Magneto: remote control over neuronal activity and behaviour

Yvette Zarb
Division of Neurosurgery

Gene expression

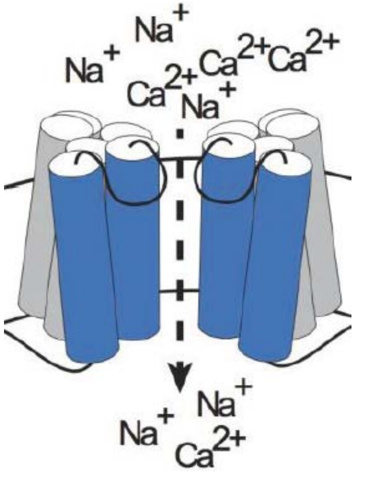
- Studying the actions specific cells and gene products is essential in understanding their physiological roles
- Regulation can be done through several approaches:
  - Chemogenetics (Tet-on system, Cre…)
  - Optogenetics (photoactivation of caged mRNA…)
- Drug kinetic-based systems are slow
- Light delivery has a limitied penetration and implanted fibres result in local activation

Nanoparticles

- Recent studies used extracellular nanoparticles (Stanley et al., 2012. *Science;* Huang et al., 2010. *Nature Nanothechnology)*
  - Controlled by radio wave activation
  - *in vitro* and *in vivo*

- Transient receptor potential cation channel (TRP) family
  - TRPV1,2,3: thermoreceptors
  - TRPV4: mechanoreceptors

- Limitations:
  - Invasive
  - Time limited, due to particle internalization
  - Acts locally



Adapted from: Wheeler et al., 2016. *Nature Neuroscience*

TECHNICAL REPORTS

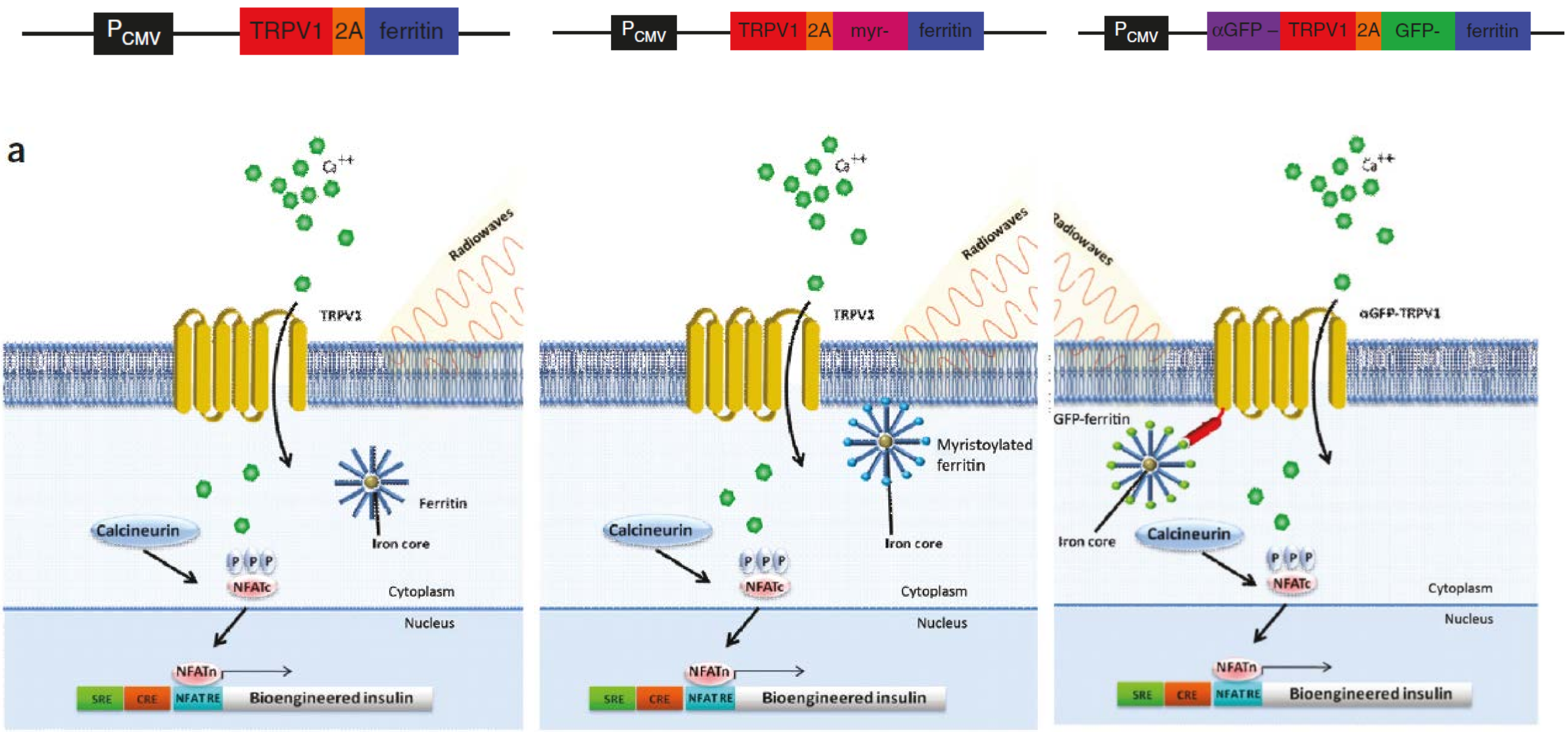
nature medicine

Remote regulation of glucose homeostasis in mice using genetically encoded nanoparticles

