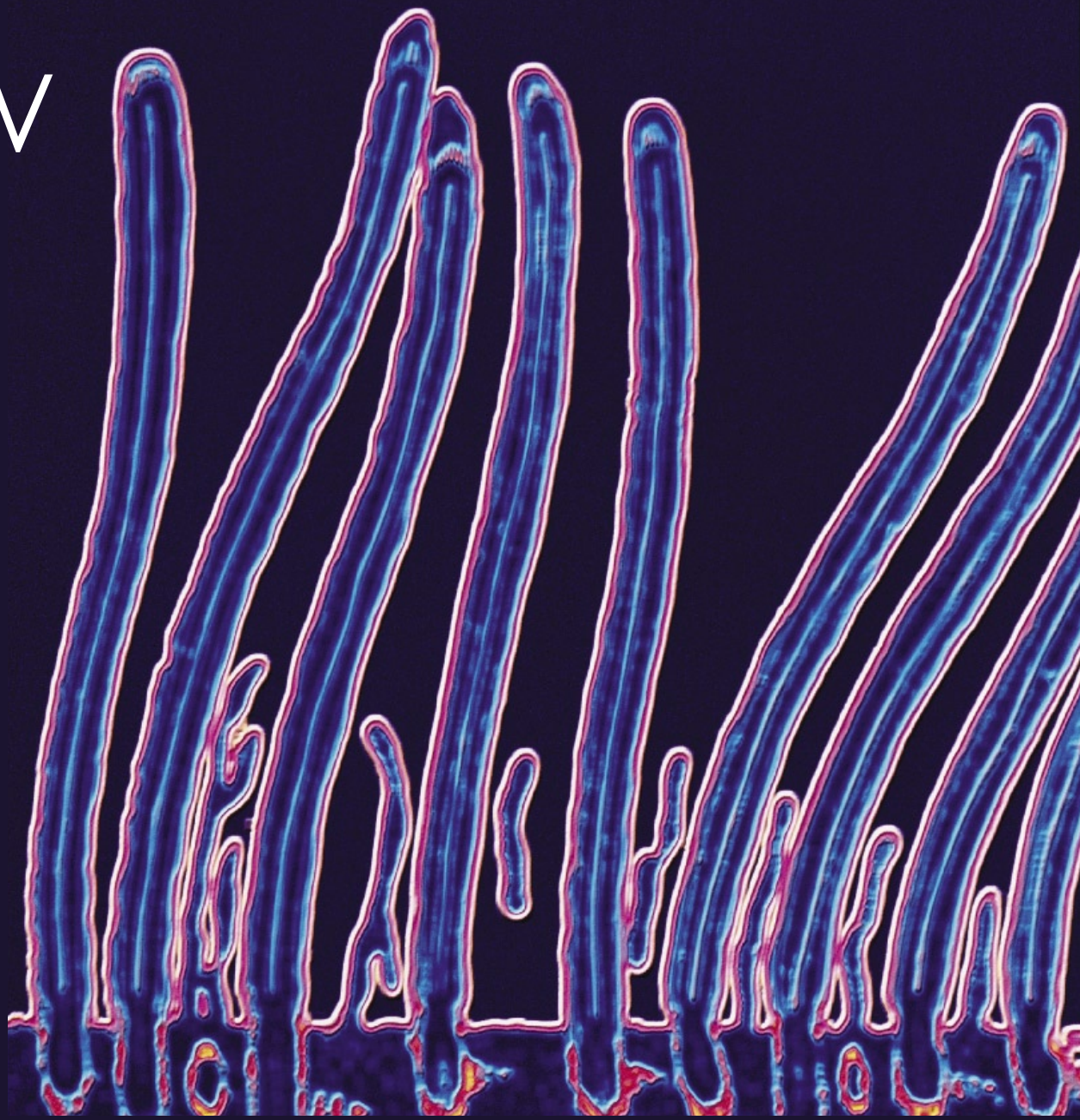
Renal cilia detect flow



Ardon Shorr

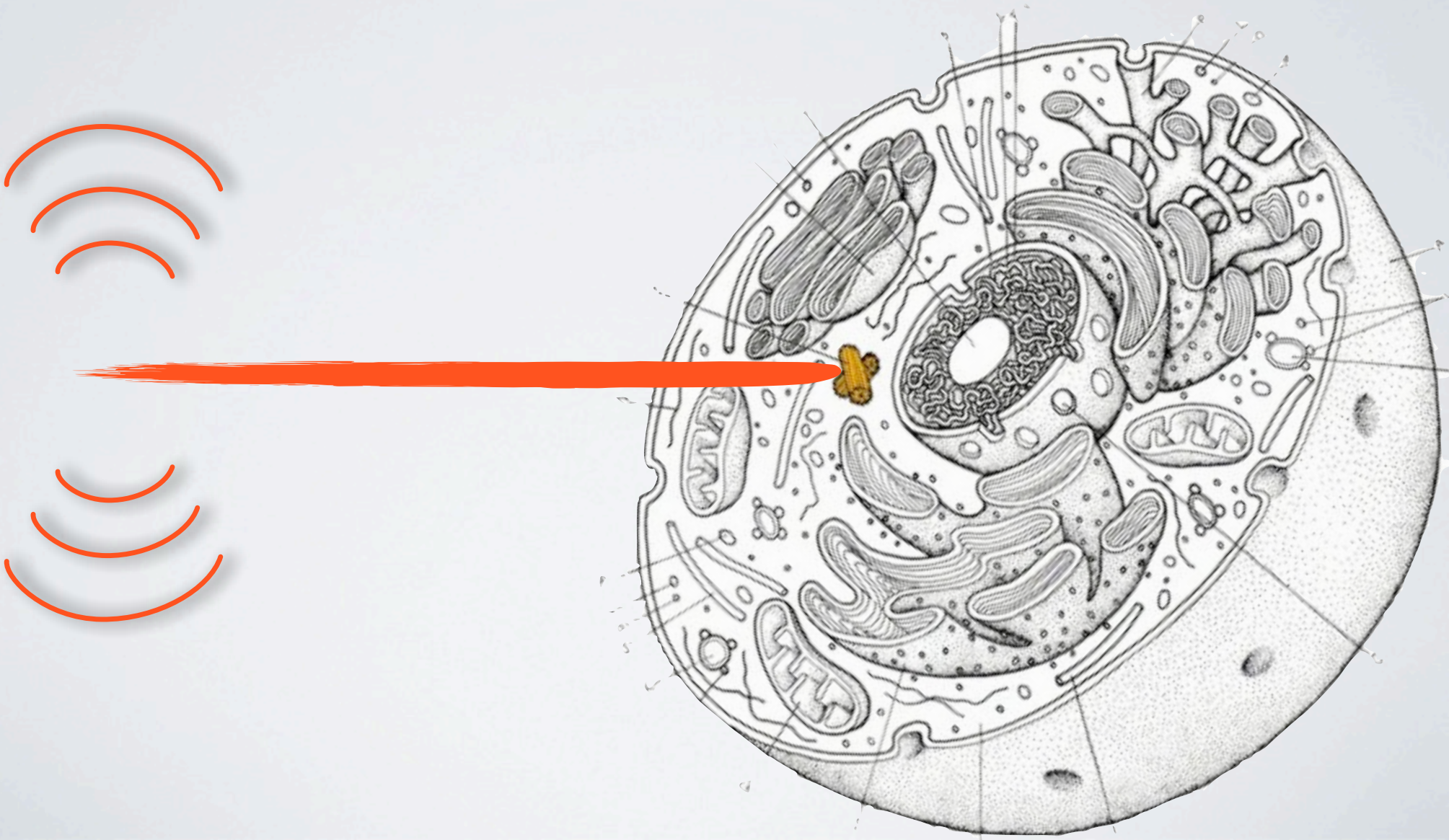
Chekhov's Gun

"If you show a gun in Act one
you better fire it in Act three."

