AFModulus Flex

a.k.a Group 5

A Biophysics Flex : Calculating Modulus from AFM data (Atomic Force Microscopy).

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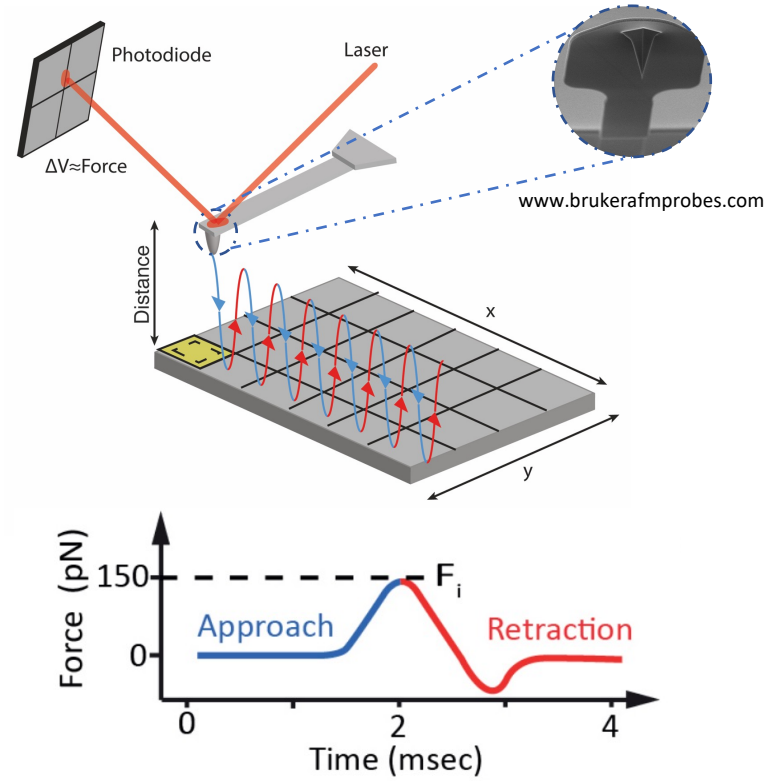
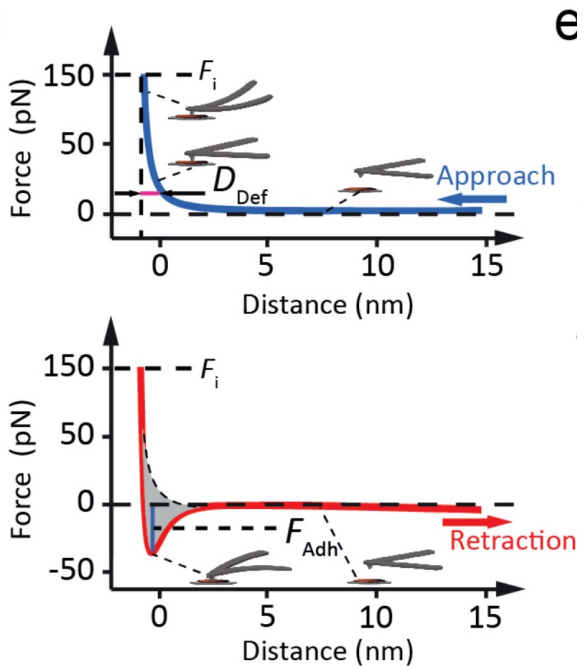
AFM in a nutshell:
How it works and what we can learn with it?

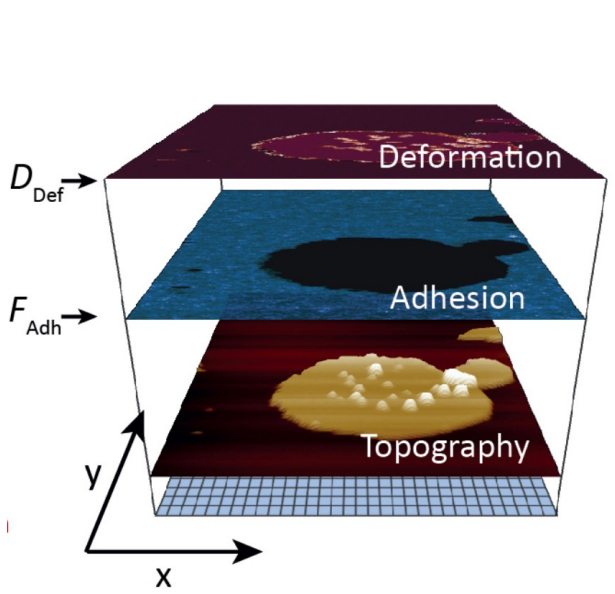
- Working principle of

Atomic force microscopy (AFM)



