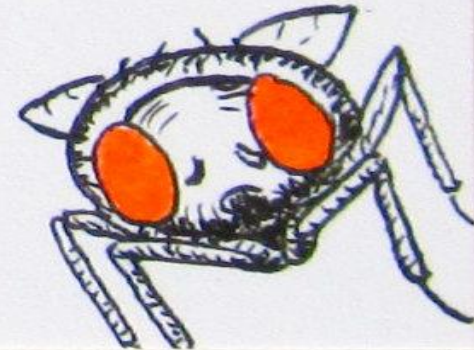
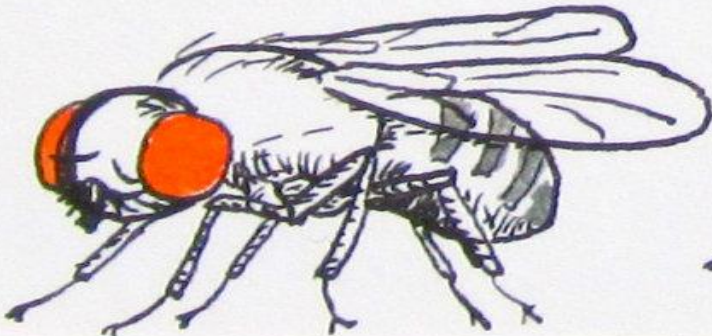
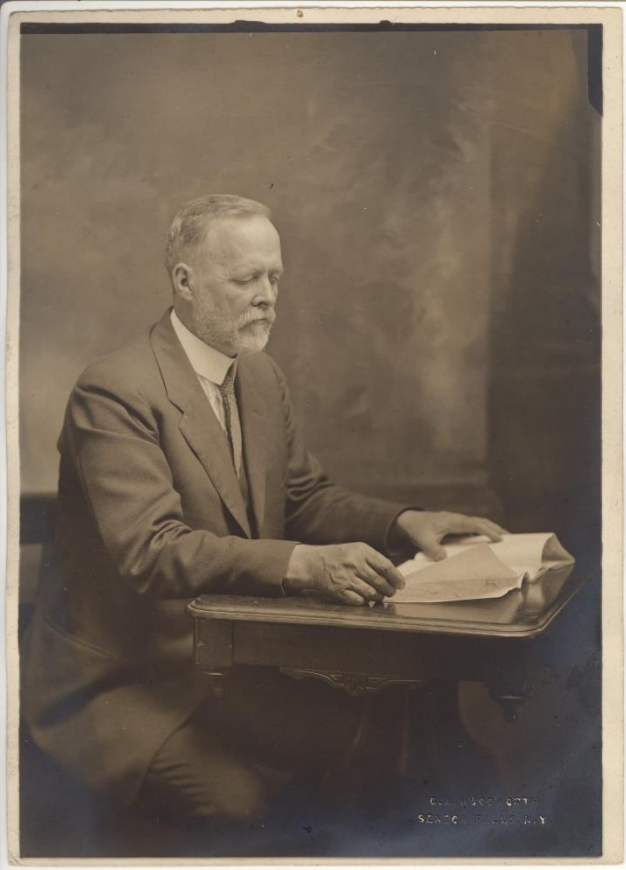


Behavioral Neurobiology of *Drosophila*

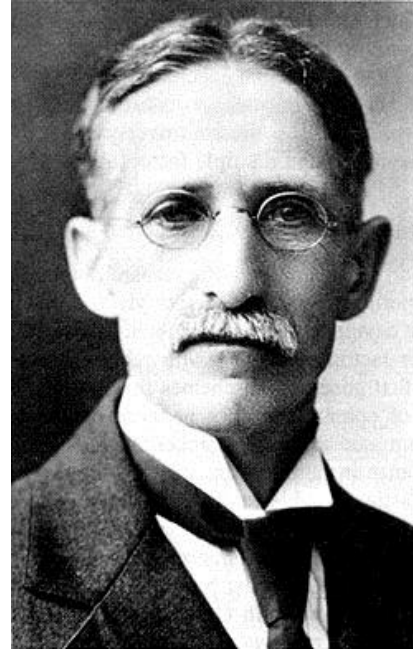


Dr. E. Axel Gorostiza
Brno, 17.03.2015

A little bit of history.



Charles W. Woodworth (USA)
Entomologist
(April 28, 1865 – November 19, 1940)



William Ernest Castle (USA)
Embryologist and geneticist
(October 25, 1867 – June 3, 1962)

Thomas Hunt Morgan (USA)
evolutionary biologist, geneticist and embryologist
(September 25, 1866 – December 4, 1945)



The Nobel Prize in Physiology or Medicine 1933 was awarded to Thomas H. Morgan
"for his discoveries concerning the role played by the chromosome in heredity".

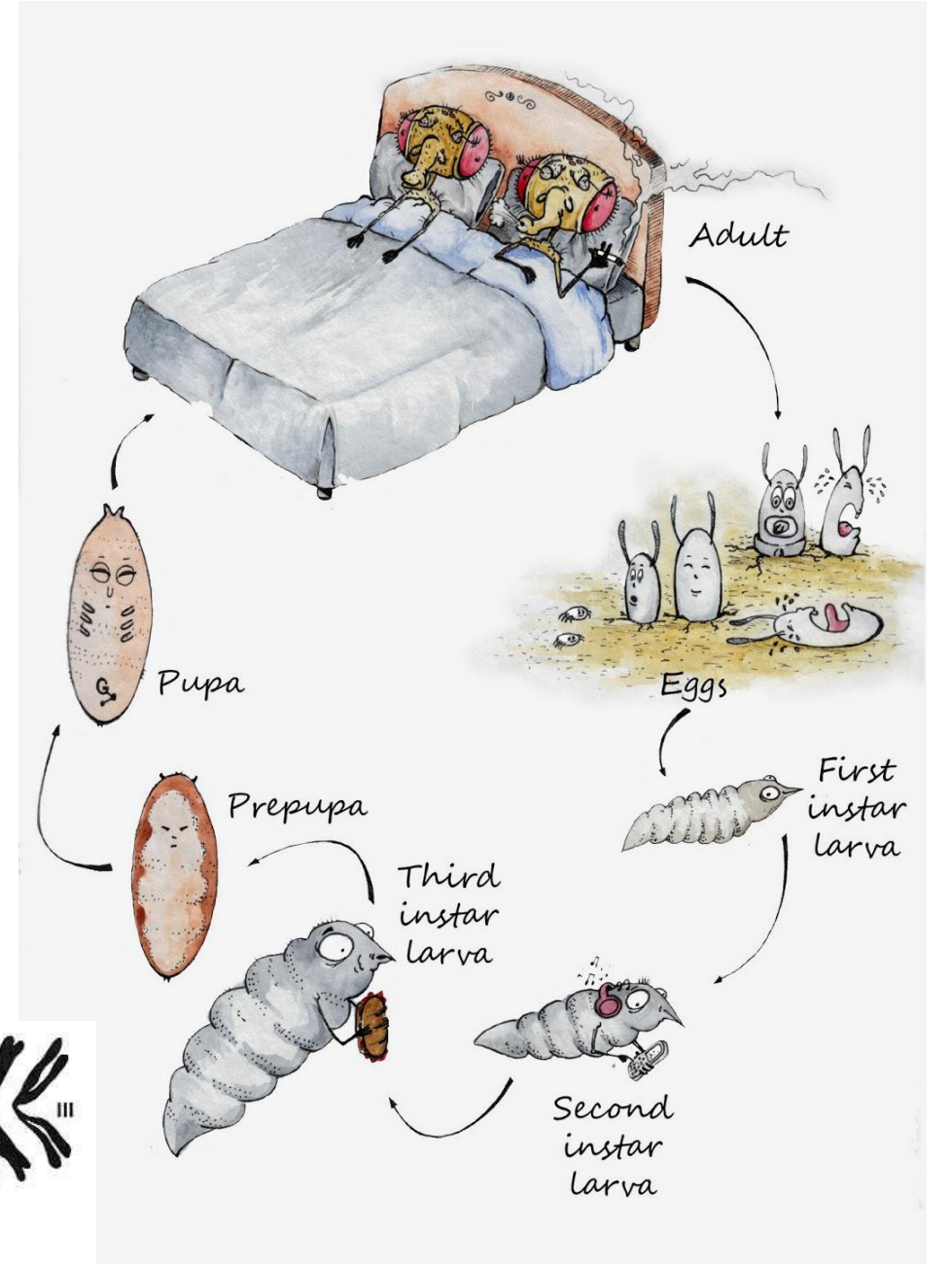
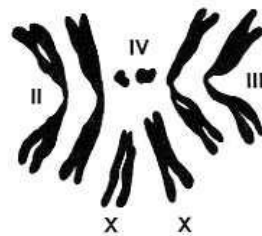
Why *Drosophila*?



<http://www.theflyroom.com/>

Fly Food Ingredients

Ingredients	Amounts (ml or g)
Water	1000 ml
Yeast (dry)	20 g
Agar	10 g
Sucrose	40 g
Corn Flour	65 g
Propionic acid	4.4 ml
10 % Nipagin in 95% EtOH	14 ml



The life cycle
Drosophila melanogaster

<http://drosophiladrawings.blogspot.de/>

But, what about behavior?

THE REACTIONS OF THE POMACE FLY (*DROSOPHILA AMPELOPHILA* LOEW) TO LIGHT, GRAVITY, AND MECHANICAL STIMULATION.

