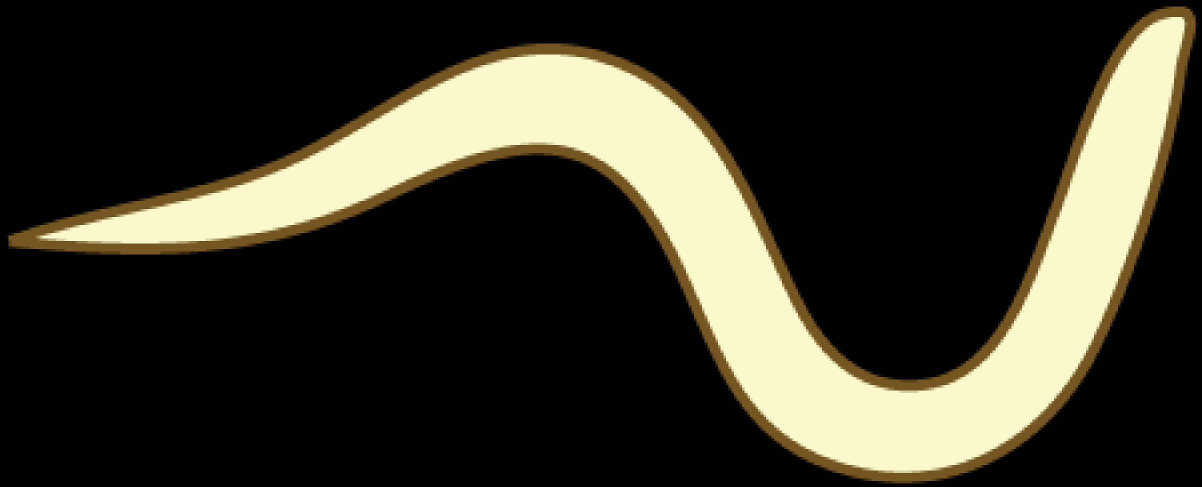


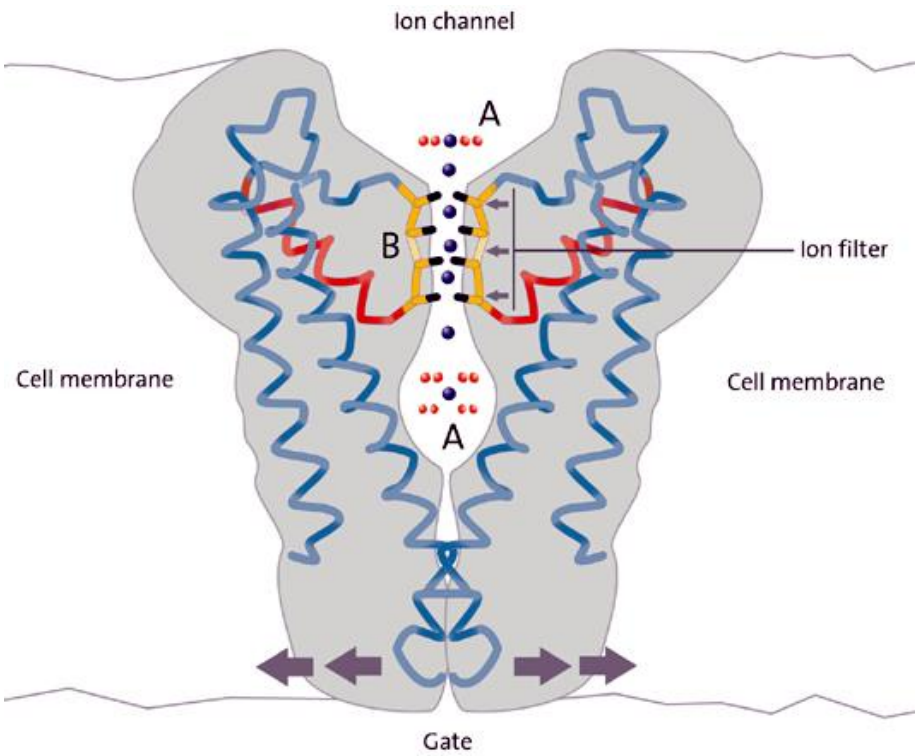
MEMS for Mechanotransduction

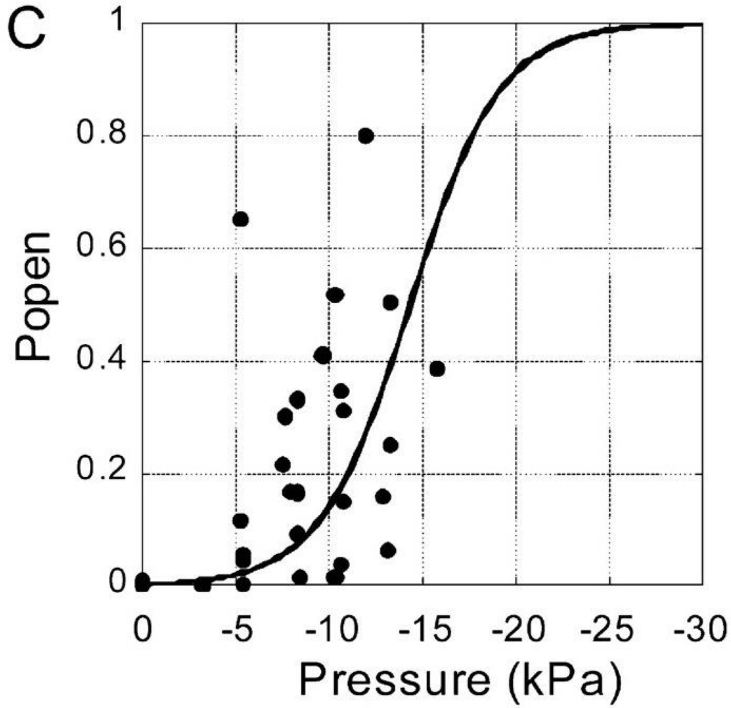
Joey Doll
Department of Mechanical Engineering
Stanford University
http://microsystems.stanford.edu

Talk Overview

- What is Mechanotransduction?
- *C. elegans* as a Model Organism
- Microscale force sensing overview
- Piezoresistive cantilevers
  - Processing techniques
  - Noise measurement results
- *C. elegans* Touch Research
  - Worm biomechanics
  - Nose touch

Cells and Ion Channels





http://fig.cox.miami.edu/~cmallery/150/memb/ion_channel_lg1.jpg

Charras et al, Biophys Journal (2004)

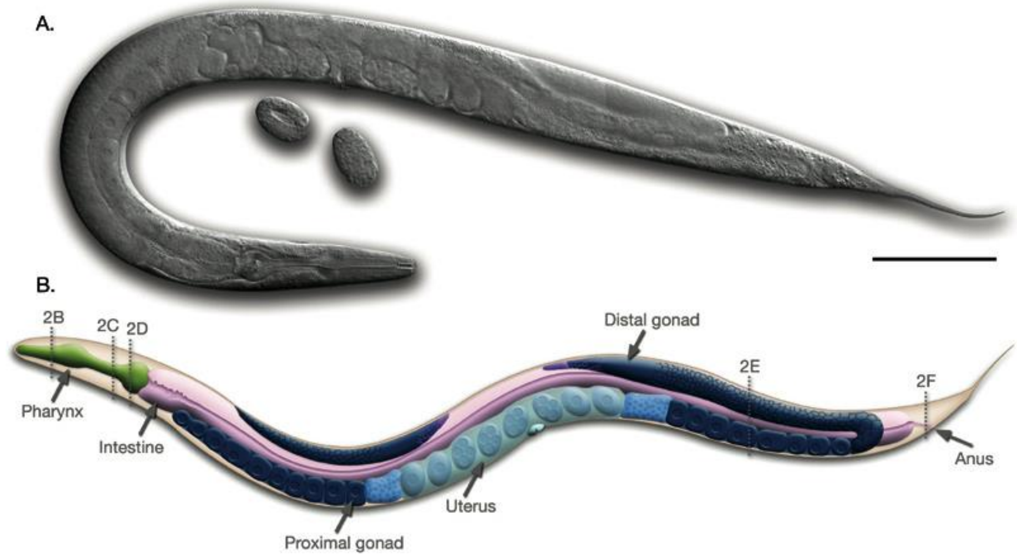
Mechanotransduction and Life

Transformation of mechanical energy into an electrochemical signal

Images from images.google.com

Caenorhabditis elegans

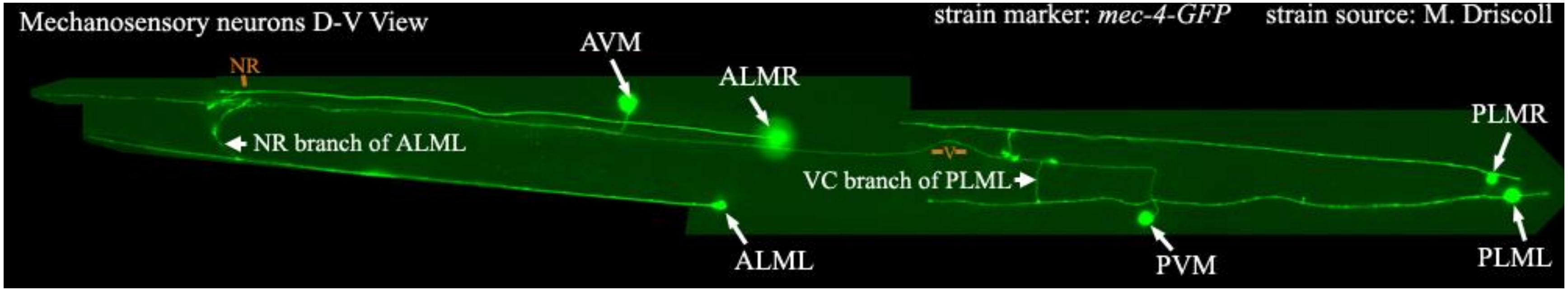
- The human brain
  - 20 – 50 billion neurons
- C. elegans
  - 1000 somatic cells
  - 302 neurons
  - 50 µm x 1 mm in size



http://www.wormbook.org

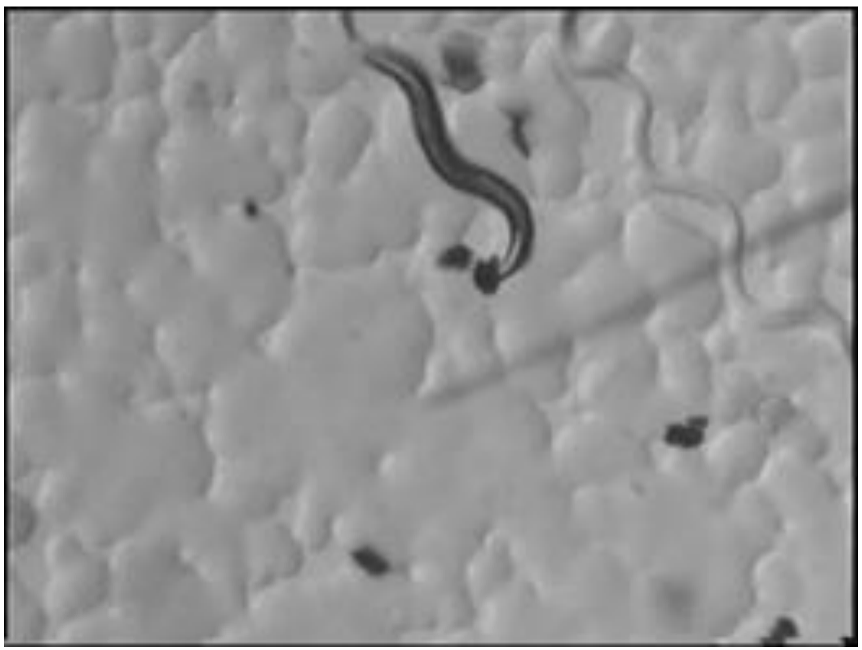
Caenorhabditis elegans

- Cell lineage and gene sequence
- 6 body wall touch receptor neurons (TRNs) that respond to light touch
- 2 nose touch neurons (ASH)
- Touch leads to change in direction, speed
  - Locomotion circuit