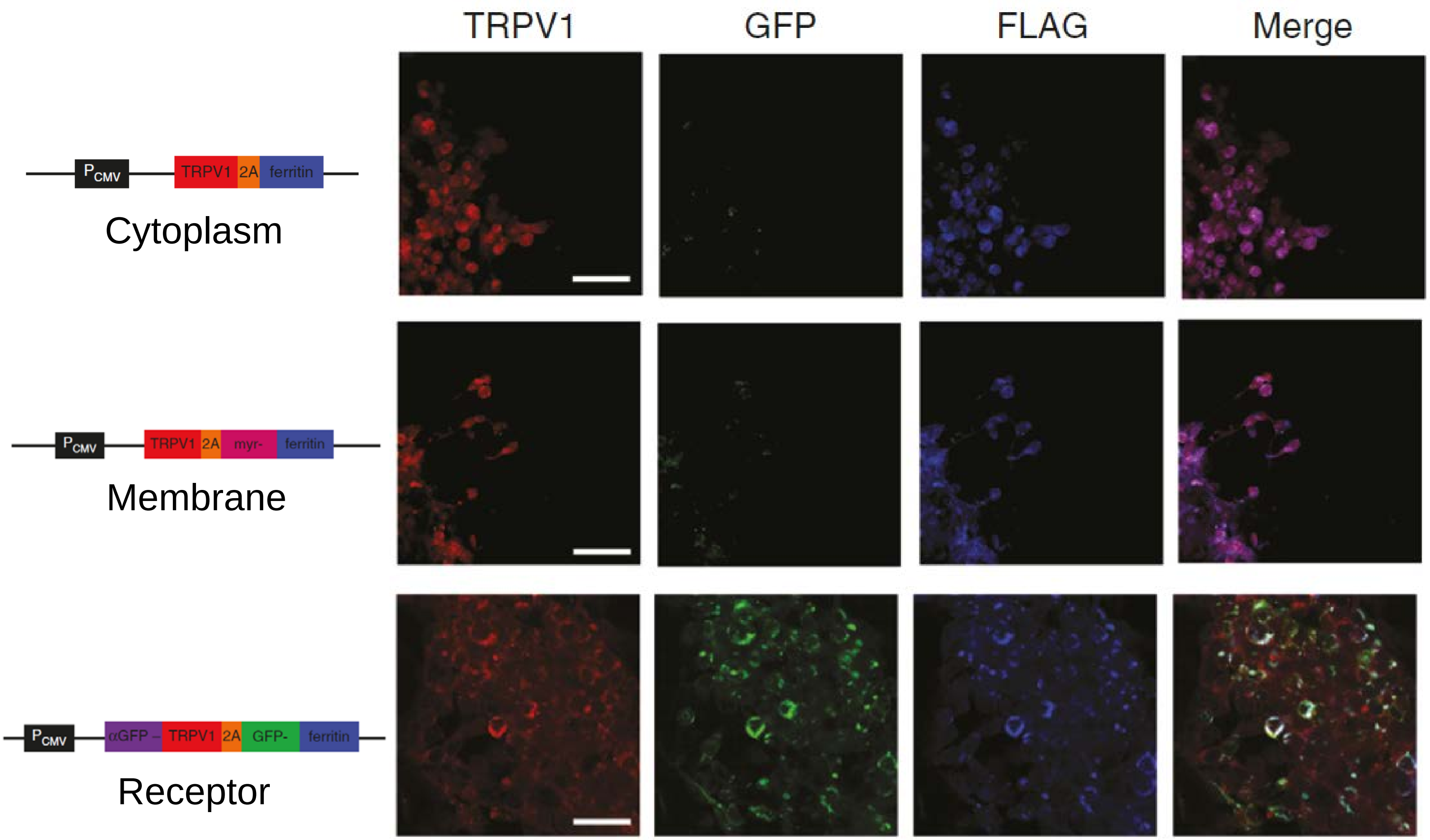
Sarah A Stanley[1,4], Jeremy Sauer[2,4], Ravi S Kane[2], Jonathan S Dordick[2] & Jeffrey M Friedman[1,3]