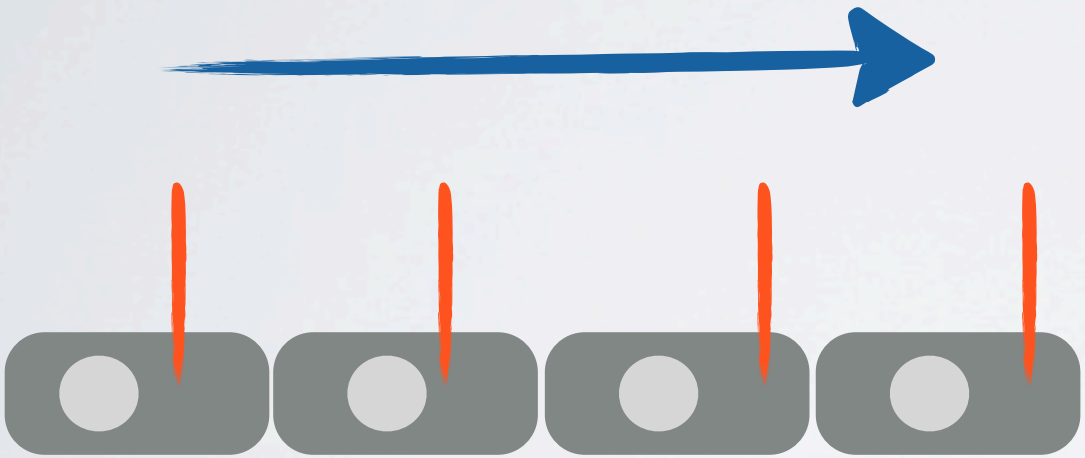
"If you say in the first chapter
that there is a rifle
hanging on the wall,
in the second or third chapter
it absolutely must go off.
If it's not going to be fired,
it shouldn't be hanging there."