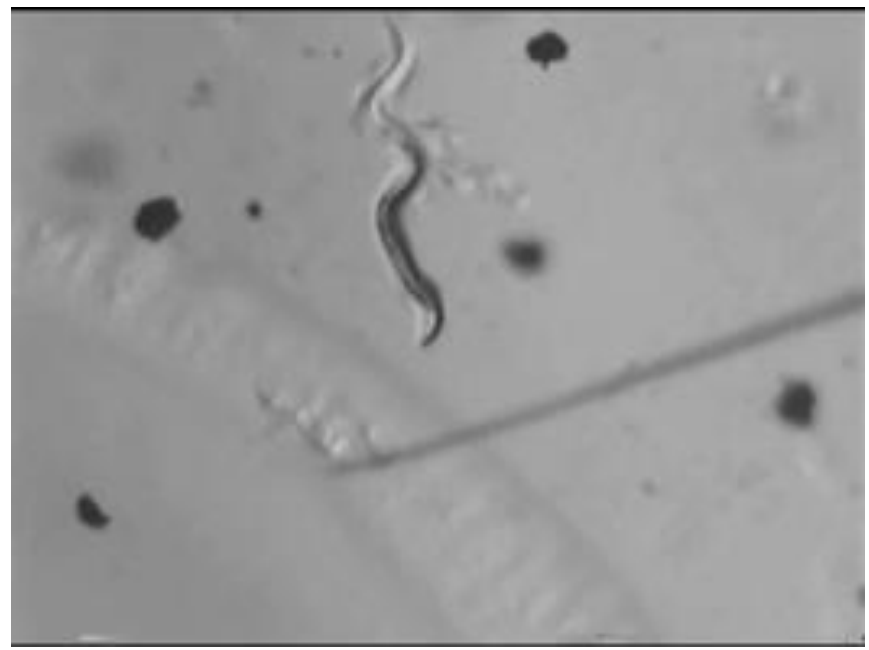


http://www.wormatlas.org/cellid/bodymechsen.htm

The Sense of Touch



Wild type

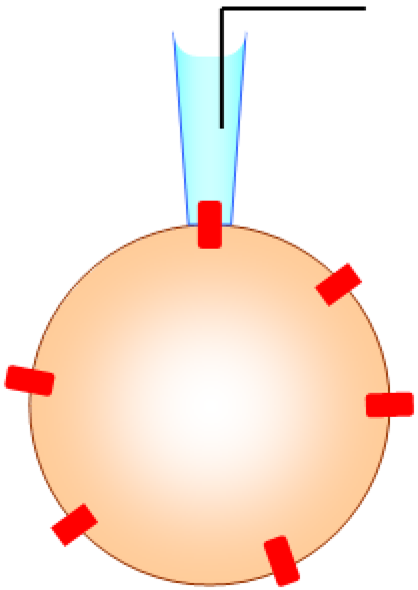


osm9 null mutant

Movies courtesy of Shana Geffeney

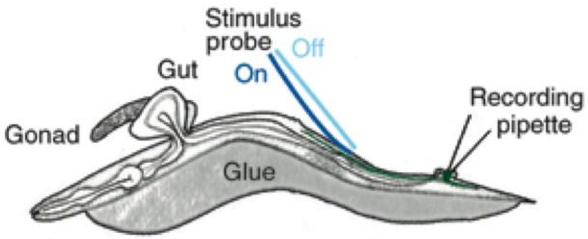
Electrophysiology Measurements

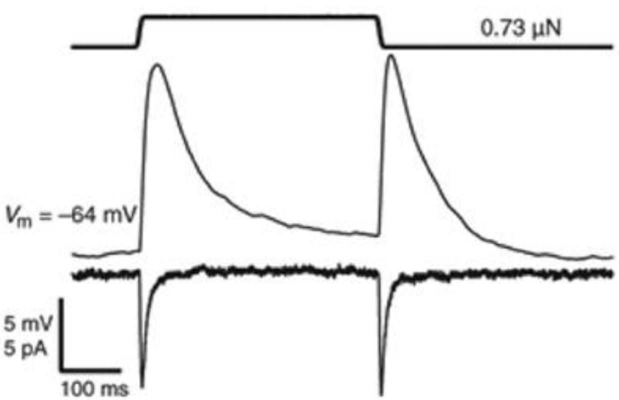
- Cells are normally at ion flux steady state
  - Δflux = Δvoltage
- Patch clamp = technique to measure the flux of charged ions through a cell membrane and change in voltage
  - Max Planck Institute, 1973
  - Other techniques too: e.g. calcium imaging via FRET

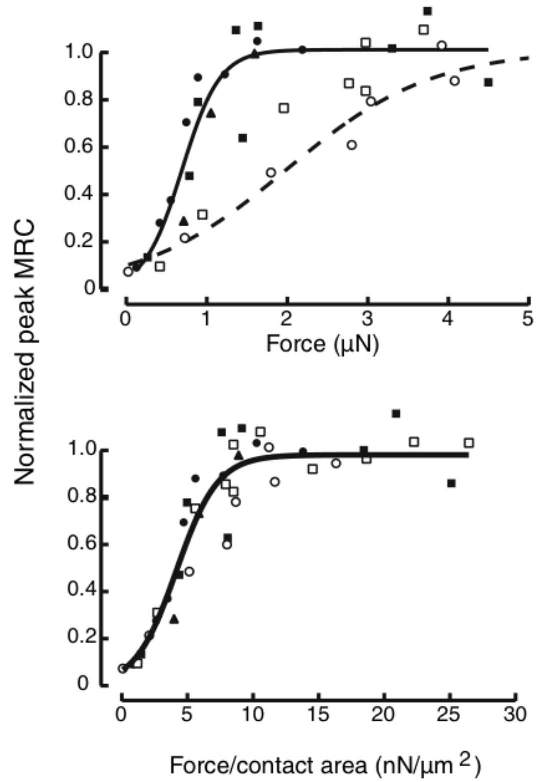


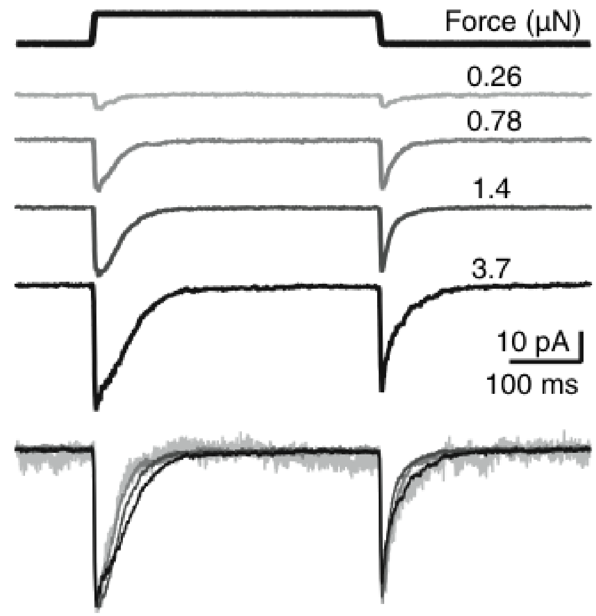
http://en.wikipedia.org/wiki/Patch_clamp

Touch Receptor Neuron Physiology









(open loop force control)

The MEC-4 DEG/ENaC channel of Caenorhabditis elegans touch receptor neurons transduces mechanical signals, O'Hagan et al, Nature Neuroscience (2005)

Talk Overview

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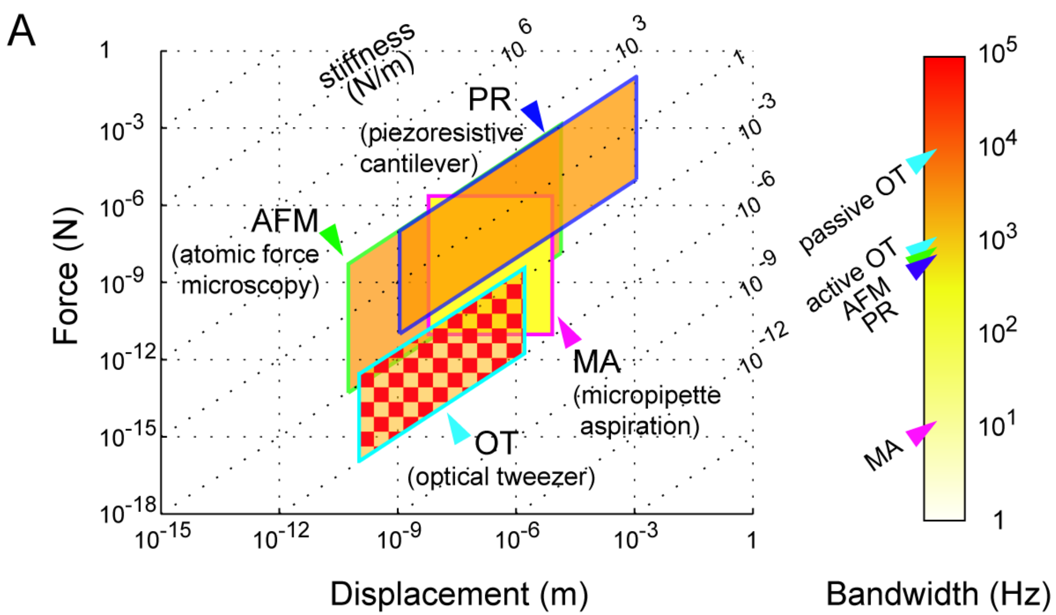
Force Sensing Techniques



Park et al, PNAS 2007

Piezoresistance

- Strain gauge: change in resistance with change in length
- Define gauge factor (GF)

$$GF = \frac{dR/R}{dL/L} = \frac{dR/R}{\varepsilon} = \frac{d\rho/\rho}{dL/L} + 1 + 2\nu$$

- Metal strain gauges: resistivity doesn't change so GF ~= 2
- For semiconductors…

$$\rho = \frac{1}{q(n\mu_n + p\mu_p)}$$

- Deformation of the crystal lattice under applied stress changes its energy band structure, altering carrier mobility and therefore resistivity

Piezoresistivity in Silicon

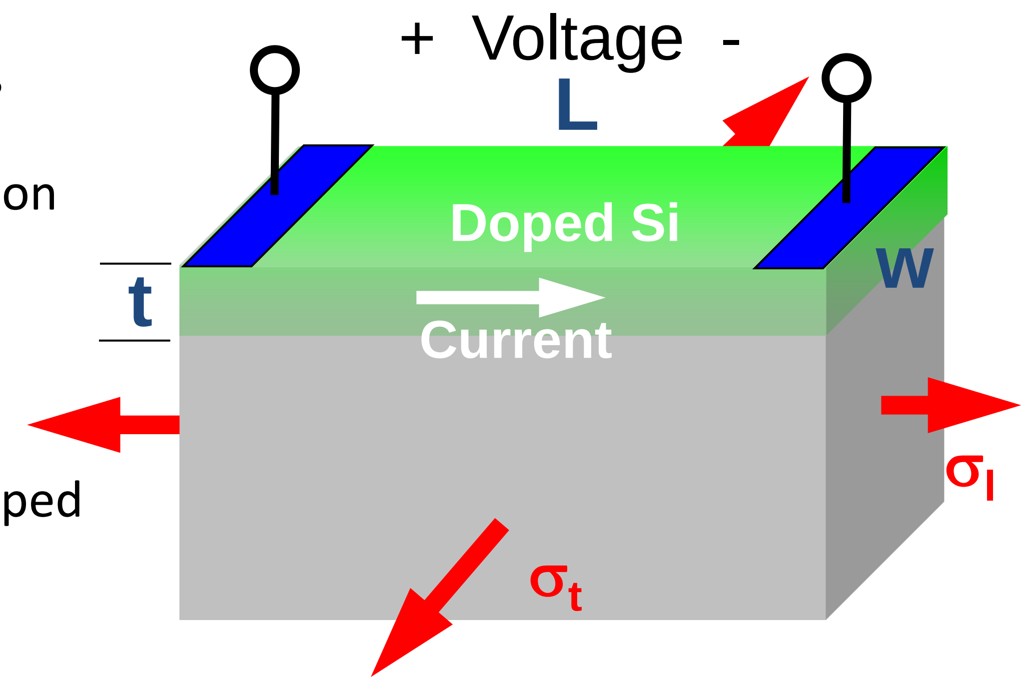
- Resistor elements fabricated by selectively introducing impurities (doping)
- Resistance of a doped silicon region is given by

$$R = \rho \frac{L}{wt}$$

- In piezoresistive materials like doped Si, ρ is stress-dependent

$$\frac{\Delta\rho}{\rho} = \pi_l \sigma_l + \pi_t \sigma_t$$

- Where π_l is the **longitudinal** piezoresistive coefficient
- And π_t is the **transverse** piezoresistive coefficient



The simplest case: piezoresistors patterned so current flows in the direction of the uniaxial stress - only longitudinal components need to be considered.

Silicon Strain Gauges?

- Strain dependencies may be derived for a silicon resistor

$$GF = \frac{dR/R}{\varepsilon} = \frac{E}{\sigma}\frac{d\rho}{\rho} + 1 + 2\nu$$

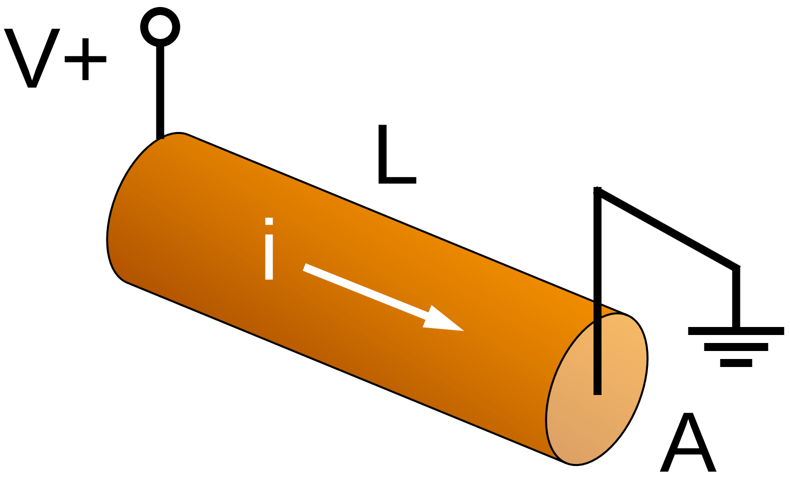
- But since $d\rho/\rho = \pi_l \sigma_l$

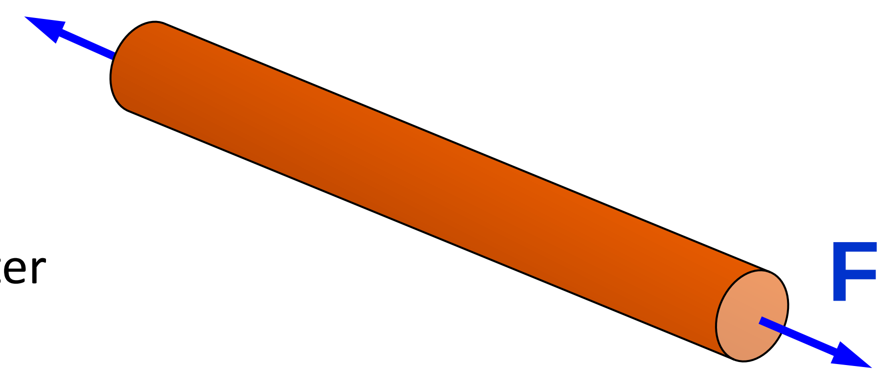
$$GF = E\pi_l + 1 + 2\nu$$

- With π_l of $100 \times 10^{-11} m^2/N$ and $E_{Si} = 190 \times 10^9 Pa$

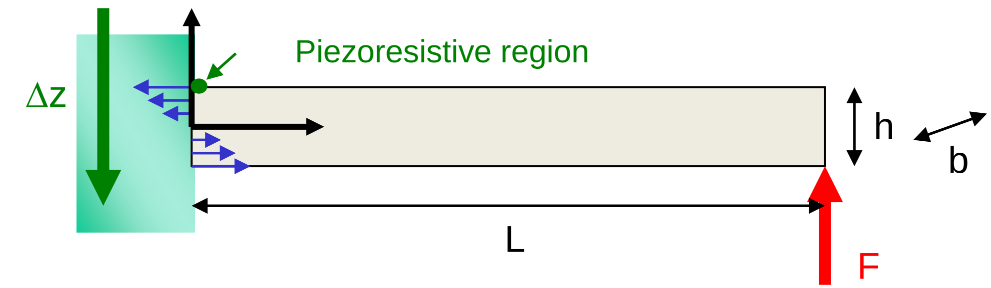
$$GF = E\pi_l + 1 + 2\nu \approx 190$$

- So the gauge factor of a Si gauge is nearly two orders of magnitude better than a metal wire!