- Investigating the **mechanical properties** of the biological samples

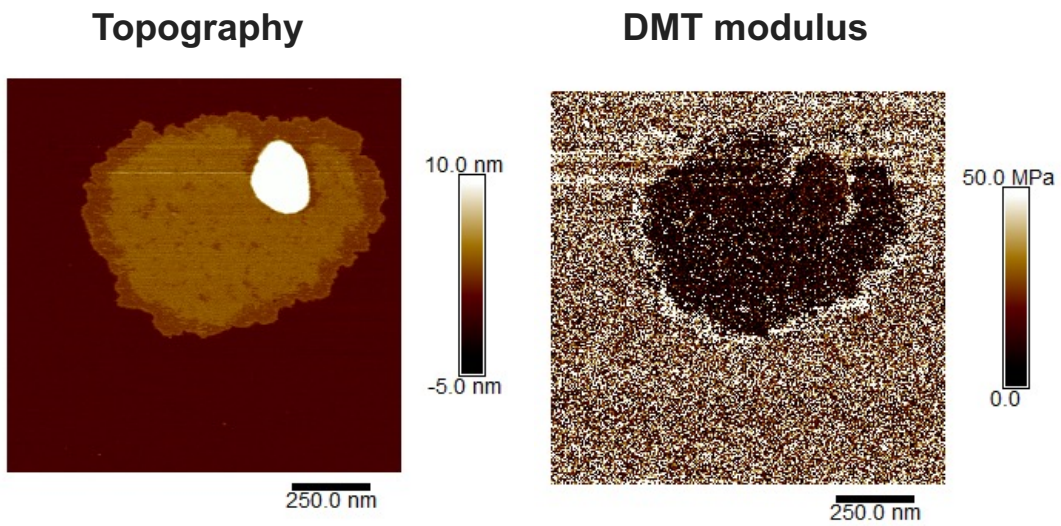




Alsteens, D. and Pfreundschuh, M., *Nature Methods*, 2015

Calculating the Young's modulus of membrane structures from AFM data

- Generating a **modulus heat map** of the imaged membrane structures

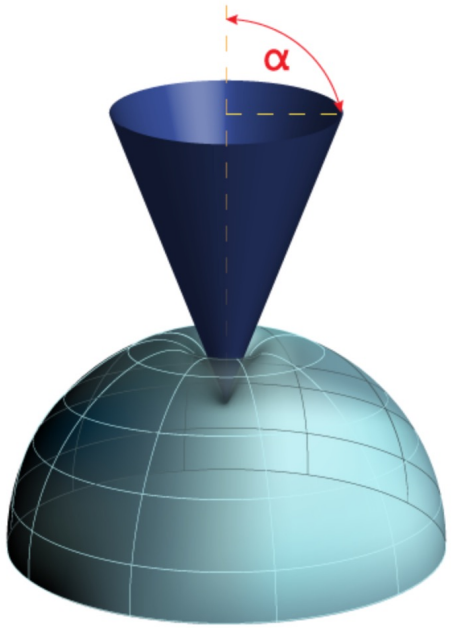


Why we can't use this modulus heat map?

- *DMT modulus is calculated with Hertzian model (spherical indenter), doesn't fit to our tip geometry*
- *The heat map is generated online, the values in extracted image are intensity values not modulus*
- *DMT modulus uses retraction curve*