FREDERIC W. CARPENTER.

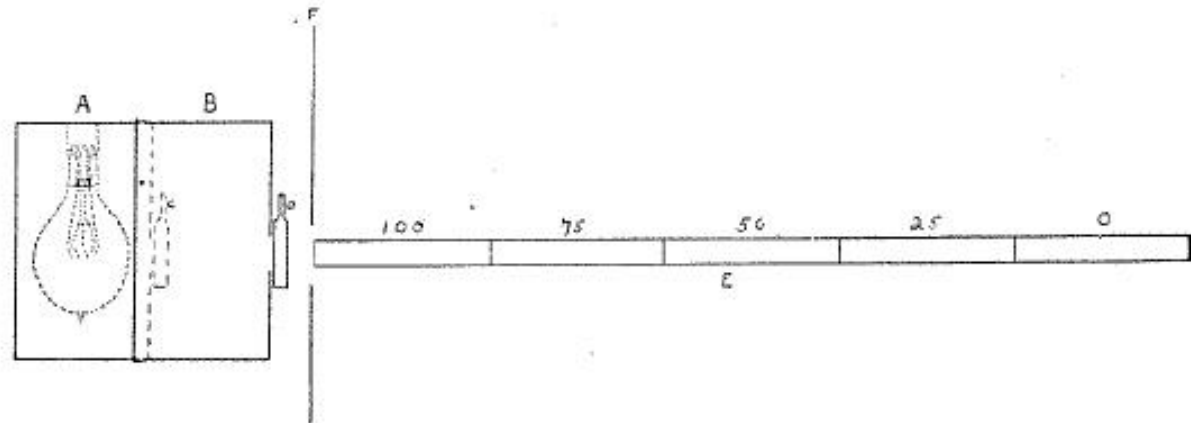
The American Naturalist, Mar., 1905, pp. 157-171

THE REACTIONS TO LIGHT AND TO GRAVITY IN *DROSOPHILA* AND ITS MUTANTS

ROBERT STANLEY McEWEN

Columbia University, New York City

J. Exptl. Zool., 1918, 49-106.



The father of Neurogenetics



Seymour Benzer

Physicist, molecular biologist and behavioral geneticist
(October 15, 1921 – November 30, 2007)

OPEN ACCESS Freely available online

PLoS BIOLOGY

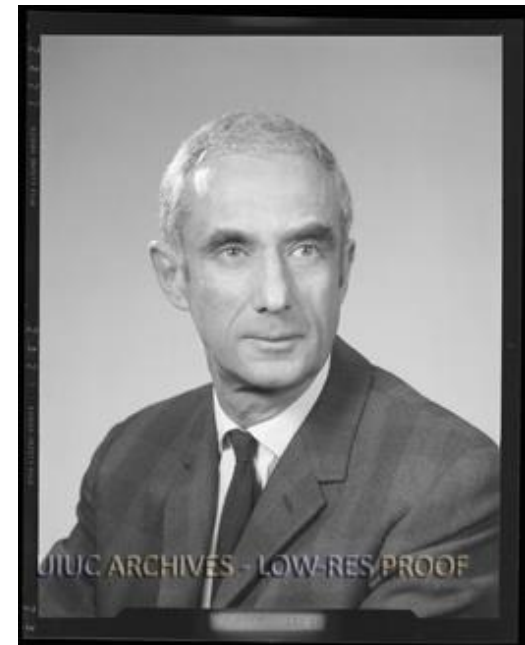
Obituary

Seymour Benzer 1921–2007

The Man Who Took Us from Genes to Behaviour

William A. Harris

Jerry Hirsch
Behavioral geneticist
(September 20, 1922 – May 3, 2008)

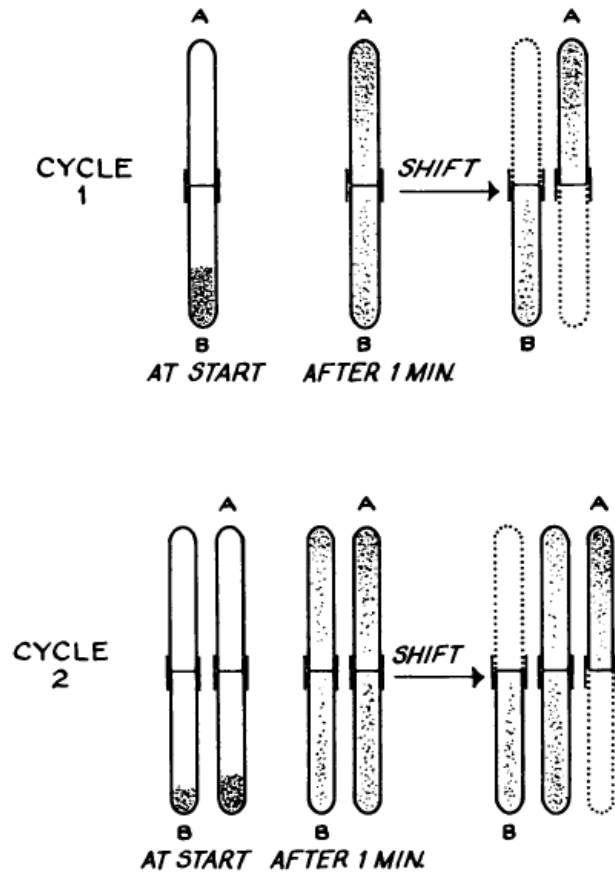


BEHAVIORAL MUTANTS OF DROSOPHILA ISOLATED BY COUNTERCURRENT DISTRIBUTION

BY SEYMOUR BENZER

DIVISION OF BIOLOGY, CALIFORNIA INSTITUTE OF TECHNOLOGY, PASADENA

Communicated June 28, 1967



Clock Mutants of *Drosophila melanogaster*

(eclosion/circadian/rhythms/X chromosome)

RONALD J. KONOPKA AND SEYMOUR BENZER

Division of Biology, California Institute of Technology, Pasadena, Calif. 91109

Contributed by Seymour Benzer, July 2, 1971

period

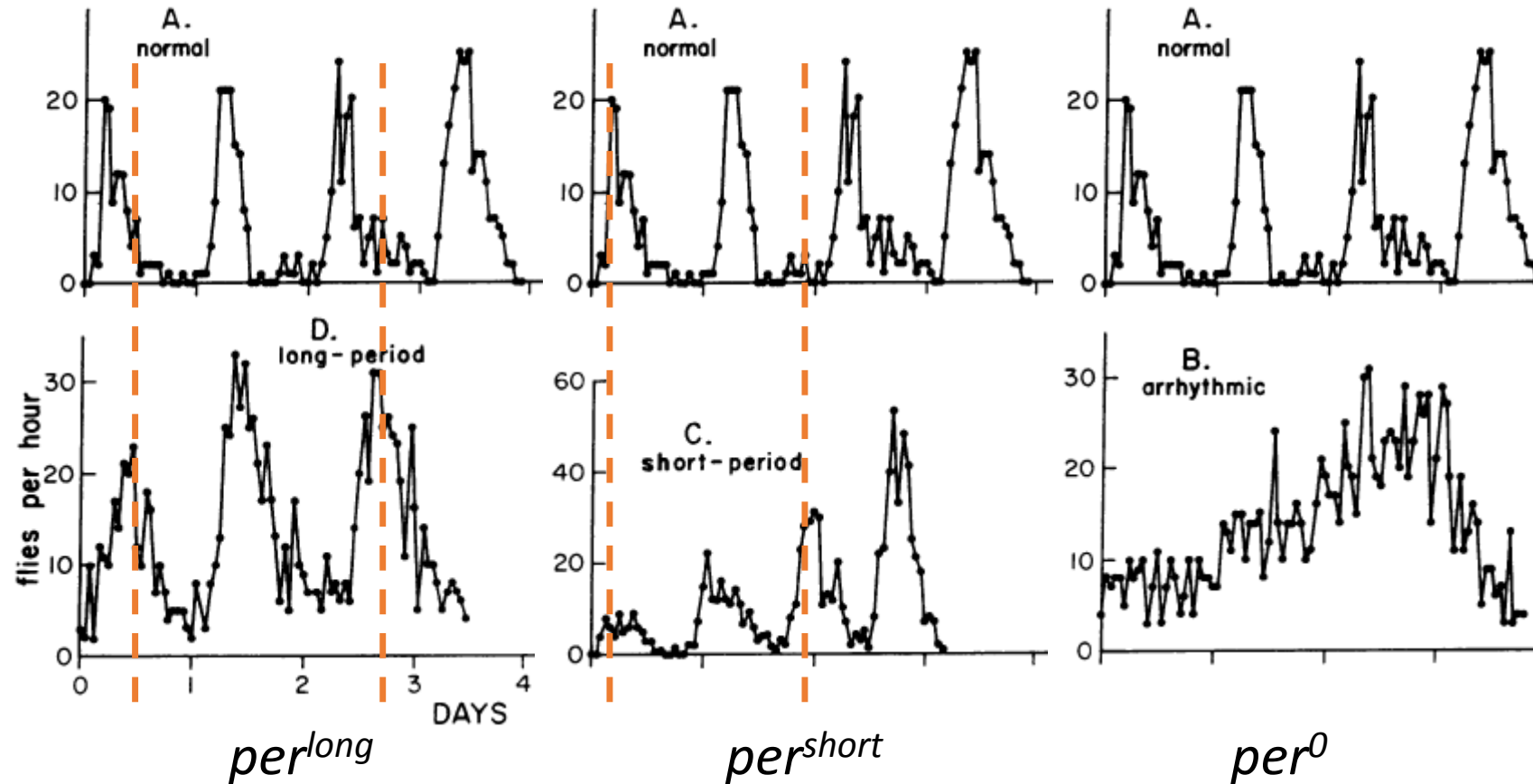


TABLE 1. Free-running period of locomotor activity

Genotype		N	Period \pm SD	Phenotype
First X chromosome	Second X chromosome			
normal (C-S)	normal (FM 7)	4	24.4 \pm 0.5	normal
arrhythmic	arrhythmic	4	arrhythmic	arrhythmic
short-period	short-period	5	19.5 \pm 0.4	short-period
long-period	long-period	4	28.6 \pm 0.5	long-period
arrhythmic	normal (FM 7)	8	25.2 \pm 0.4	\sim normal
short-period	normal (FM 7)	5	21.9 \pm 0.4	intermediate
long-period	normal (FM 7)	5	25.5 \pm 0.5	\sim normal
short-period	arrhythmic	6	19.5 \pm 0.4	short-period
long-period	arrhythmic	5	30.6 \pm 1.3	long-period
short-period	long-period	6	22.9 \pm 0.4	\sim normal