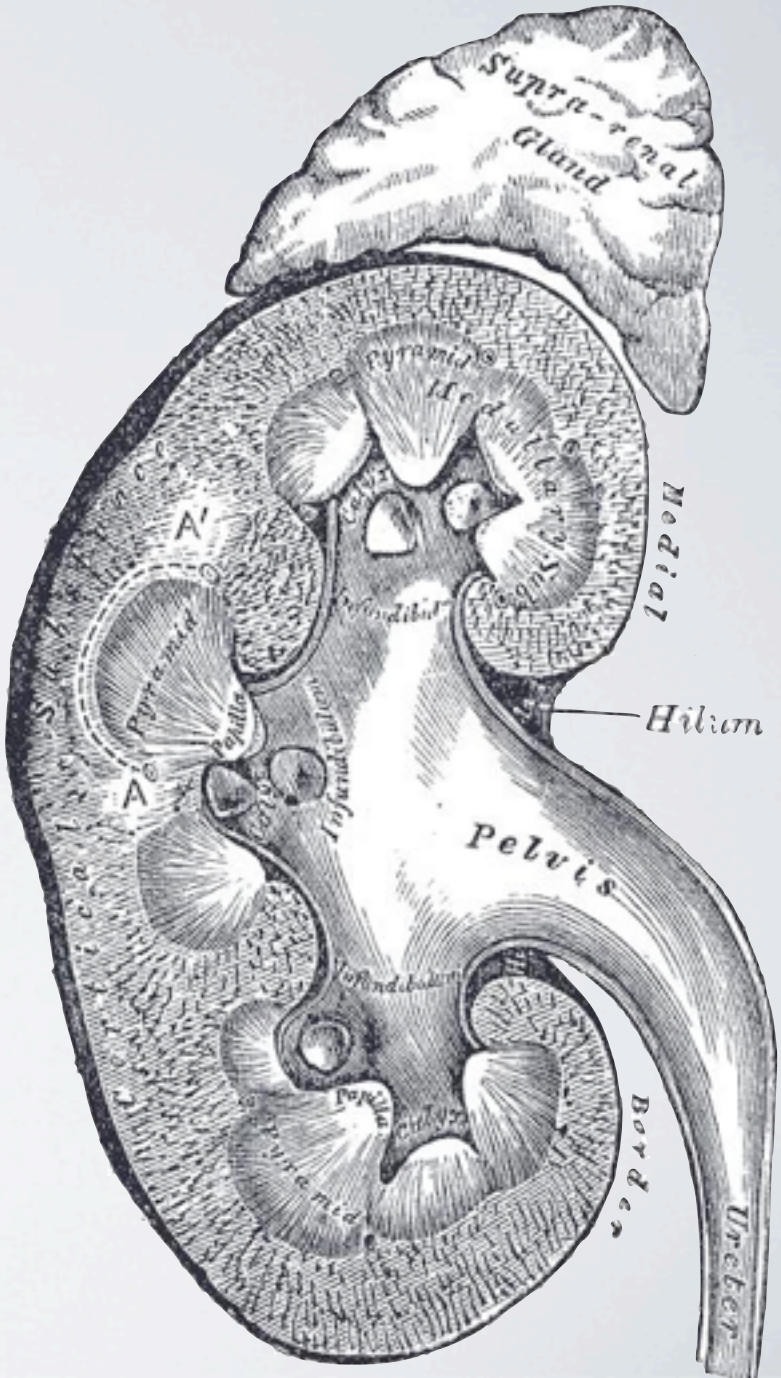
Evolutionary conservation

Primary cilia are ancient, with an unknown function

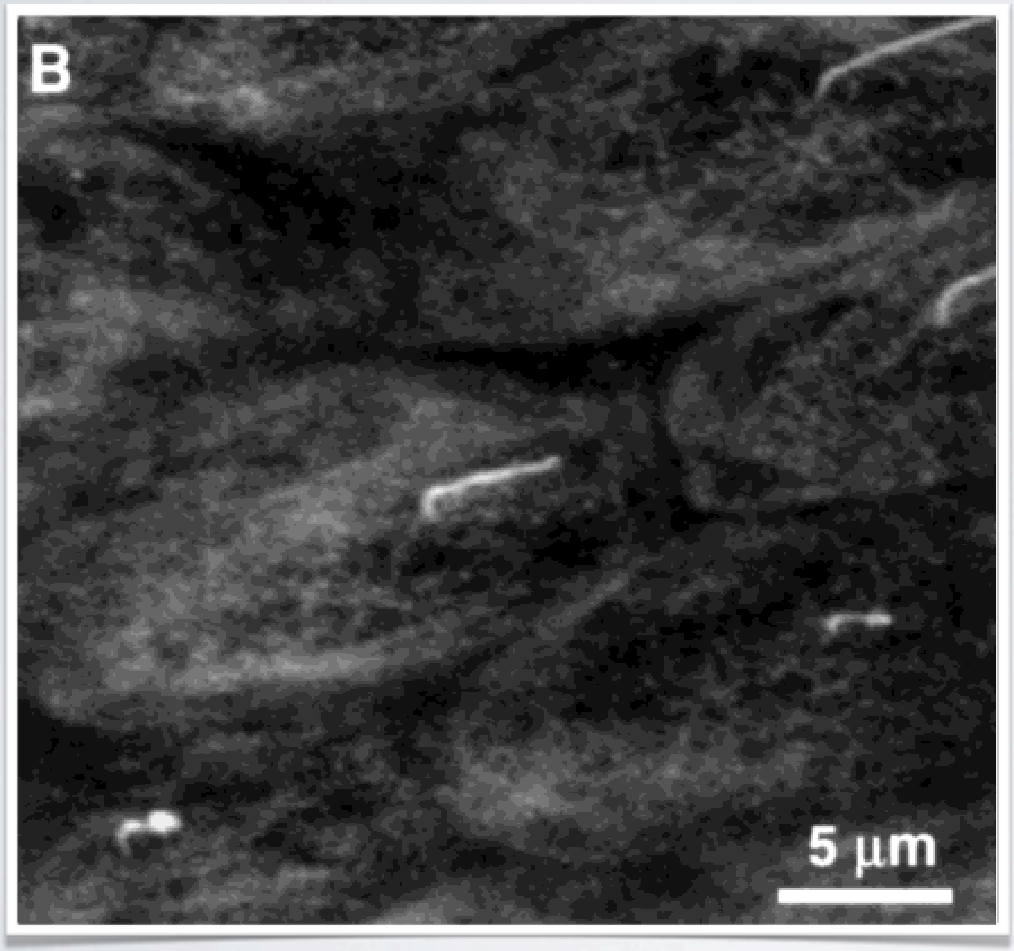


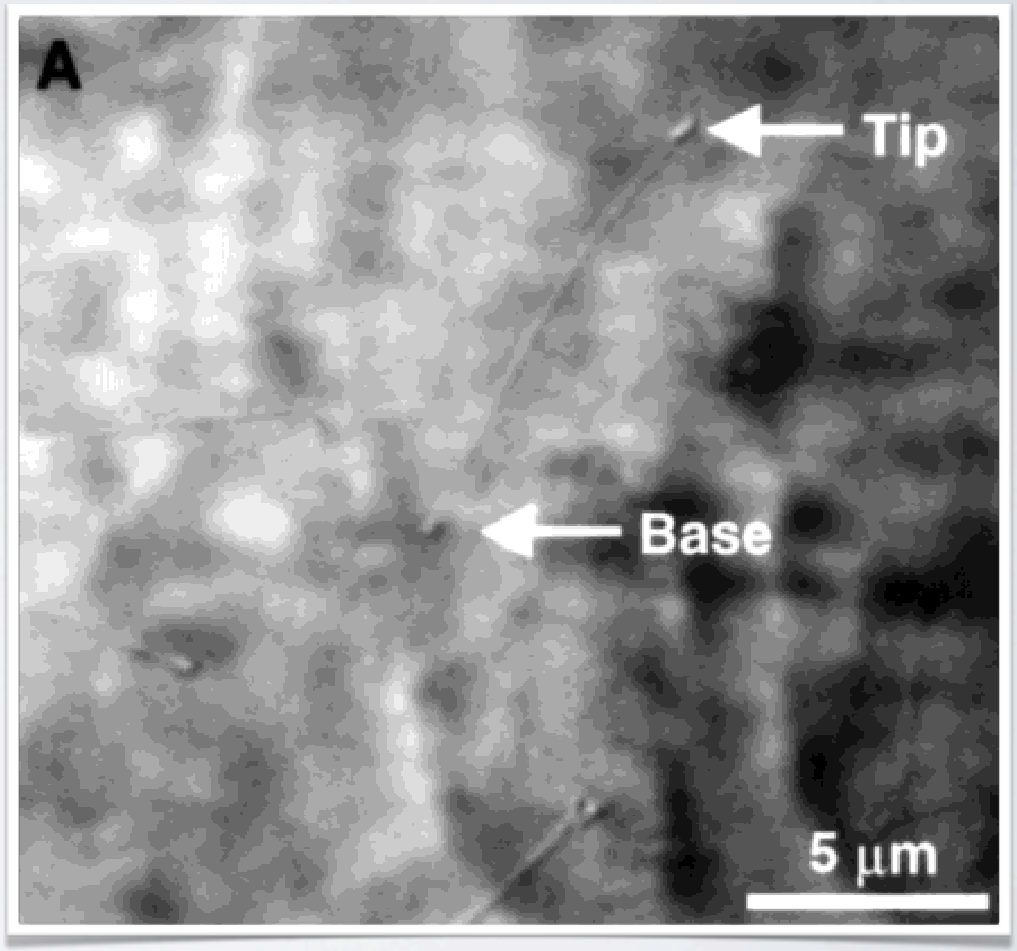
Kidney epithelia each express a single primary cilium