- **Fitting the Sneddon model** to calculate the Young's modulus

Sneddon Model (conical indenter)



$$E = \frac{F * \pi * (1 - \nu^2)}{2 * \tan(\alpha) * \delta^2}$$

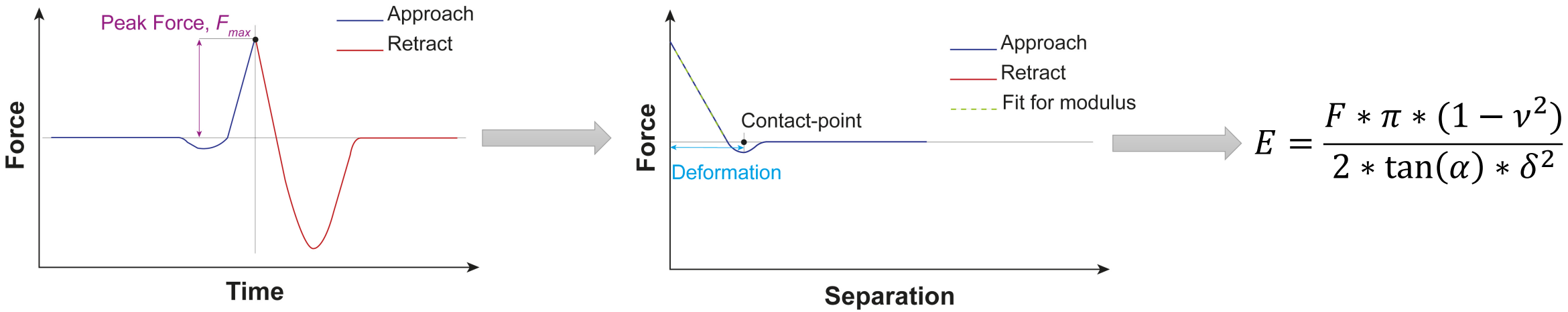
Linearized equation

- E = Young's modulus (fit parameter)
- F = Force
- δ = Indentation depth
- ✓ v = Poisson's ratio (sample dependent, typically 0.2 - 0.5)
- ✓ α = Half-angle of the indenter

Acquiring F and δ values from force-distance curves

What we need?

- Reading the $\mathbf{F_{max}}$ from F vs t curve
- Estimating the **δ** from the F vs separation of approach curve
- Plug into Sneddon model to calculate modulus values!

