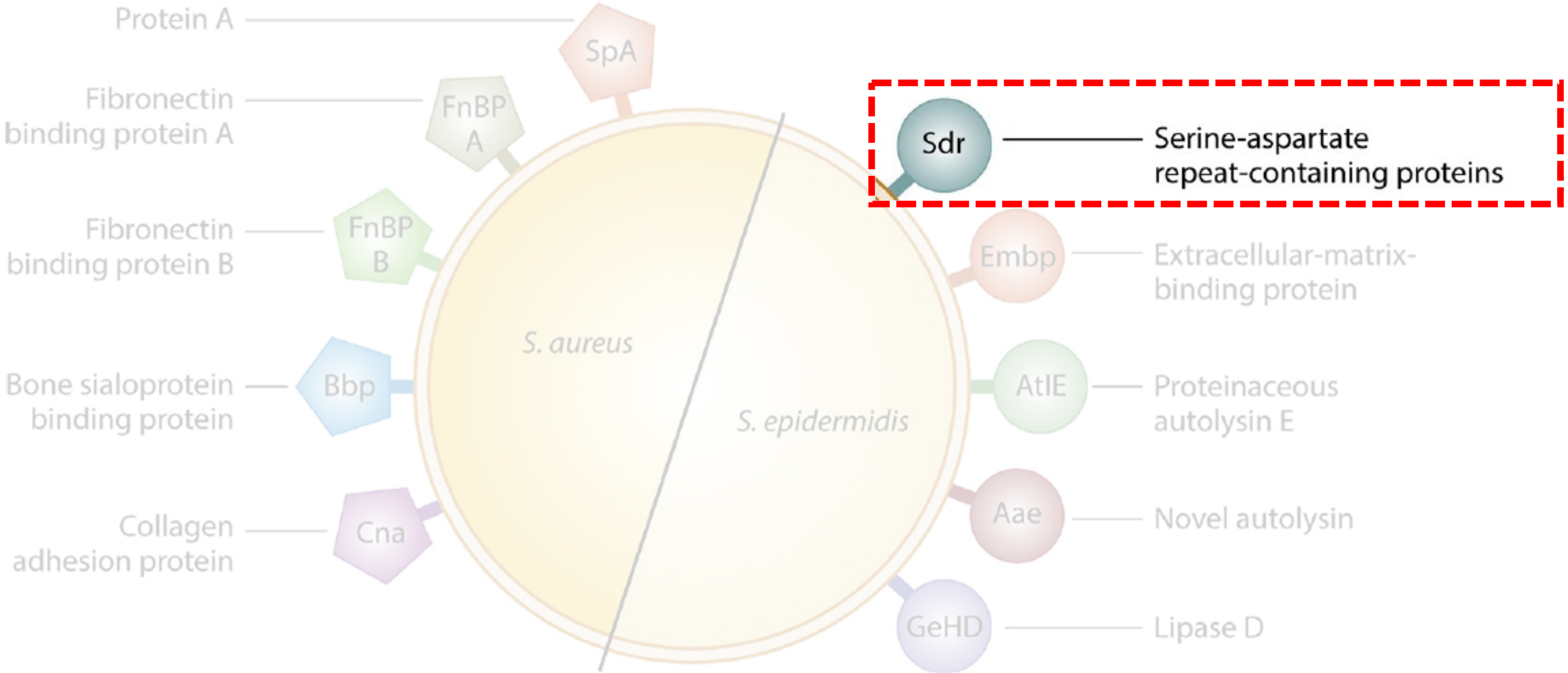

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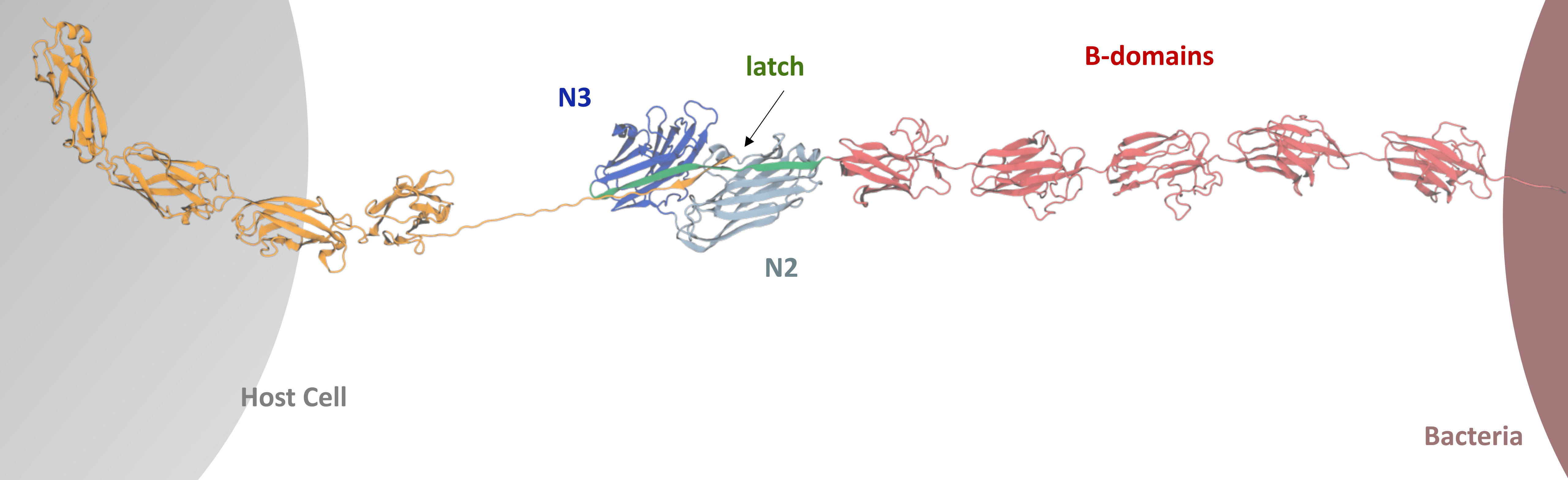
Adapted from: Kavanagh, et. al.; Clinical Microbiology Reviews, 2018

How is the molecular complex formed by these proteins?

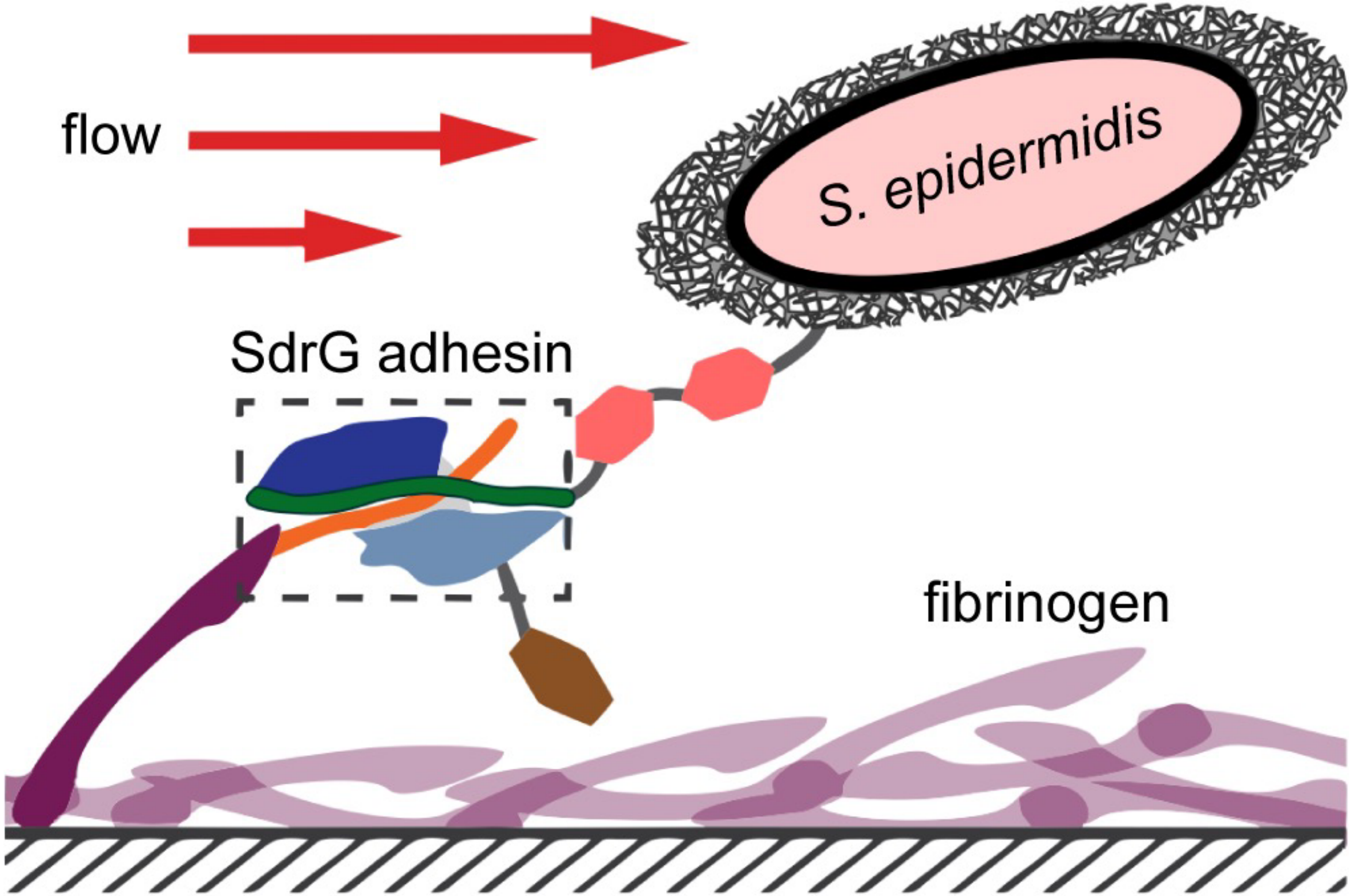
(Dock – Lock – Latch) DLL Mechanism

Yves Dufrêne
UCLouvain



Data not published

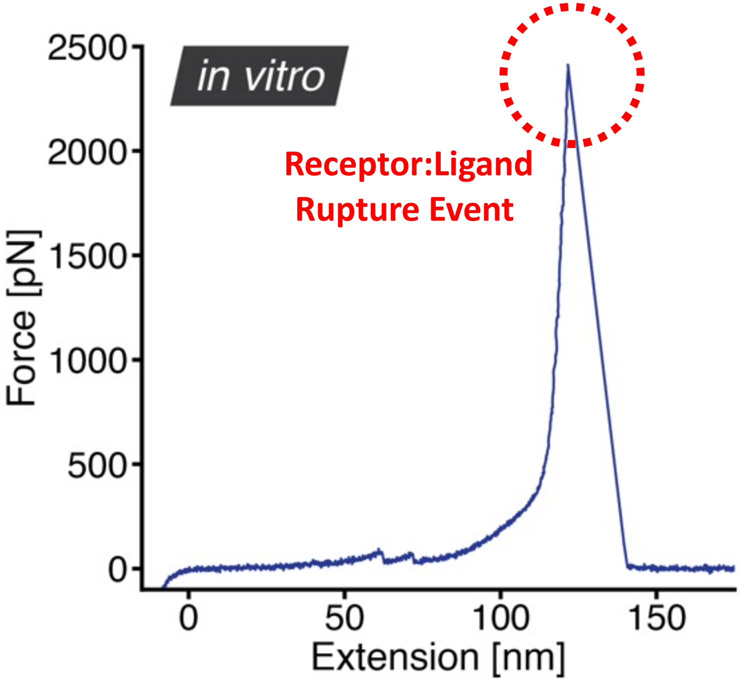
Staphylococci Adhesins



The Hyperstable SdrG:Fgb interaction

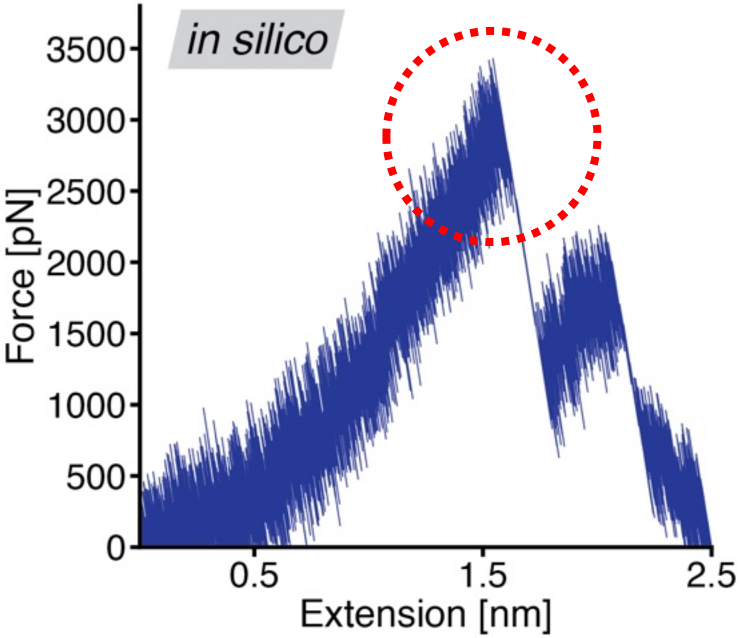
Lukas F. Milles

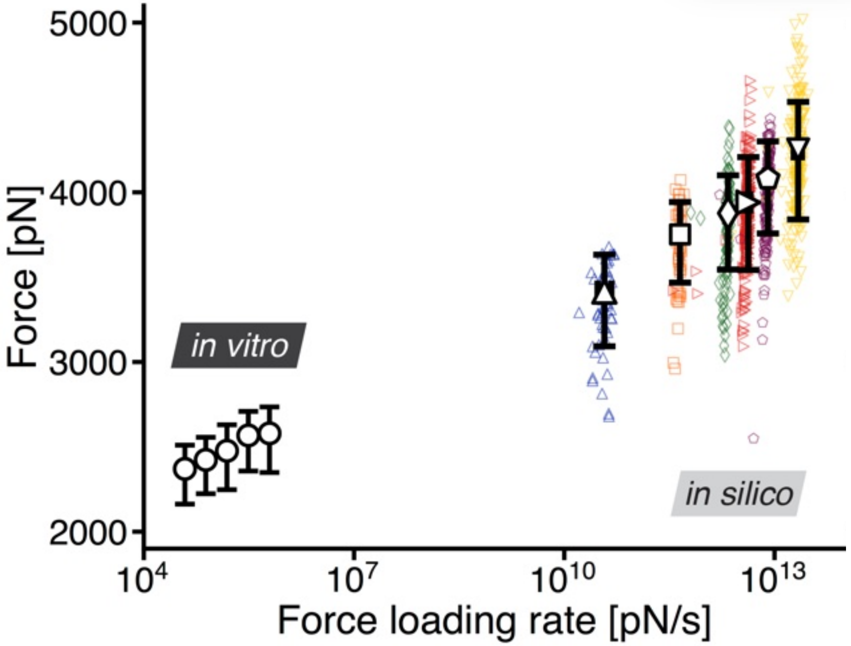
Hermann Gaub
(LMU)