Constructs used *In vitro*

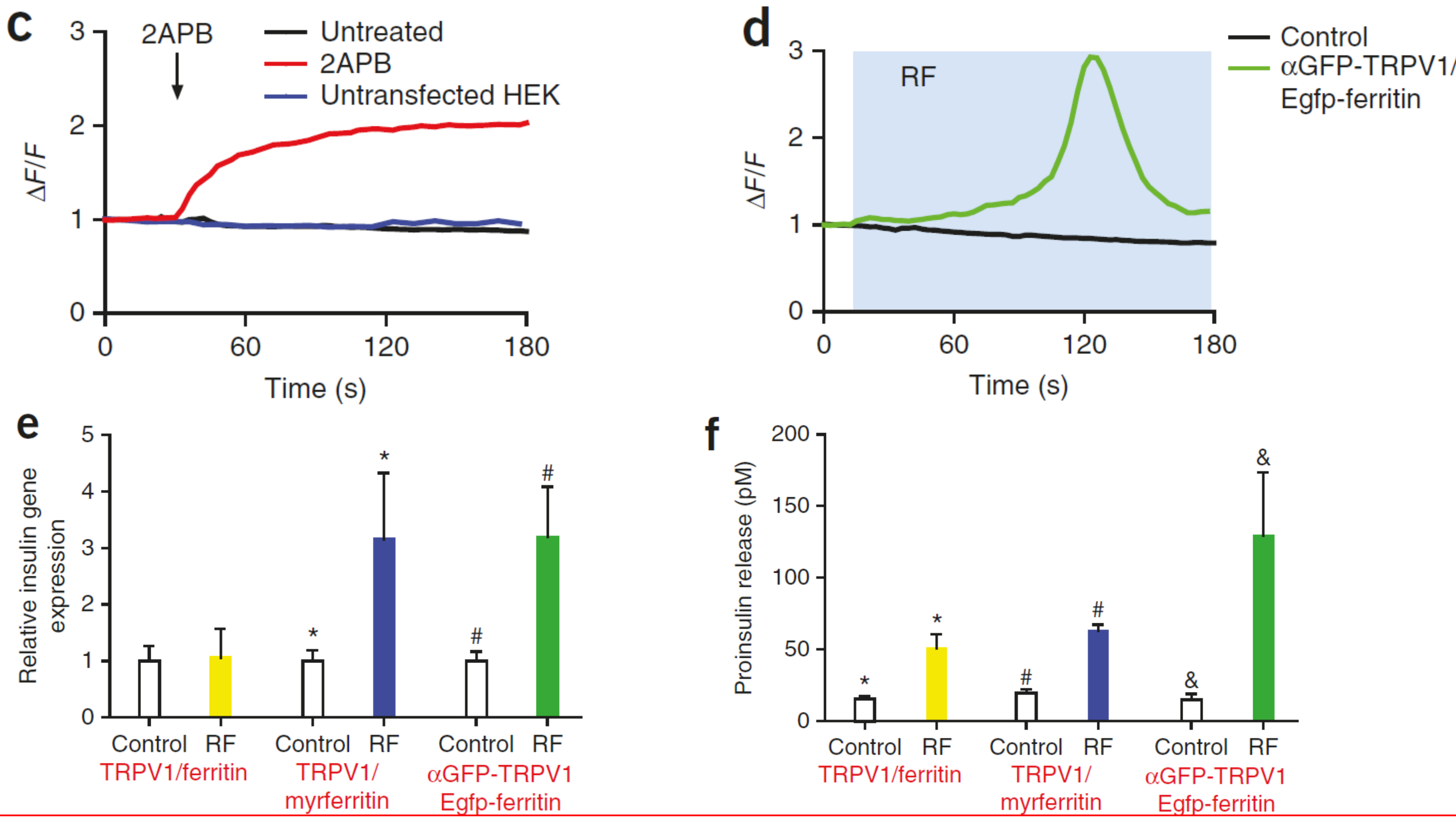
Construct 1 Construct 2 Construct 3



Confirmation of predicted location of the constructs

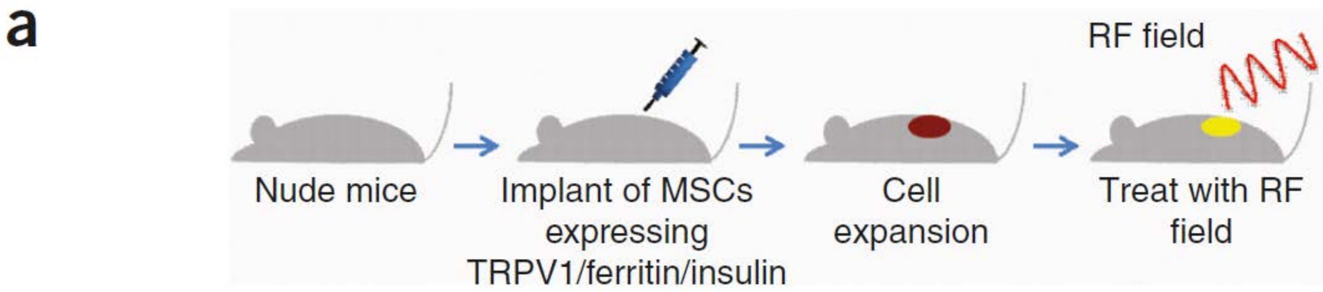


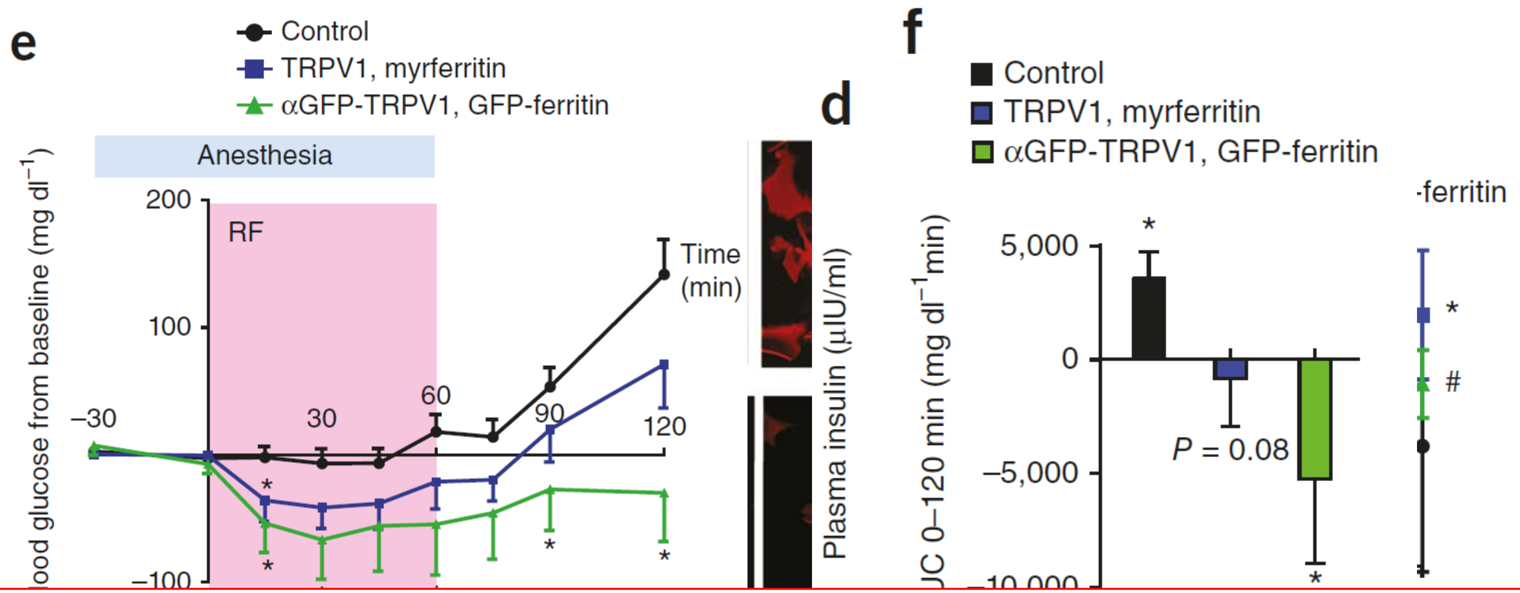
Efficiency in transducing an RF signal to gene expression



Direct tethering of ferritin to TRPV1 is able to transduce the RF signal more efficiently