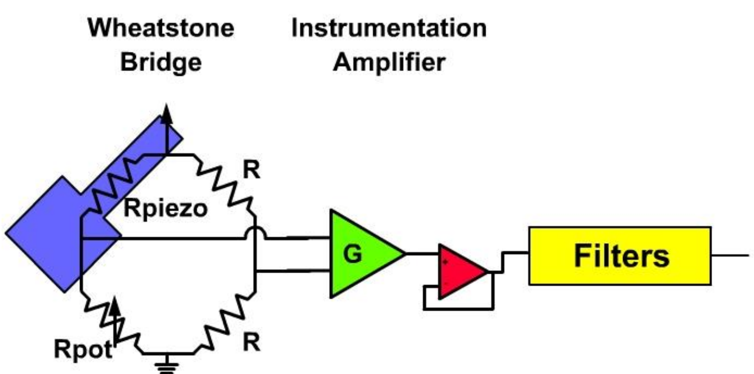


Piezoresistive Force Sensing

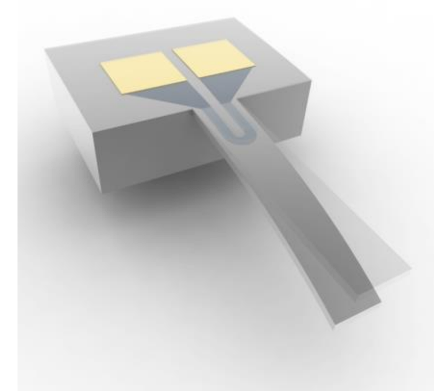


$$F = k\Delta z \qquad (k = \frac{3EI}{L^3} = \frac{Ebh^3}{4L^3})$$

$$V_{out} \cong G\frac{V_B \Delta R}{4R} \cong G\frac{3V_B \pi_L L}{2bh^2}F = GSF \qquad (S = \frac{3V_B \pi_L L}{2bh^2})$$

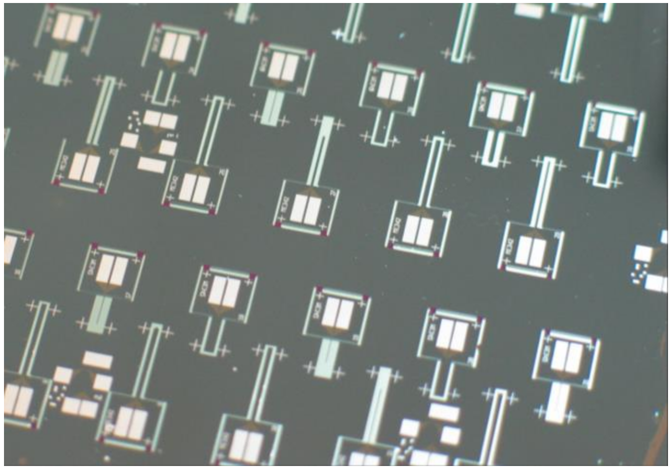


$$\frac{\Delta R}{R} \cong \pi_L \sigma \cong \frac{6\pi_L L}{bh^2}F$$



Piezoresistive Force Sensing

- Orientation is critical
- Electronic force sensing (no optics)
- Piezoresistivity varies with doping, temperature, light
- Design should maximize stress
- Small setup area + integration with other systems
- Fabrication
  - Ion implantation (thick cantilevers)
  - Epitaxy (thin cantilevers)



Example

Width: 80~400um

Length: 2000~6000um

Thickness: 15um

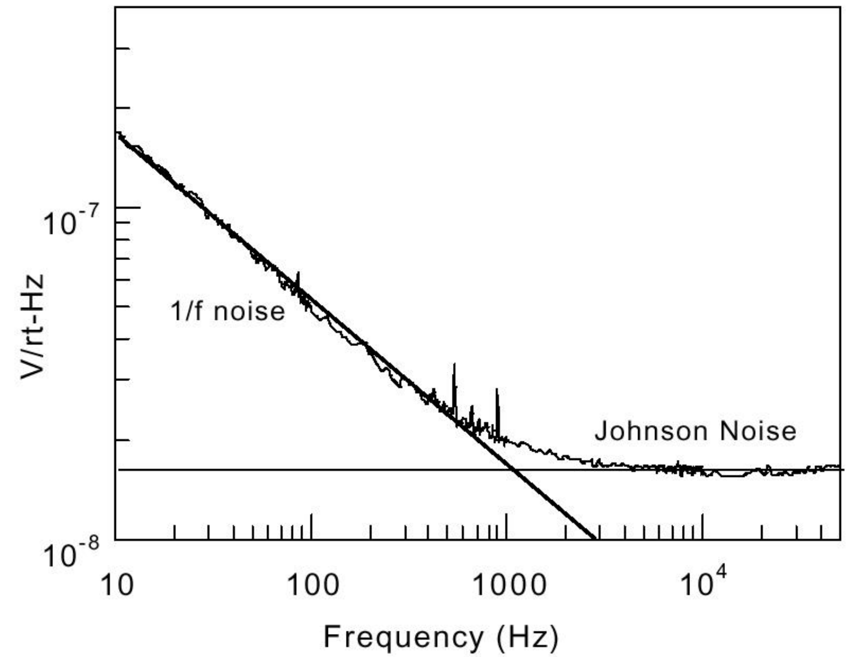
k: 0.1~2N/m

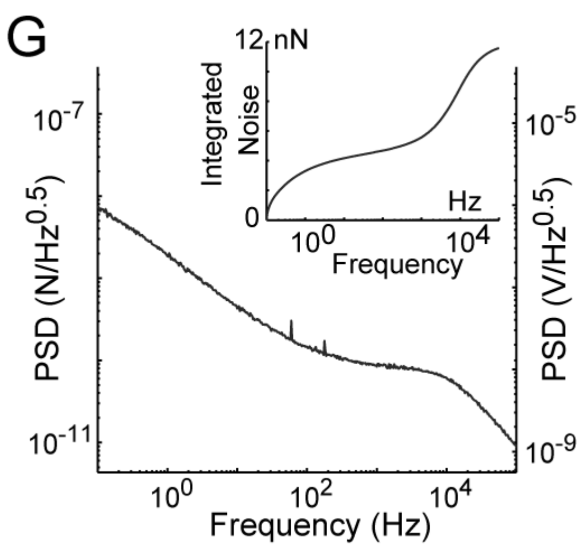
ω: 0.6~5.3 kHz

S: 3000-21000 uN/V

Force Resolution

- Sensitivity and noise trade off with process, geometry
  - Johnson noise
  - 1/f noise
  - Amplifier and background





$$\sigma_{\min} = \frac{\sqrt{4kRT\left(f_{\max} - f_{\min}\right) + \frac{\alpha V_b^{2}}{fN} \ln\left(\frac{f_{\max}}{f_{\min}}\right) + S_b}}{\frac{V_b \overline{\pi}}{2}}$$

Piezoresistive coefficient scaling

Tradeoff between sensitivity (π) and dopant concentration (R, N)

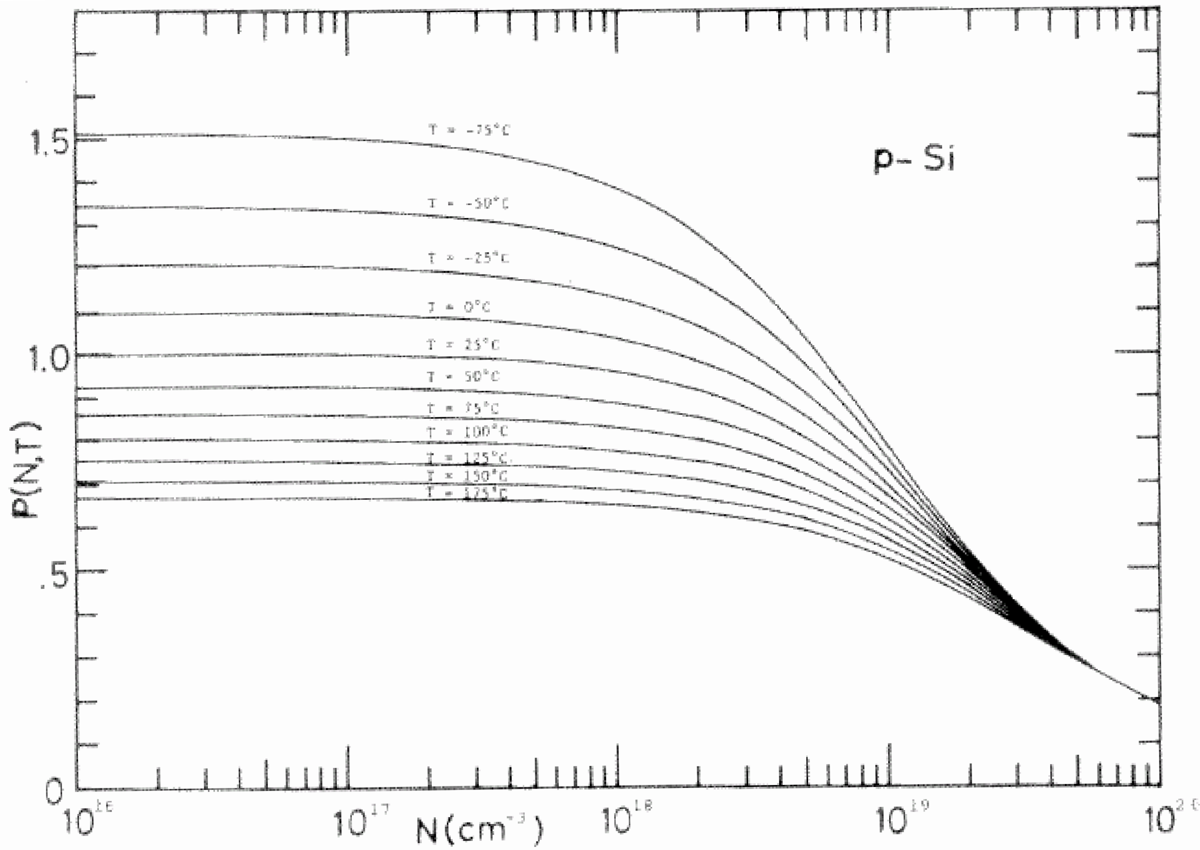
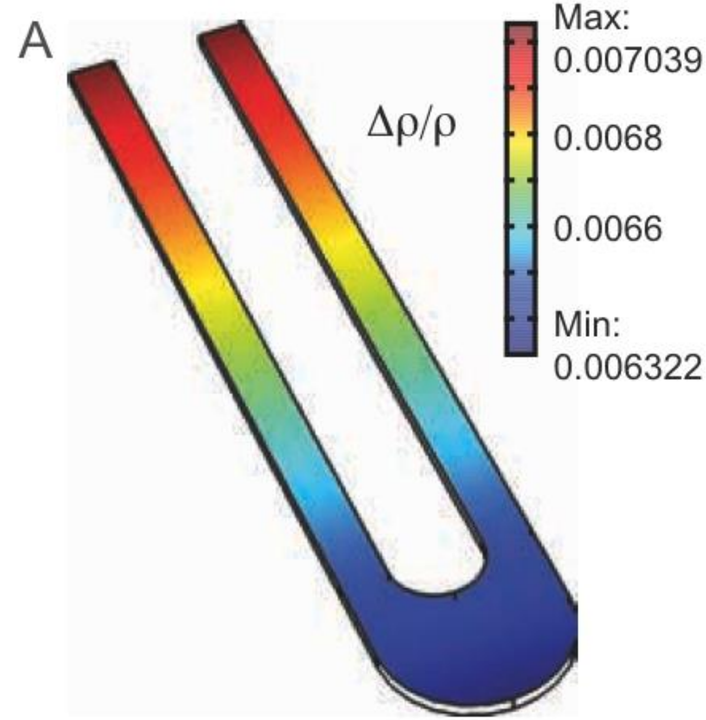


Fig. 9. Piezoresistance factor $P(N, T)$ as a function of impurity concentration and temperature for p-Si.