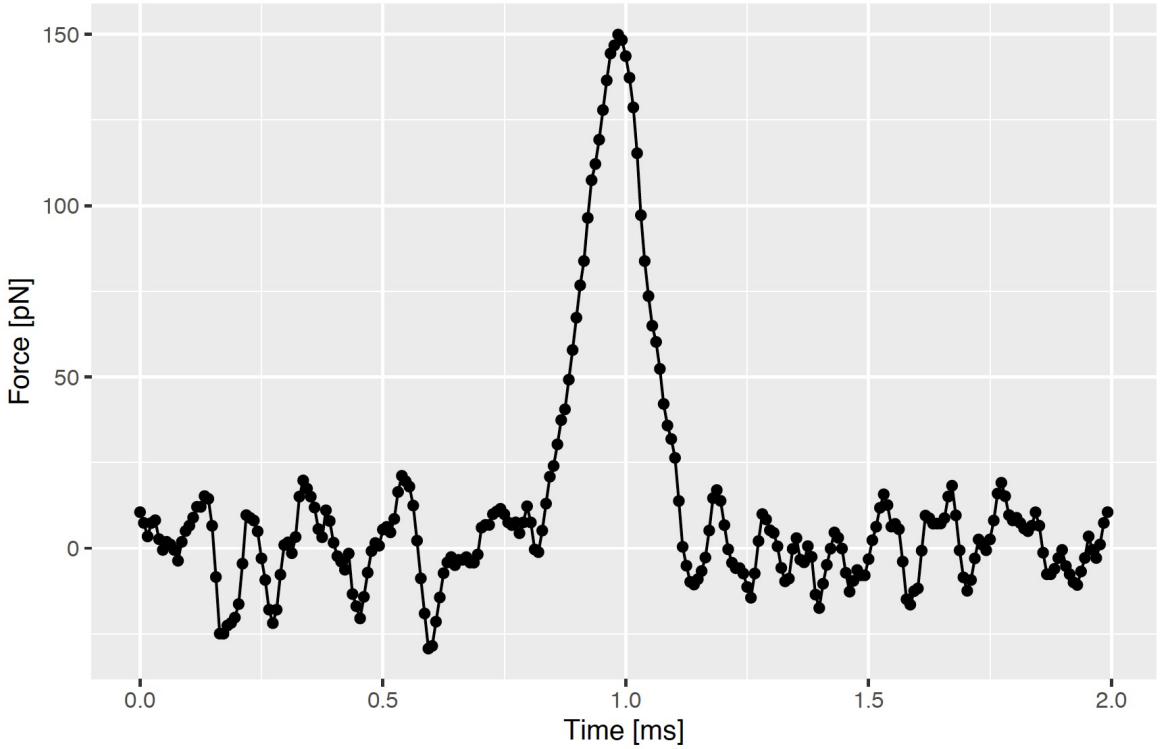


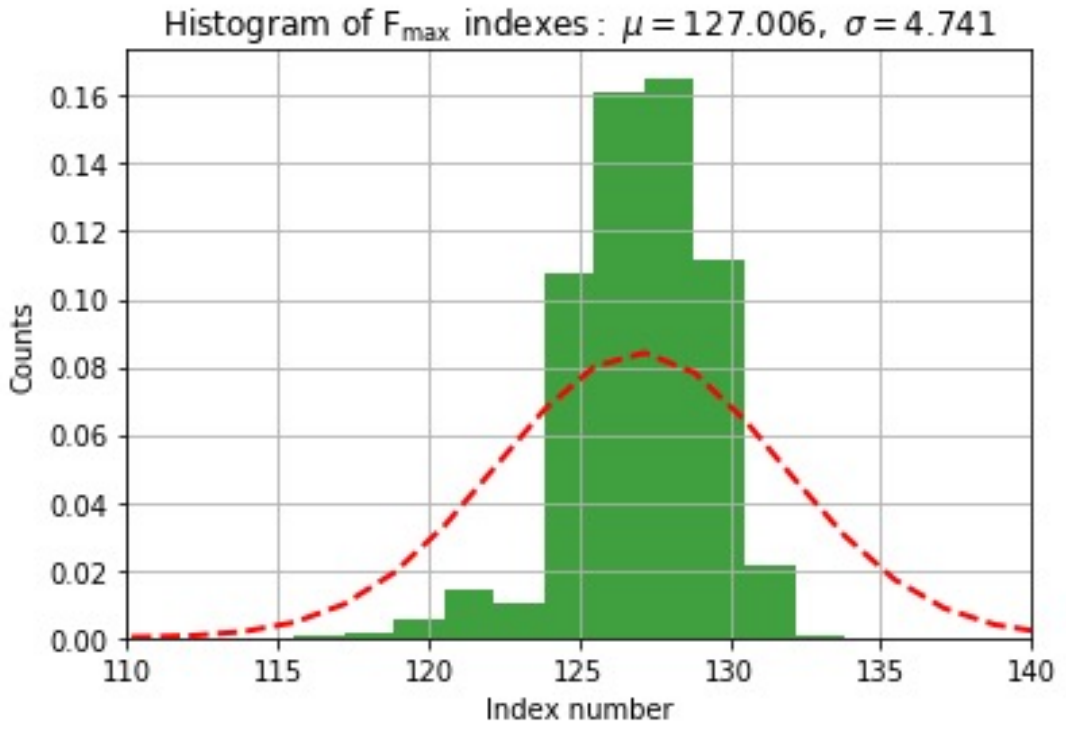
$$E = \frac{F * \pi * (1 - \nu^2)}{2 * \tan(\alpha) * \delta^2}$$