Receptor:Ligand Rupture Event

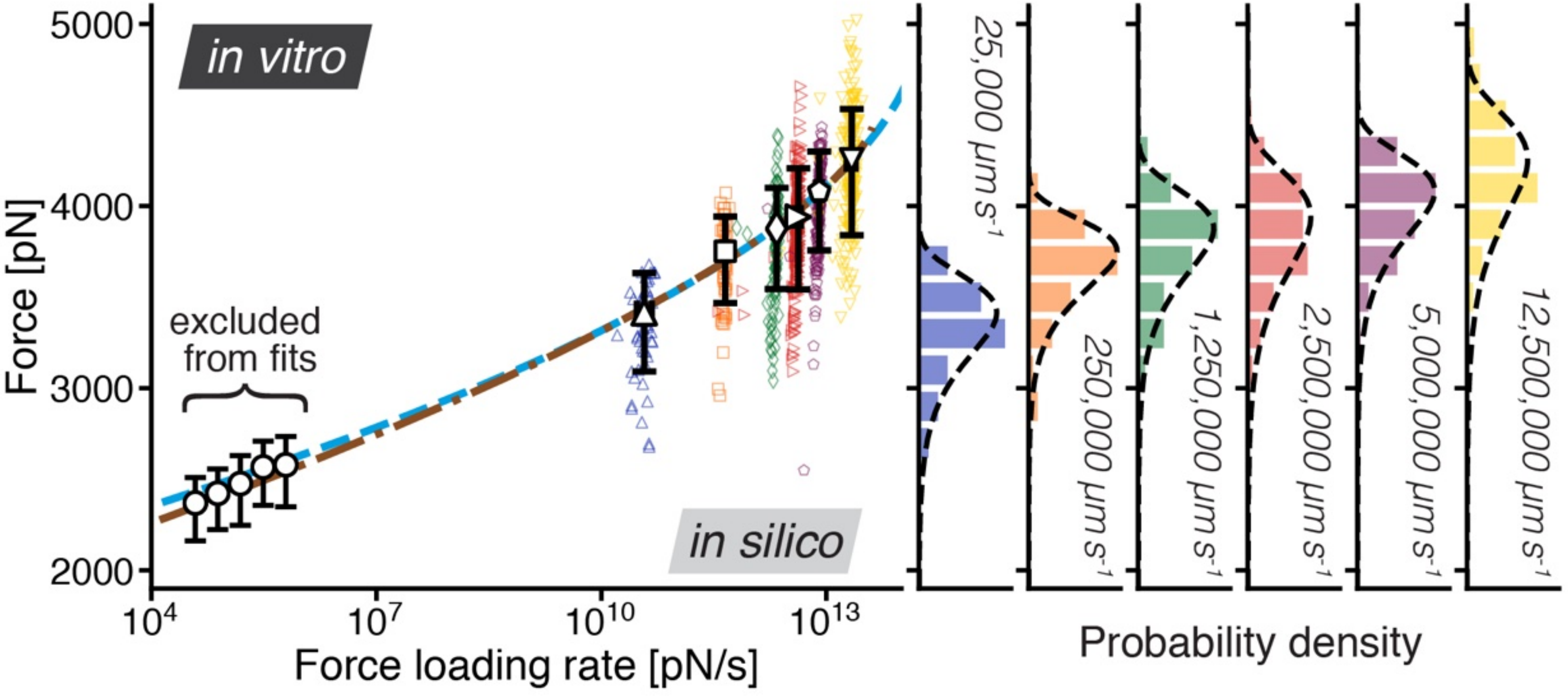
Receptor:Ligand Rupture Event





LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

Dudko-Hummer-Szabo (DHS) Theory: Simulation and experiments agreement



Two Force regimes:

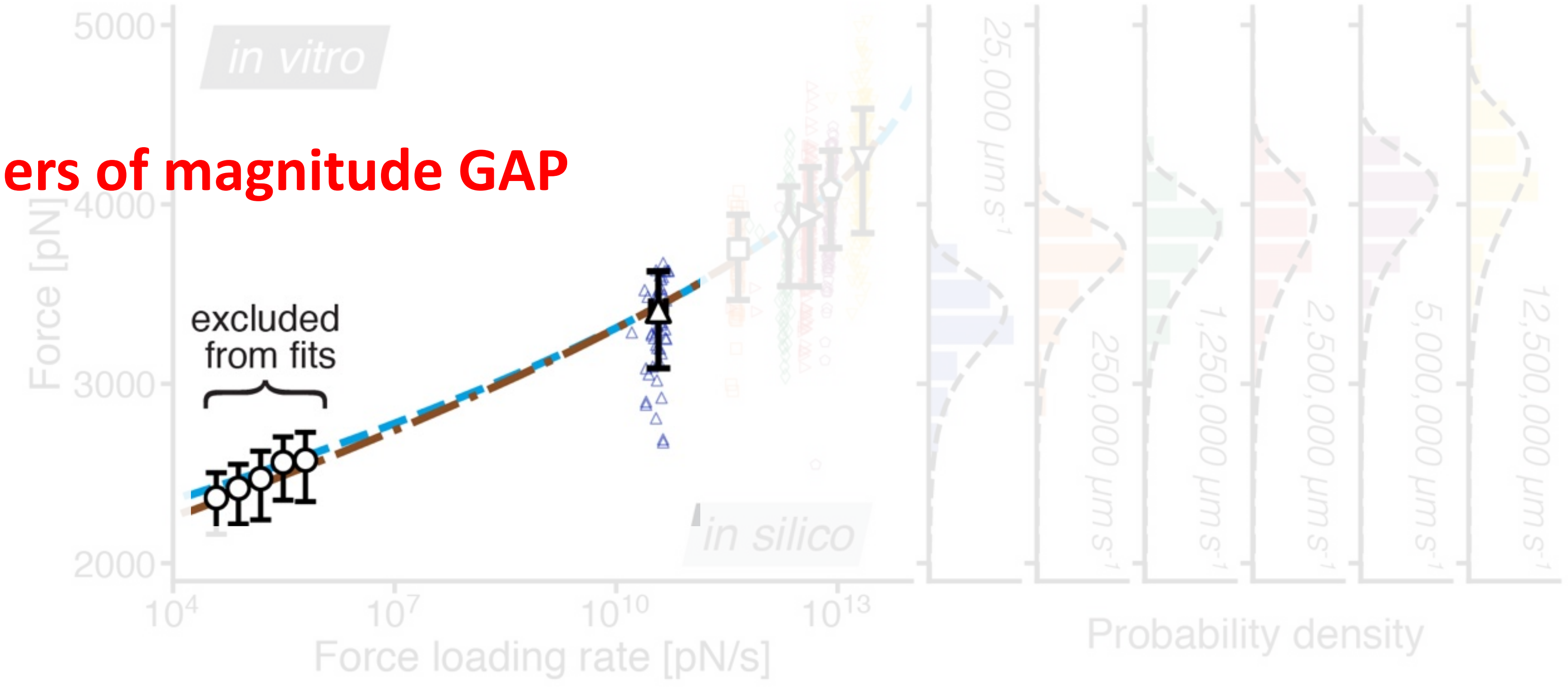
1. Experiments are in the diffusive regime for unfolding (primarily driven by diffusion over the barrier)
2. Simulations are likely in a transition point to a deterministic regime (driven by fast extension with no contribution from diffusion)

$$< F(r) >= \frac{\Delta G}{\nu \Delta x}\left\{1-\left[\frac{k_B T}{\Delta G} ln\left(\frac{k_0\, k_B T}{r\, \Delta x} exp\left(\frac{\Delta G}{k_B T}+\gamma\right)\right)\right]^{\nu}\right\}$$

Dudko, et. al.; Physical Review Letters, 2006
Bullerjahn, et. al.; Nature Communications, 2014
T Verdorfer, RC Bernardi, et. al.; **Combining in Vitro and in Silico Single-Molecule Force Spectroscopy to Characterize and Tune Cellulosomal Scaffoldin Mechanics.** JACS, 2017
LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

Dudko-Hummer-Szabo (DHS) Theory: Simulation and experiments agreement

5 orders of magnitude GAP



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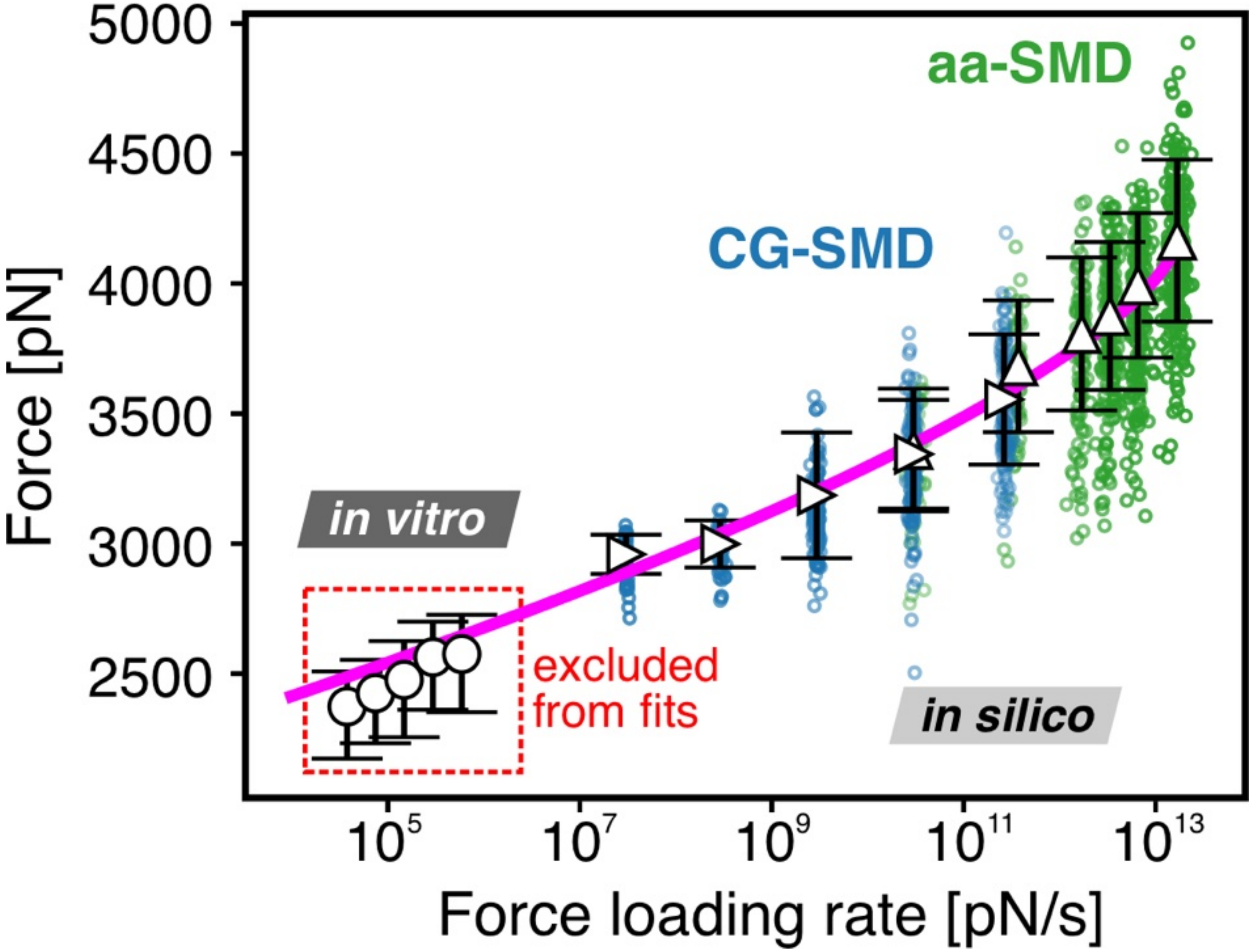
LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