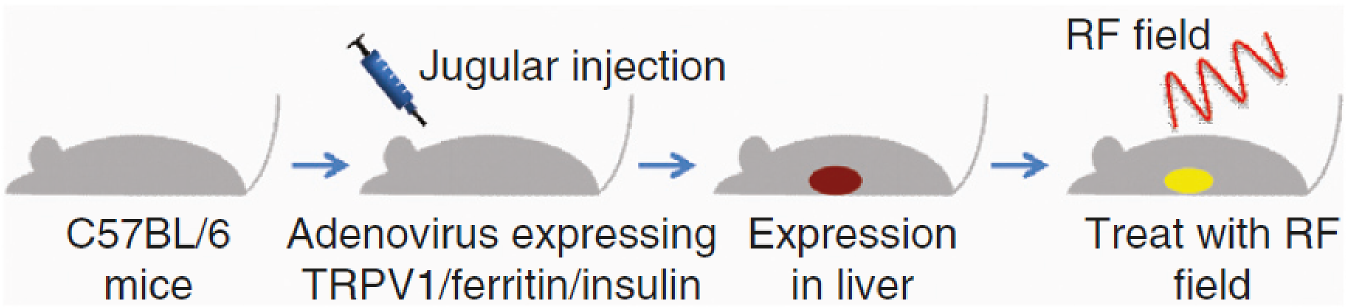
Construct testing *in vivo*

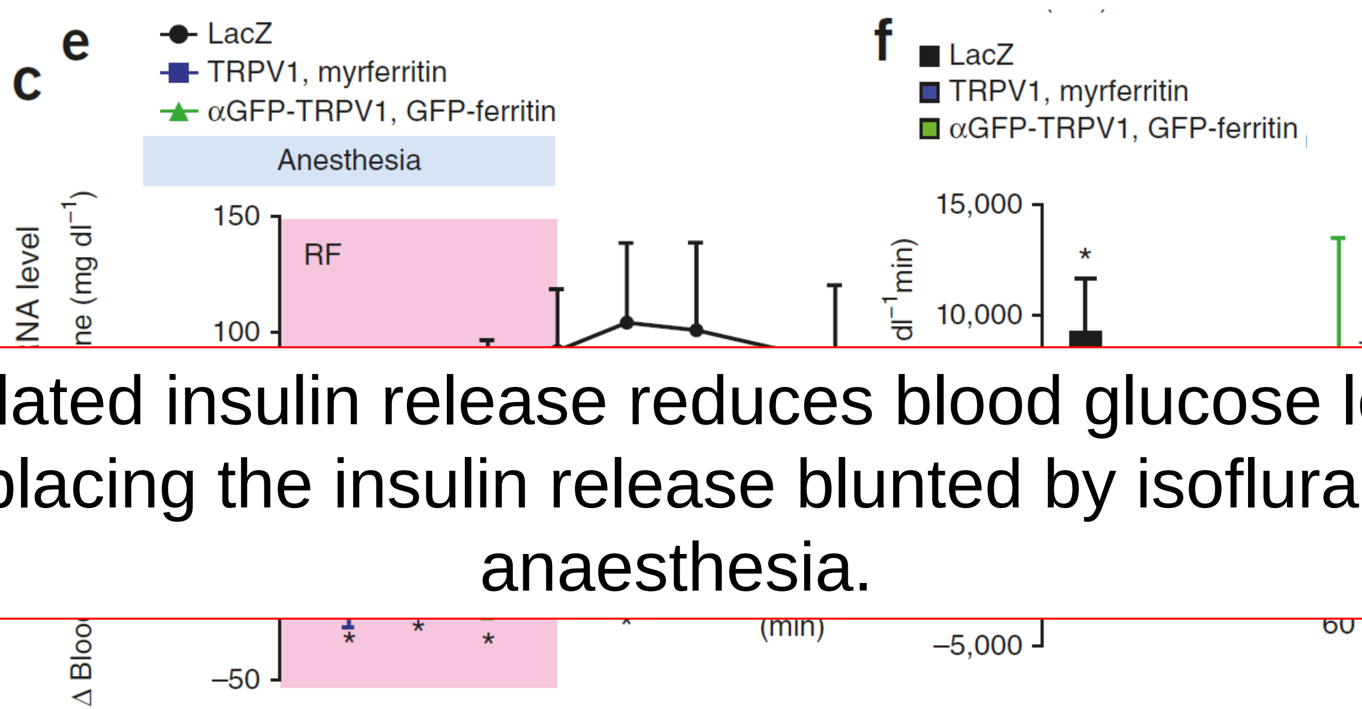




This system regulates gene expression and protein release *in vivo*.

Replicating results *in vivo* using adenoviruses expressing contructs





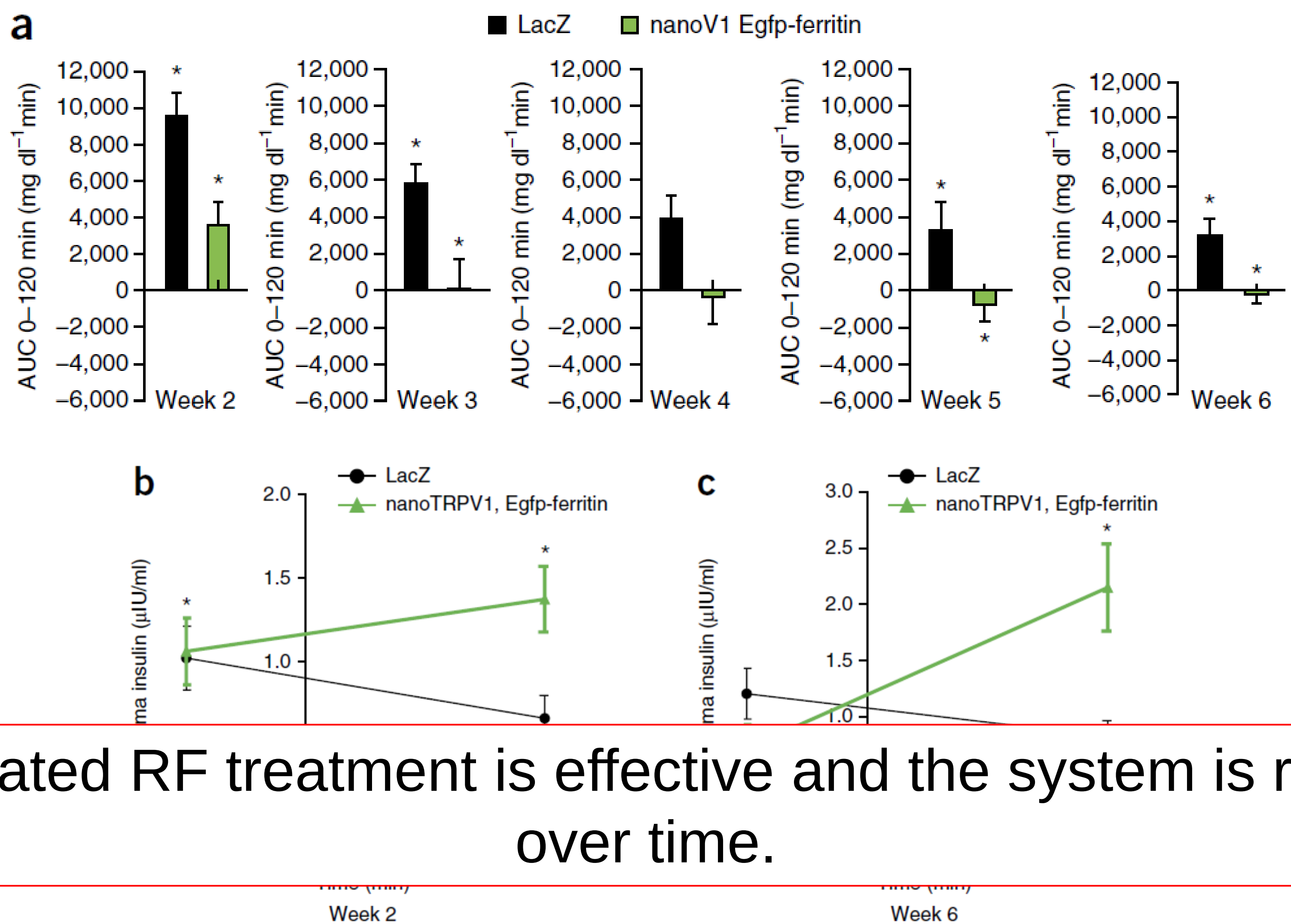
RF-regulated insulin release reduces blood glucose levels by replacing the insulin release blunted by isoflurane anaesthesia.

Repeated RF treatment to regulate protein delivery

- Ensure effectiveness of TRPV1 and genetically encoded nanoparticles over time

- Treated with 1 hour of RF once a week during weeks 2-6 after injection

Repeated RF treatment to regulate protein delivery



Repeated RF treatment is effective and the system is robust over time.

Remote activation using a magnetic field

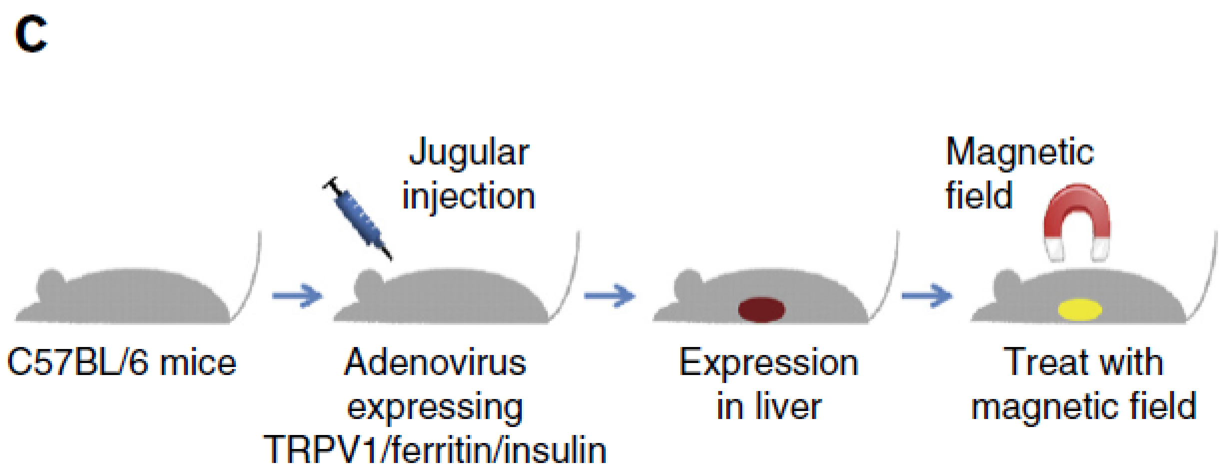
- RF treatment could gate TRPV1 by:
  - Particle heating
  - Mechanical stimulus