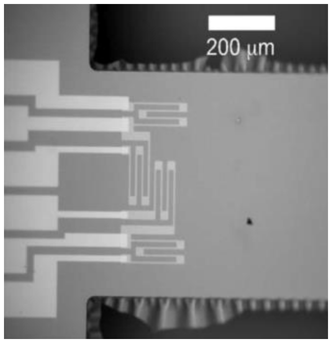
Piezoresistor Design

- Choices
  - Cantilever dimensions
  - Piezoresistor dimensions
  - Dopant concentration
  - Bias voltage
- Given constraints
  - Frequency range
  - Measurement bandwidth

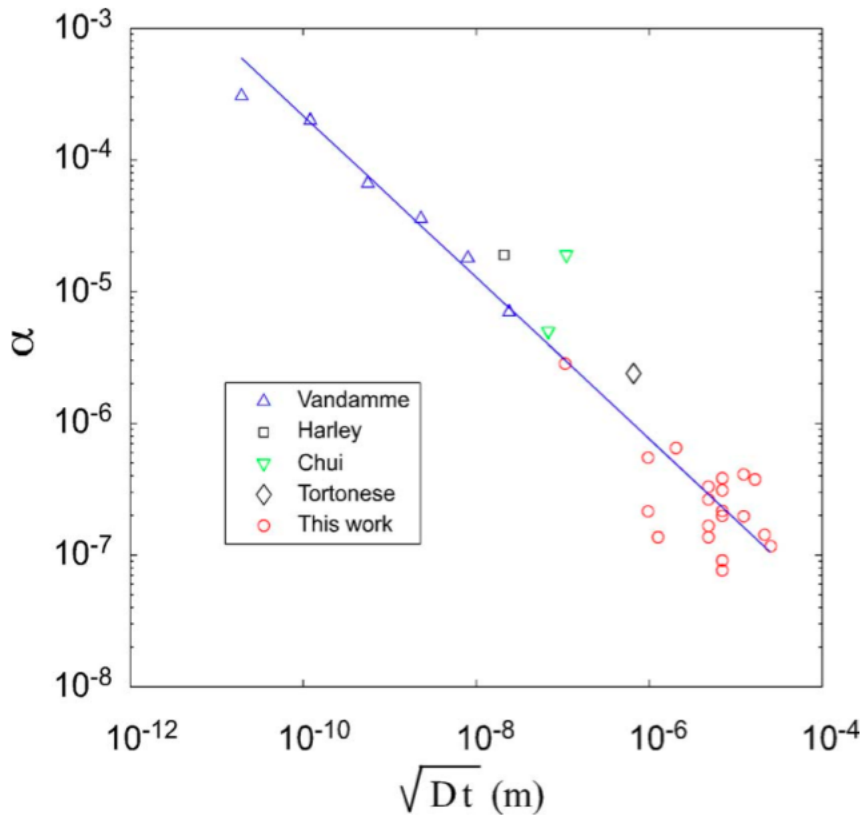


Park et al, Hilton Head 2008

Example: Low frequency design



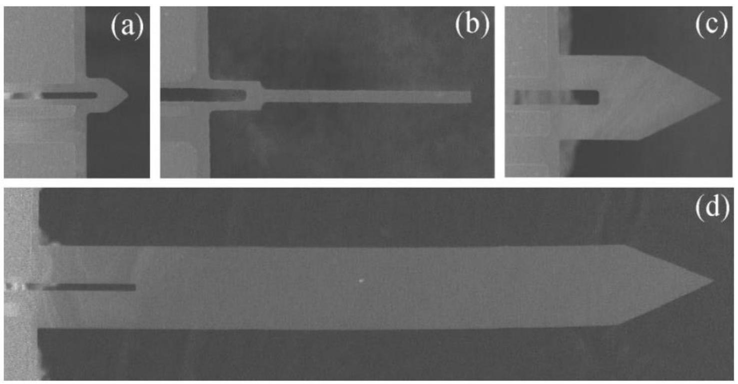
	This work	
	Full active bridge	1/4-active bridge[a]
Boron dose (cm^{-2})	5×10^{16}	1×10^{14}
Peak concentration (cm^{-3})	2.7×10^{19}	6.2×10^{17}
Anneal temperature (°C)	1100	1000
Anneal time (min)	50	52
Spring constant (N/m)	2.1	17
Sensitivity (V/N)	330	179
ω_n (kHz)	1.7	3.7
Resistance (kΩ)	1.8	16.8
Johnson noise (nV/$\sqrt{\text{Hz}}$)	5	16
Corner frequency (Hz)	0.6	20
$1/f$ noise at:		
10 Hz (nV/$\sqrt{\text{Hz}}$)	5[h]	22
10 Hz (nV/V $\sqrt{\text{Hz}}$)	0.4[h]	6
1 Hz (nV/V $\sqrt{\text{Hz}}$)	1.2[h]	20
0.1 Hz (nV/V $\sqrt{\text{Hz}}$)	3.7	NA



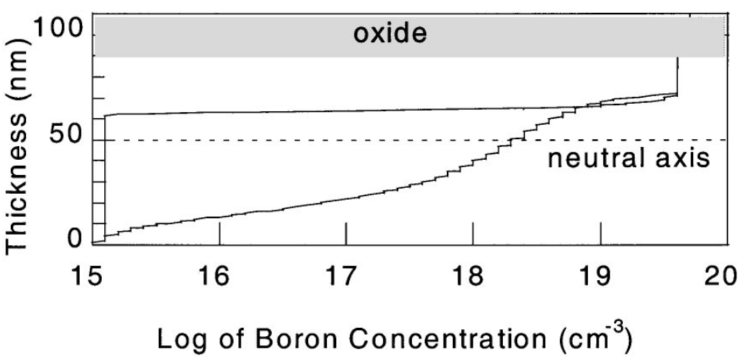
- 140dB dynamic range
- 100pN resolution from 0.1Hz to 100Hz

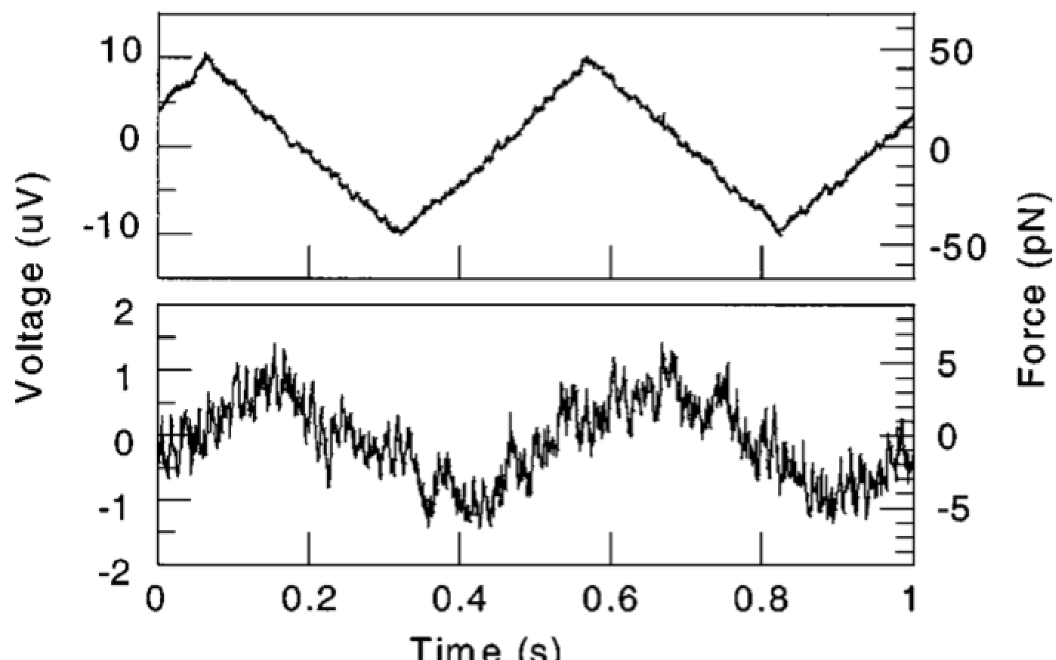
Low 1/f noise full bridge microcantilever with longitudinal and transverse piezoresistors, Mallon et al, APL (2008)

Example: High frequency design



- 89nm thick
- 500fN resolution from 10Hz to 1kHz





High-sensitivity piezoresistive cantilevers under 1000A thick, Harley and Kenny, APL (1999)

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- ***C. elegans* Touch Research**
  - Worm biomechanics
  - Nose touch

C. elegans Touch Research

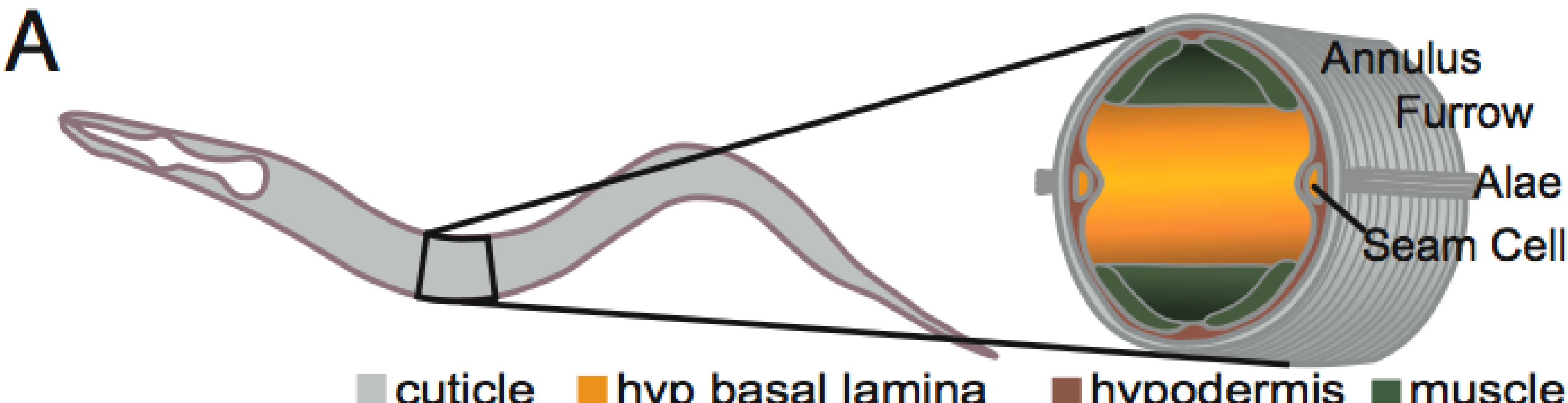
- **Worm biomechanics**
- Behavioral response to nose touch
- Touch electrophysiology (worm level)
- Touch electrophysiology (cell level)
- Modeling (mechanical and molecular)

Sung-Jin Park

Analysis of nematode mechanics by piezoresistive displacement clamp
Sung-Jin Park, Miriam Goodman and Beth Pruitt, *PNAS* 2007

Worm Biomechanics

- What is the modulus of a worm?
- What contributes to the stiffness?
  - Elastic deformation of cuticle
  - Hydrostatic pressure

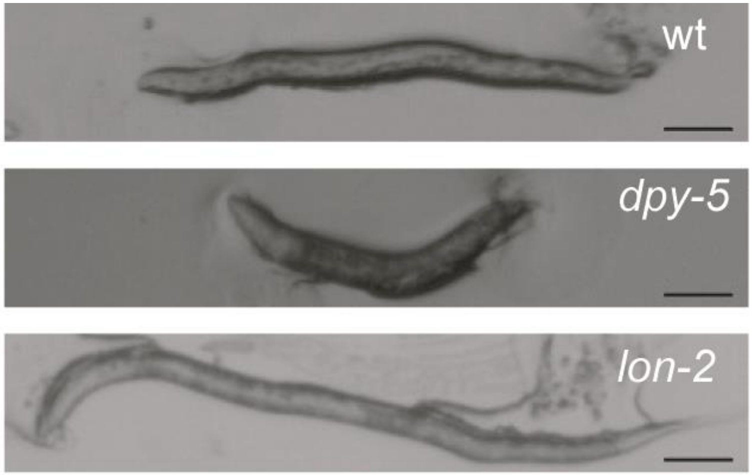


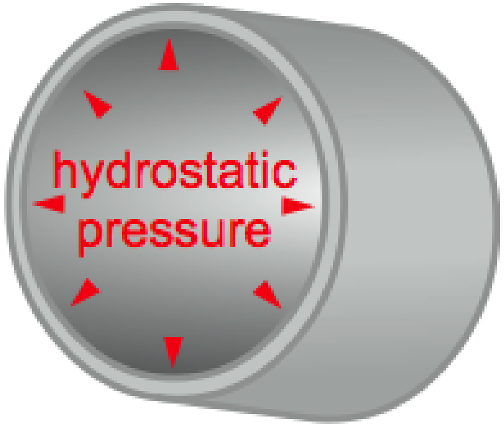
Park et al, *PNAS* 2007

Experimental Approach

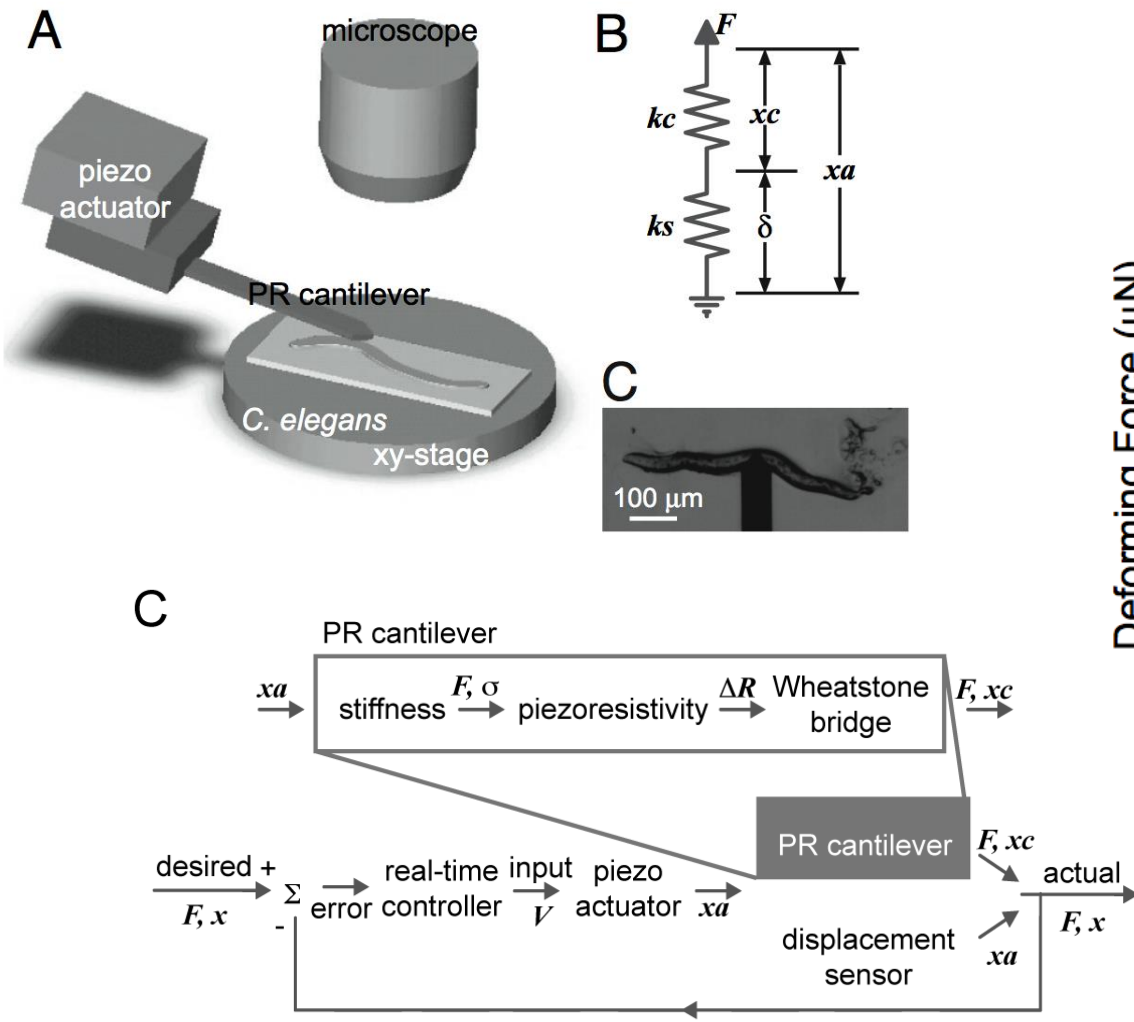
1. The effect of cuticle mutations and body shape can be studied by examining cuticle mutants