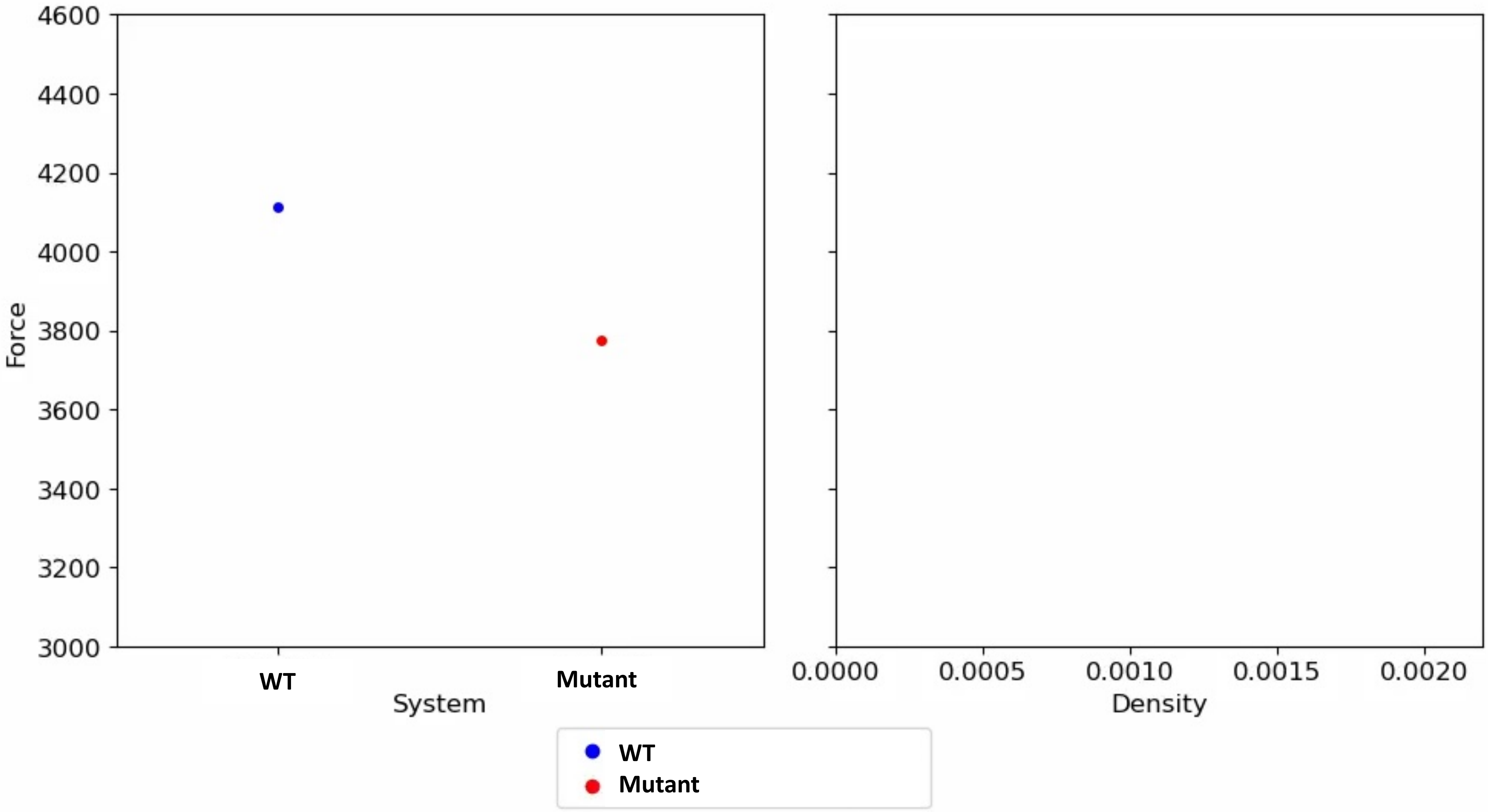
Bridging the Gap between Experiments and Simulations

Diego EB Gomes



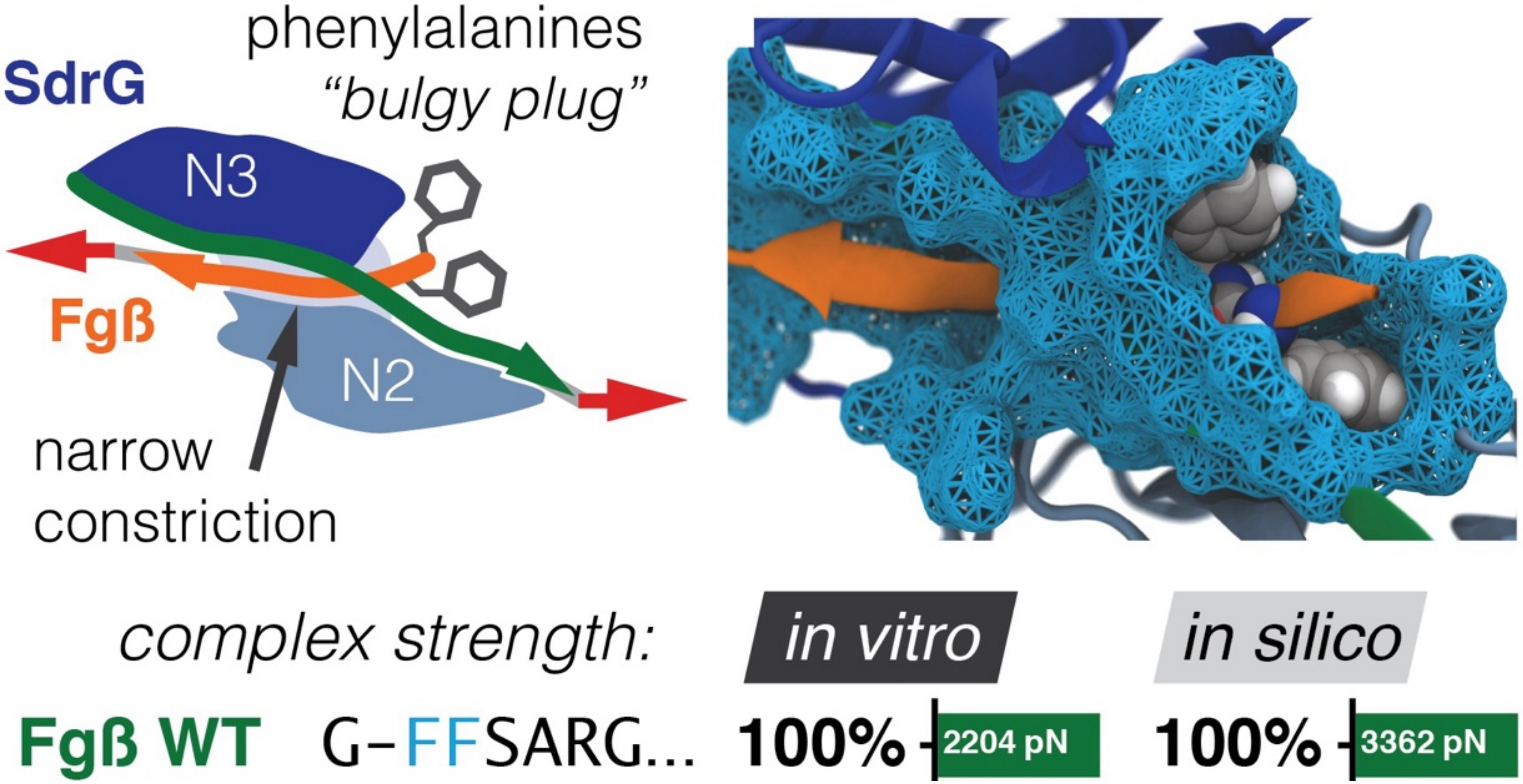
DEB Gomes, …, RC Bernardi; **Bridging the gap between in vitro and in silico single-molecule force spectroscopy.** bioRxiv, 2022

Why do we need so many simulation replicas?



The mechanism of the hyperstable SdrG:Fgβ interaction

Hidrogen bond network



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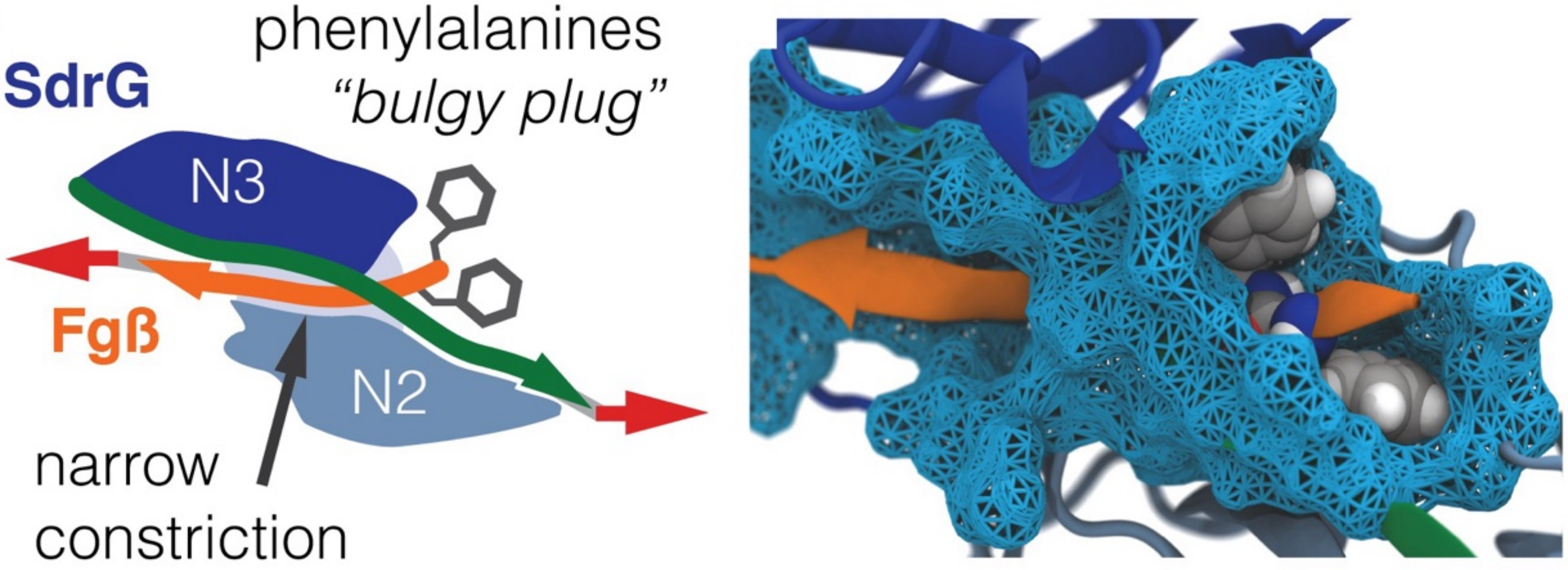
rcbernardi@auburn.edu

LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

Hidrogen bond network



complex strength:		*in vitro*		*in silico*	
Fgß WT	G-FFSARG...	**100%**	2204 pN	**100%**	3362 pN
FgßF3	GFFFSARG...	101%	2236 pN	104%	3486 pN

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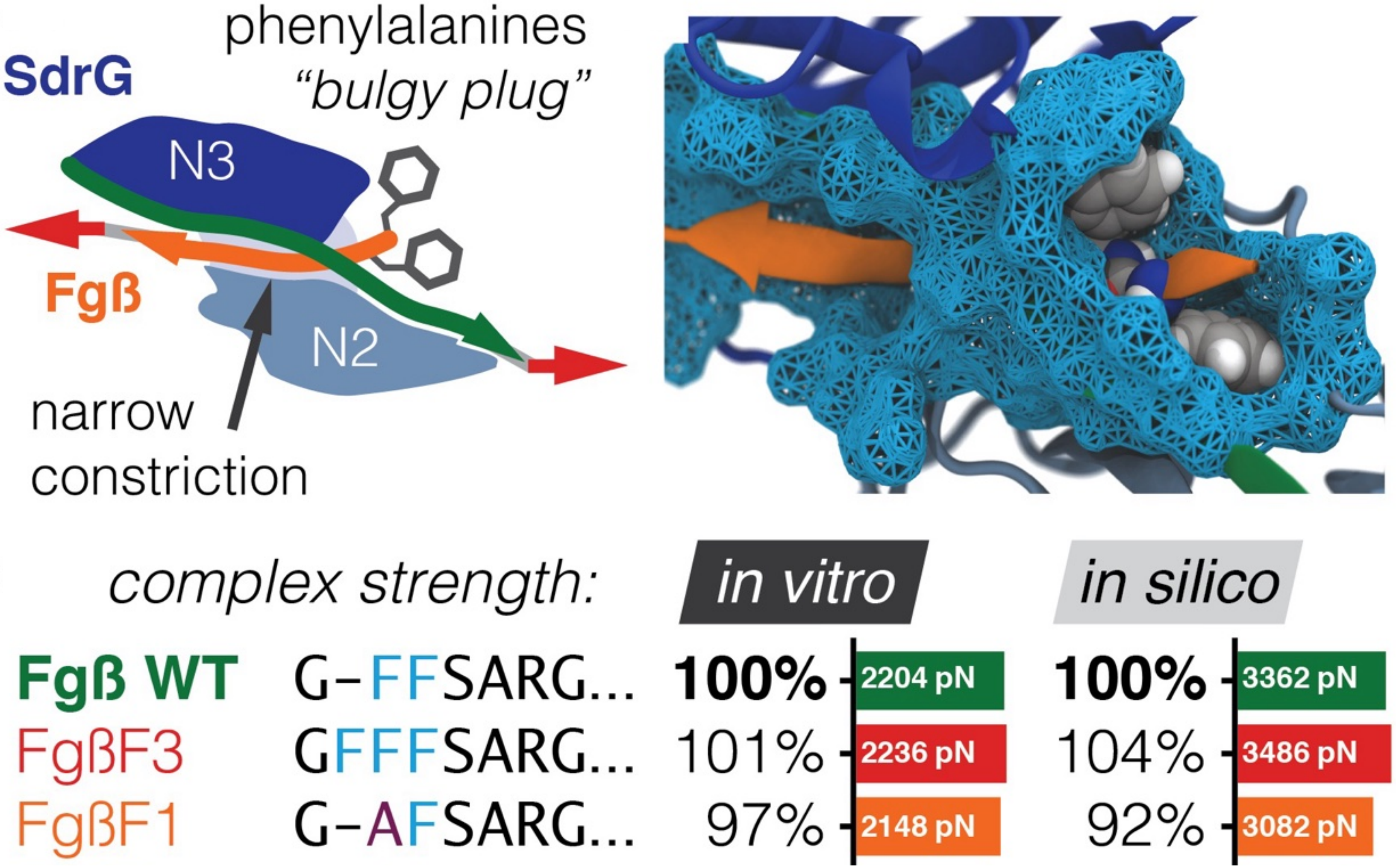