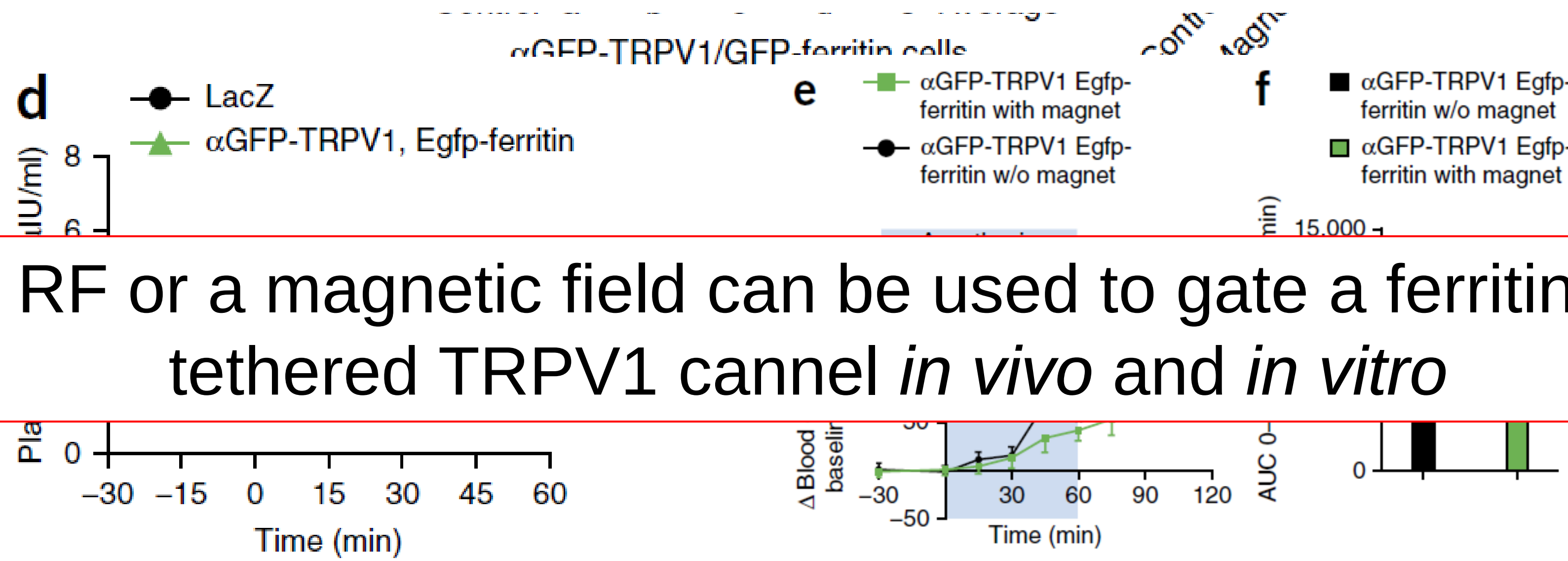
- TRPV family members have been implicated in mechanosensing

- Tethered ferritin nanoparticles have superparamagnetic properties

- Could an external magnetic field be converted into a mechanical force as adjacent particles align with the field?

Remote activation using a magnetic field





RF or a magnetic field can be used to gate a ferritin-tethered TRPV1 cannel *in vivo* and *in vitro*

Conclusion

- Genetically encoded ferritin-tethered TRPV1 system enables:
  - Remote, robust and repeated temporal control of gene expression *in vivo*
- RF or magnetic fields can be used
- Adenovirus-mediated transgene expression is limited to several weeks
  - More durable vectors
  - Development of transgenic mice
- Regulating gene expression without chemical toxicity
- TRPV1 gates Ca^{2+}, thus can be used to regulate neuron activity

nature neuroscience

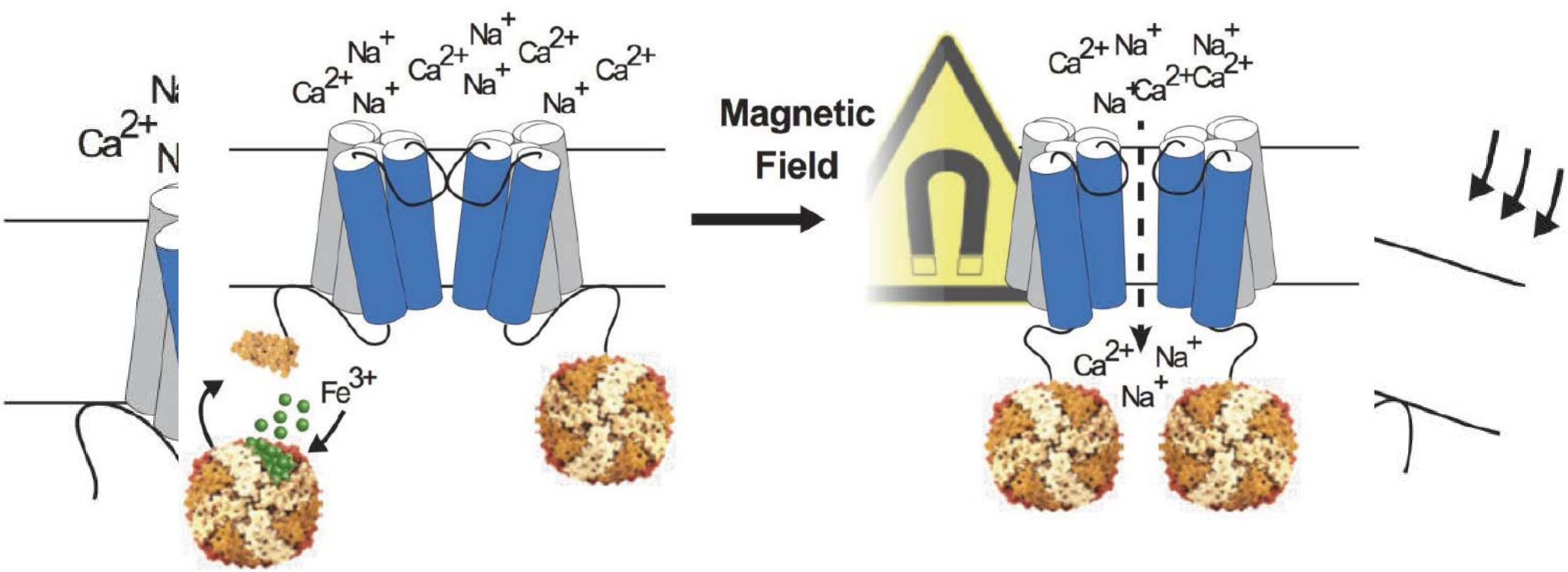
Genetically targeted magnetic control of the nervous system