Primary cilia are microtubule structures





The Journal of
Membrane
Biology

Bending the MDCK Cell Primary Cilium Increases Intracellular Calcium

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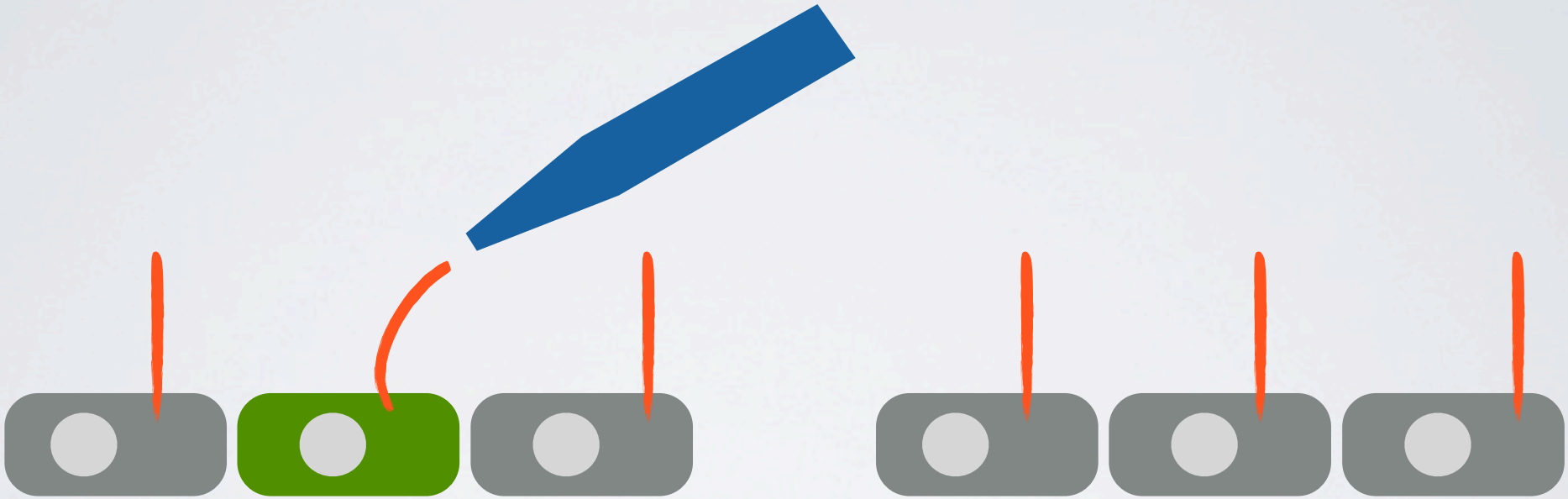
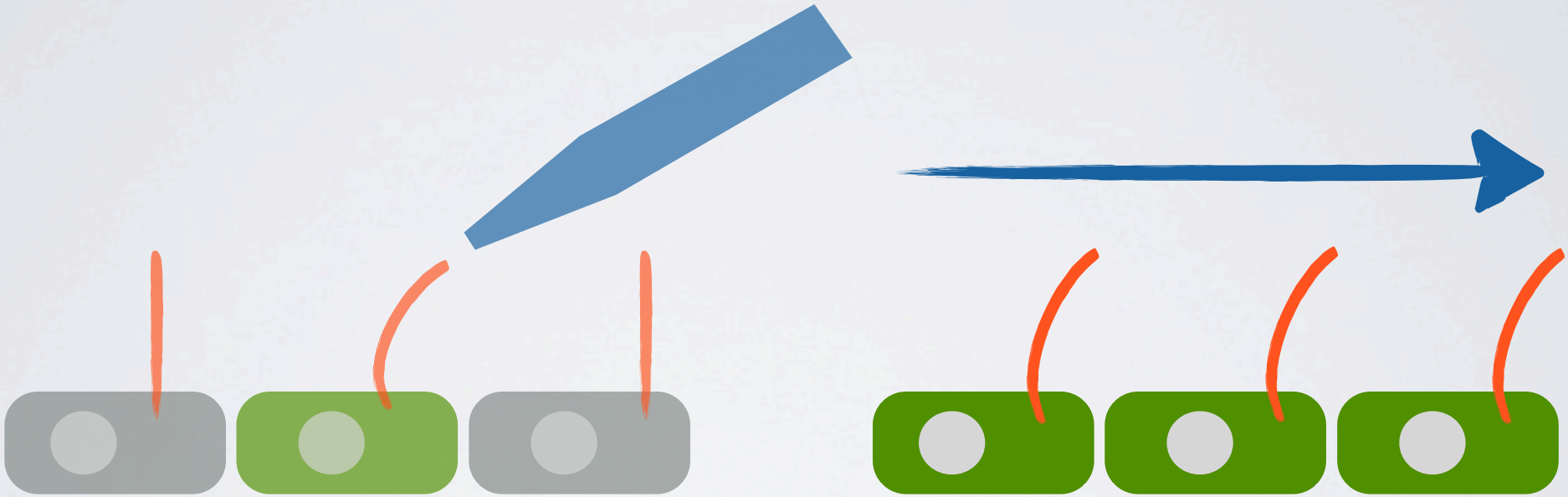
NIH, NHLBI, LKEM, 10 Center Drive, Bldg. 10, Bethesda, MD 20892-1603, USA

Bending cilia increases calcium inside the cell

Cilia can be bent by **suction**

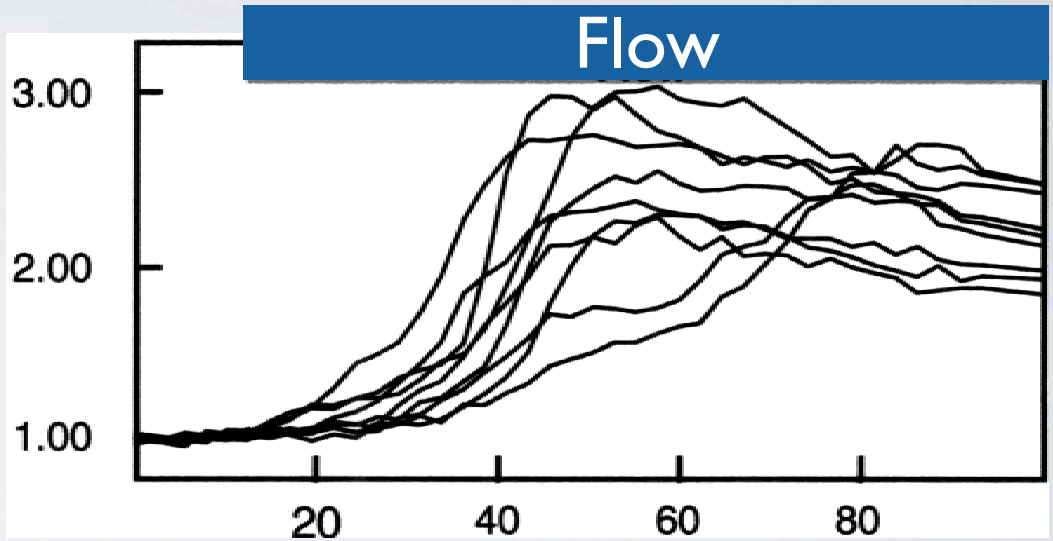


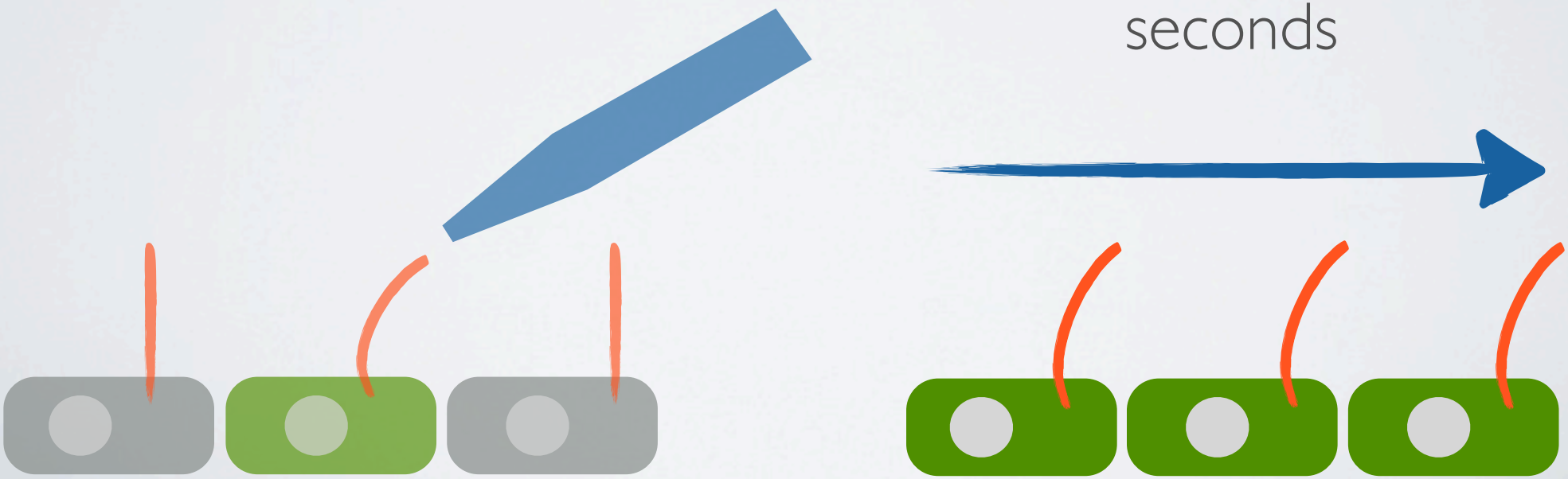
Cilia can be bent by suction or **laminar flow**