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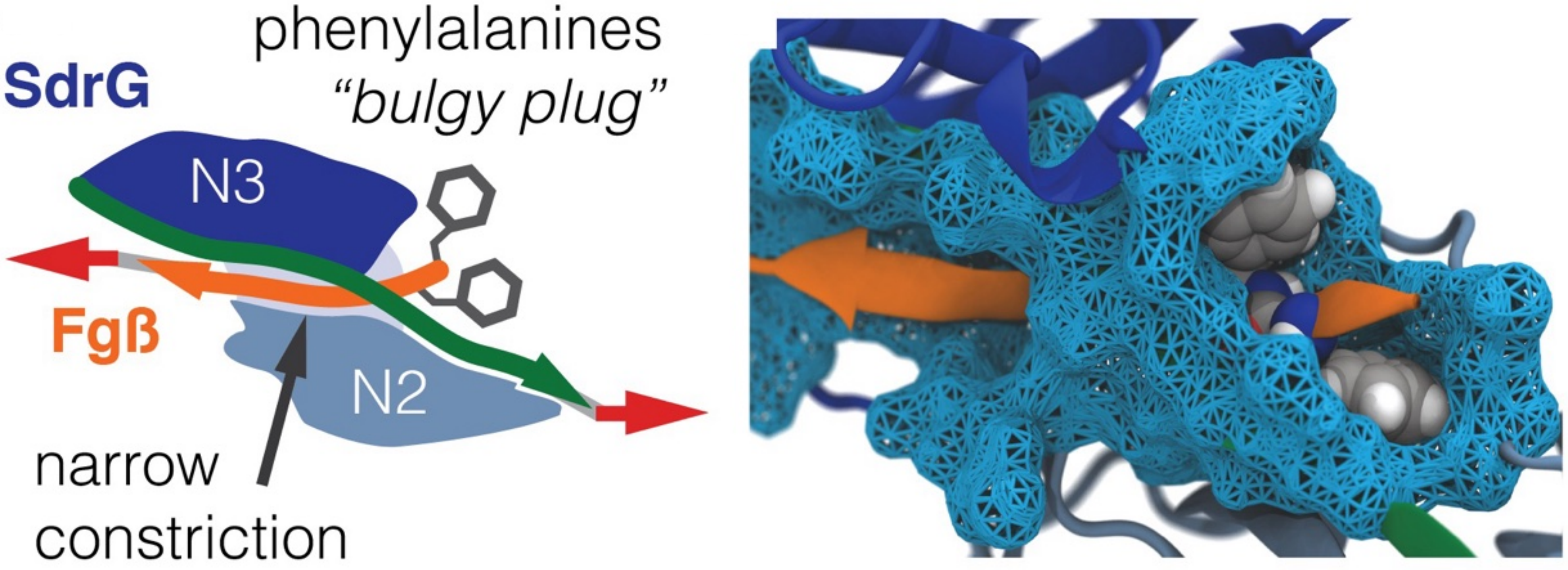

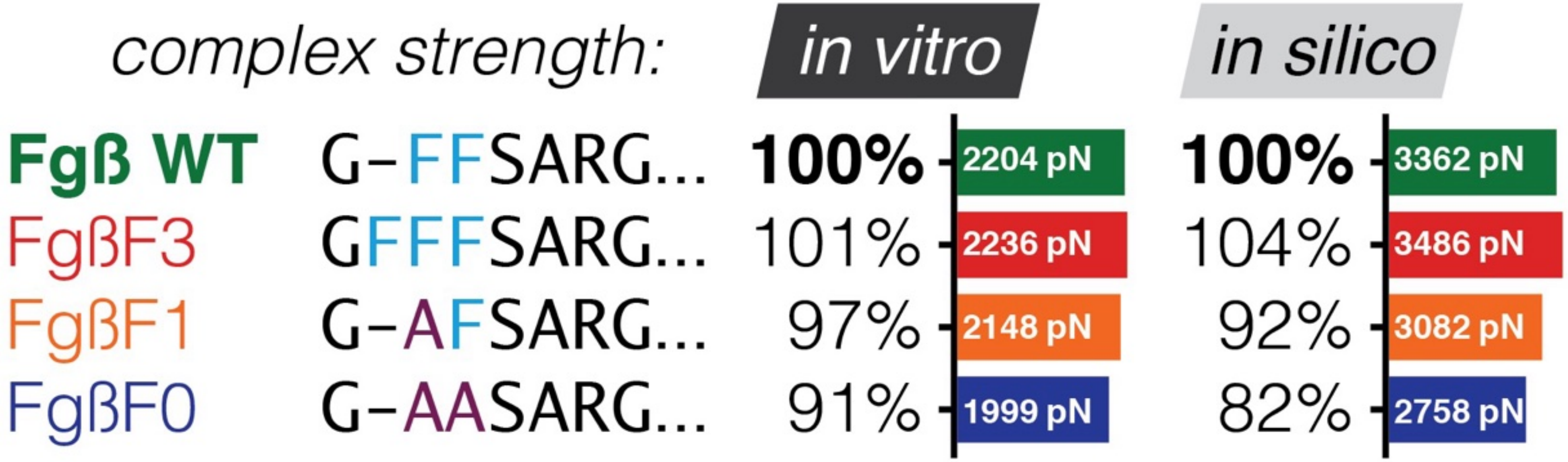

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FgßF1	G-AFSARG...	97%	2148 pN	92%	3082 pN
FgßF0	G-AASARG...	91%	1999 pN	82%	2758 pN

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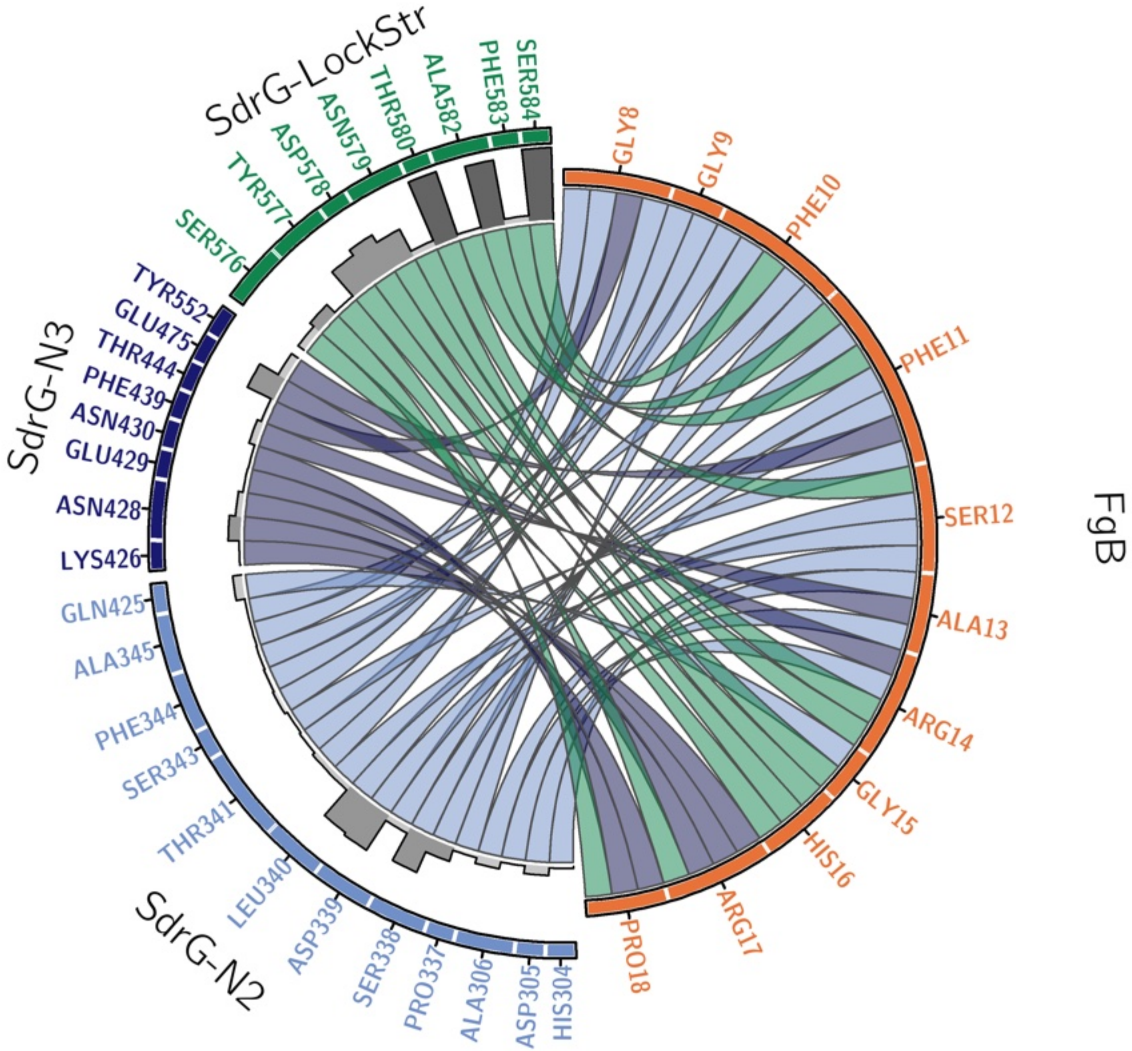
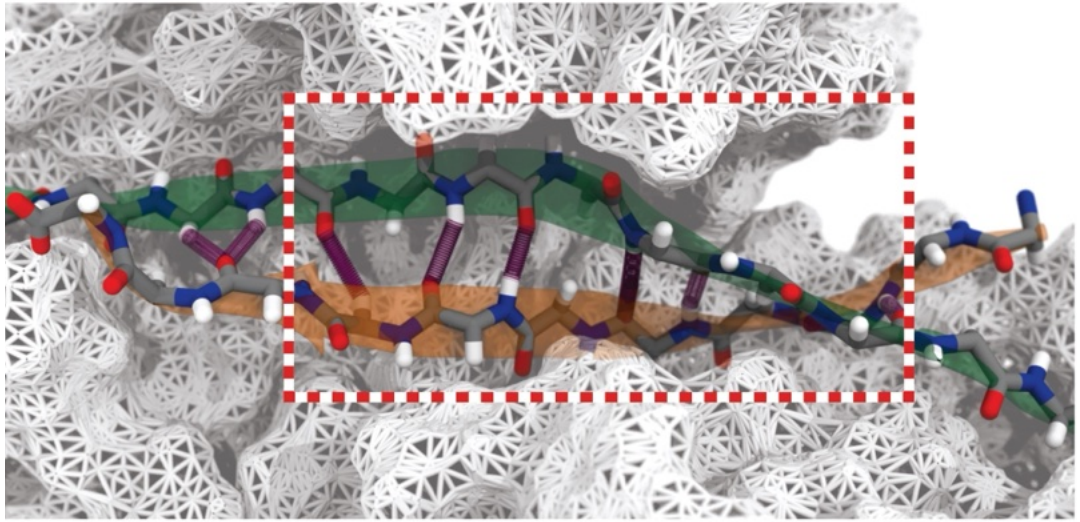
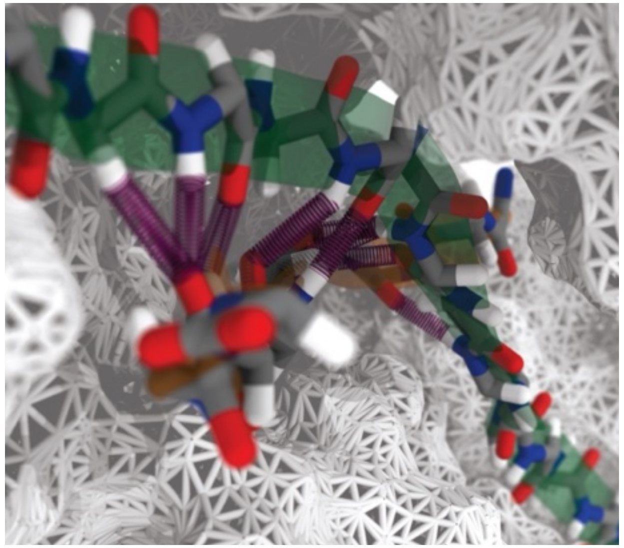
LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

Hidrogen bond network







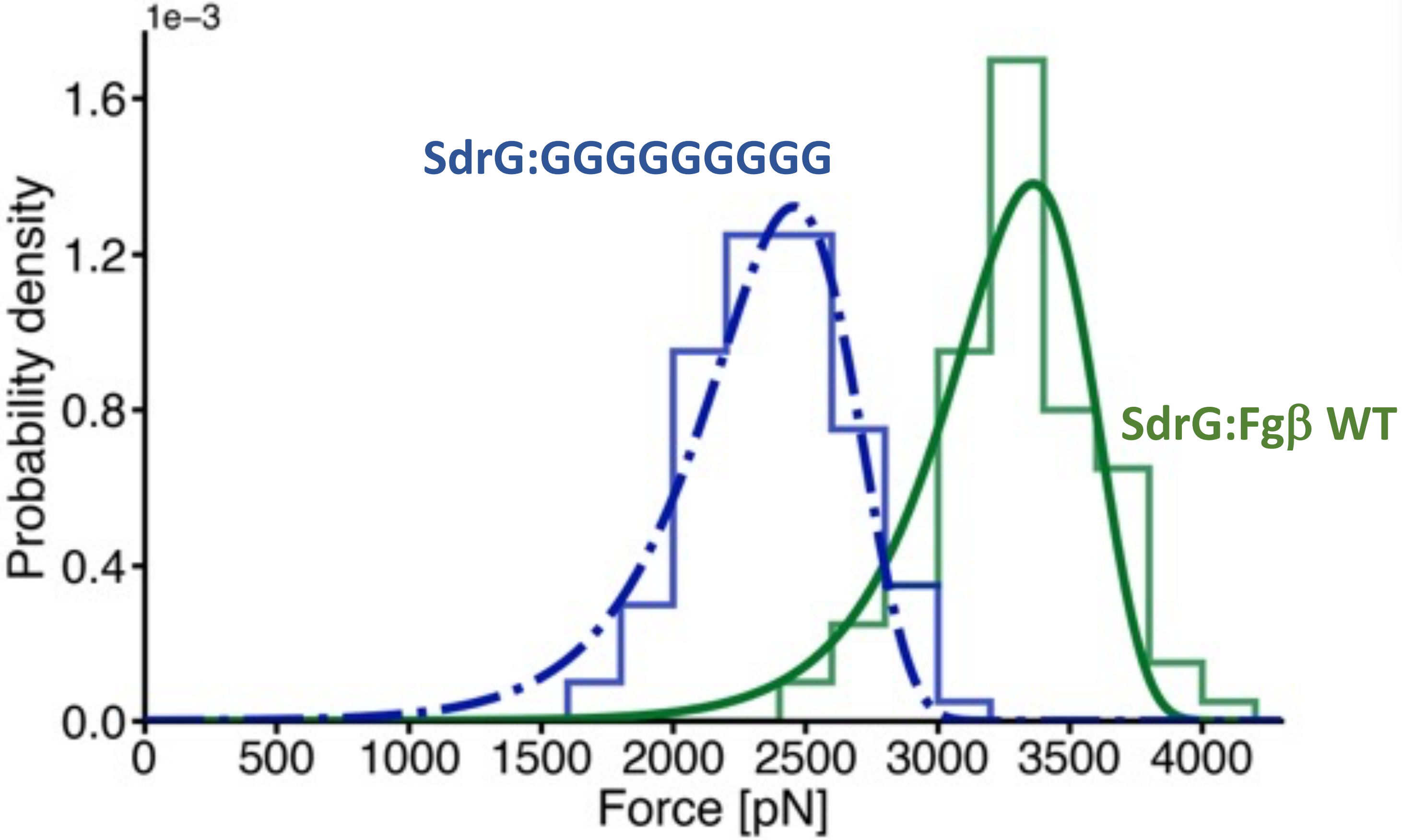
A bond with a twist!