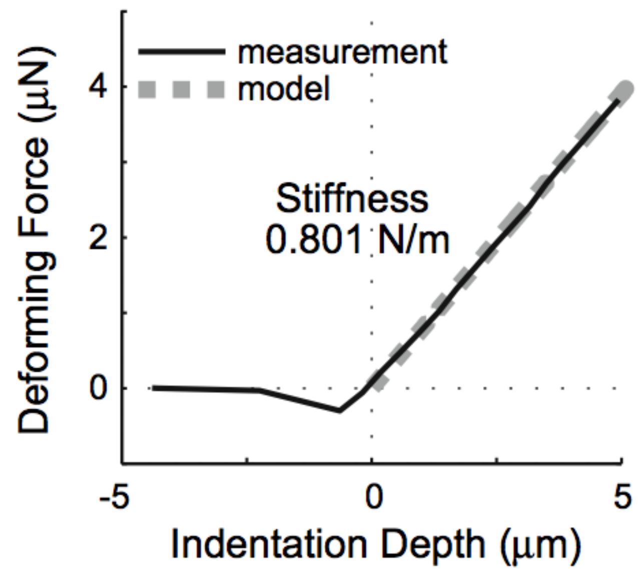
2. The stiffness contribution of internal hydrostatic pressure can be studied by puncturing worms or exposing them to osmotic shock





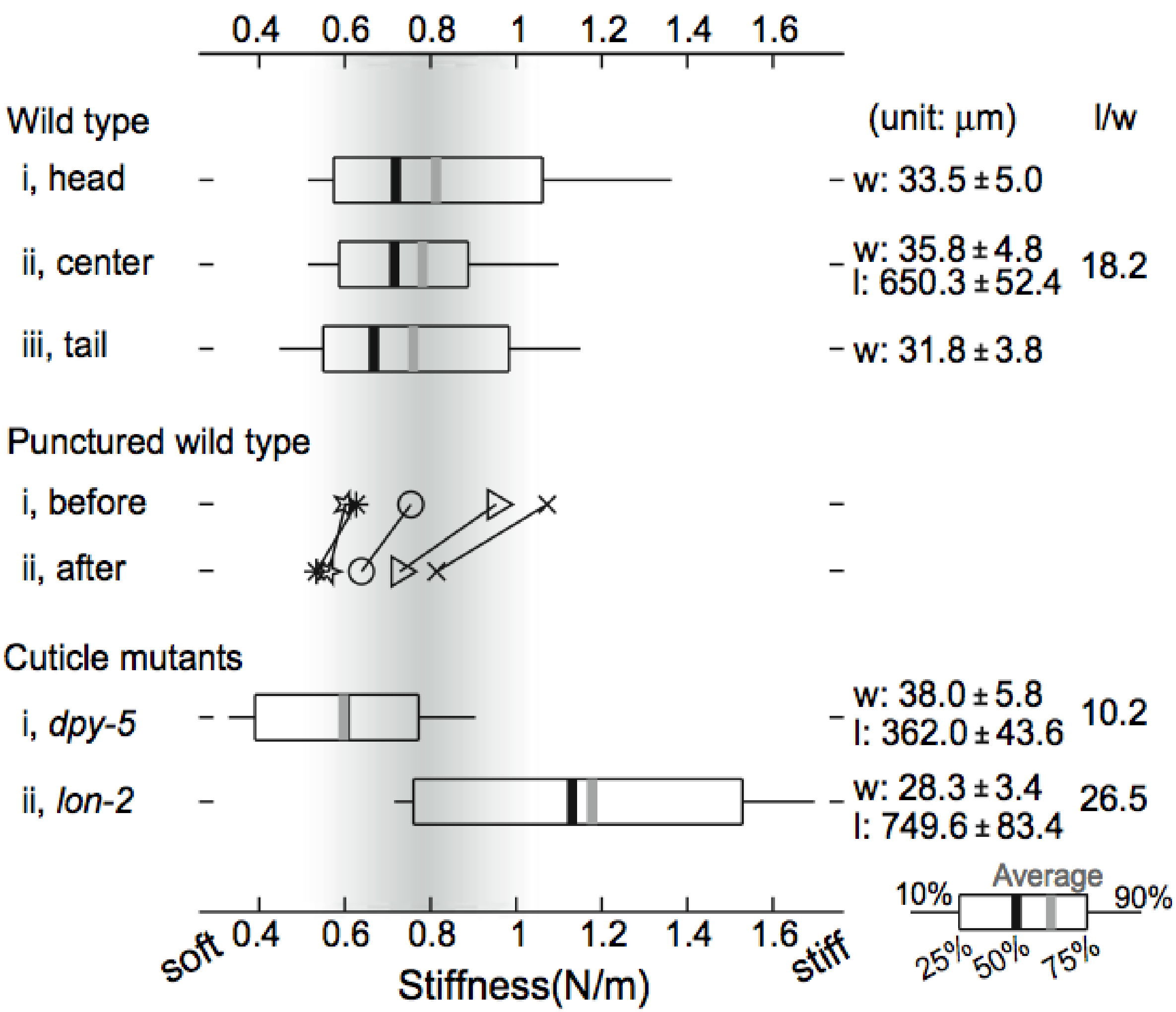
Experimental Setup and Analysis





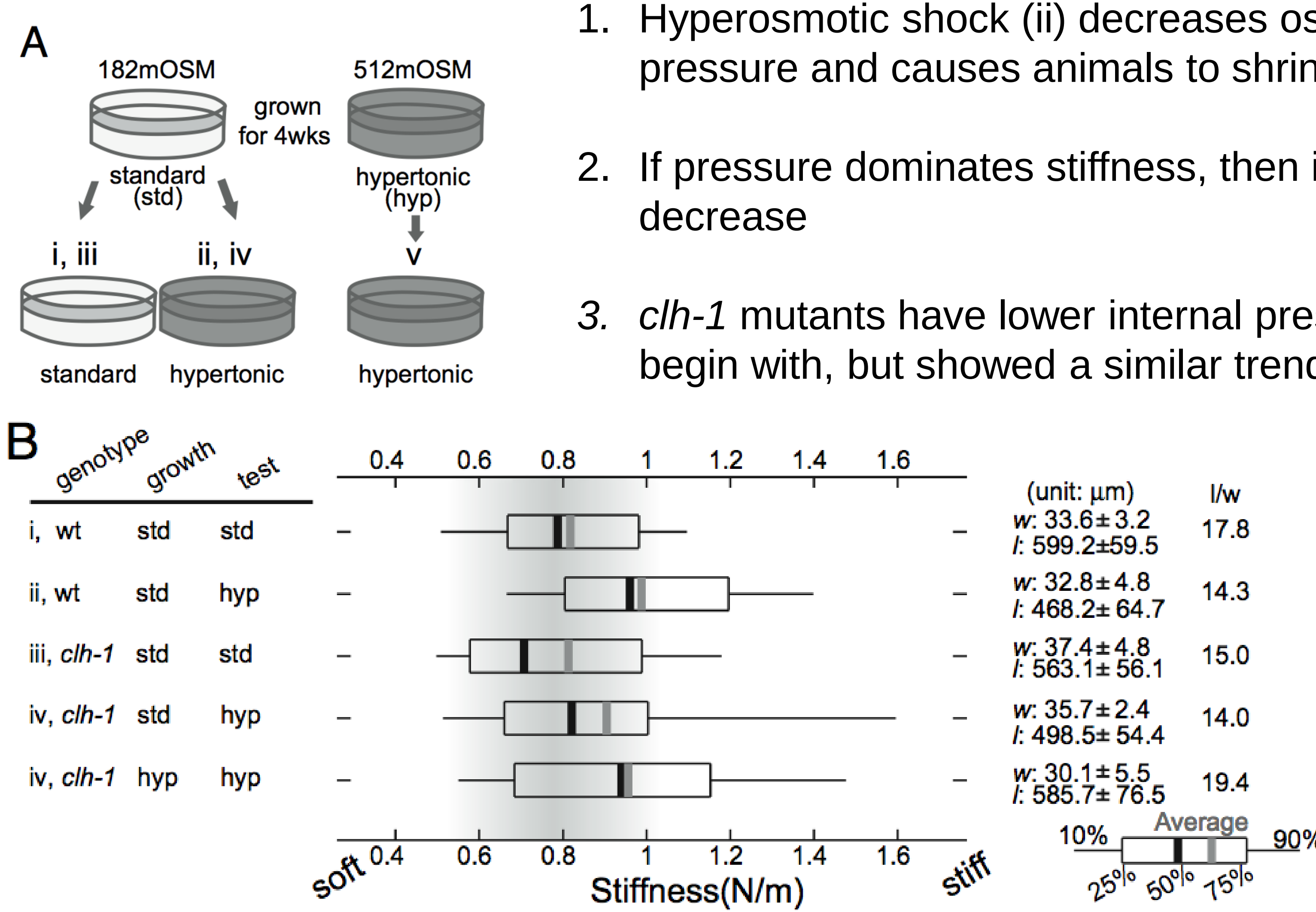
Experiments performed with worm glued on agar

Experimental Results



1. Puncturing slightly reduced stiffness
2. If hydrostatic pressure were dominant, dpy-5 would be stiffer than lon-2 (assuming constant hydrostatic pressure)
3. However, the opposite situation was found to be true

Experimental Results



1. Hyperosmotic shock (ii) decreases osmotic pressure and causes animals to shrink

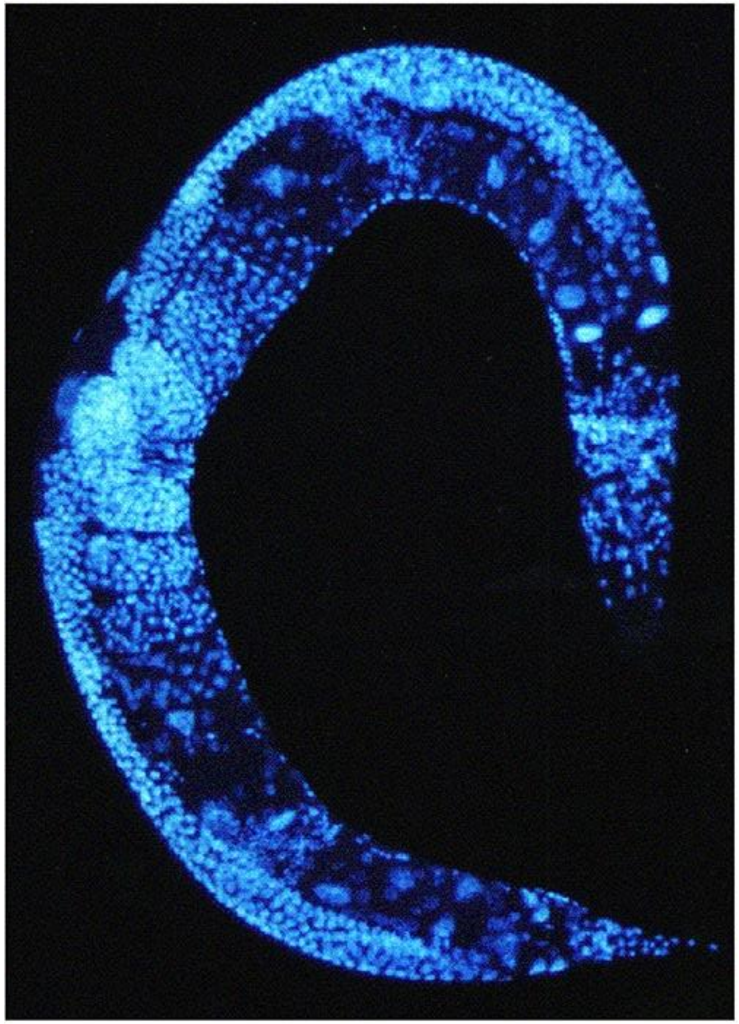
2. If pressure dominates stiffness, then it should decrease

3. *clh-1* mutants have lower internal pressure to begin with, but showed a similar trend