Drosophila as a model for neurodegenerative diseases

Neuron, Vol. 10, 839–850, May, 1993, Copyright © 1993 by Cell Press

Defective Glia in the *Drosophila* Brain Degeneration Mutant *drop-dead*

Robert L. Buchanan and Seymour Benzer
California Institute of Technology
Division of Biology
Pasadena, California 91125

***Spongecake* and *eggroll*: two hereditary diseases in *Drosophila* resemble patterns of human brain degeneration**

Kyung-Tai Min and Seymour Benzer

Current Biology 1997, 7:885–888
<http://biomednet.com/elecref/0960982200700885>

Drosophila as a model for neurodegenerative diseases

1) To model human diseases.

[Nature](#). 2000 Mar 23;404(6776):394-8.

A *Drosophila* model of Parkinson's disease.

[Feany MB](#)¹, [Bender WW](#).

[J Vis Exp](#). 2014 Aug 17;(90). doi: 10.3791/51625.

Methods to characterize spontaneous and startle-induced locomotion in a rotenone-induced Parkinson's disease model of *Drosophila*.

[Liao J](#)¹, [Morin LW](#)¹, [Ahmad ST](#)².

2) To find genes involved in neurodegeneration or diseases.

[PLoS One](#). 2008 Oct 8;3(10):e3332. doi: 10.1371/journal.pone.0003332.

A functional misexpression screen uncovers a role for *enabled* in progressive neurodegeneration.

[Rezával C](#)¹, [Berni J](#), [Gorostiza EA](#), [Werbajh S](#), [Fagilde MM](#), [Fernández MP](#), [Beckwith EJ](#), [Aranovich EJ](#), [Sabio y García CA](#), [Ceriani MF](#).

3) To find modulators, interacting genes or treatments related to diseases.

[Neuroscience](#). 2015 Mar 6. pii: S0306-4522(15)00207-9. doi: 10.1016/j.neuroscience.2015.02.048. [Epub ahead of print]

Attenuation of Neuromotor Deficits by Natural Antioxidants of *Decalepis hamiltonii* in Transgenic *Drosophila* Model of Parkinson's Disease.

[Jahromi SR](#)¹, [Haddadi M](#)², [Shivanandappa T](#)¹, [Ramesh SR](#)³.

Again, why *Drosophila*?



Table 5. Enhancer trap alleles

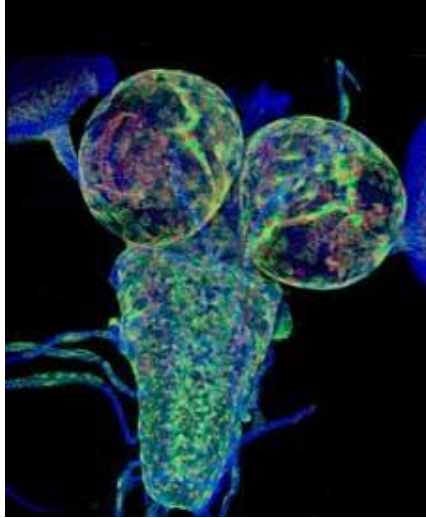
CG#	GENE	LINE	SITE	CHR	Strand	phenotype	T1	Insert	Met	Stop	T2	Insert	Met	Stop	T3	Insert	Met	Stop	T4	Insert	Met	Stop	Type
CG7147	kuz	CB02000	13532398	2L	+	lethal	RA	1	2	12	RB	1	2	12									4
CG7892	nmo	CB02015	7938638	3L	+	lethal	RA	1	3	9	RB	1	3	9	RE	1	3	9	RC	1	3	9	4
CG3758	esg	CB02017	15311950	2L	+	lethal	RA	1	1	1													4
CG30403		CB02022	16756893	2R	+	lethal	RA	0.5	1	13													30
CG9936	skd	CB02029	20943006	3R	+	lethal	RA	1.5	2	13	RC	1.5	2	13									28
CG2411	ptc	CB02030	3710594	2R	+	lethal	RA	1	1														30
CG17654	Eno	CB02039	1729440	2R	+	lethal	RA	3	3	4	RE	3	3	4	RC	3	3	4	RD	3	3	4	4
CG3619	Di	CB02040	1515179	2R	+	lethal	RA	0.5	0	6	RB	0.5	1	6									30
CG1621	CG1621	CB02042	2853998	2R	+	lethal	RA	1	1														30
CG12891	CPT1	CB02043	5539025	2R	+	lethal	RA	1	1														4
CG4043	Rp46	CB02050	6238635	3R	+	lethal	RA	1	1														30
CG5677	CG5677	CB02054	20046300	3R	+	lethal	RA	1	1														4
CG9062	CG9062	CB02056	6348682	2R	-	lethal	RA	1	1			1	1	9									4, 1
CG10823	CG10823	CB02057	17122275	3R	+	lethal	RA	1	1			1	1	4									30
CG9415	Xbp1	CB02061	16187154	2R	+	lethal	RA	1	1			1	1	2									4
CG30498	bca	CB02070	2622679	2R	+	lethal	RA	1	1			1	1	2									30
CG15864	CG15864	CB02071	4076201	3R	+	lethal	RA	1	1			1	1	10									5
CG31314	Neu3	CB02076	10523056	3R	+	lethal	RA	1	1			1	1	10									30
CG8128	CG8128	CB02087	15428524	X	+	lethal	RA	1	1			1	1	14	RD	1.5	2	8					4
CG17117	hth	CB02095	6440060	3R	-	semi-lethal	RA	0.5	1	14	RB	0.5	1	14	RD	1.5	2	8					28
CG5887	desat1	CB02105	8269757	3R	+	lethal	RA	1	1			1	1	5	RE	1.5	2	5	RB	1.5	2	5	28
CG30497	CG30497	CB02106	2840573	2R	+	lethal	RA	1	1			1	1	5									4
CG17342	Lk6	CB02120	7590202	3R	+	lethal	RA	0.5	1														30
CG4570	CG4570	CB02124	6682635	3R	+	lethal	RA	1	1			1	1	4									4
CG1427	CG1427	CB02125	1425793	3R	+	lethal	RA	0.5	2			1	1	4									30
CG3428	CG3428	CB02131	9445286	3L	+	lethal	RA	1	1			1	1	4									4
CG14478	CG14478	CB02133	12521410	2R	+	lethal	RA	1	1			1	1	4									4
CG7820	[(3)87Df	CB02135	8856406	3R	+	lethal	RA	0.5	1			1	1	4									30
CG2161	Rpa	CB02139	1438299	3R	-	lethal	RA	1	2			1	2	8									4
CG31241	CG31241	CB02140	14081648	3R	+	lethal	RA	1	2			1	2	8									4
CG17328	CG17328	CB02149	16277703	2L	+	lethal	RA	0.5	1			1	1	4									30
CG12284	th	CB02150	13L	3L	+	lethal	RA	0.5	1			1	1	4									30
CG1837	CG1837	CB02168	11449163	X	+	lethal	RA	0.5	1			1	1	4									3A

Genome Res. 2001 Jun;11(6):1114-25.

A systematic analysis of human disease-associated gene sequences in *Drosophila melanogaster*.