Michael A Wheeler[1,2], Cody J Smith[1,7], Matteo Ottolini[3,7], Bryan S Barker[2,3], Aarti M Purohit[1], Ryan M Grippo[1], Ronald P Gaykema[3], Anthony J Spano[1], Mark P Beenhakker[4], Sarah Kucenas[1,5], Manoj K Patel[3], Christopher D Deppmann[1,5,6] & Ali D Güler[1]

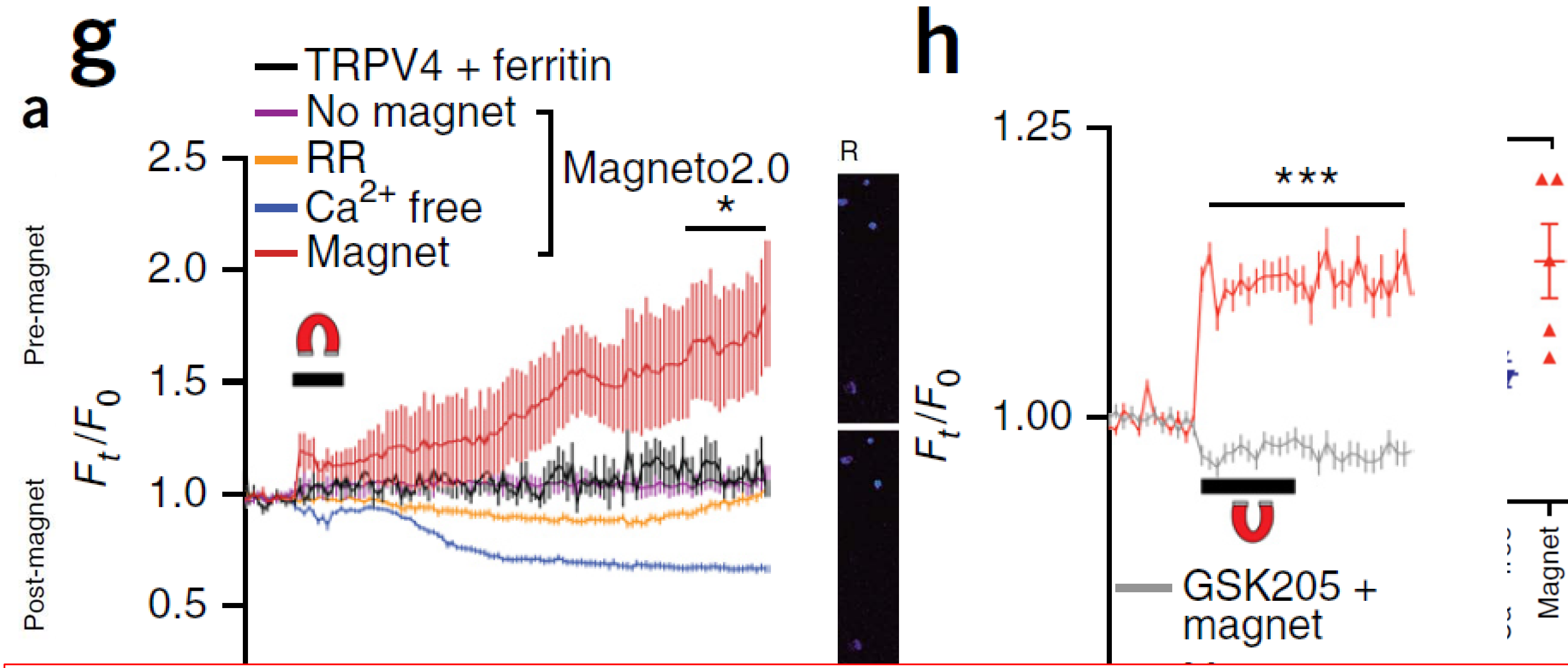
Design of a magnetically sensitive cation channel

- Single-component magnetogenetic actuator

- TRPV4: responds to pressure



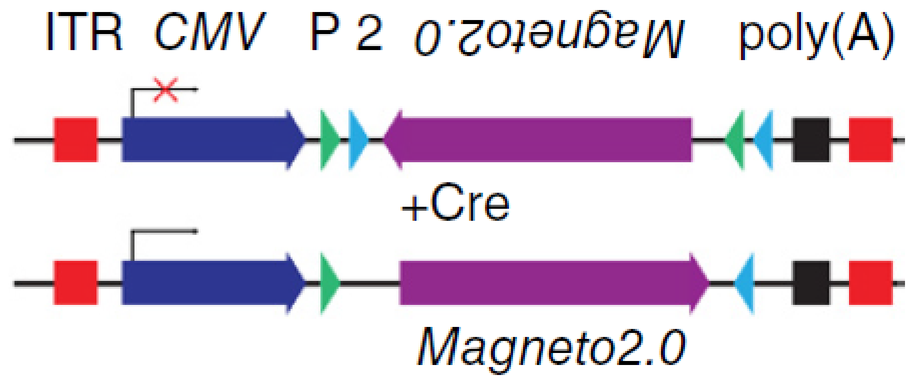
Remote control of calcium signalling using Magneto 2.0 *in vitro*

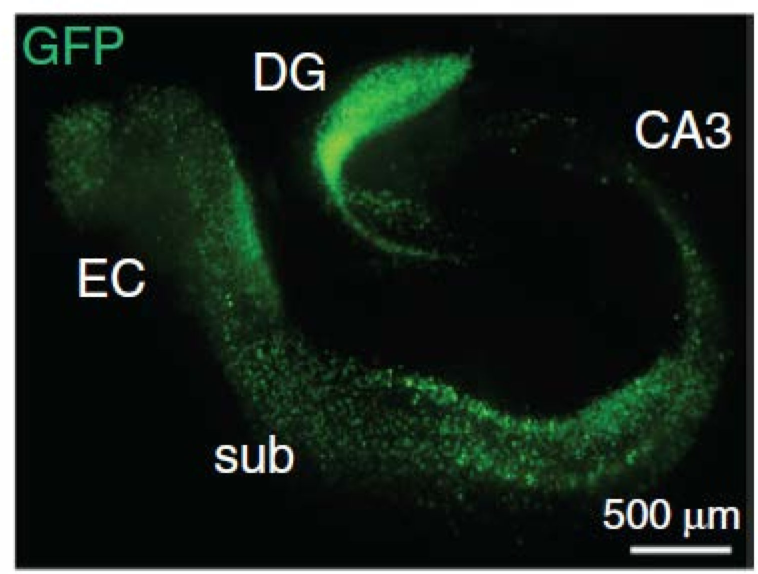


Magneto2.0 is a magnetically sensitive, genetically encoded actuator that can manipulate cellular activity *in vitro*