Worm Biomechanics Conclusions

1. C. elegans can be modeled as an elastic shell

2. Animal stiffness is dominated by cuticle stiffness rather than internal hydrostatic pressure



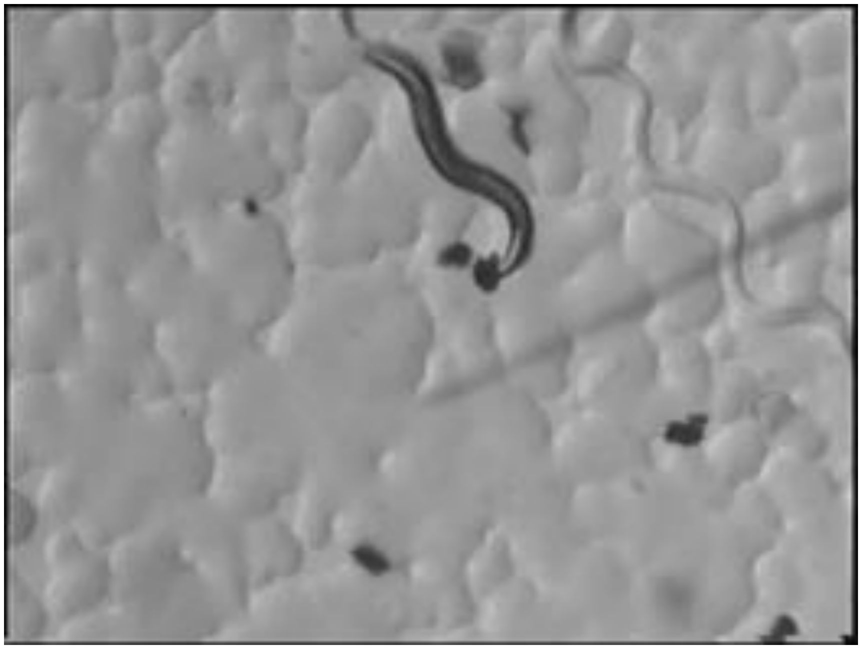
http://wormbook.sanger.ac.uk/chapters/www_intromethodscellbiology/cellfig3.jpg

C. elegans Touch Research

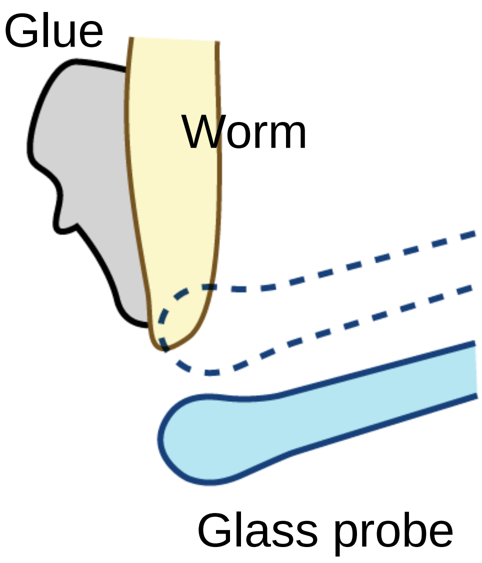
- Worm biomechanics
- **Behavioral response to nose touch**
- Touch electrophysiology (worm level)
- Touch electrophysiology (cell level)
- Modeling (mechanical and molecular)

Behavioral Response to Nose Touch

Behavioral (Active)



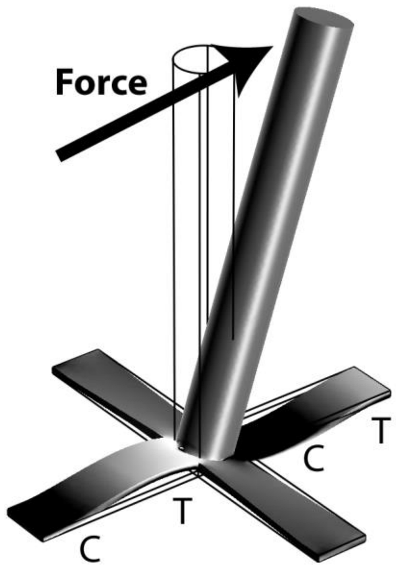
Electrophysiological (Passive)

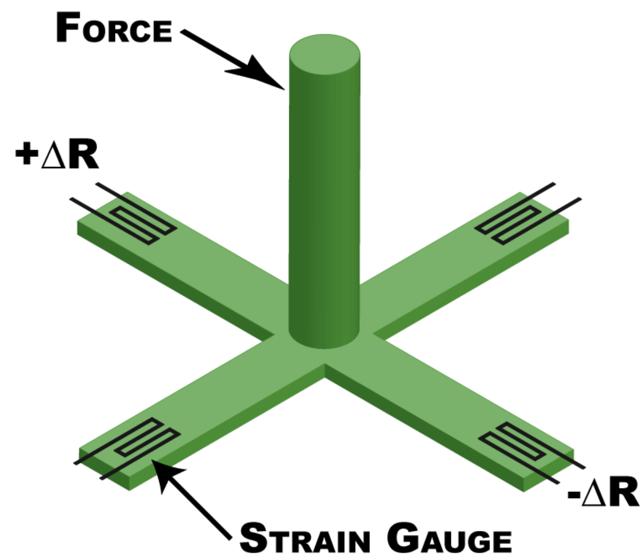


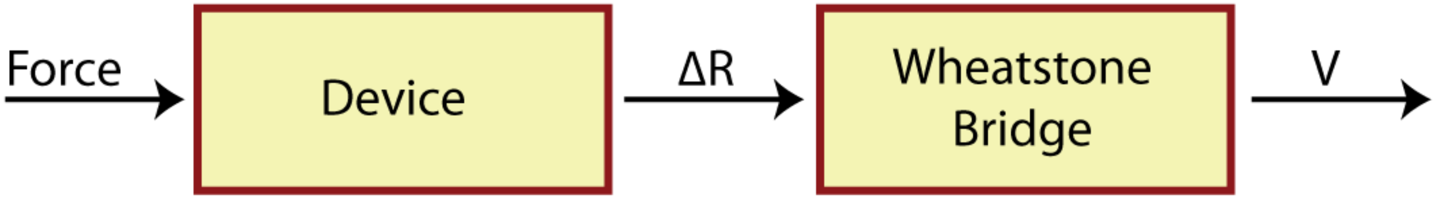
Questions to Answer

1. What size of force elicits a behavioral response?
2. How does this value compare to the forces applied during physiological experiments?

Device Design

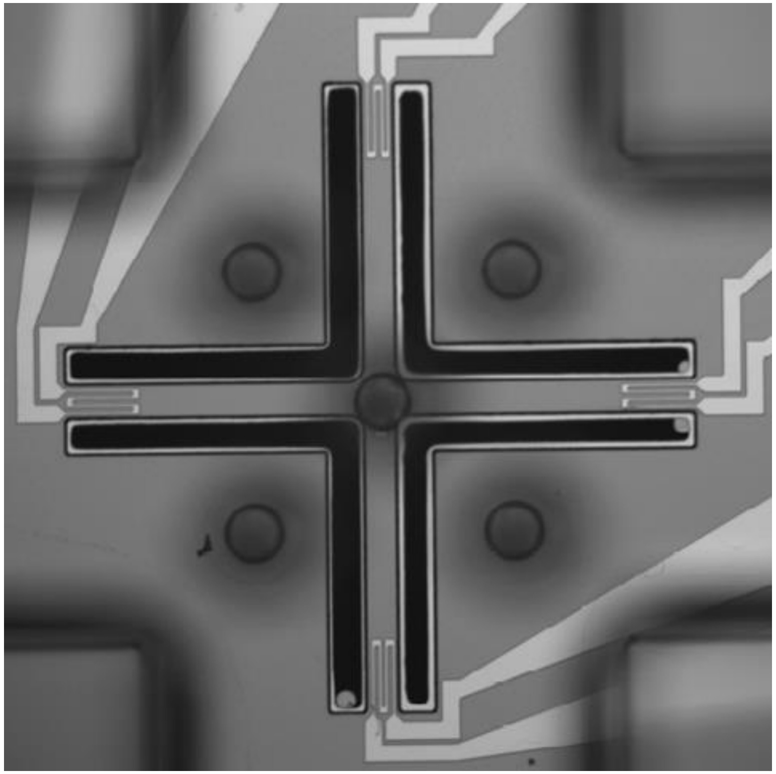


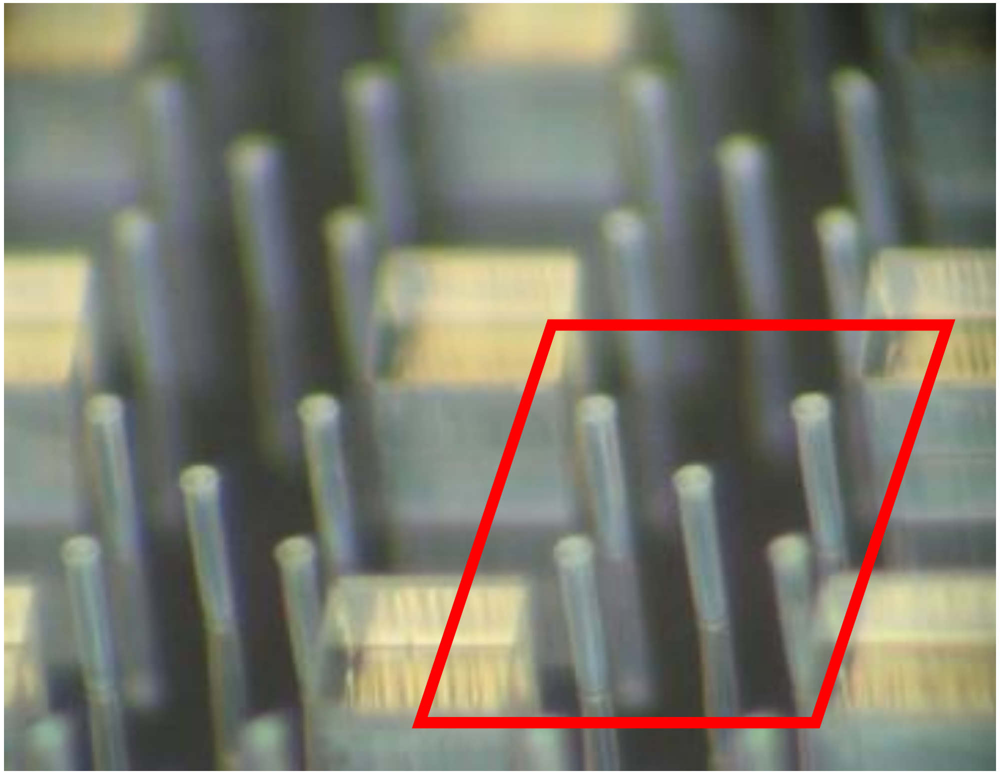




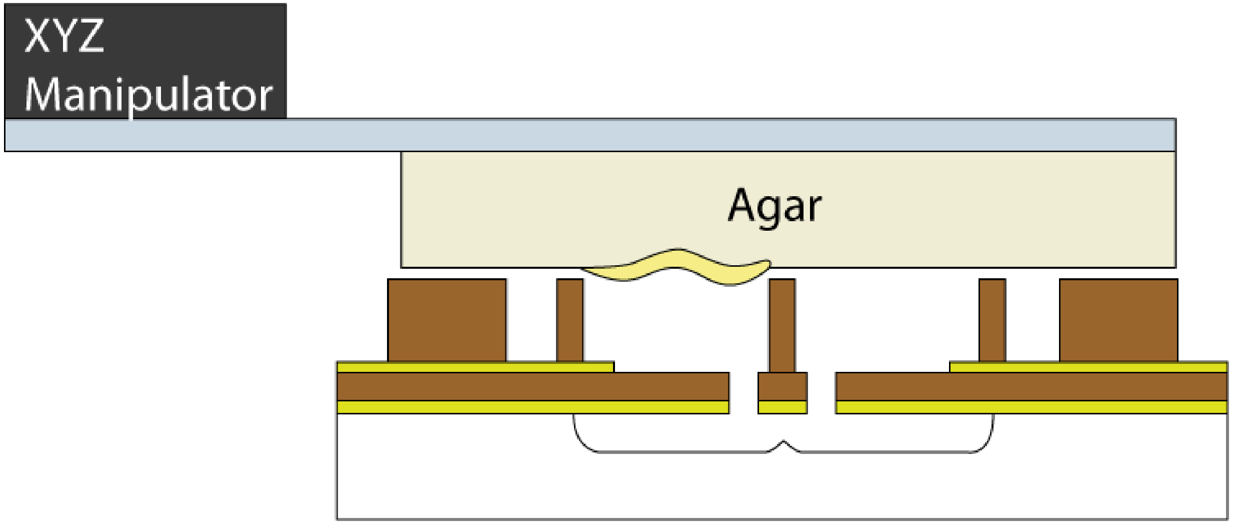
Doll JC, Harjee N, Klewja N, Kwon R, Coulthard SM, Goodman MB, Pruitt BL, Biological measurements of C. elegans touch sensitivity with microfabricated force sensors. Proceedings of MicroTAS (2007).