*And we need to do this for 256*256 curves…*

Acquiring F and δ values from force-distance curves

Locating the F_{max} on F vs t curve



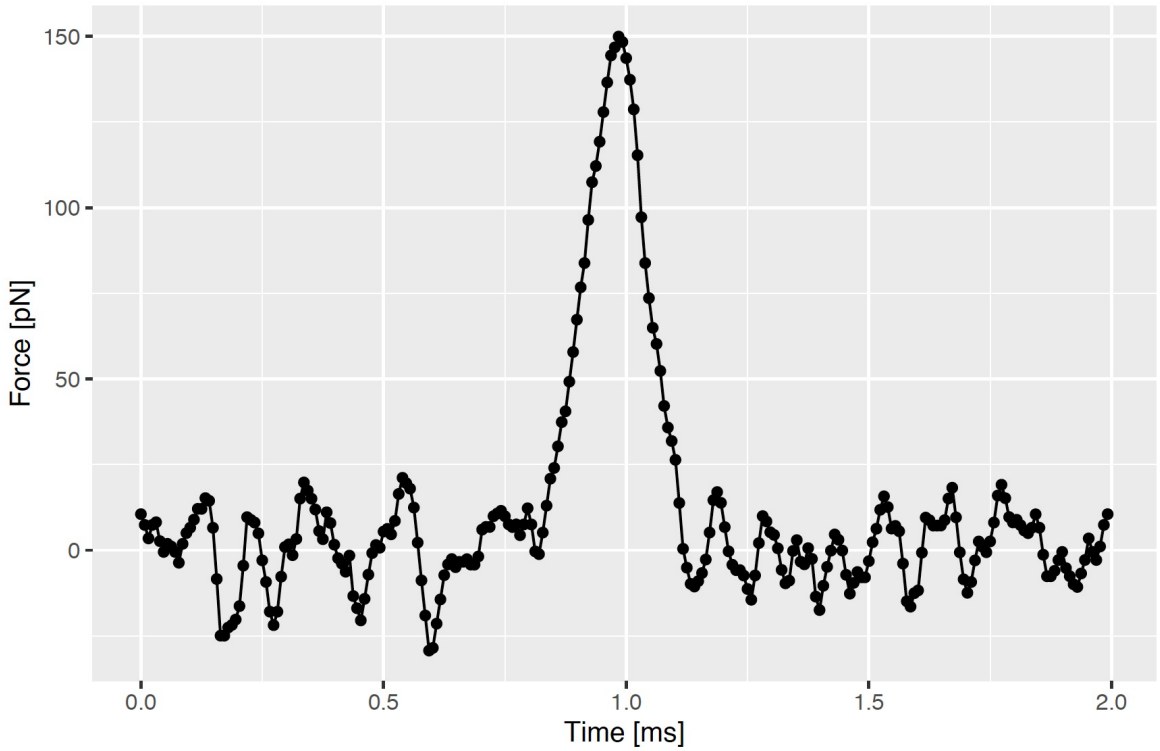


- Ideally, approach and retraction curves should be equal halves of the F vs t curve. But background removal of the cantilever motion (sinusoidal fashion) by the software doesn't work perfectly always.

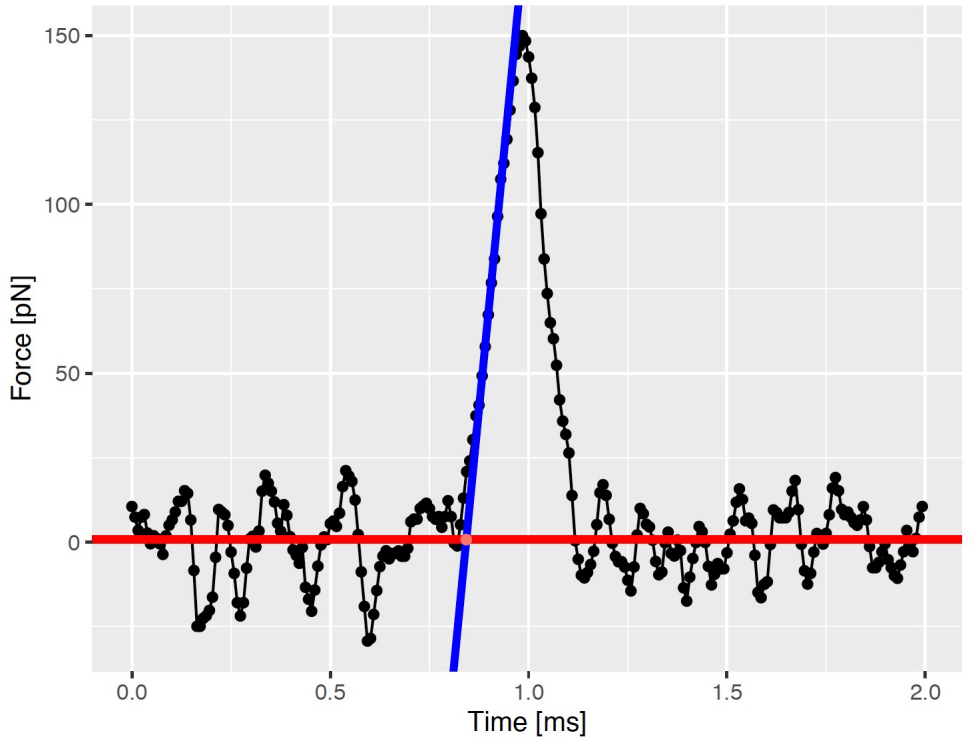
- Therefore, we can't define the index of the **F_{max}** as middle point *(Index no.128)* for all curves.