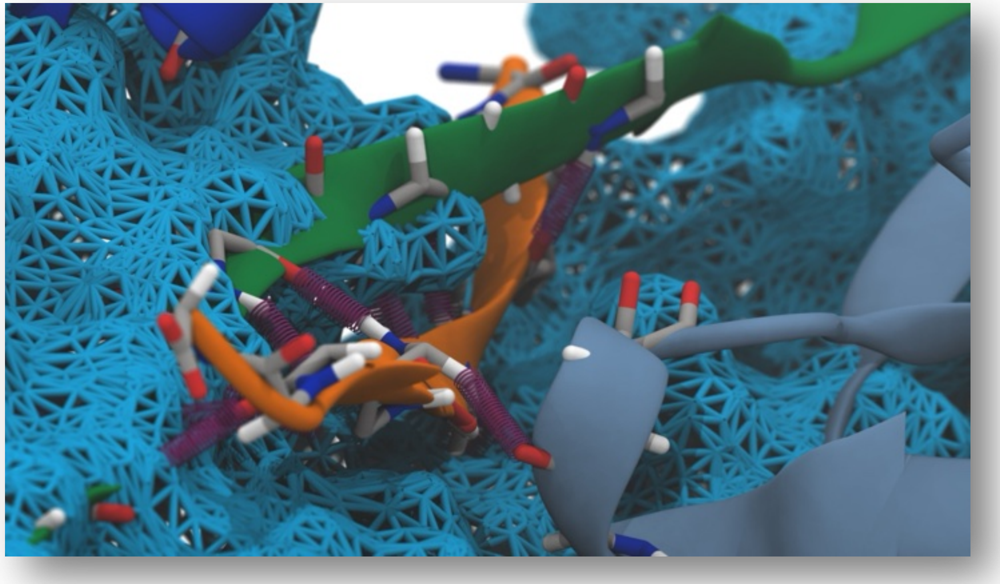
The corkscrew shape makes the complex hyperstable

LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

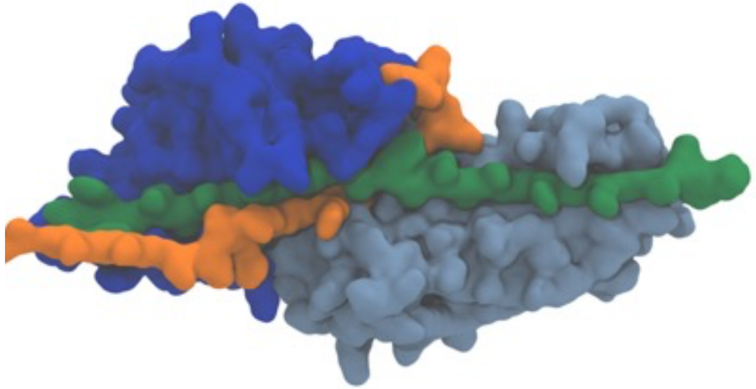
Side-chain Independence: Sequence Independence

Amino acid sequence is very important for binding affinity, but once it is bound even a polyglycine would become hyperstable





strong confinement



LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

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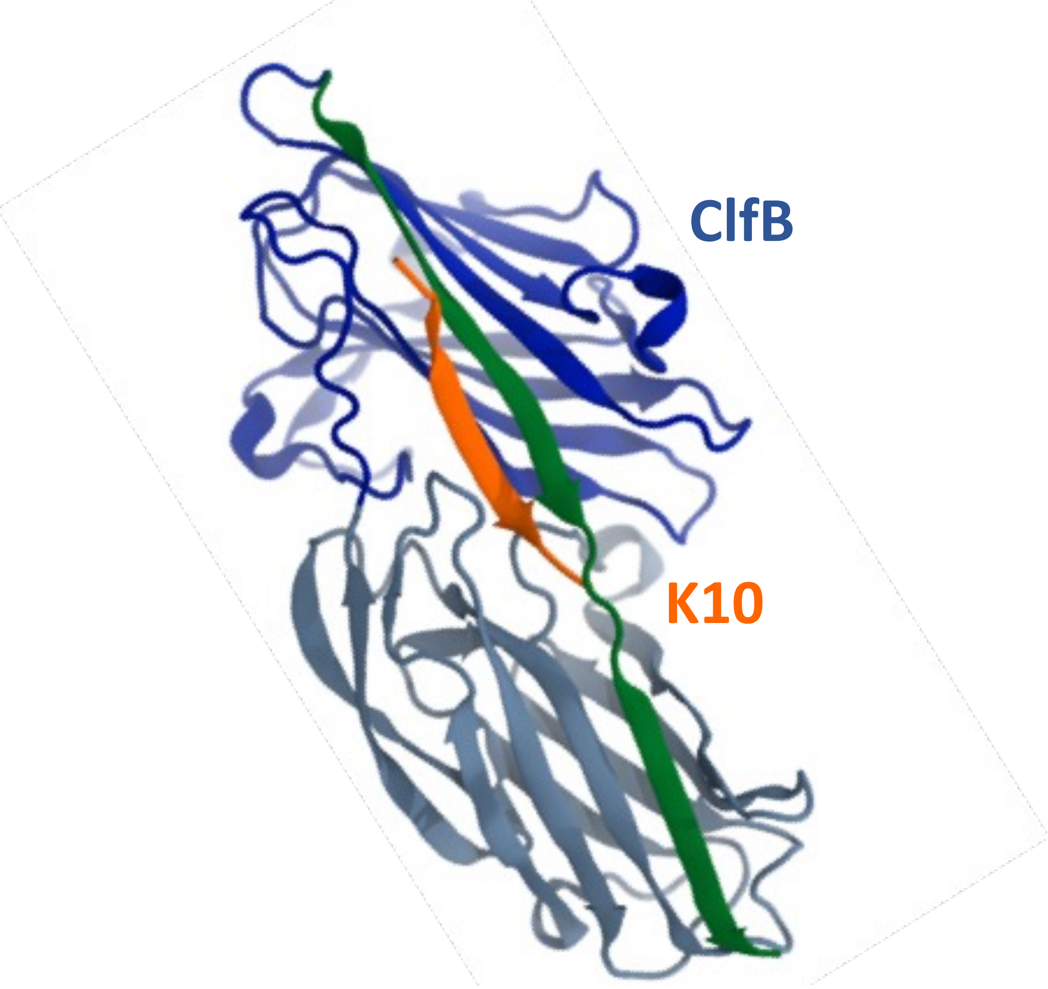
Side-chain Independence: Sequence Independence

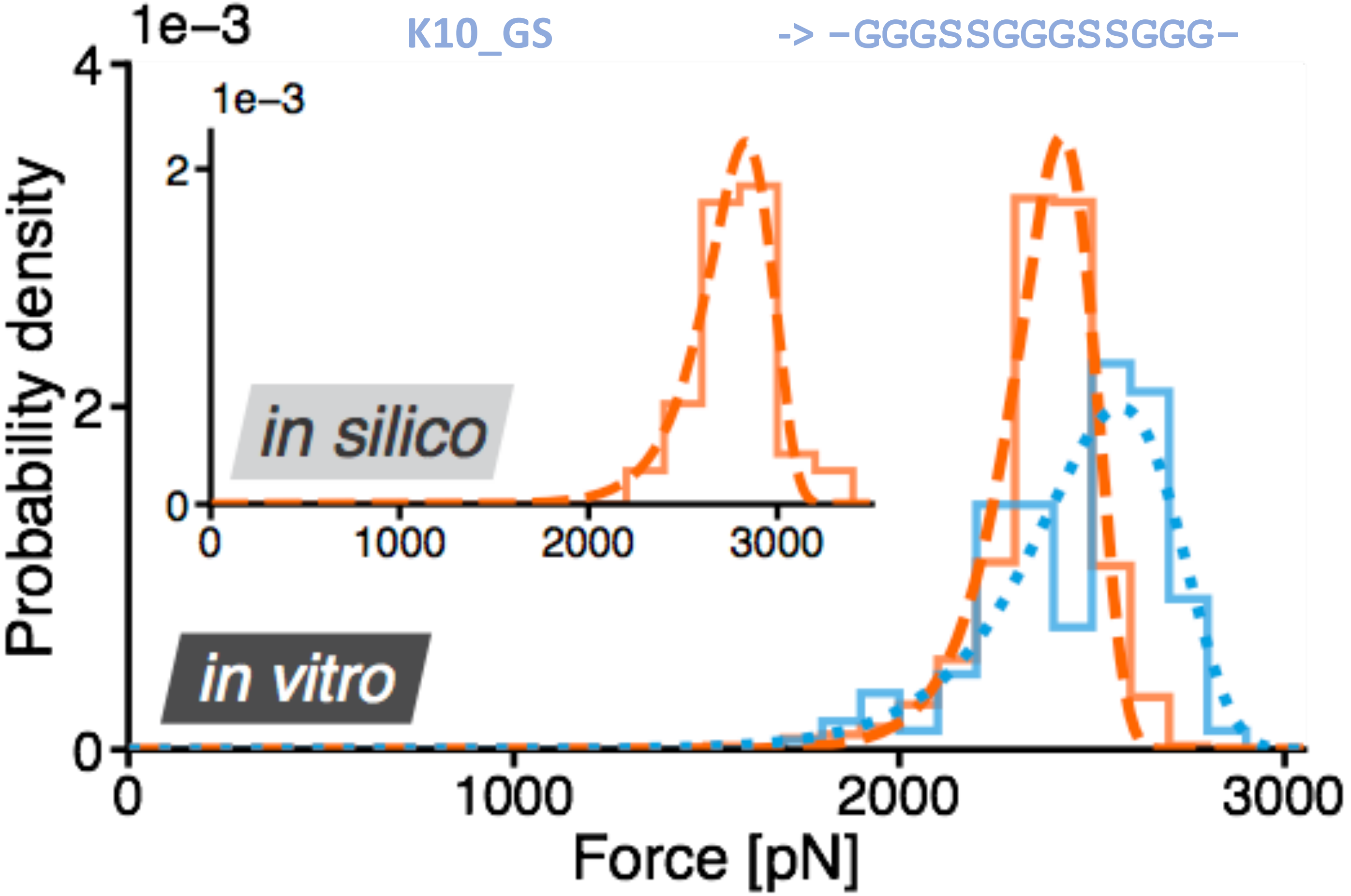
A sequence of GS-repeats is sufficient to withstand 2nN



ClfB from *S. aureus*, same Dock, Lock & Latch mechanism, binds C-Terminal peptides:

Keratin K10 -> `YGGGSSGGGSSGGGH`

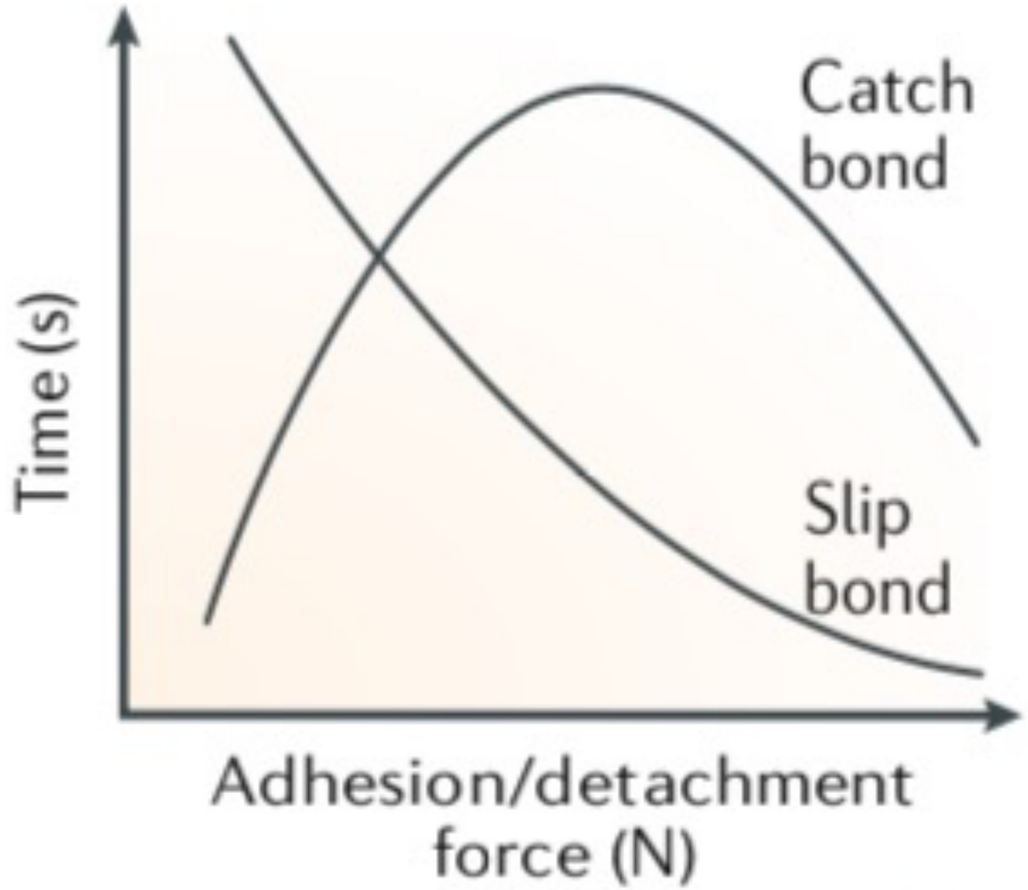
K10_GS -> `-GGGSSGGGSSGGG-`

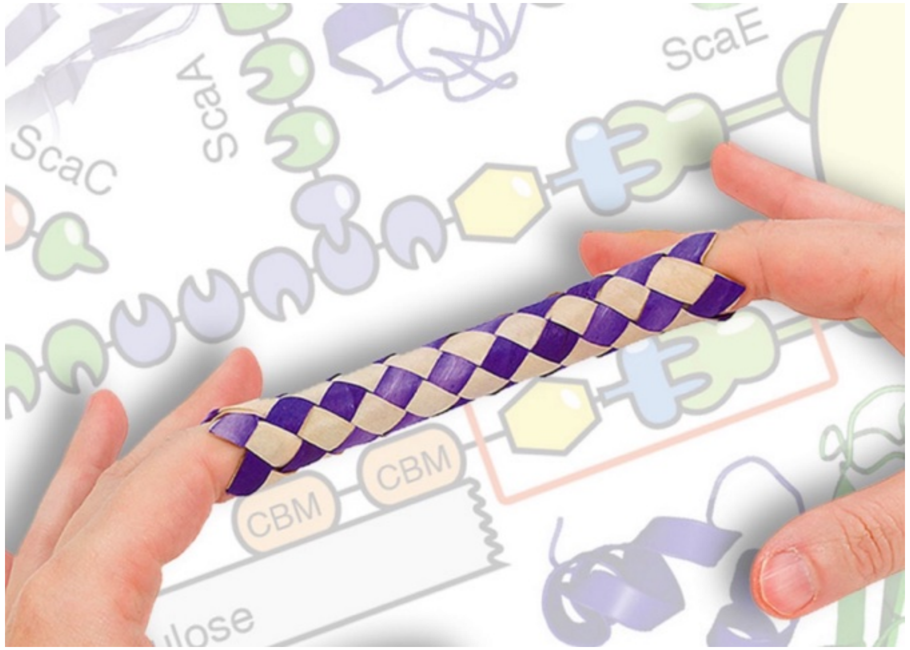


LF Milles, K Schulten, HE Gaub, RC Bernardi; **Molecular mechanism of extreme mechanostability in a pathogen adhesin.** Science, 2018

But is this really a catch-bond?

Catch Bonds





Molecular Finger Trap Puzzle

Common Affinity
High Mechanostability

Rafael C. Bernardi
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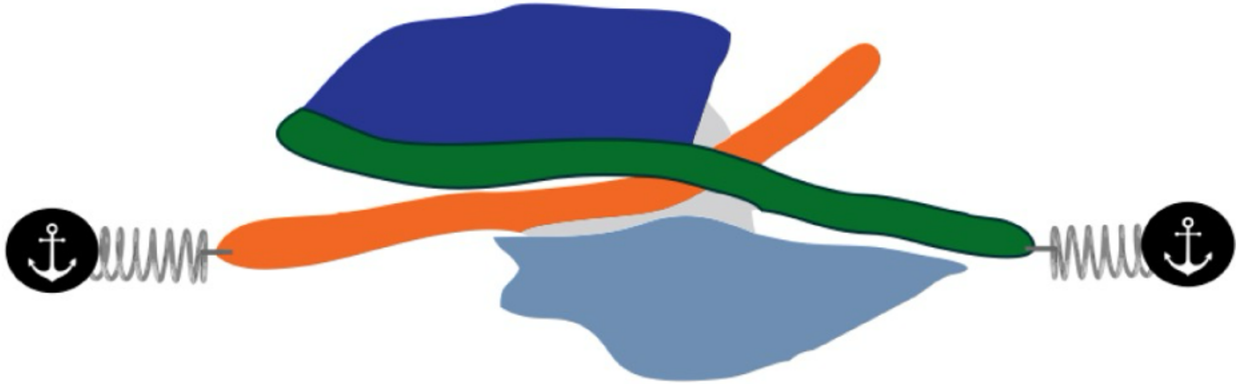
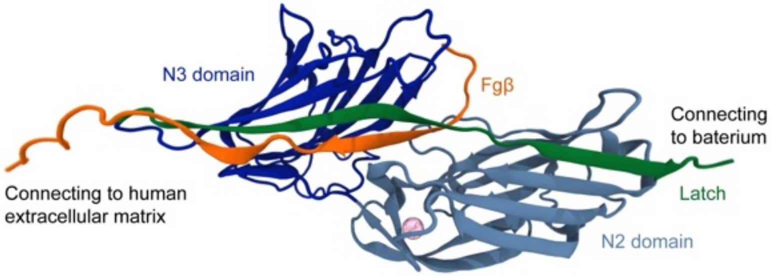
YF Dufrêne & A Persat; Nature Reviews Microbiology, 2020

Mechanics of Staphylococci Adhesins

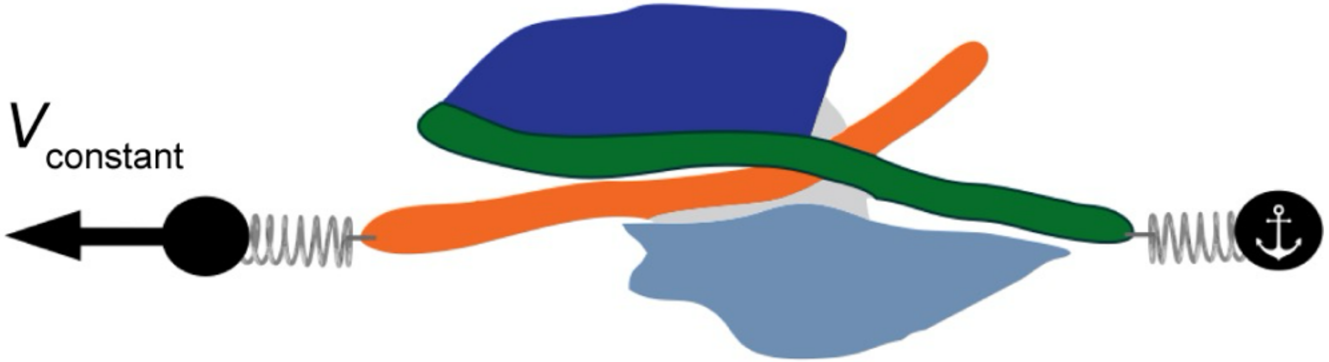
magnetic tweezer based SMFS

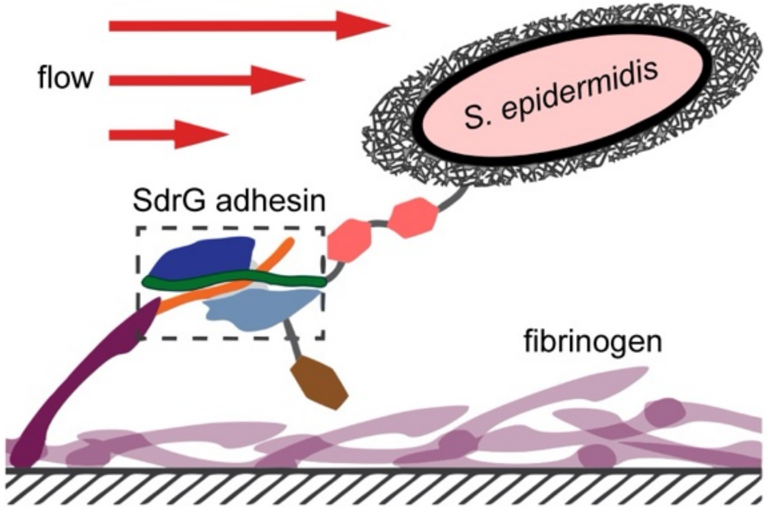


Experimental data: Huang et. al. JACS, 2022



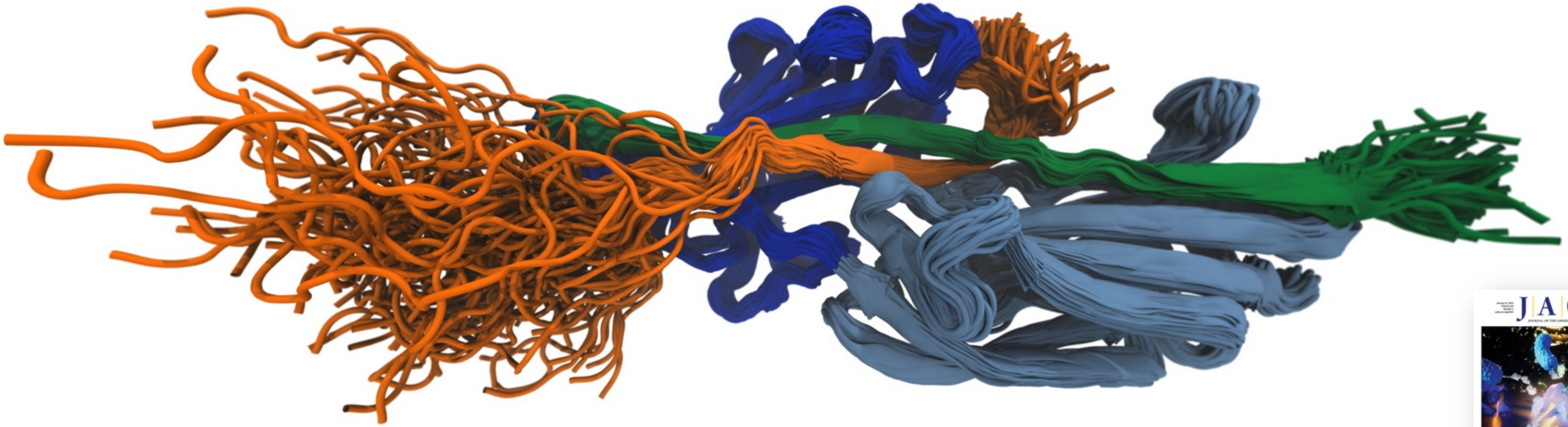
atomic force microscopy based SMFS





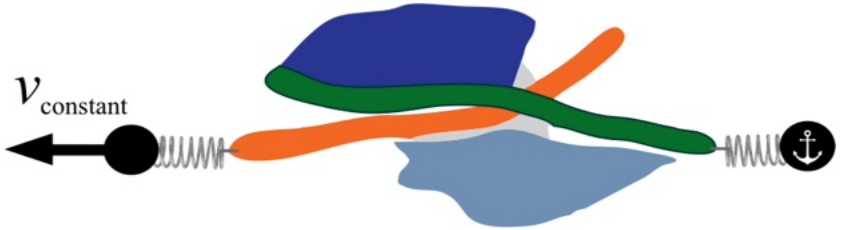
Experimental data: Milles et. al. Science, 2018

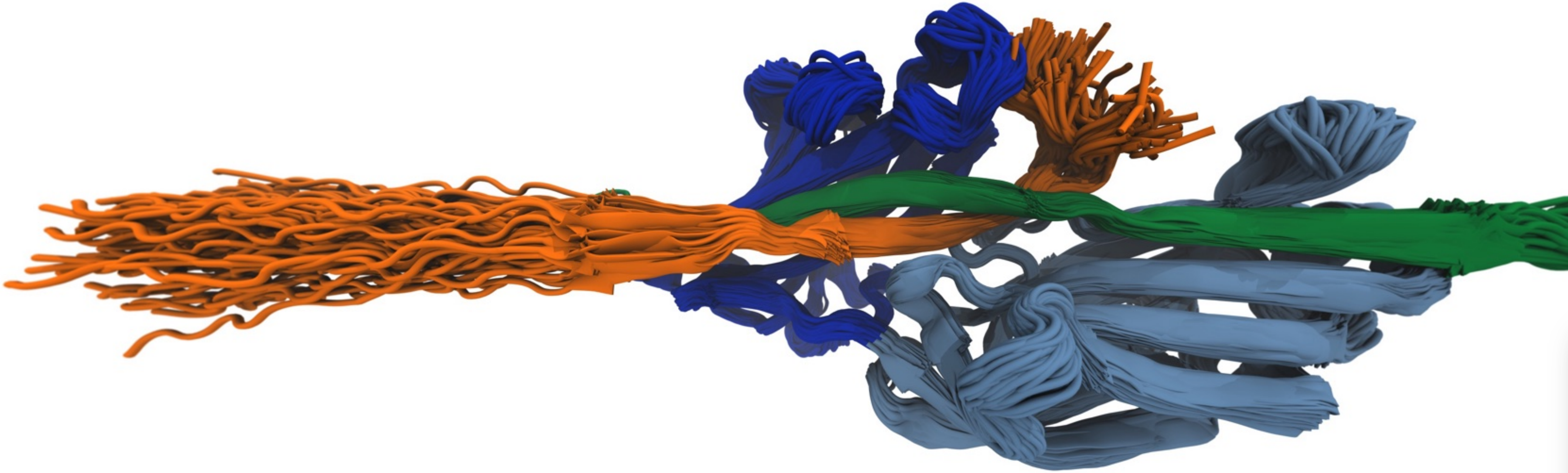
Mechanical vs Thermal Unbinding



MCR Melo, …, RC Bernardi; **Molecular origins of force-dependent protein complex stabilization during bacterial infections.** JACS, 2023