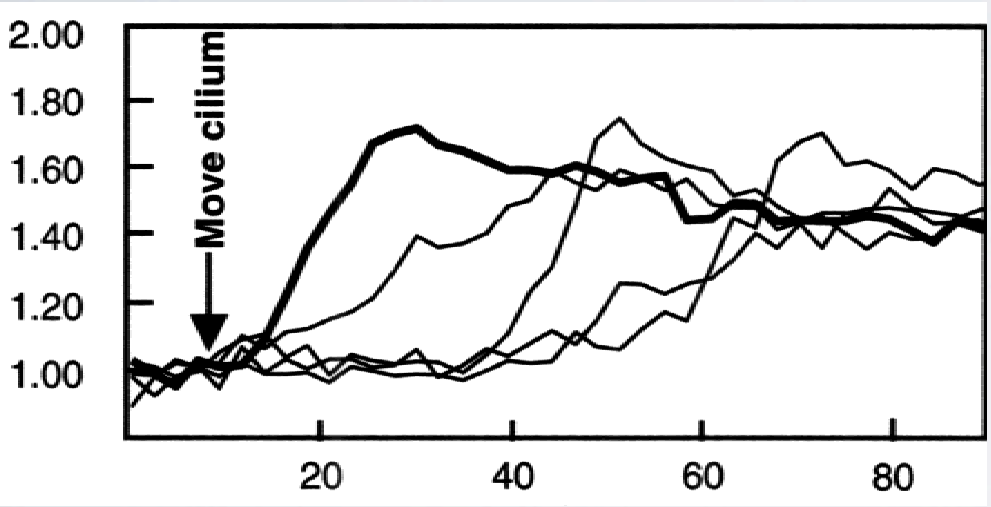
Flow triggers an increase in calcium

1.74 ± 0.07 fold
($p < 0.01$, $n = 37$).

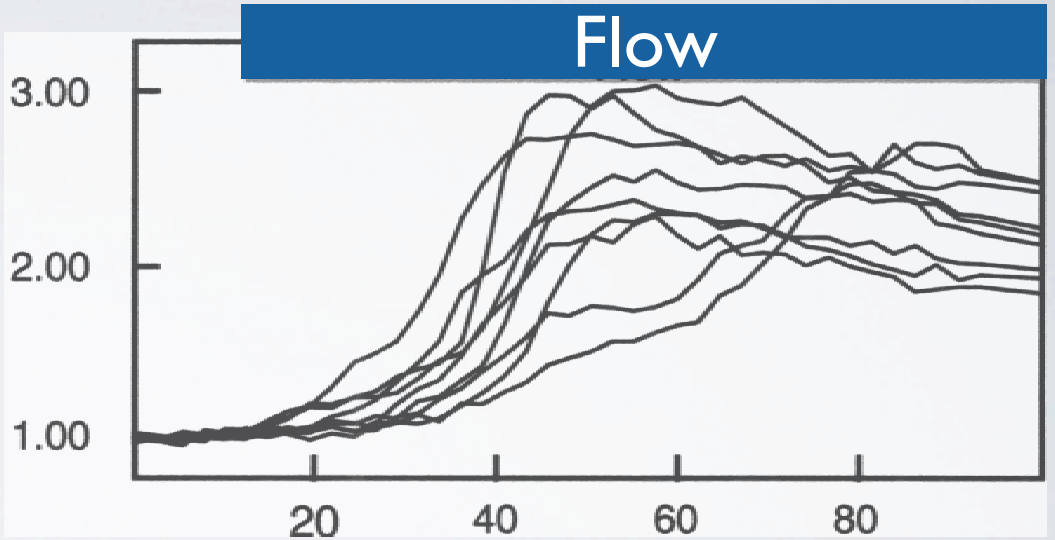




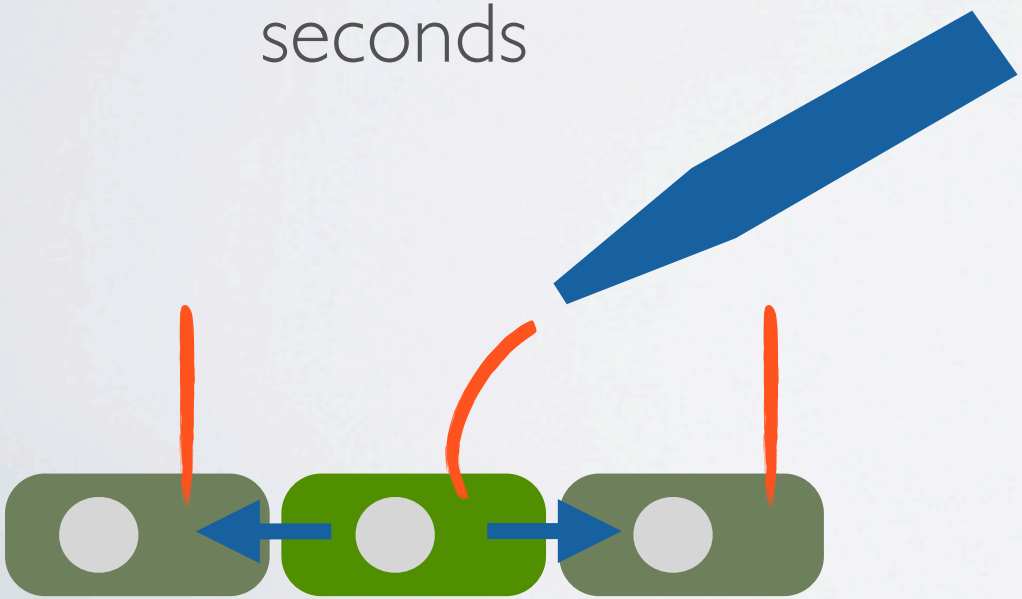
Suction triggers an increase in calcium in one cell that spreads to neighbor cells

