C. *elegant* Neurobiology Techniques
History of *C. elegans*

- 1963 – Introduced and used by Sydney Brenner
- 1974 – First mutants published by Brenner
- 1982 – “Genetic control of programmed cell death.” Horvitz *et. al.*
- 1994 – First use of GFP in animals. Chalfie *et. al.*
- 1998-1999 – *C. elegans* is the first animal to have its genome sequenced. *C. elegans* Sequencing Consortium
The Worm as a Model for Neurobiology

- Powerful genetic model
  - Short life span
  - Hermaphrodite
  - Day 5 adults experience neurodegeneration
- Simple nervous system – 302 neurons
  - Complete connectome
- In vivo modeling
  - Transparent
Outline

• *In vivo* Imaging Techniques
  – Single neuron imaging
  – Microfluidics

• Behavior
  – Mechanosensation assay
  – Sensory memory assay

• Behavior to Molecule

• Genetic Screens

“If you had the wiring diagram, could you compute behavior?” – Sydney Brenner
In vivo Imaging
Visualizing Cell Activity \textit{in vivo}

Dawitz et. al., 2011

Doser et. al., unpublished
Optogenetics

TULIPS

A
Plasma membrane
PH domain
GFP
LOVpep
ePDZ
mKate2

Blue light

D
Mitochondrion
hsPEX3
mKate2
LOVpep
ePDZ
GFP
Kinesin

Harterink et. al., 2016
Chromophore-Assisted Light Inactivation

CALI
Chromophore-Assisted Light Inactivation

\[ h\nu \]

\[ ^1\text{O}_2 \]

\[ \text{miniSOG} \]

\[ ^1\text{O}_2 \]

\[ \text{CaMKII} \]

\[ \text{CALI} \]

C

\[ h\nu \rightarrow \text{CALI} \]

\[ \text{CALI} \]

\[ \text{knmo} \]

34 s

(1)

\[ \text{kymo} \]

34 s

(1')

\[ \text{CALI} \]

\[ \text{Anterograde} \]

\[ \text{Retrograde} \]

Transport events (normalized)

\[ {\text{Before}} \]

\[ {\text{After}} \]

***

Hoerndli et. al., 2015
Microfluidics to Increase Throughput

H. Lu – Georgia Tech

Gabel, 2008

Guo et al., 2008

Chung et al., 2008

Guo et al., 2008

Chung et al., 2008
Behavior

• **Mechanosensation assay**
  – Touch assays

• **Sensory**
  – Drop assay
  – Chemotaxis
Behavior to Study Memory

- Diacetyl
- Ethanol
- Naive
- Conditioned
- 1Hr Starvation
- Worms select preferred odor

**WT**

- PTP-3
- ΔPTP-3A
- ΔPPase

Pierce et. al., unpublished
From Behavior to Molecule

Diacetyl

Ethanol

Naive

1 Hr Starvation

Worms select preferred odor

Conditioned
From Behavior to Molecule

Naive

Conditioned

Diacetyl
Ethanol

1Hr Starvation
Worms select preferred odor

Worms select preferred odor

From Behavior to Molecule

Naive

Conditioned

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Worms select preferred odor

Worms select preferred odor

From Behavior to Molecule

Naive

Conditioned

Diacetyl
Ethanol

1Hr Starvation
Worms select preferred odor

Worms select preferred odor
Forward Genetic Screens

Forward Genetic Screen in *Caenorhabditis elegans* Suggests F57A10.2 and acp-4 As Suppressors of C9ORF72 Related Phenotypes

Xin Wang1,2*, Limin Hao2, Taixiang Saur2, Katelyn Joyal2, Ying Zhao3, Desheng Zhai2, Jie Li4, Mohitar Pribadi5, Giovanni Coppola6, Bruce M. Cohen7 and Edgar A. Buttner2

1Virginia Tech, 2The Chinese University of Hong Kong, 3National Yang Ming University, 4National Taiwan University, 5National Tsing Hua University, 6University of Southern California, 7University of Texas, San Antonio

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Forward Genetic Screens

A

EMS Mutagenesis

Raise F2

F2 Adult Day 1

Screen #1

15 min.

Discard developmental motor deficit mutants

F2 Adult Day 3 (and Day 5)

Screen #2

60 min.

Keep adult-onset motor deficit mutants

Kawamura et. al., 2019

Hadziselimovic et. al., 2014
Does ROS Signaling Modulate GLR-1 Transport?
How do Phosphatases Regulate Synaptic Plasticity?
References


• https://www.wormatlas.org/
• https://www.wormbase.org/
• http://www.wormbook.org/
• https://www.hoerndlilab.org/
If a neuron wore pants, would it wear them
Like this

like this?

or like this?

THANK YOU!!!