Mechanical vs Thermal Unbinding





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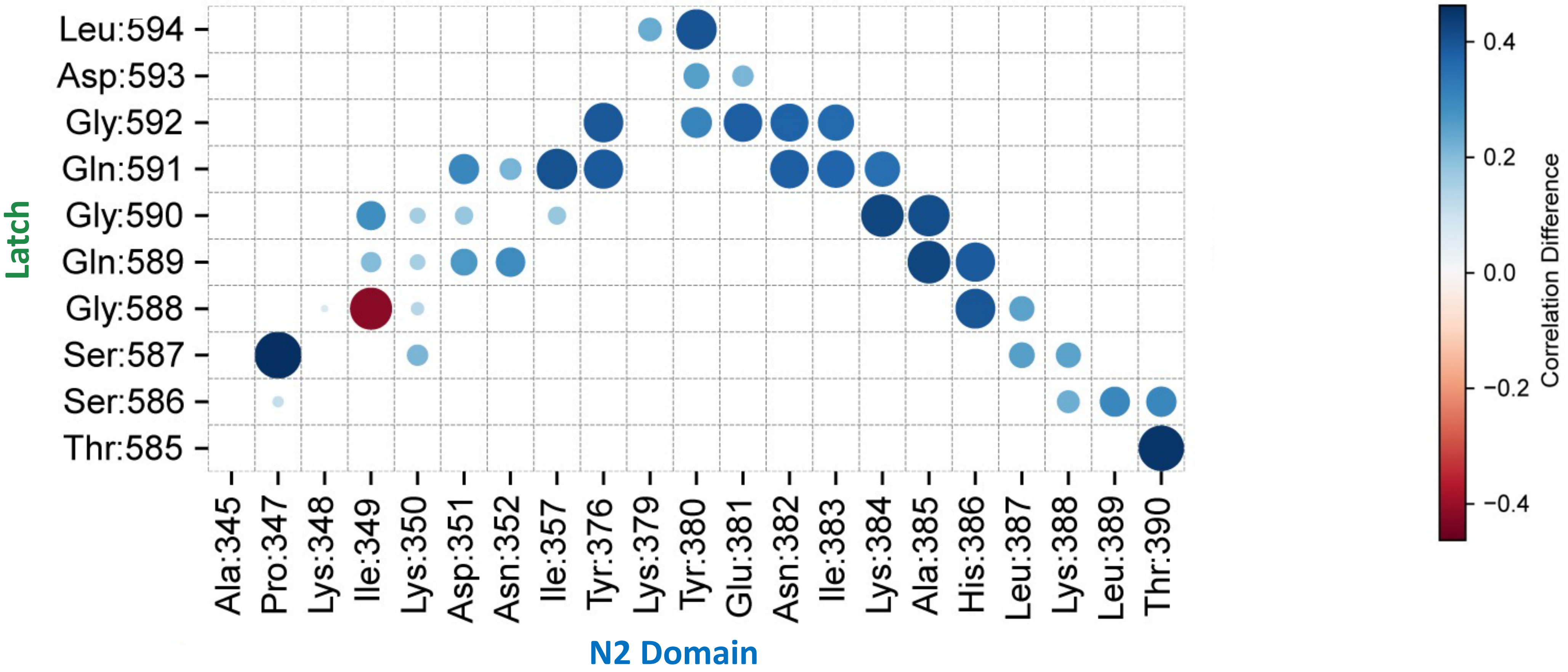
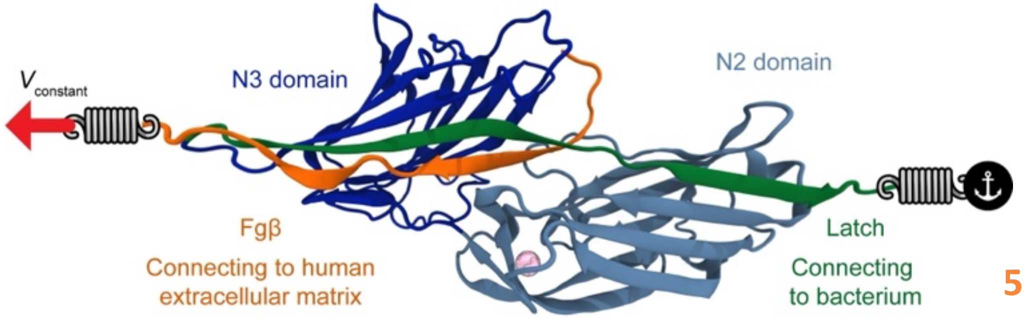
The interface between the Latch and the protein gets more rigid

Marcelo CR Melo

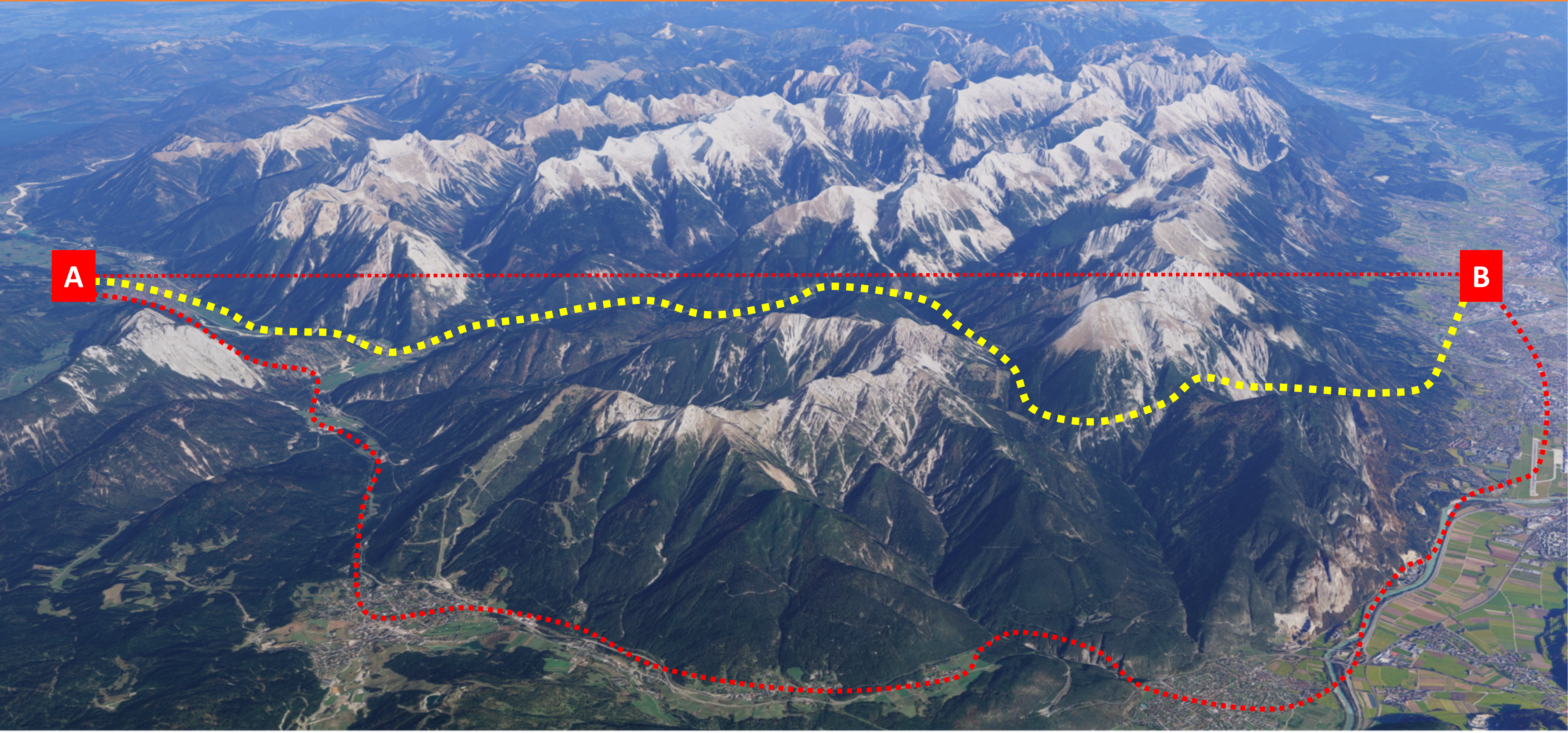


BLUE = Stronger correlation in the AFM-like pulling
RED = Stronger correlation in the MT-like force clamping



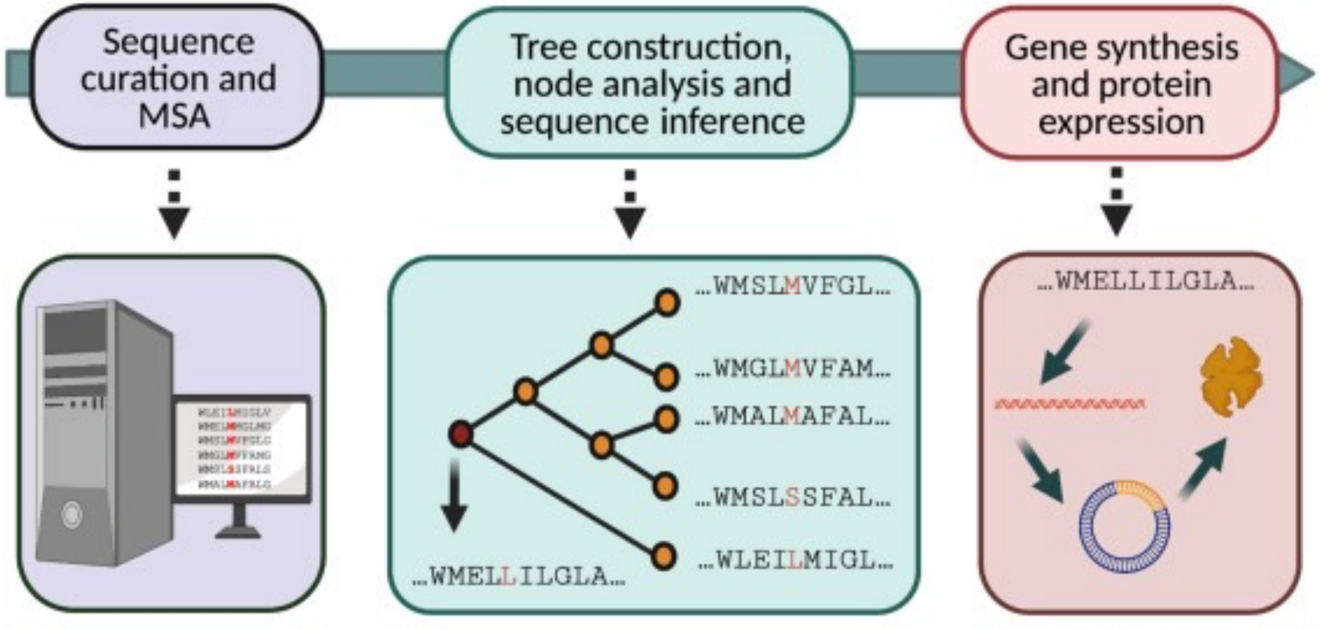
MCR Melo, …, RC Bernardi; **Molecular origins of force-dependent protein complex stabilization during bacterial infections.** JACS, 2023

Catch-Bonds: Free Energy Landscape View of the Mechanoactive Bond

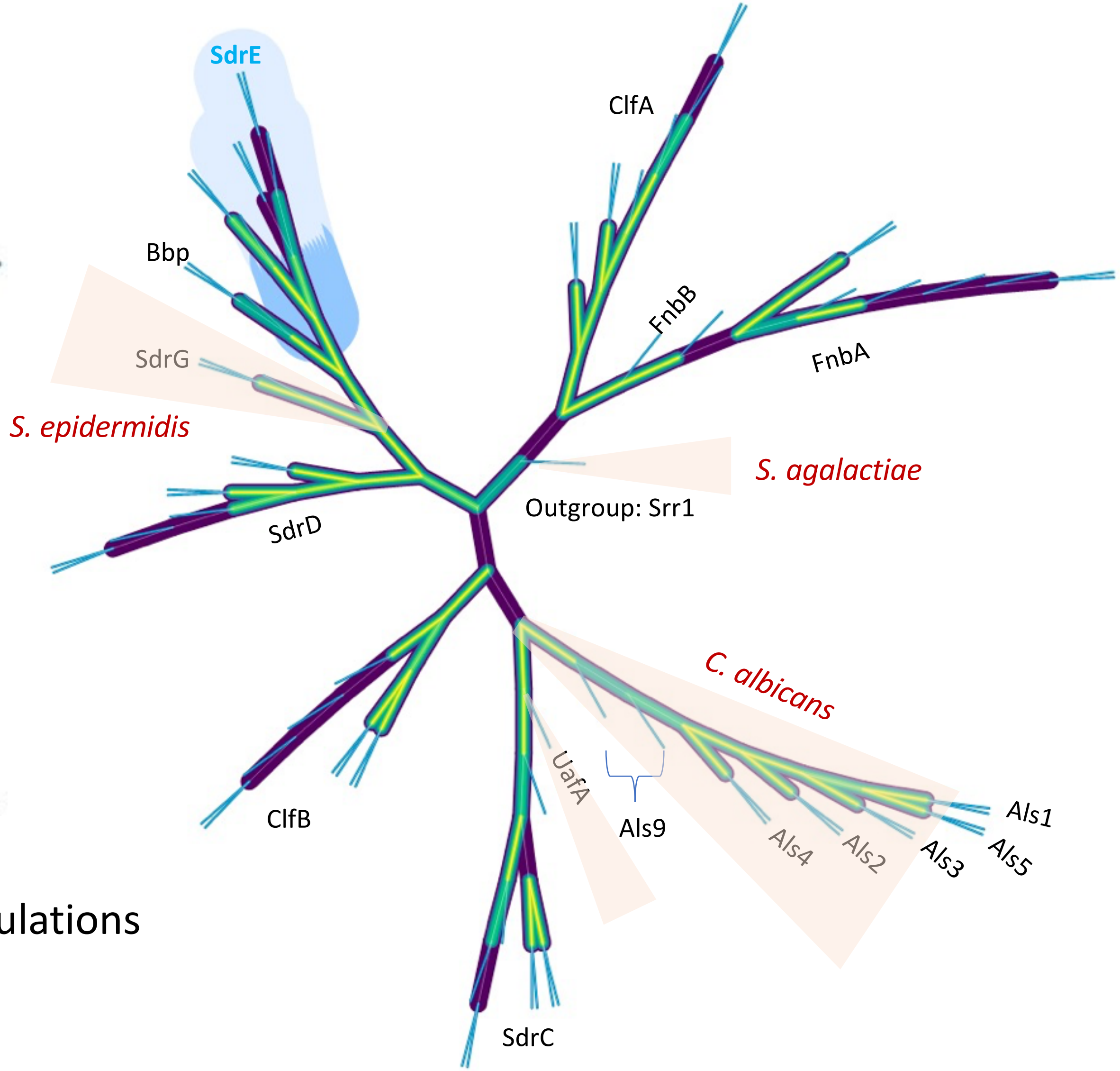


How adhesins became so force-resilient?