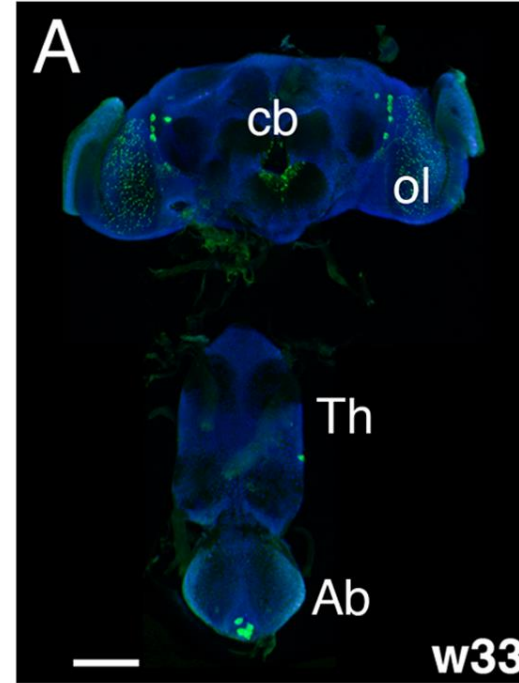
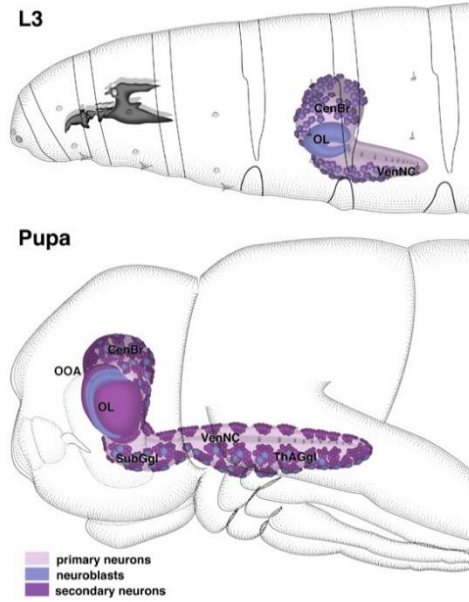
Reiter LT¹, Potocki L, Chien S, Gribskov M, Bier E.

Again, why *Drosophila*?



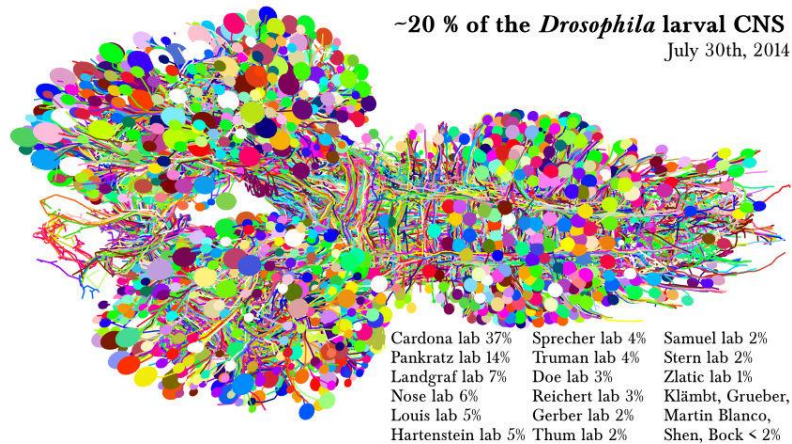
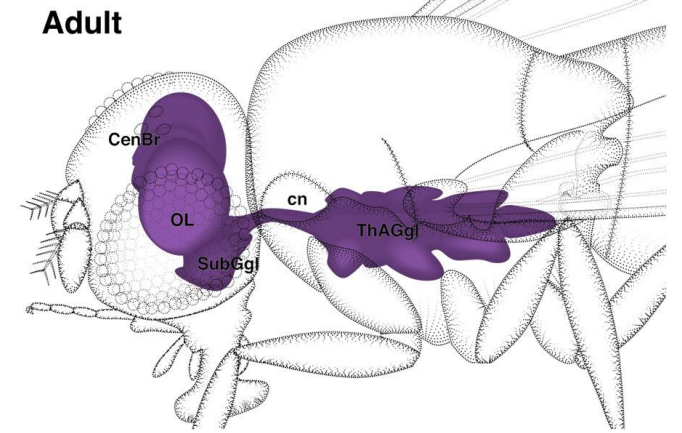
Schmidt I. *et al*, The Journal of Neuroscience, 2012