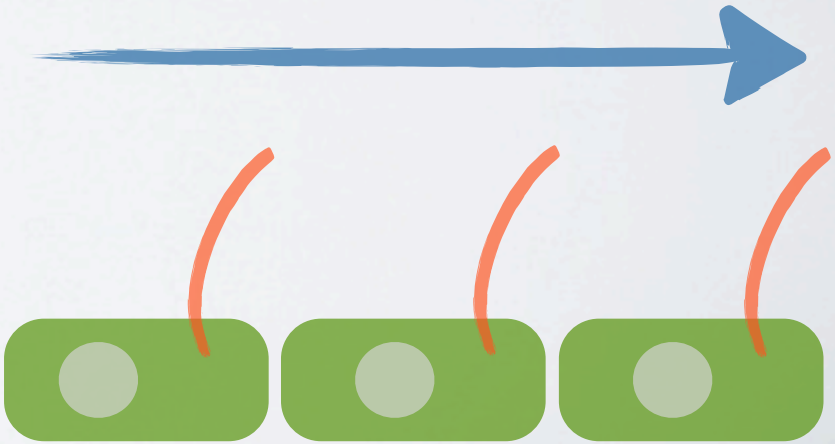


seconds

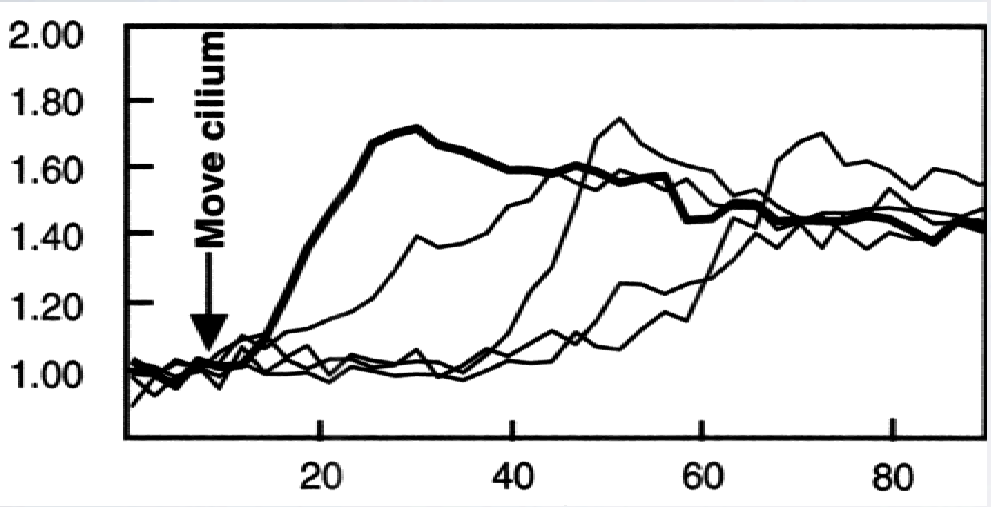


seconds

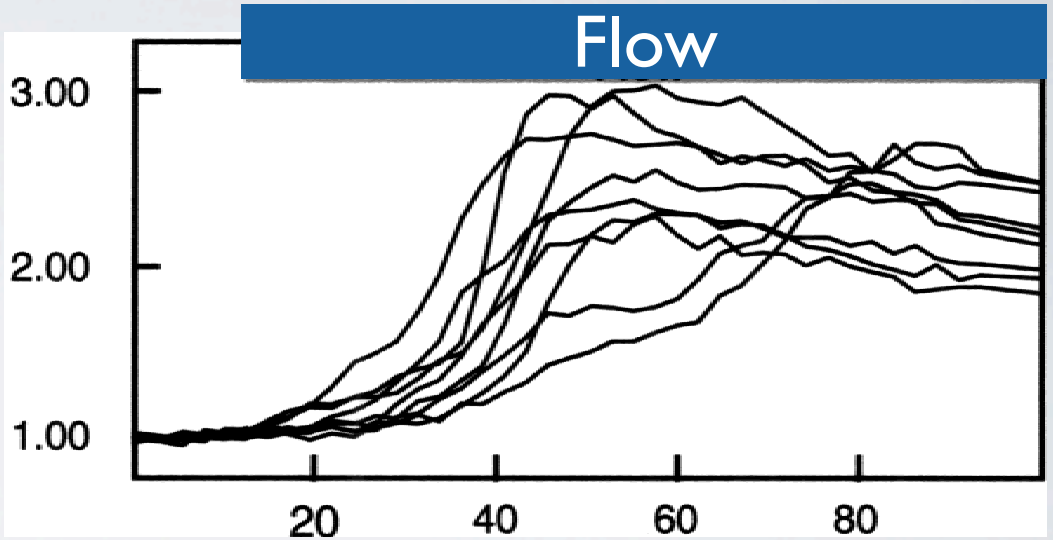




Controls show this response is not an artifact



seconds

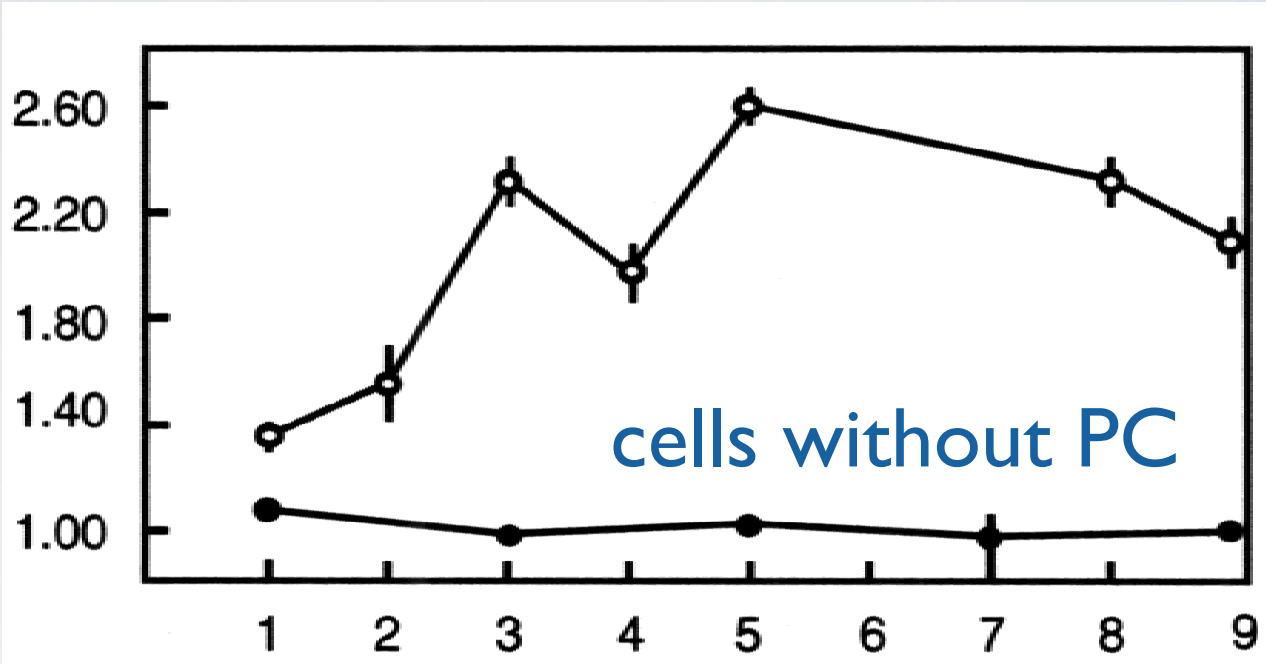


seconds

- Ionomycin serves as a positive control
- Control experiments used a pH-sensitive dye:
  - no change in cell thickness, volume, intracellular pH
- Bleaching or dye leakage would diminish fluorescence