Ancestral sequence reconstruction



Trends in Biochemical Sciences



- Ancestral sequences to be modelled and refined by MD simulations
- SMD to investigate mechanostability
- Identify key mutations that confer this property

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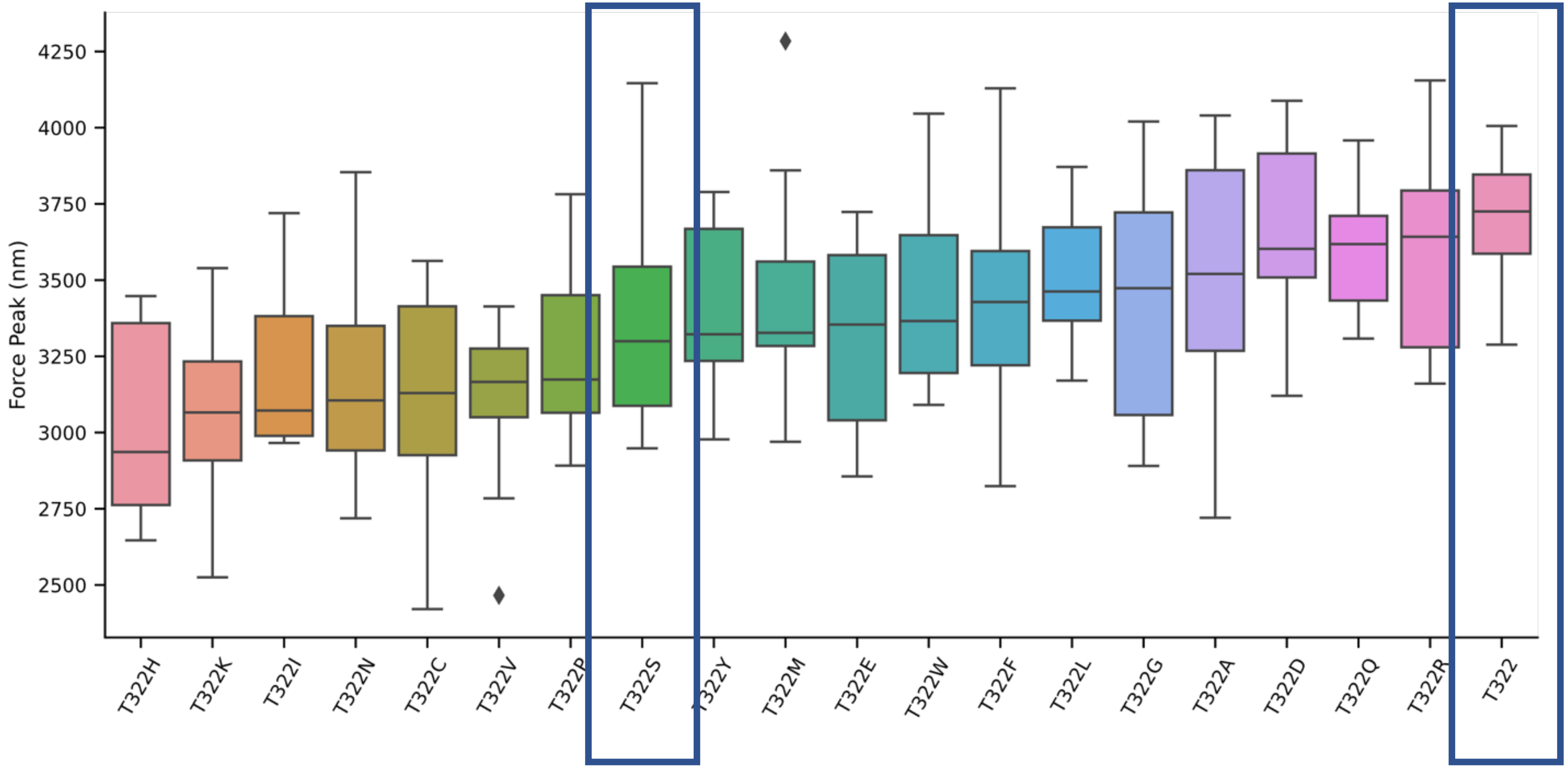
rcbernardi@auburn.edu

PSFC Gomes, **Data not published**

Ancestral sequence reconstruction

Priscila SFC Gomes

Strain R1 mutants

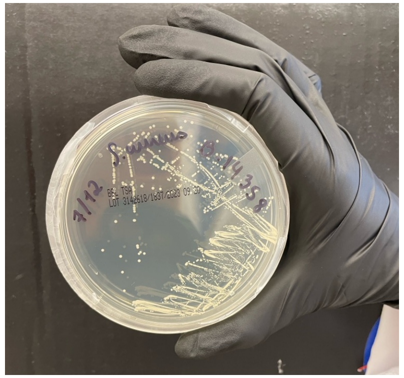


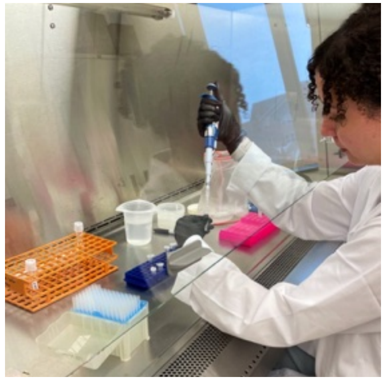
MSSA mutation

MRSA mutation

PSFC Gomes, **Data not published**

Evolution of antimicrobial resistance





Sequencing

Historical strains 19th century:

S. aureus subspecies aureus Rosenbach 1884

- DNA extraction using E.Z.N.A. Bacterial DNA kit (OMEGA Bio-Tek)

UniProt

Isolates with MSSA and MRSA phenotypes:

- Strain NCTC 8325 (early 60s)
- Strain N315 (1982)
- Strain Mu50 (1997)

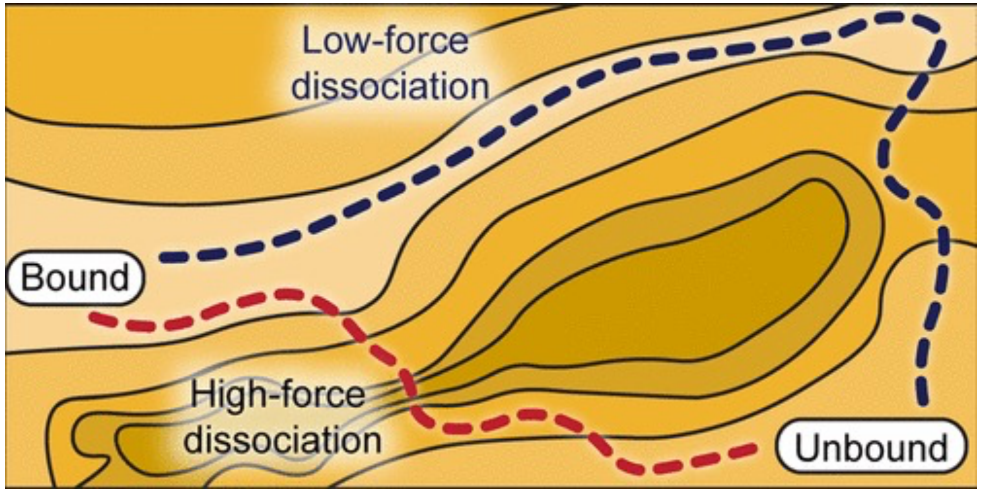
Protein sequences:

- 3D model construction
- All-atom MD simulations
- Force Resilience distribution

PSFC Gomes, **Data not published**

Take home Message

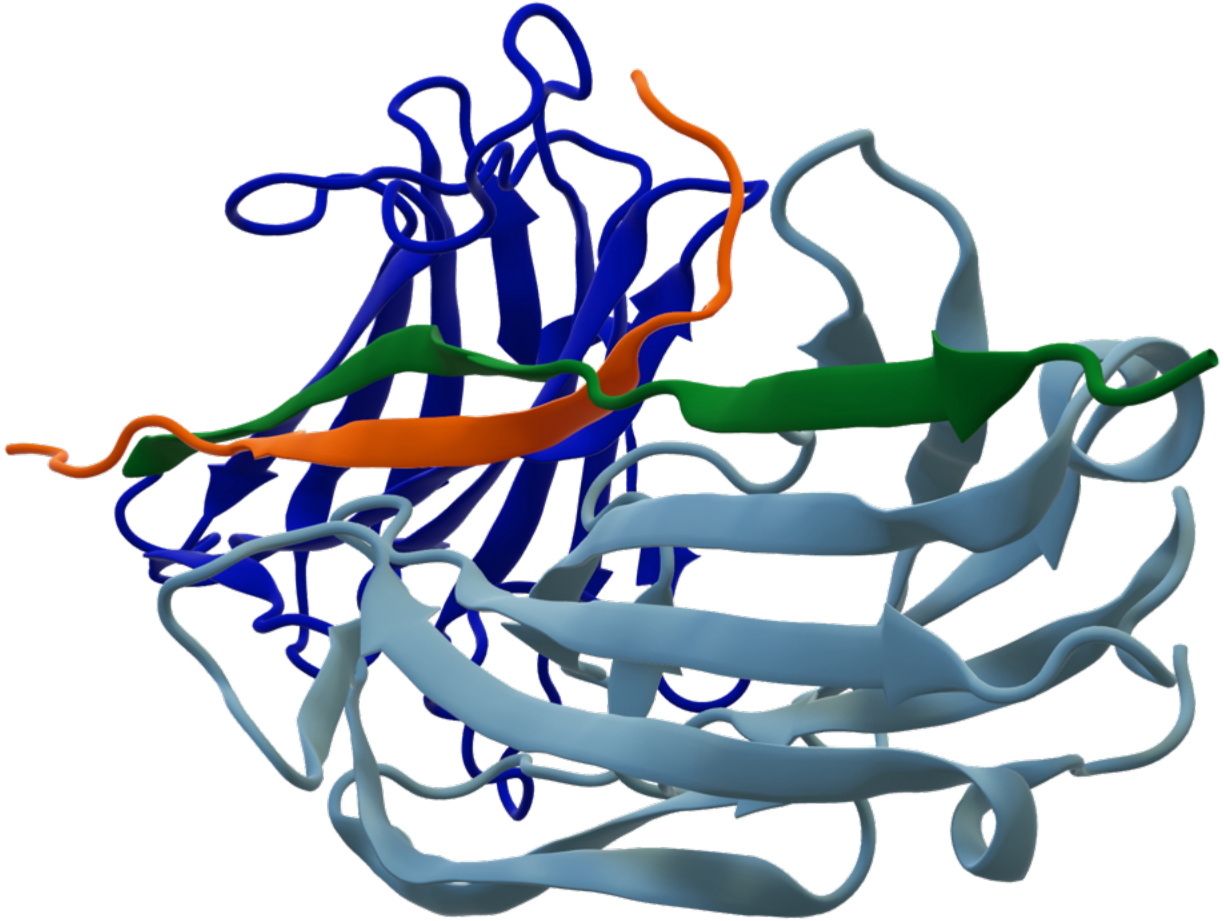
- Equilibrium binding affinity is frequently unrelated to mechanical strength;



- We can measure force resilience using Single Molecule Force Spectroscopy approaches, both *in vitro* and *in silico*;
- We can use MD trajectories to analyze how force propagation pathways “activate” catch-bonds;
- Pathogenic bacteria uses catch-bonds to cling to our extracellular matrix;
- Dynamical Network Analysis can be used to investigate unbinding pathway;
- Network nodes are a good descriptor to predict force-resilience.
- Bacteria are evolving to adhere even better to our cells.

Take home Message

May the Force be with you!



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Acknowledgments

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Prof. Klaus Schulten
Prof. Zan Luthey-Schulten

Ludwig Maximilians University of Munich, Germany
Prof. Hermann Gaub

University of Basel & ETH Zurich, Switzerland
Prof. Michael Nash

Ultrech University, The Netherlands
Prof. Jan Lipfert

Weizmann Institute, Israel
Prof. Ed Bayer

Catholic University of Louvain, Belgium
Prof. David Alsteens
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Funding

NSF

MCB-2143787
CAREER: In Silico Single-Molecule Force Spectroscopy

NIH National Institutes of Health

R24 GM-145965
Resource for Macromolecular Modeling and Visualization

AUBURN UNIVERSITY

College of Sciences and Mathematics

Thank you for your attention!