L1: 10000 neurons



Shafer OT, Taghert PH, PLoS ONE, 2009

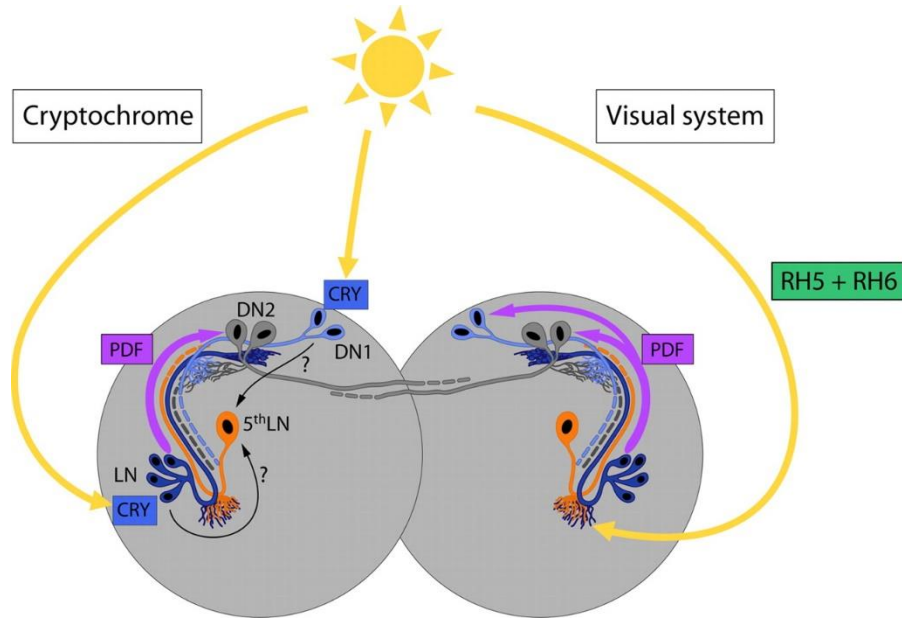
100000 neurons



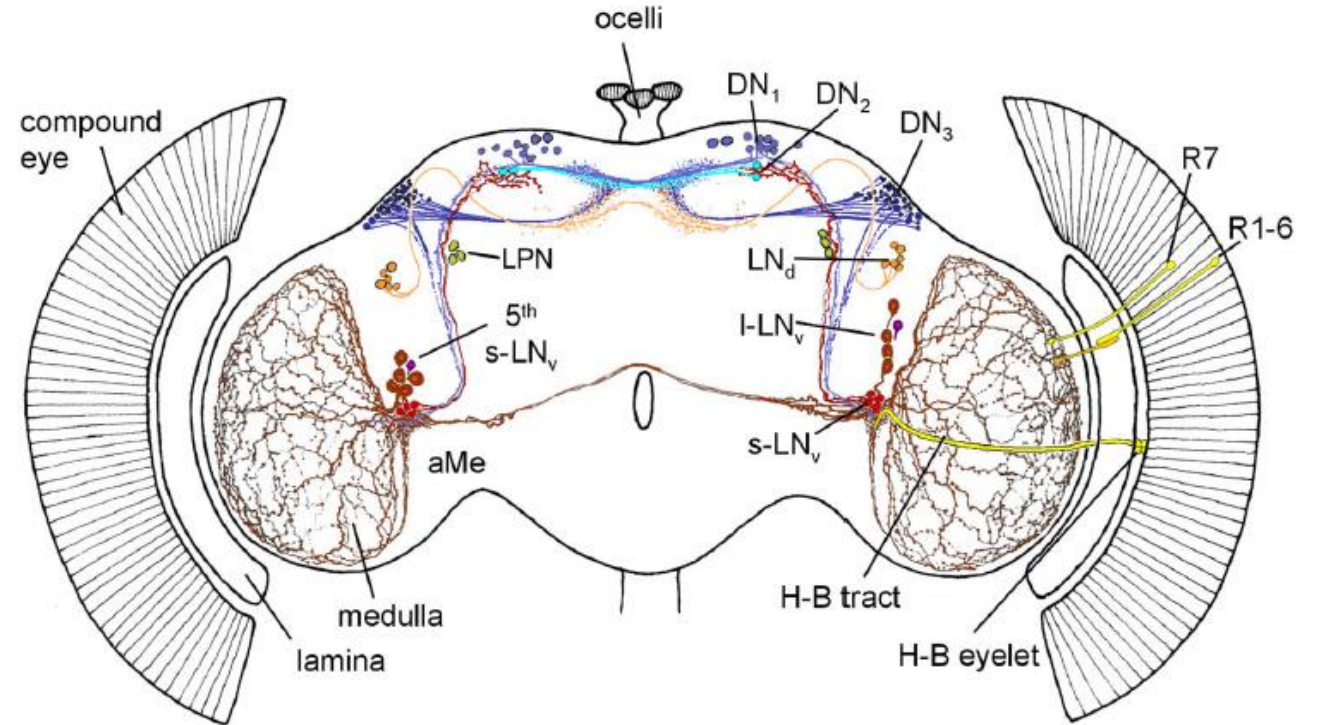
L1: 3 million synapses estimated

<http://albert.rierol.net/lab.html>

Metamorphosis: new body, new neurons, new behaviors



André Klarsfeld et al., The Journal of Neuroscience 2011

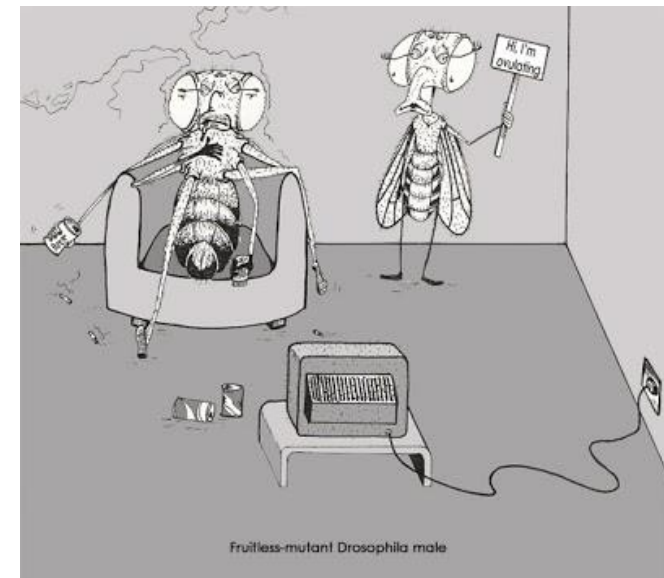
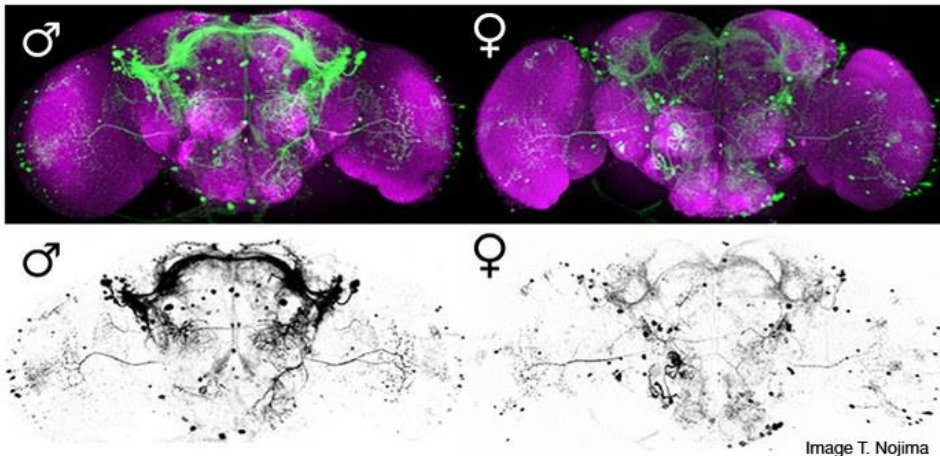
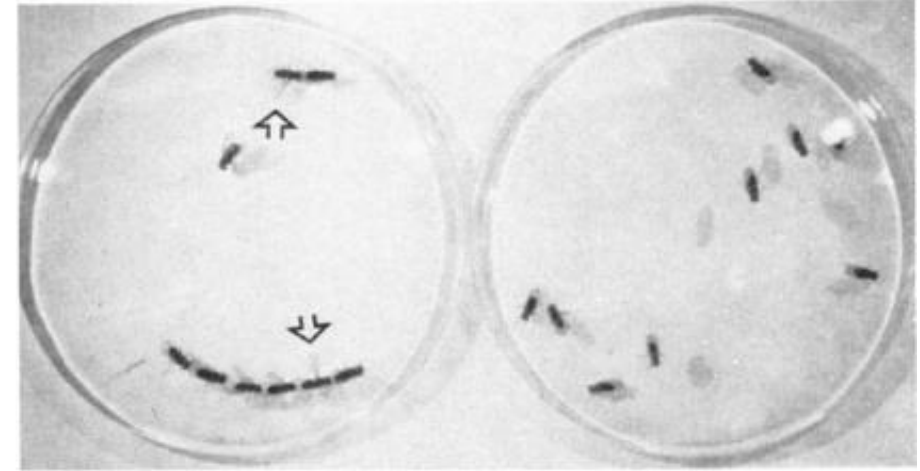


C. Helfrich-Förster et al., The Journal of Comparative Neurology 2007

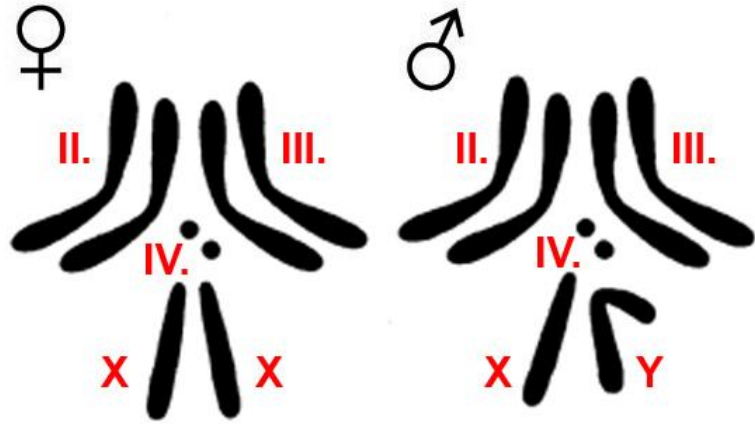
Genetic tools: balancers

fruitless

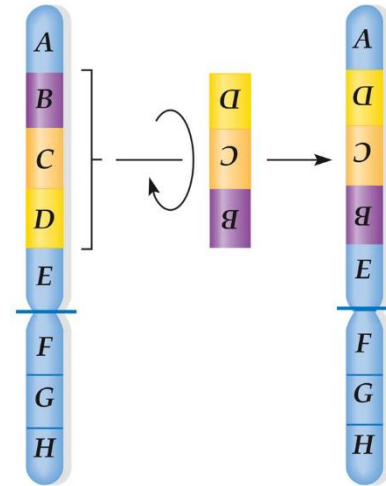
- ✓ Sex determination
- ✓ Development structures necessary for courtship:
 - Muscle development
 - Neurons
- ✓ Aggressive behavior
- ✓ Male courtship behavior:
 - wing vibration
 - song production
 - mating
 - etc.
- ✓ etc.



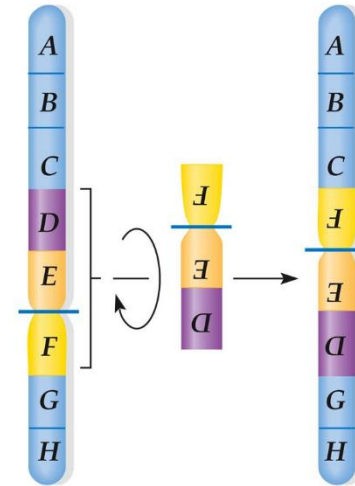
Genetic tools: balancers



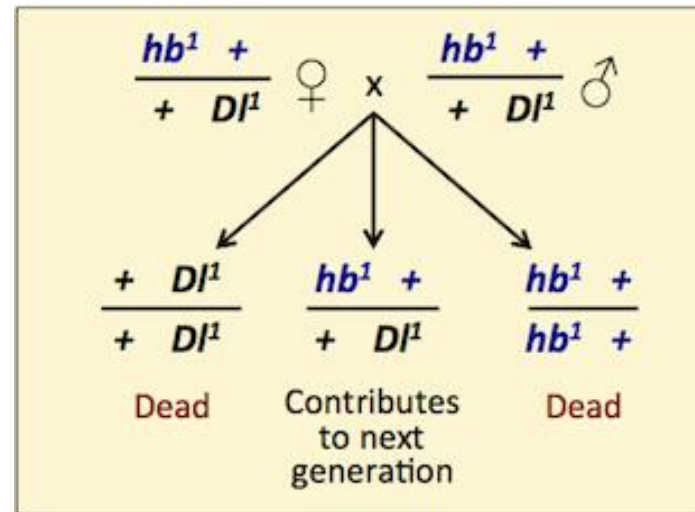
a) Paracentric inversion (does not include centromere)



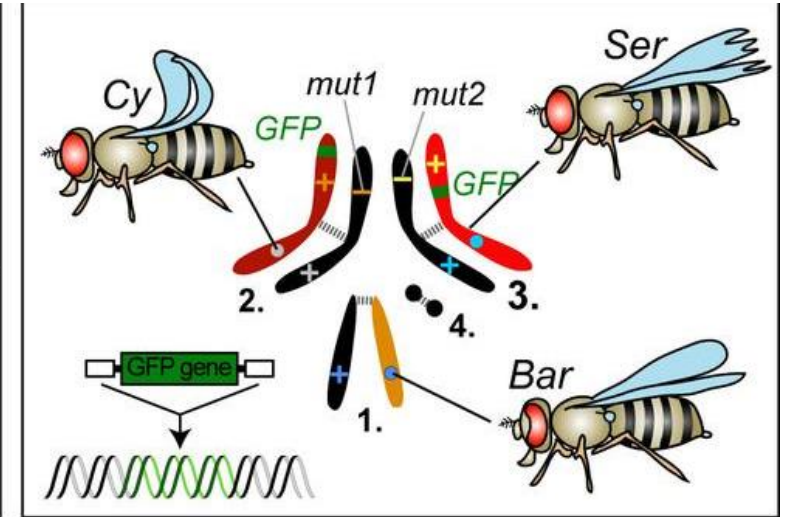
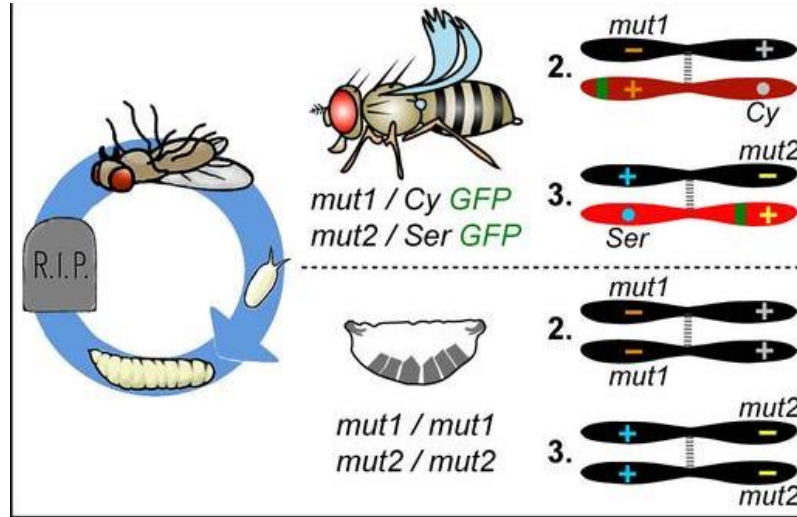
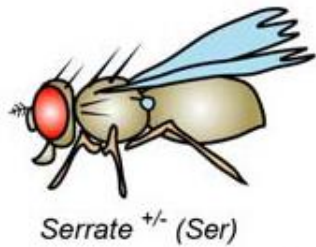
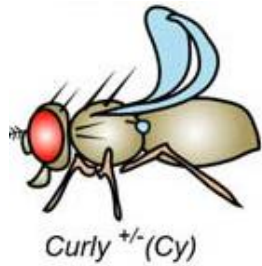
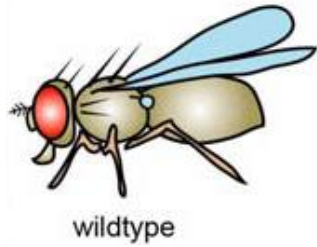
b) Pericentric inversion (includes centromere)