Response depends on cilia and flow rate

relative fluorescence

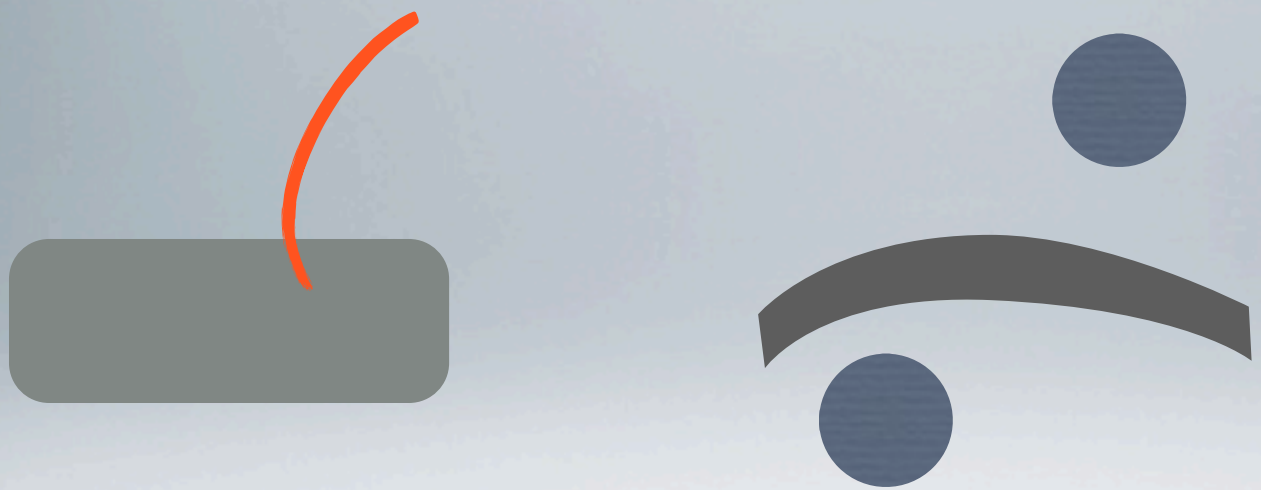


flow μL/sec

- Ionomycin serves as a positive control
- Control experiments used a pH-sensitive dye:
  - no change in cell thickness, volume, intracellular pH
- Bleaching or dye leakage would diminish fluorescence

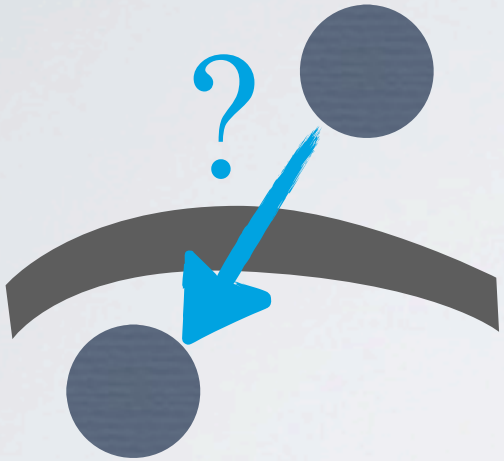
Bending cilia increases calcium inside the cell

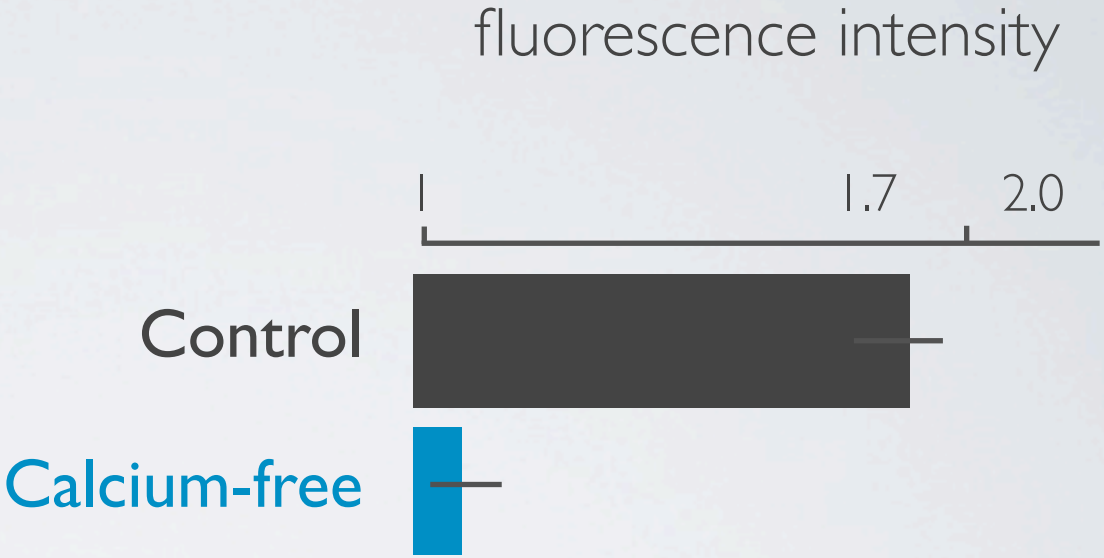
- Flow or suction increases calcium inside the cell
- Bending one cilium spreads a signal to neighbors
- This response requires a primary cilium
- This response is "dose-dependent"