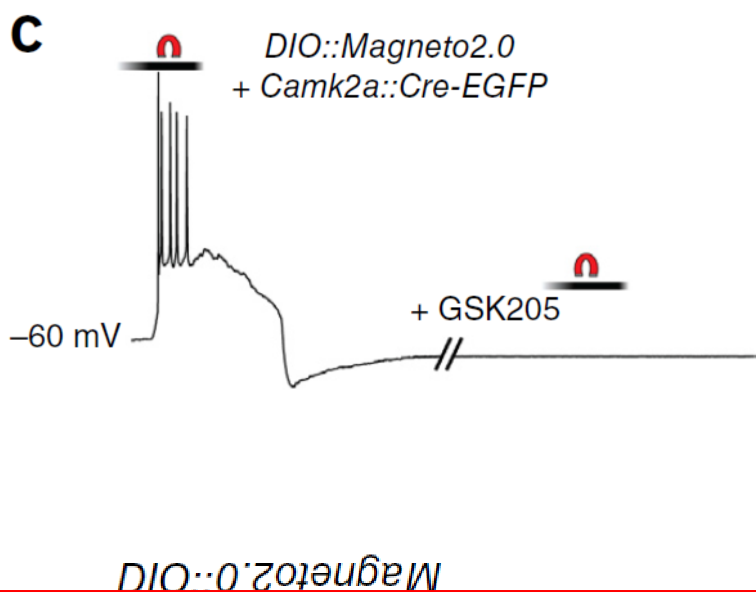
Magneto 2.0 in mouse brain slices

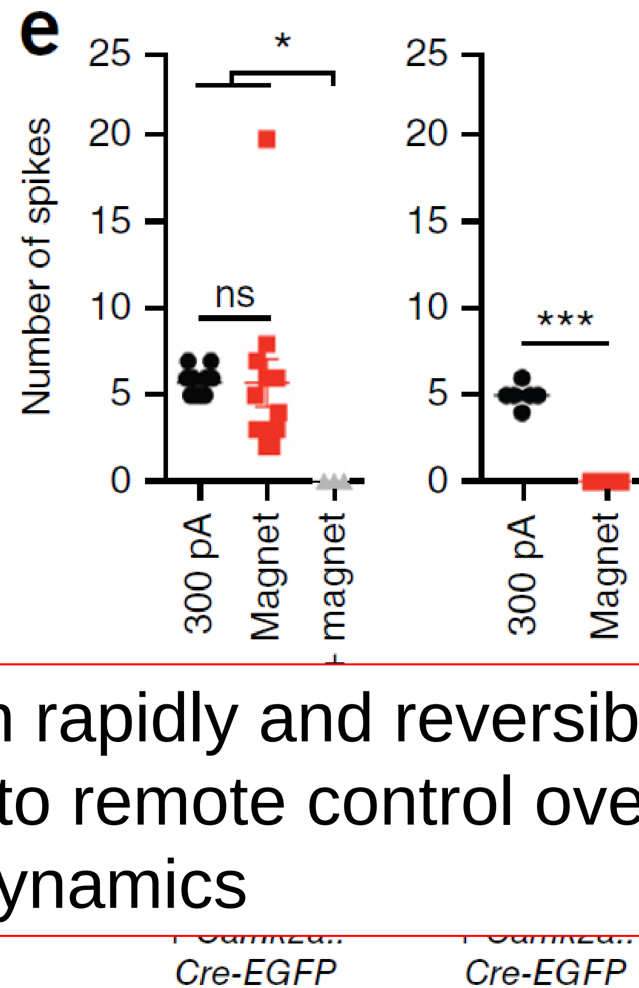
- To determine temporal kinetics
- Does it have a rapid activation in response to magnetic fields in live tissue?





Electrophysiological characterization in mouse brain slices





Activation of Magneto 2.0 can rapidly and reversibly depolarize neurons, leading to remote control over neural circuit dynamics

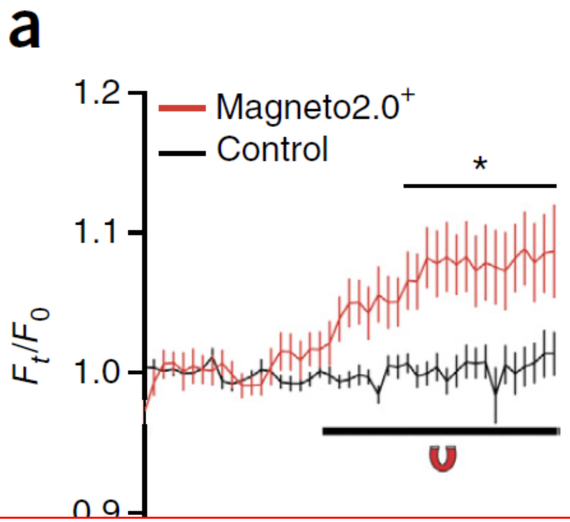
Magnetic control over zebrafish tactile behaviours

- Validation of Magneto2.0 *in vivo*

- Remotely modulate simple behaviour of zebrafish

- Transient expression of of Magneto2.0 in Rohon-Beard sensory neurons
  - Regulatory sequences of the *neurog1* promoter

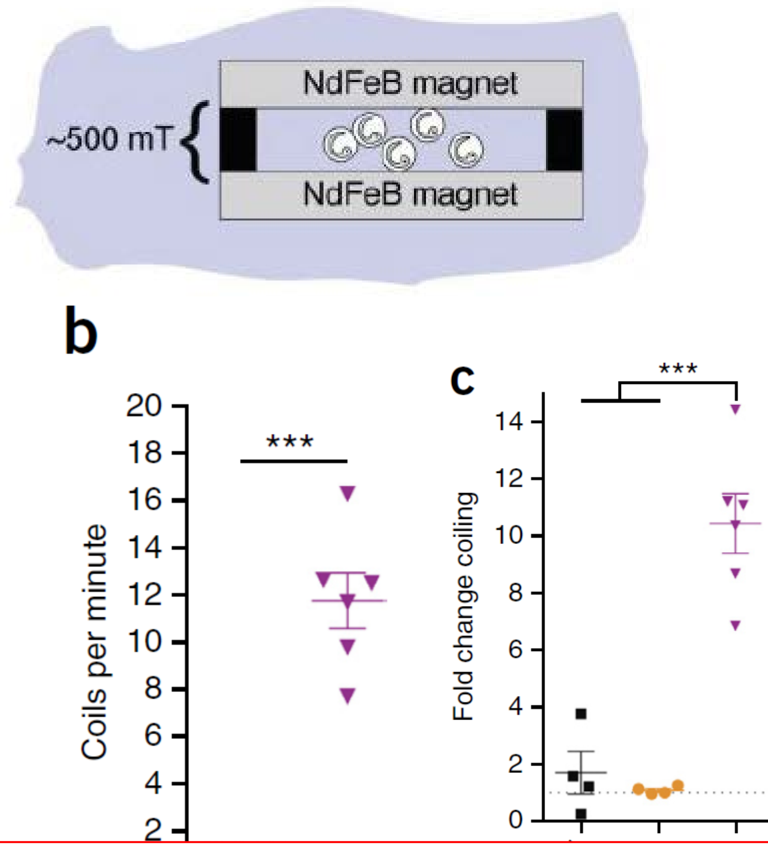
- Detection of activated neurons through the genetically encoded



Magnetic stimulation reliably activates Magneto2.0$^+$ neurons *in vivo*

Magnetic control over zebrafish tactile behaviours

- Is this sufficient to modulate behaviour in zebrafish?