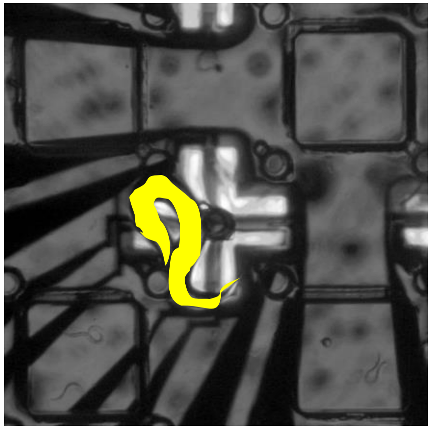
Finished Device



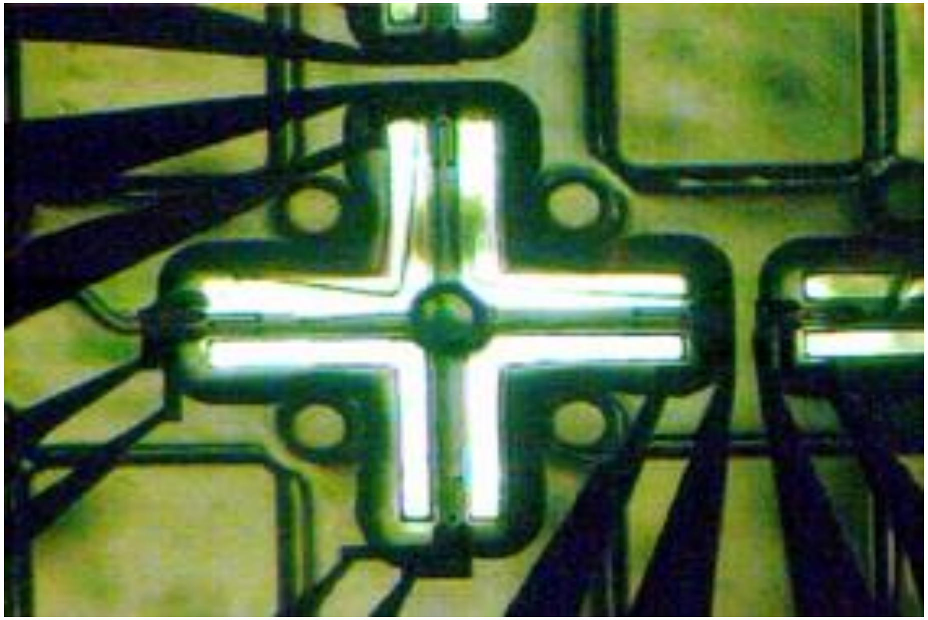


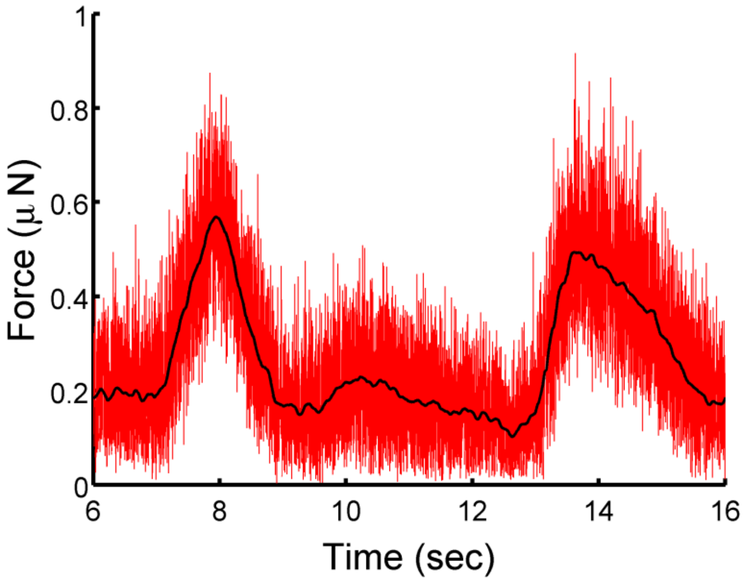
Experimental Setup



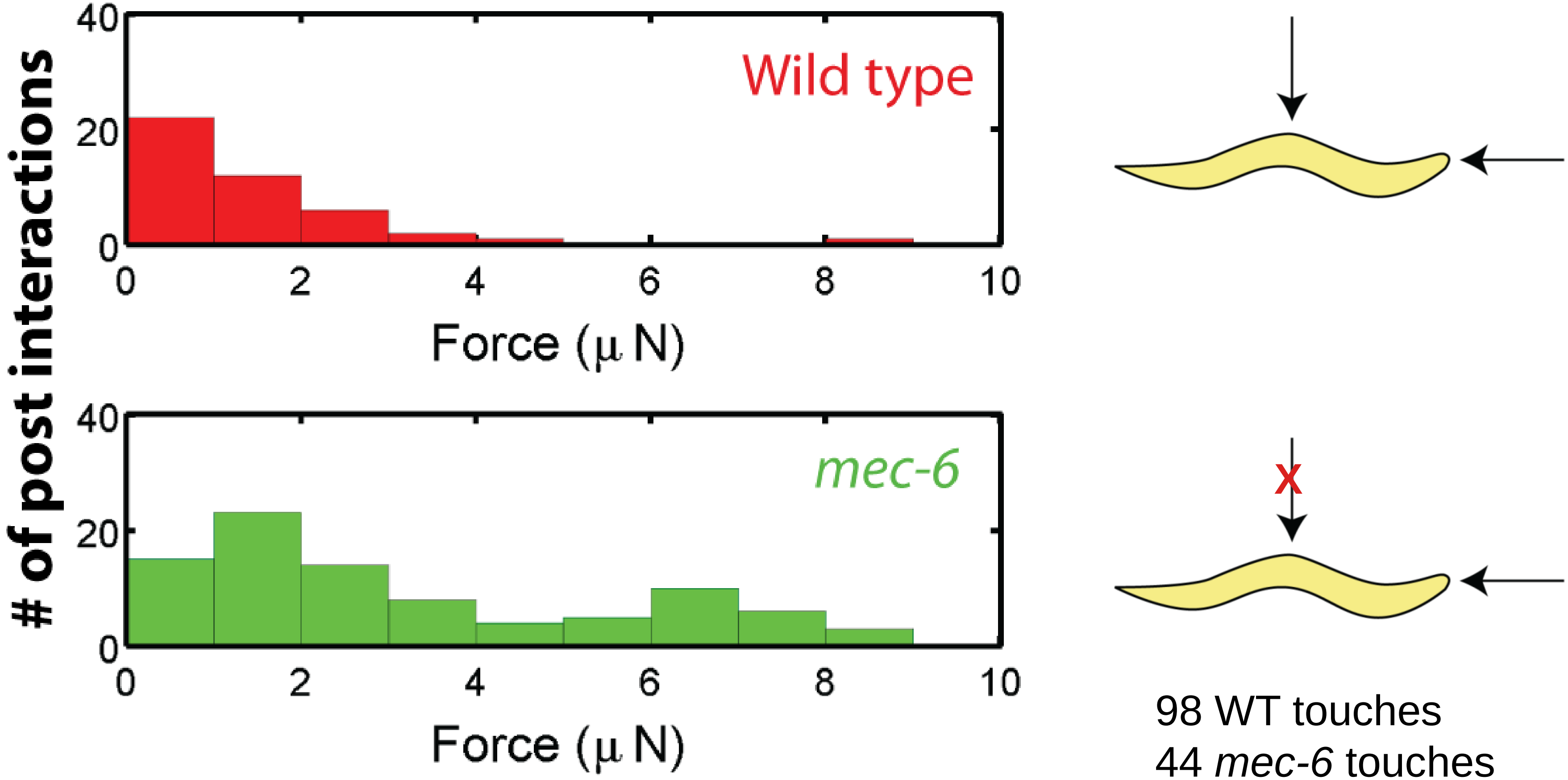


Worm Measurement Setup





Experimental Results



98 WT touches
44 *mec-6* touches

C. elegans Touch Research

- Worm biomechanics
- **Behavioral response to nose touch**
- Touch electrophysiology (worm level)
- Touch electrophysiology (cell level)
- Modeling (mechanical and molecular)

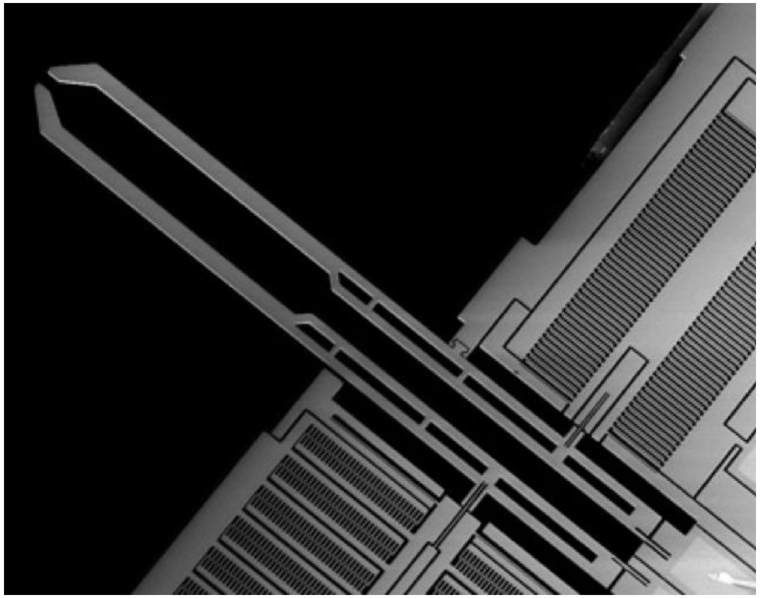
Prof. Brad Nelson
Institute of Robotics and Intelligent Systems
ETH Zurich

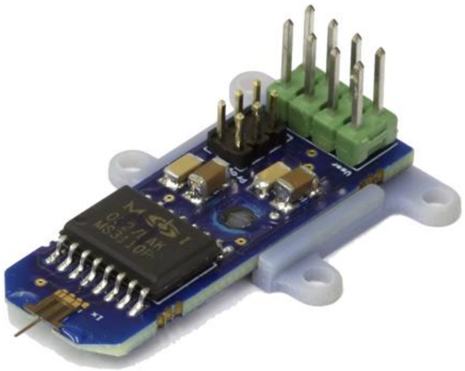
Felix Beyeler

Simon Muntwyler

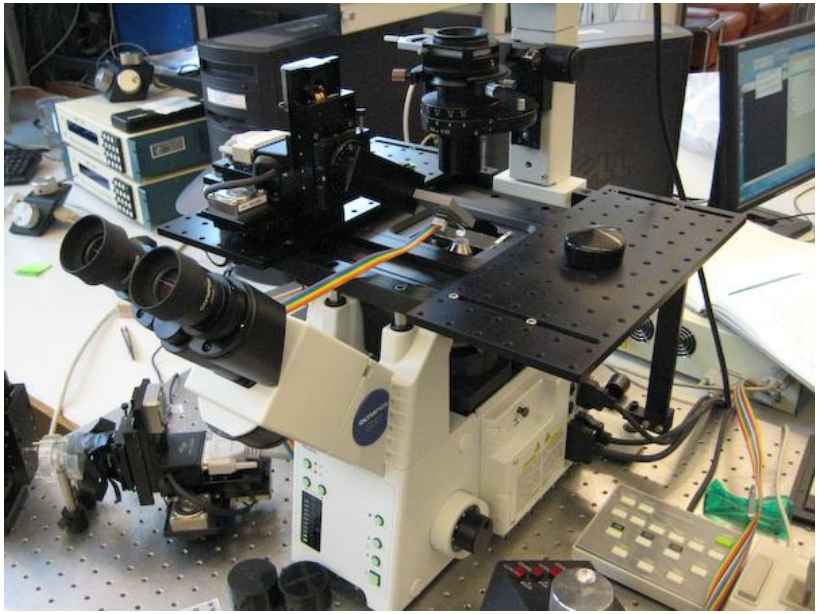
Force Measurement Probe

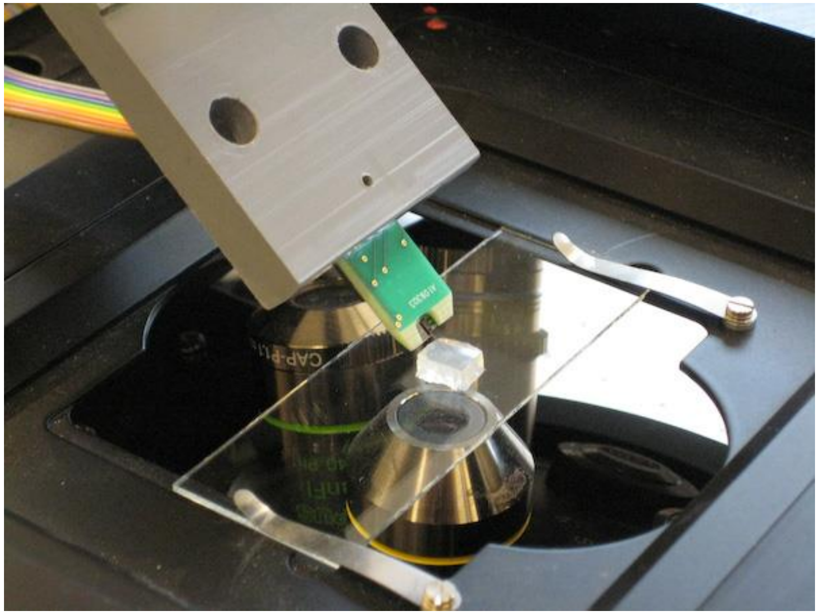
- In-plane electrostatic force sensor
- Force resolution ~10nN
- Resonant freq ~350Hz
- Actuator + sensor for gripping
  - Broke off actuator for these measurements