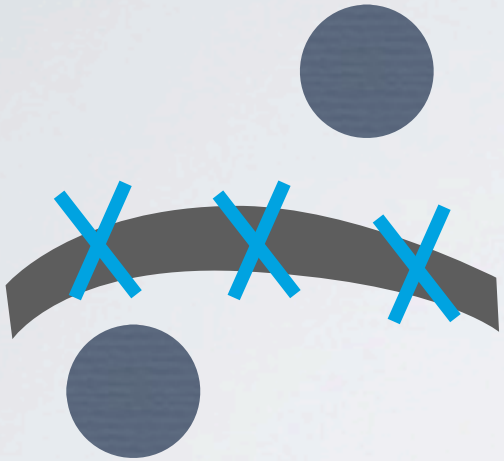
This calcium comes from two sources

Bending-induced calcium response depends on extracellular calcium

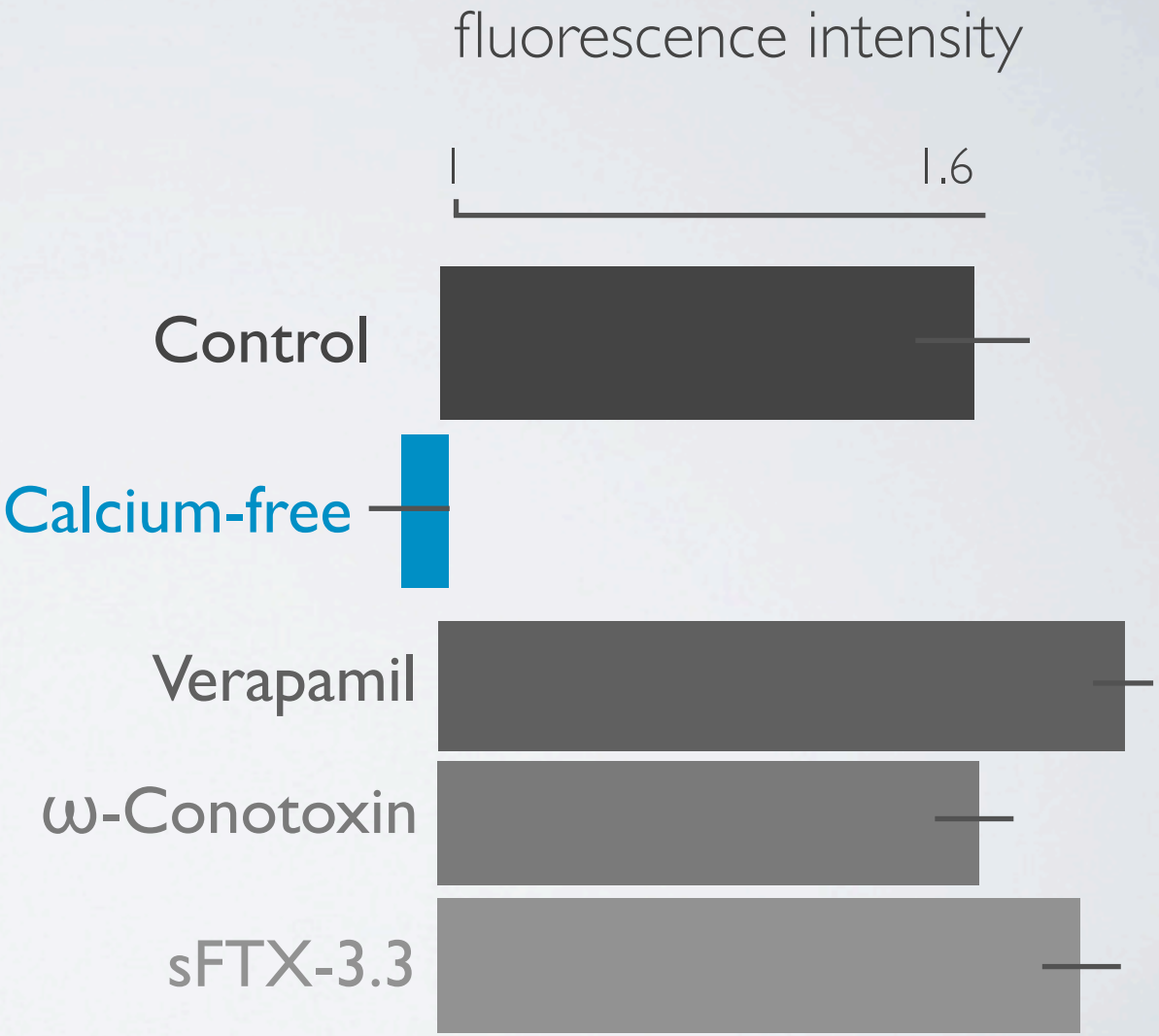




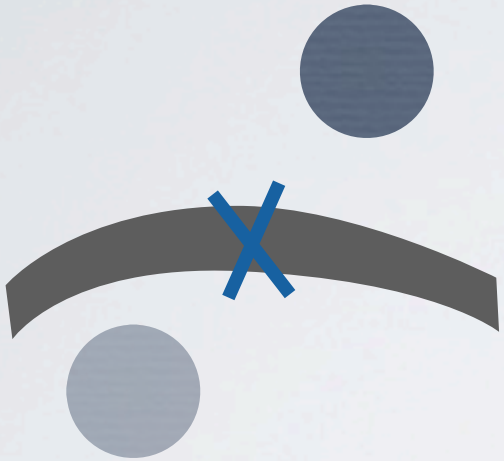
Bending response does not depend on calcium channels



Calcium channel blockers

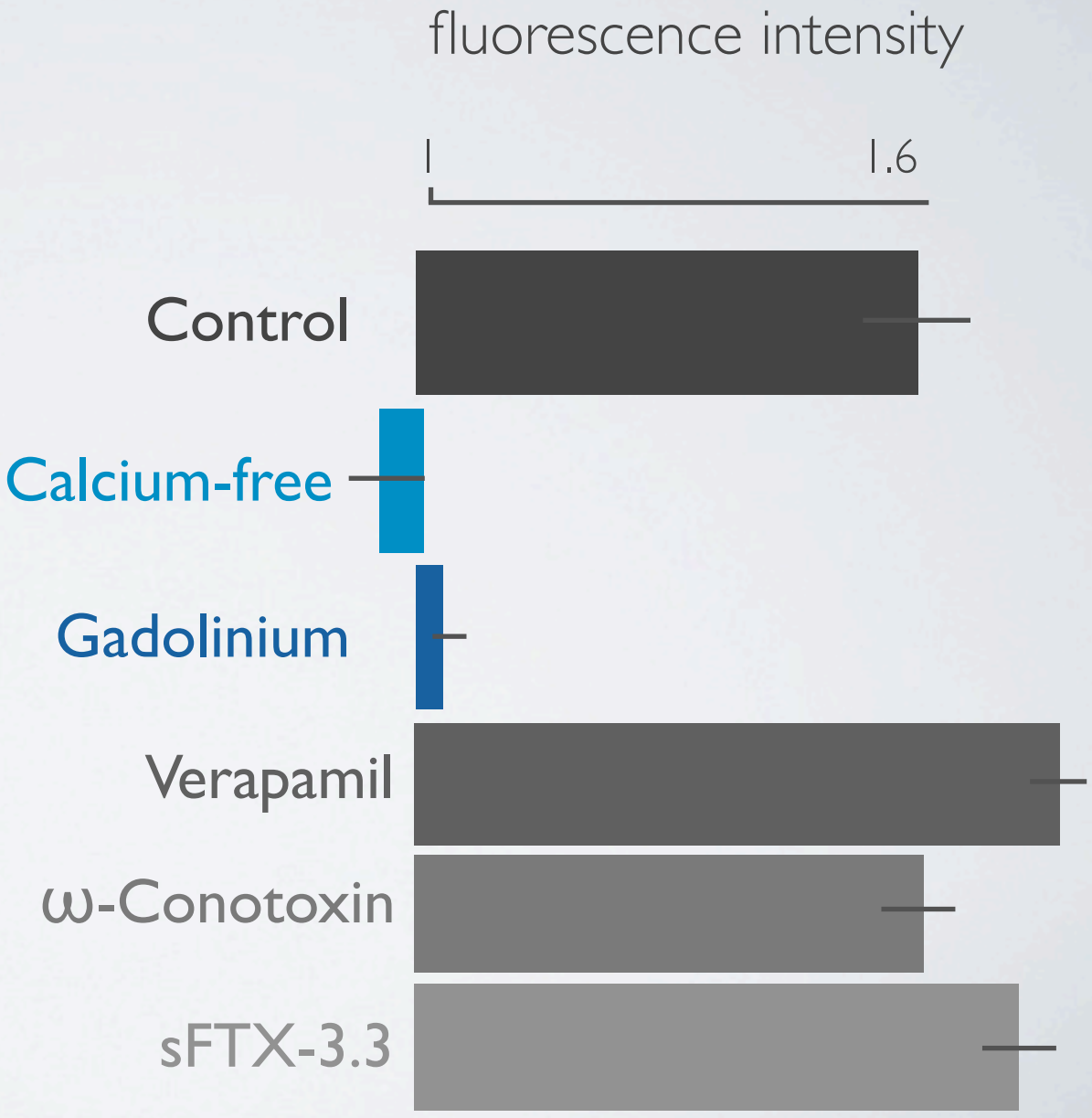


Bending response depends on stretch channels



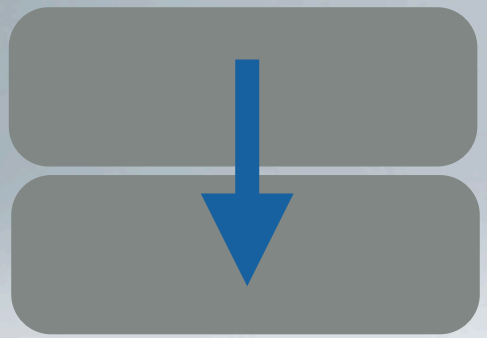
Stretch channel blocker

Calcium channel blockers



This calcium comes from two sources

- Cilia response depends on extracellular calcium
  - and stretch channels
- But calcium doesn't have to get in
- So it liberates intracellular calcium



This signal spreads by gap junctions

Critique

Renal cilia detect flow

Ardon Shorr