Acquiring F and δ values from force-distance curves

Locating the F_{max} on F vs t curve

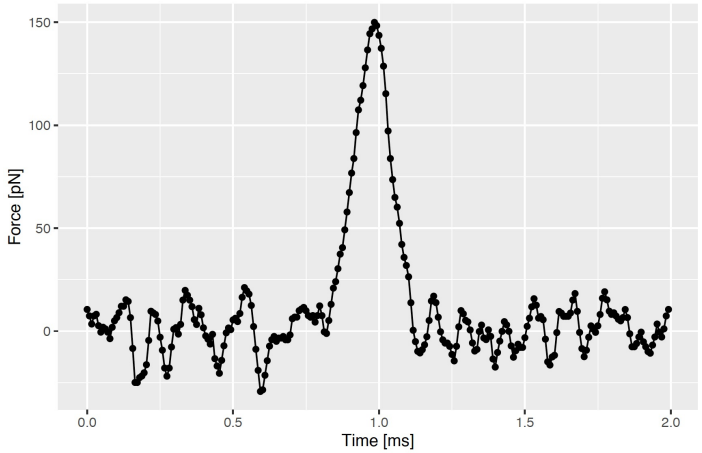


Estimating the contact point index



Acquiring F and δ values from force-distance curves

Locating the F_{max} on F vs t curve



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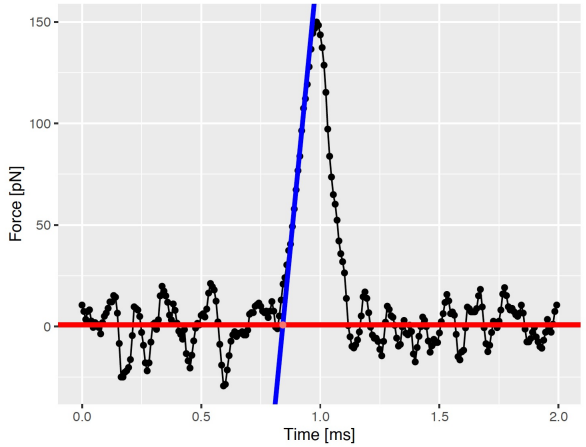

t (ms)	F (pN)	Separation (nm)	Z (nm)	Idx. No
0.7968752	9.285193	5.743021	54.3702	103
0.8046877	4.486228	5.317121	54.79222	104
0.8125002	−3.460504	4.906089	55.19931	105
0.8203128	−4.315822	4.510174	55.59122	106
0.8281253	1.920297	4.129611	55.96771	107
0.8359378	9.73452	3.764632	56.32856	108
0.8437503	17.55164	3.415457	56.67356	109
0.8515628	20.64595	3.082294	57.00248	110
0.8593753	26.89366	2.765347	57.31514	111
0.8671879	33.93204	2.464802	57.61135	112
0.8750004	37.03531	2.180847	57.89092	113
0.8828129	45.65499	1.913647	58.1537	114
0.8906254	54.27776	1.663365	58.39951	115
0.8984379	63.69127	1.430154	58.62822	116
0.9062504	73.10788	1.21415	58.83968	117
0.914063	80.16477	1.015486	59.03377	118
0.9218755	92.7382	0.8342832	59.21038	119
0.929688	103.7396	0.670647	59.36938	120
0.9375005	108.4431	0.5246776	59.5107	121
0.945313	115.5127	0.3964627	59.63424	122
0.9531255	124.1609	0.2860808	59.73993	123
0.9609381	132.8122	0.1935965	59.82771	124
0.9687506	140.6791	0.1190686	59.89752	125
0.9765631	143.036	0.06253838	59.94931	126
0.9843756	146.1836	0.02404153	59.98307	127
0.9921881	144.611	0.003601313	59.99877	128

$$\delta = Sep._{idx\_contact} - Sep._{idx\_Fmax}$$

Acquiring F and δ values from force-distance curves

Estimated F_{max}, δ

Sneddon Model (conical indenter)

$$E = \frac{F * \pi * (1 - \nu^2)}{2 * \tan(\alpha) * \delta^2}$$

E = Young's modulus (fit parameter)