Copyright © 2006 Pearson Benjamin Cummings. All rights reserved.



Genetic tools: balancers



chromosome 2			chromosome 3		
F1	$mut1$	$GFP \text{ Cy}$	F1	$mut2$	$GFP \text{ Ser}$
$mut1$	$mut1 / mut1$ 	$mut1 / CyO \text{ GFP}$ 	$mut2$	$mut2 / mut2$ 	$mut2 / Ser \text{ GFP}$
$GFP \text{ Cy}$	$mut1 / CyO \text{ GFP}$ 	$CyO / CyO \text{ GFP / GFP}$ 	$GFP \text{ Ser}$	$mut2 / Ser \text{ GFP}$ 	$Ser / Ser \text{ GFP / GFP}$

Genetic tools: transgenic flies

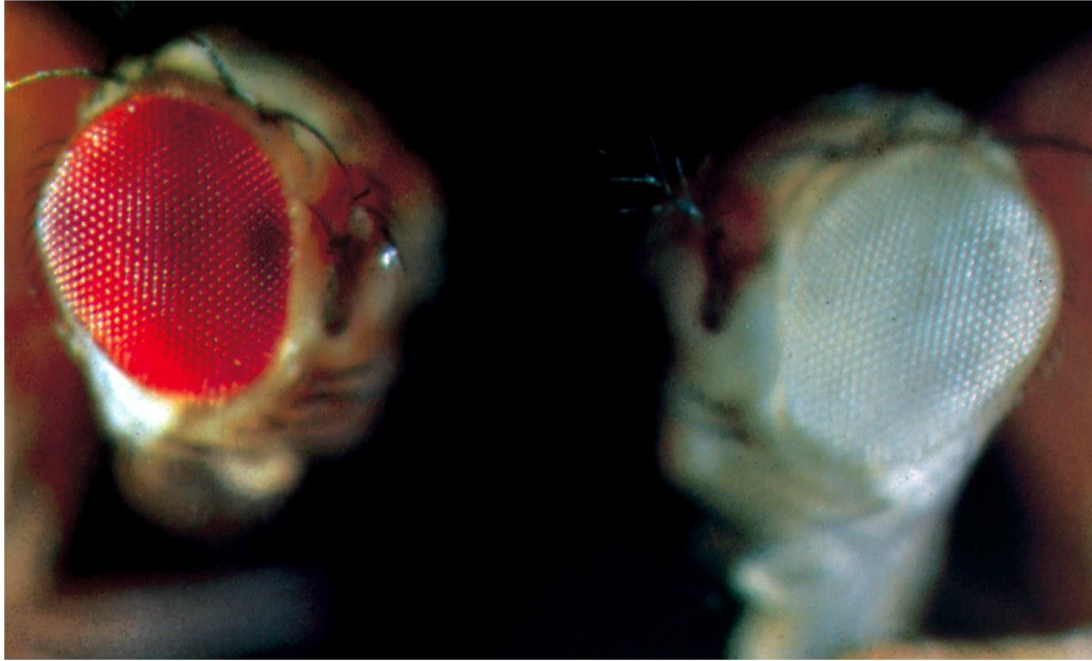
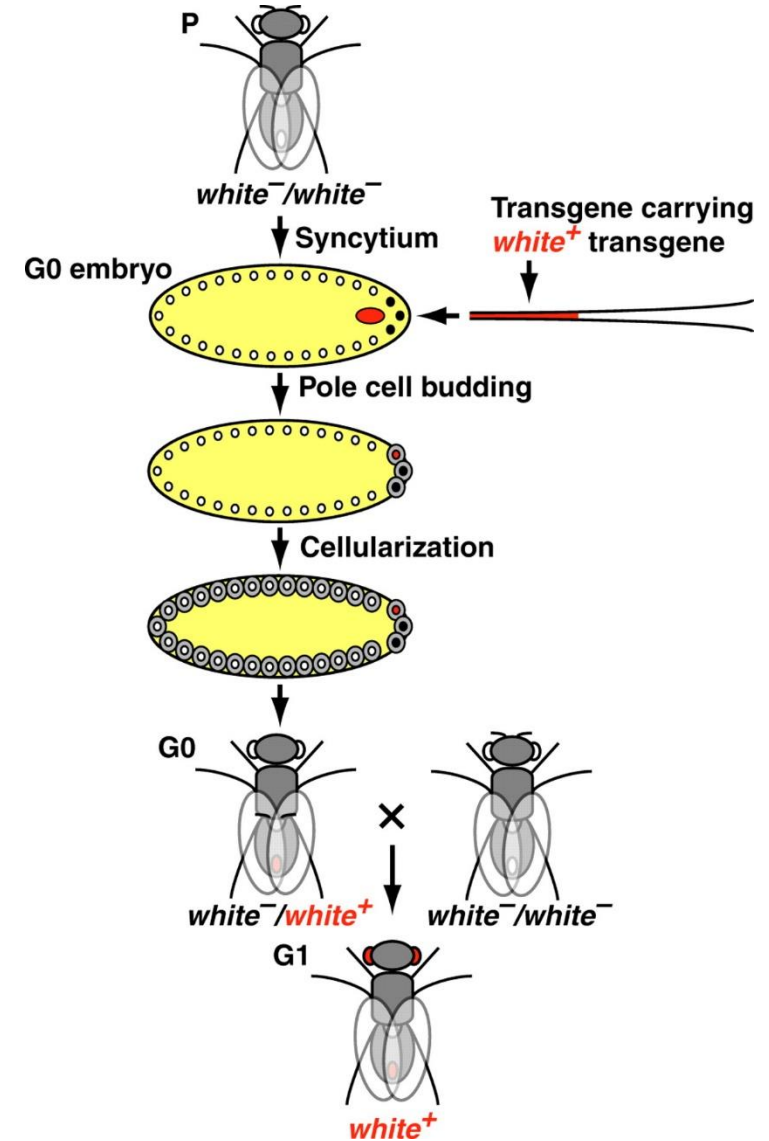
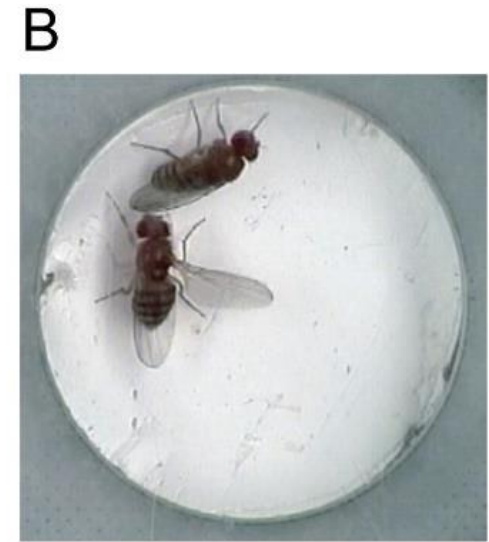
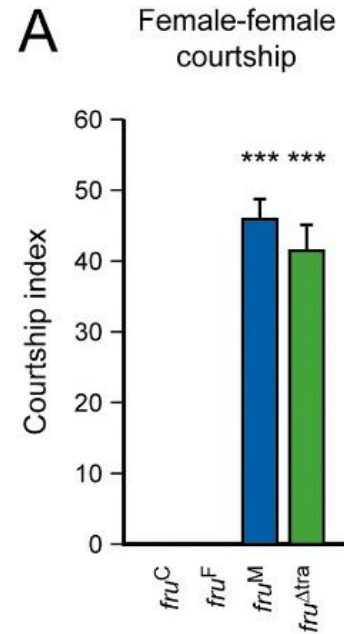
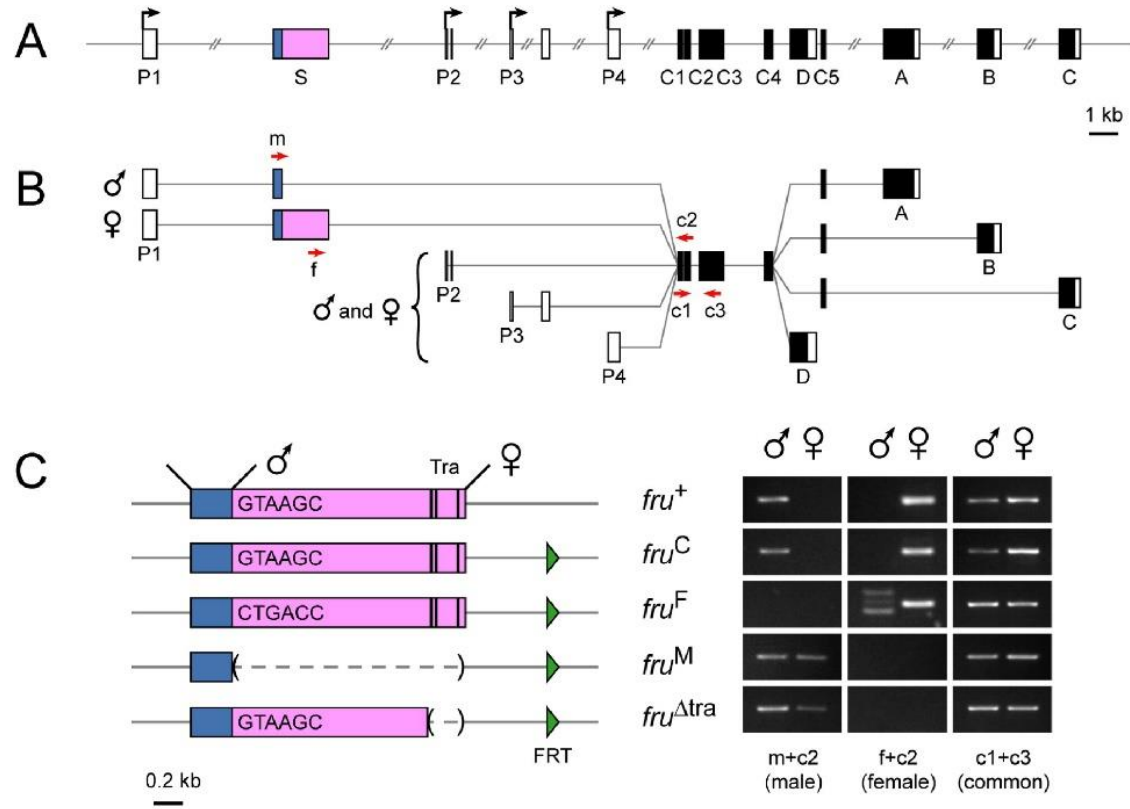