- Hypothesis: if Rohon-Beard neurons are activated, coiling behaviour is induced

Magneto2.0 is a viable candidate for remotely controlling neuronal activity and animal behaviour *in vivo*

Remote control of neural activity in freely behaving mice

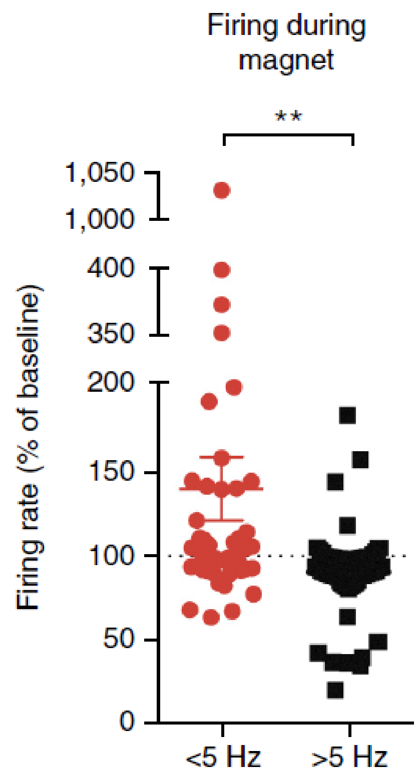
- Transduction with AAV1, expressing Magneto2.0 in a Cre dependent manner

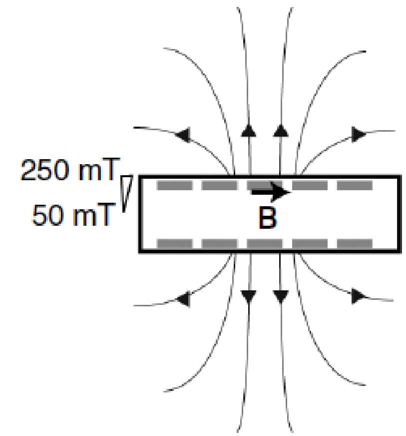
- To test the activation of a nucleus in the striatum
- Extracellular single-unit recordings electrode using a tetrode microdrive
- Examine the neural firing in freely behaving mice under the effect of magnetic stimulation

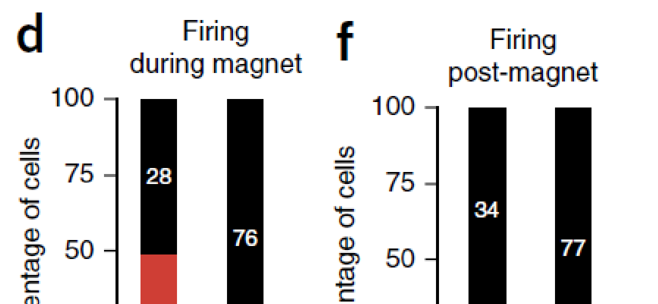
http://users.atw.hu/braintelemeter/Pati 7%20copy.jpg

Remote control of neural activity in freely behaving mice

- Approx. 50% of medium spiny neurons (MSNs) express DR1
- Recorded cells:
  - Slow spiking (<5Hz) – MSN
  - Fast spiking (>5Hz) – GABAergic neurons



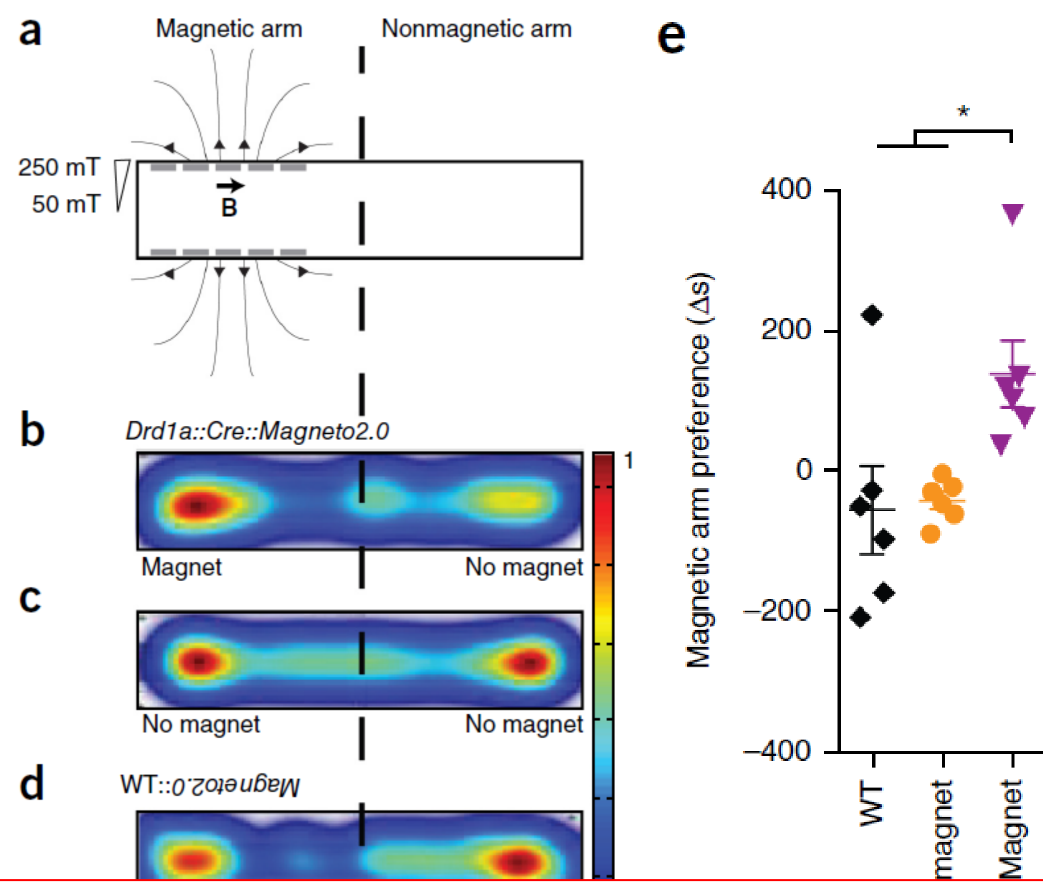




Magneto2.0 is capable of controlling neural firing in deep brain regions in response to magnetic fields.

Control of D1R-mediated striatal reward valence

- Does Magneto2.0 control of neural activity translate to control over behaviours?
- Regulation of dopamine signalling (reward behaviour)
- Real-time place preference (RTPP) assay



Magneto2.0 can be used to control complex mammalian behaviours in freely moving mice

Conclusion

- Demonstrates magnetic control of the nervous system using genetically encoded magnetic actuators
- Limitation:
  - Since the design is based on TRPV4, this system is sensitive to endogenous stimuli that activate TRPV4
- With further optimization of this method will aide to better understand neural development, function and pathology.

Thanks