- ✓ F = Force
- ✓ δ = Indentation depth
- ✓ v = Poisson's ratio
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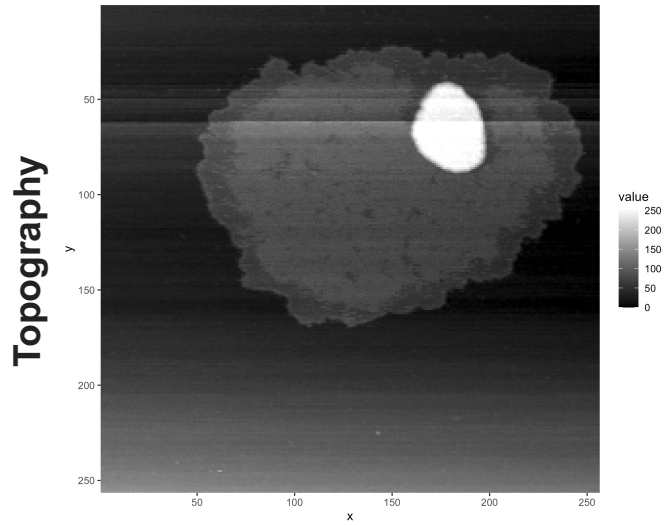
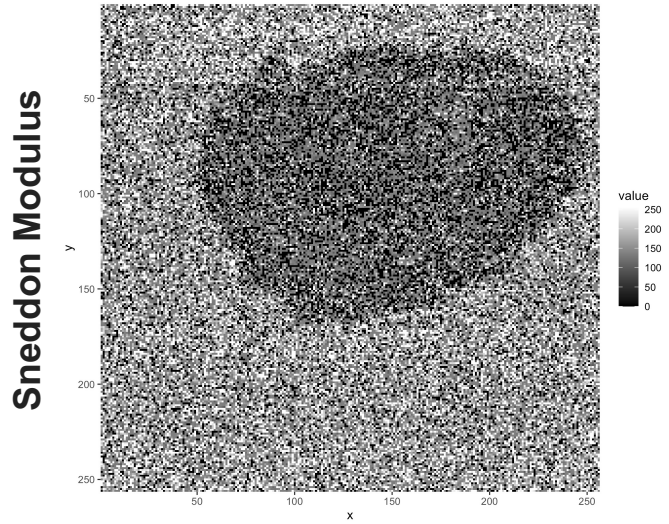
...

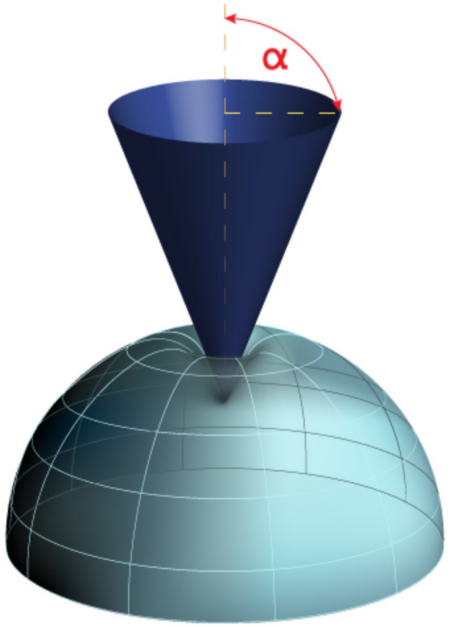
It takes some time to treat 256x256 force curves





How our estimations can effect the calculated modulus value?

Sneddon Model (conical indenter)



$$E = \frac{F * \pi * (1 - \nu^2)}{2 * \tan(\alpha) * \delta^2}$$

Linearized equation

E = Young's modulus (fit parameter)

F = Force

δ = Indentation depth

v = Poisson's ratio (sample dependent, typically 0.2 - 0.5)

α = Half-angle of the indenter

- Our approach estimates indentation depth from a conversion table
- Poisson ratio of the surface (mica) and subtance (membrane) are not the same
- Proper calibration of the cantilever is necessary for reliable force readouts

Thank you for your listening!

Check out our AFModulus Flex on GitHub!

https://github.com/audreyyeoCH/AFModulus_Flex