Figure 2-18
Introduction to Genetic Analysis, Tenth Edition
© 2012 W. H. Freeman and Company

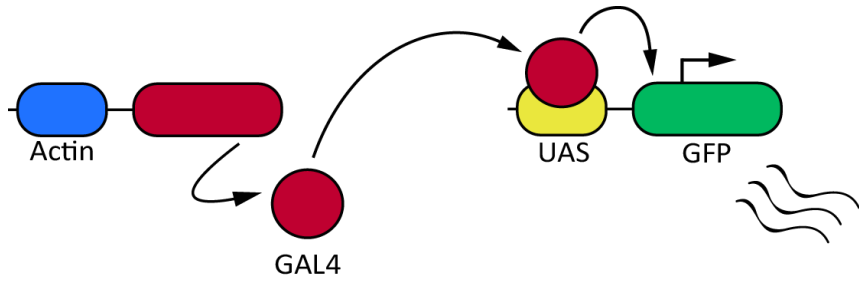
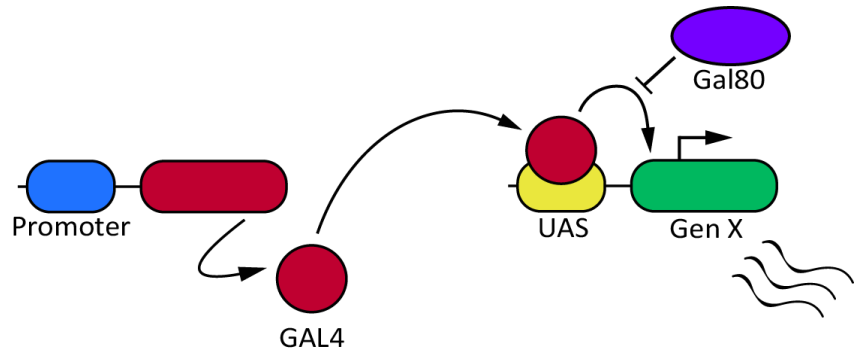
Drosophila transgenesis: *white*⁺ transgene DNA (red) is injected into generation zero *Drosophila* embryos (G0) of less than 1 hour old, which have been obtained from a parental (P) generation.



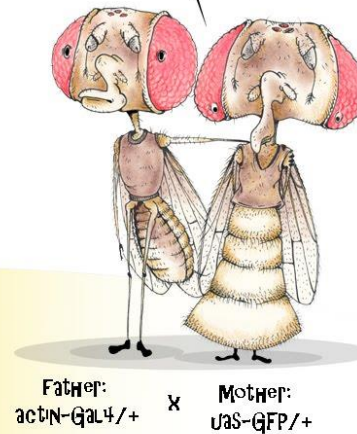
Genetic tools: transgenic flies



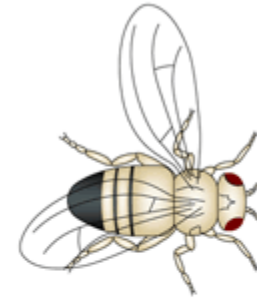
Genetic tools: GAL4/UAS and other tools



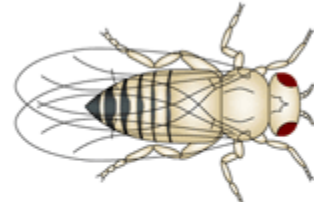
What Have We done Wrong?



Enhancer-trap GAL4



UAS-gene X



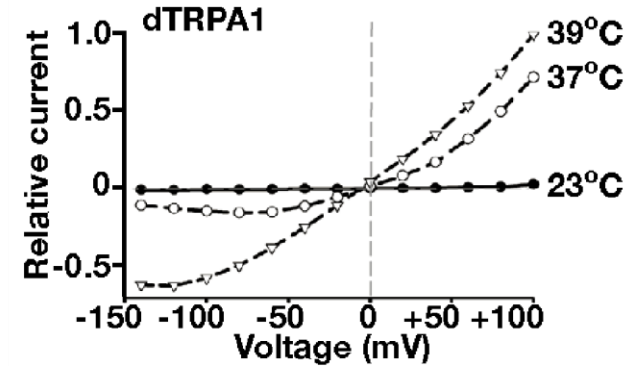
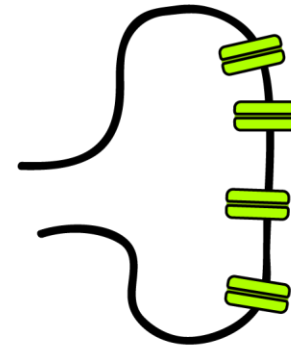
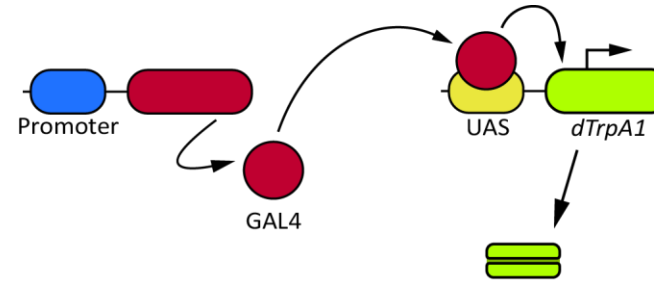
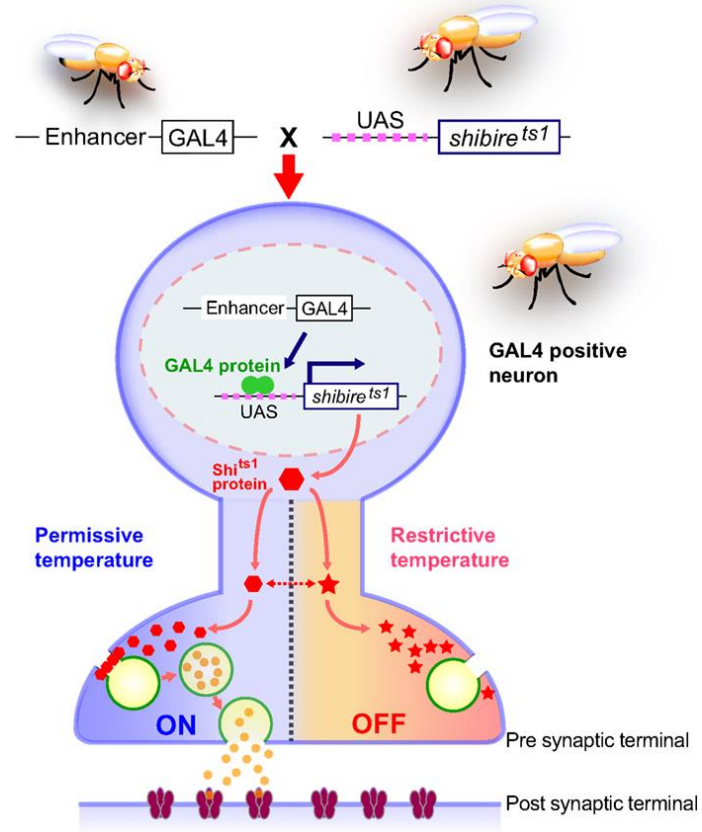
Genomic enhancer → GAL4
Tissue-specific expression of GAL4