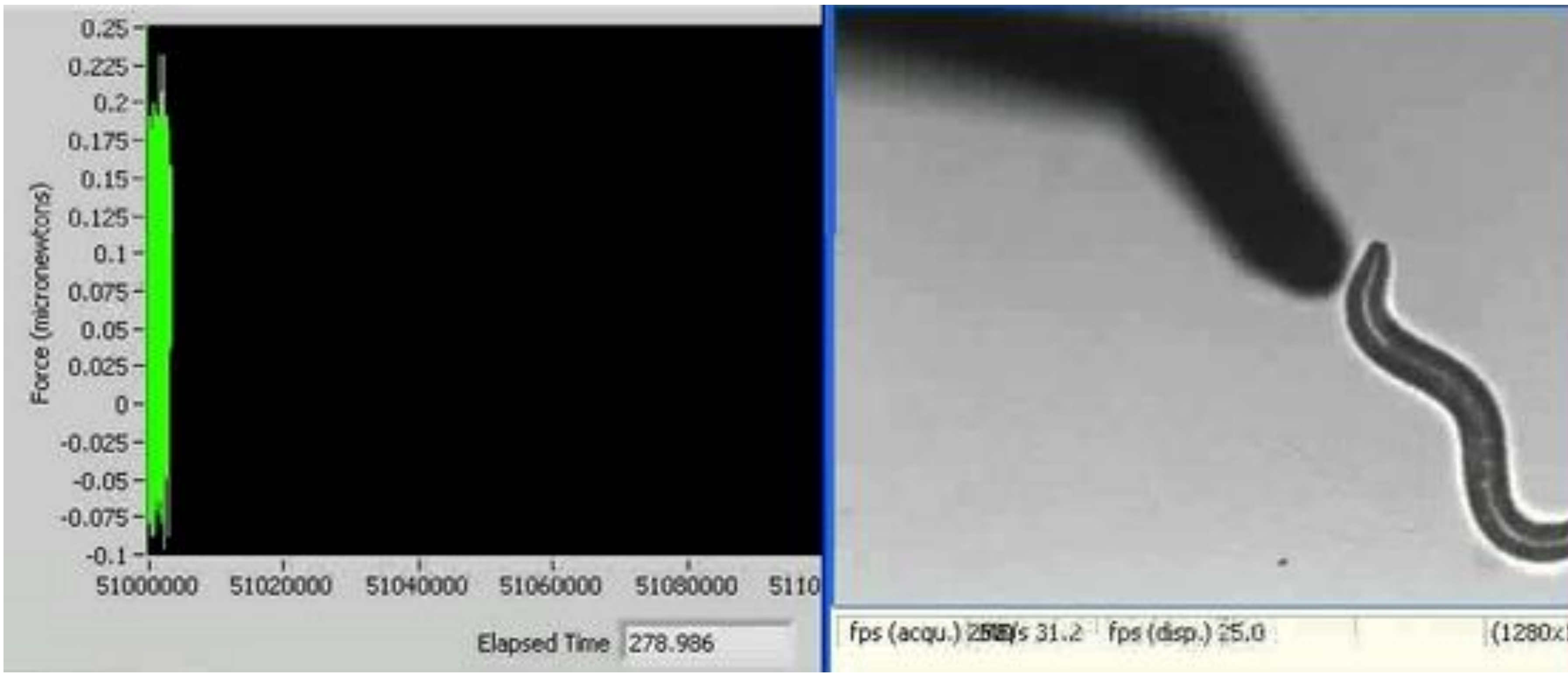
Experimental Setup





Worms on top of piece of agar, inverted microscope (100x mag), motorized stage. Sensor mounted on 3-axis motor (manual or computer control). Acquisition with Labview.

Force Measurement in Action



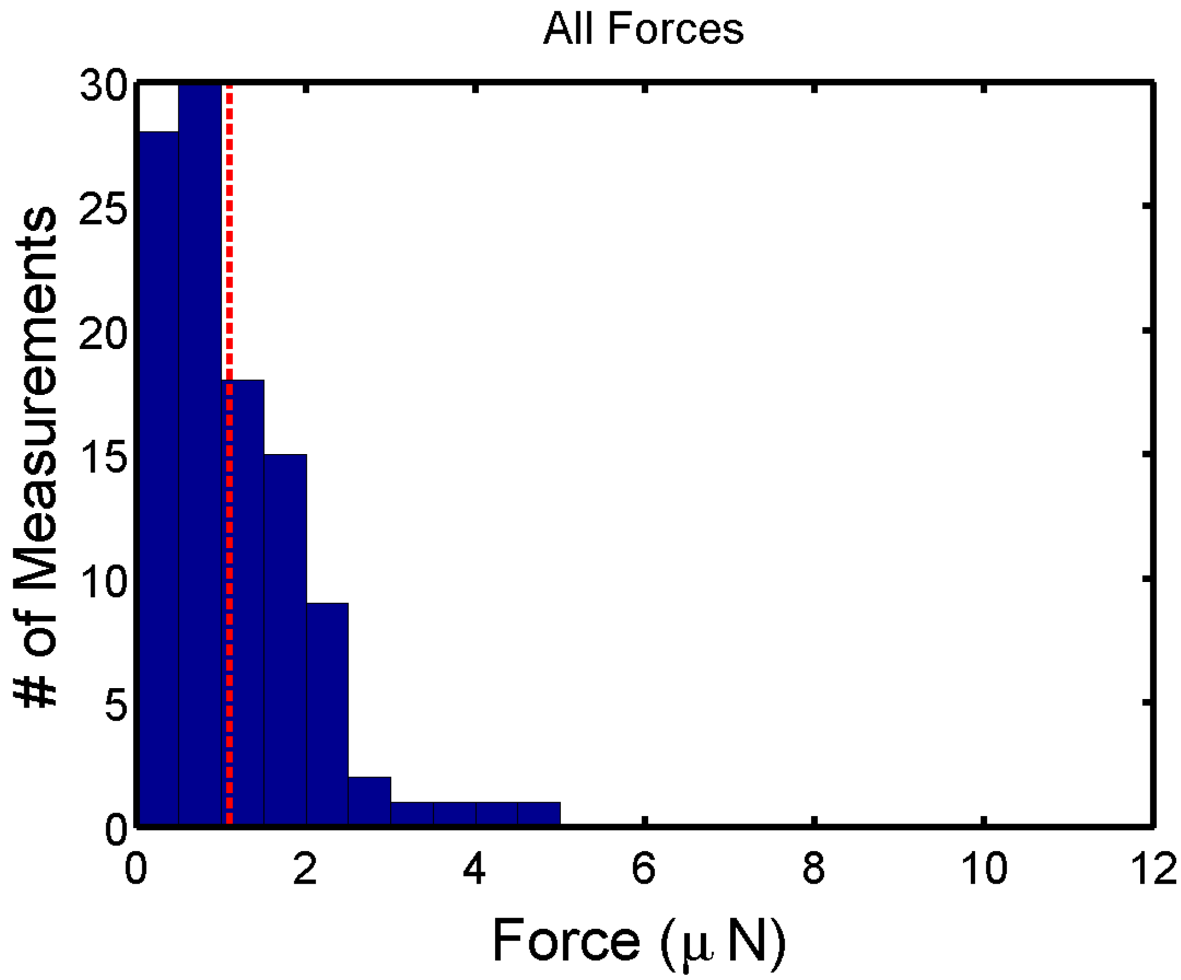
Note the push (-) and pull (+) of the worm on the probe

Preliminary Nose Touch Results

8 worms
106 nose touches

All Touches
Mean = 1.10 uN
STD = 0.91 uN

Mean of ind. worms
Mean = 1.11 uN
STD = 0.38 uN



C. elegans Touch Research

- Worm biomechanics
- Behavioral response to nose touch
- Touch electrophysiology (worm level)
- Touch electrophysiology (cell level)
- Modeling (mechanical and molecular)

Acknowledgements

- Fellowships from
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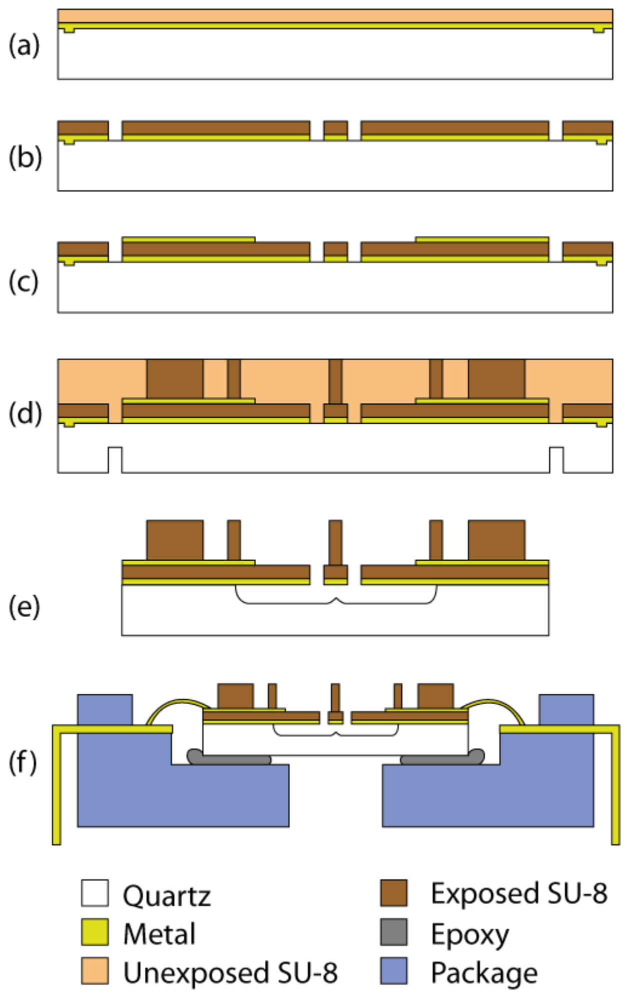
  - NIH R01

Stanford Microsystems group (2007)

Thank You

Fab Process

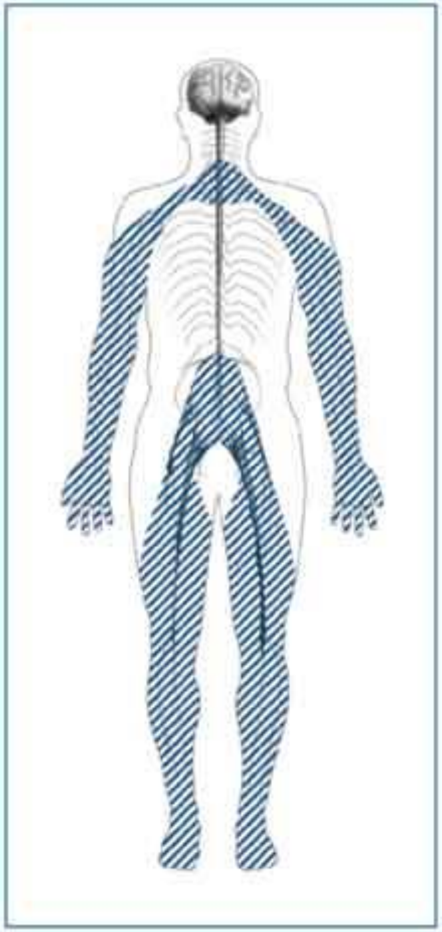


(a) Sputter Cr/Au adhesion layer and spin 5µm SU-8 on quartz.
(b) Pattern cantilever arms and metal.
(c) Deposit and pattern strain gauges.
(d) Deposit and expose SU-8 pillar layer. Wafer saw from the backside.
(e) Develop SU-8 and release in HF.
(f) Glue device to package and wire bond.

Devices were fabricated with 200, 300, 400 and 500µm long cantilever arms. Force pillars were 350µm tall and 70µm in diameter.

One Reason to Study Touch

- Healthcare
  - 11M diabetics in US
  - 59% suffer from Diabetic Peripheral Neuropathy (DPN)
  - 66k lower limp amputations annually
  - $4.6-13.7B annual cost of DPN

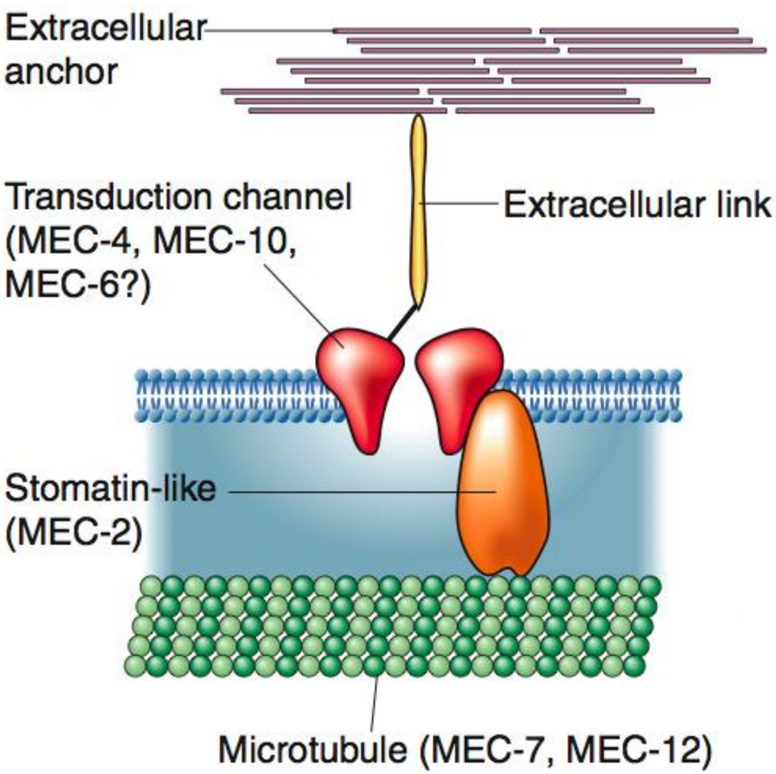


http://diabetes.niddk.nih.gov/dm/pubs/neuropathies/

Gordois, A. et al. The Health Care Costs of Diabetic Peripheral Neuropathy in the U.S. Diabetes Care 26, 1790-1795 (2003).

Work to Date

- Known
  - Channel identity
- Unknown
  - Adaptation mechanism
  - Gating mechanism
  - Opening/closing kinetics
  - Only *in vivo* experiments



(Model as of 2005, still changing)

Metazoan mechanotransduction mystery finally solved, Ronan and Gillespie, Nature (2005)

Force Sensing Techniques

- **Capacitive**, $\Delta C \propto$ gap (geometry &/or properties, sensitivity scales as Area, L^2)
  - pN in commercial parts
- **Tunneling**, $\Delta I \propto$ gap (geometry &/or properties)
  - pN in custom (soon commercial) parts
- **Optical**, $\Delta V \propto$ geometry (sensitivity $\propto$ reflected intensity, ultimately Area, L^2)
  - pN in Commercial AFM
  - aN in Custom Parts e.g. optical tweezers
- **Piezoresistive**, $\Delta R \propto$ strain, temperature, light
  - nN in commercial parts
  - pN in custom cantilevers
  - Sensitivity $\propto$ dimensions, orientation, number carriers
- Measured forces get smaller as scale down sensors
  - e.g. Pressure Sensors, F ~ Area (L^2)
  - e.g. Inertial Sensors, F ~ Volume (L^3)