GAL4 → UAS → gene X
Transcriptional activation of gene X

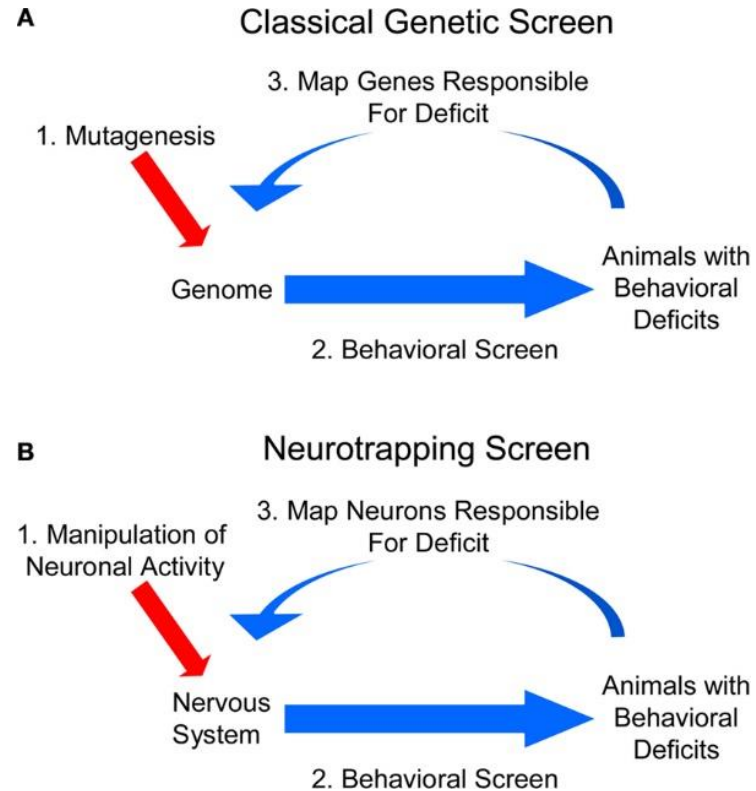
Nature Reviews | Genetics

Genetic tools: GAL4/UAS and other tools

Neuronal inhibition and activation



Genetic tools: GAL4/UAS and other tools

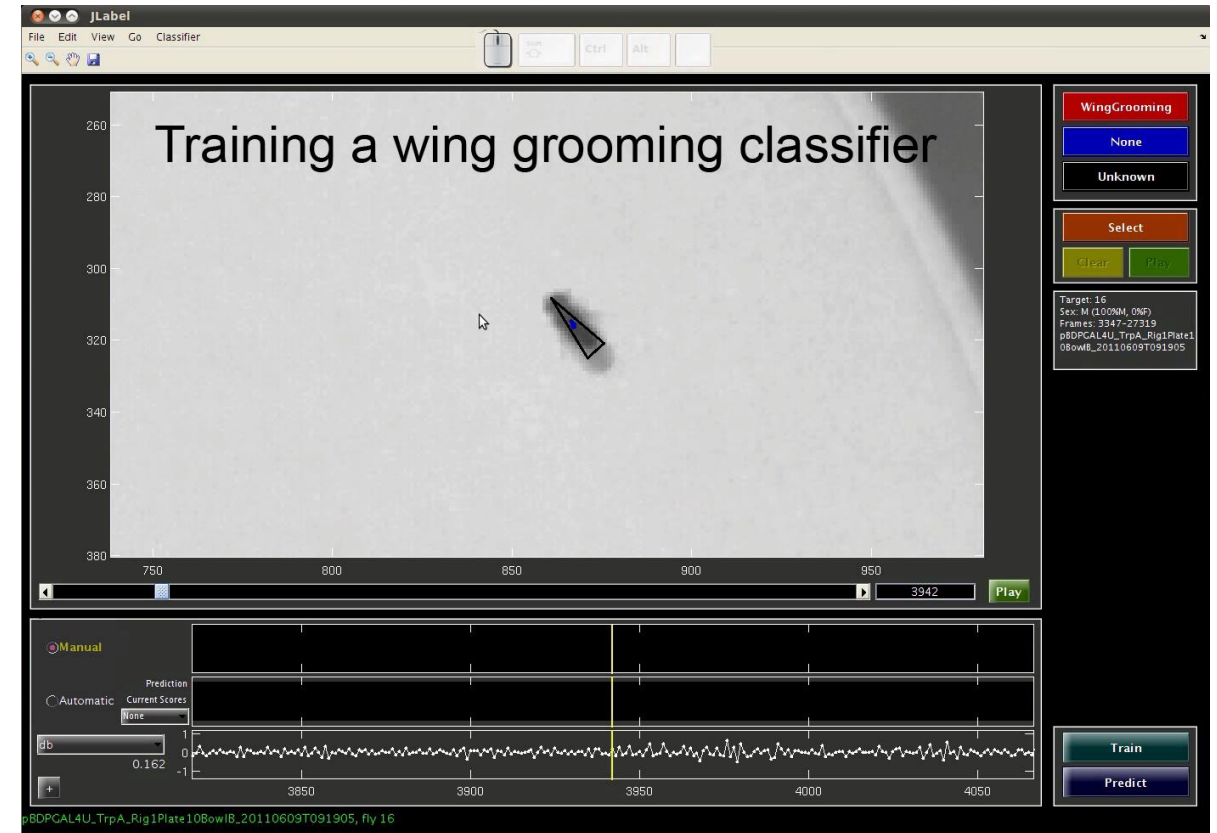
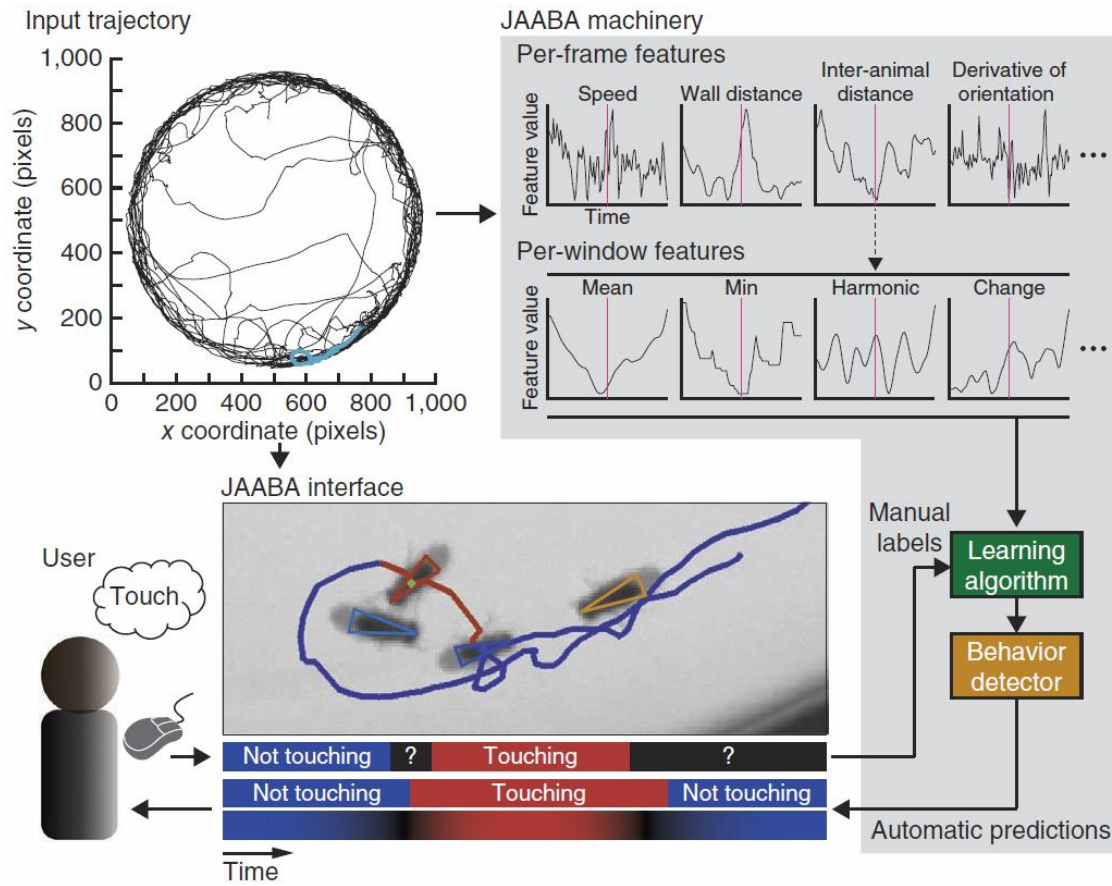


JAABA: interactive machine learning for automatic annotation of animal behavior

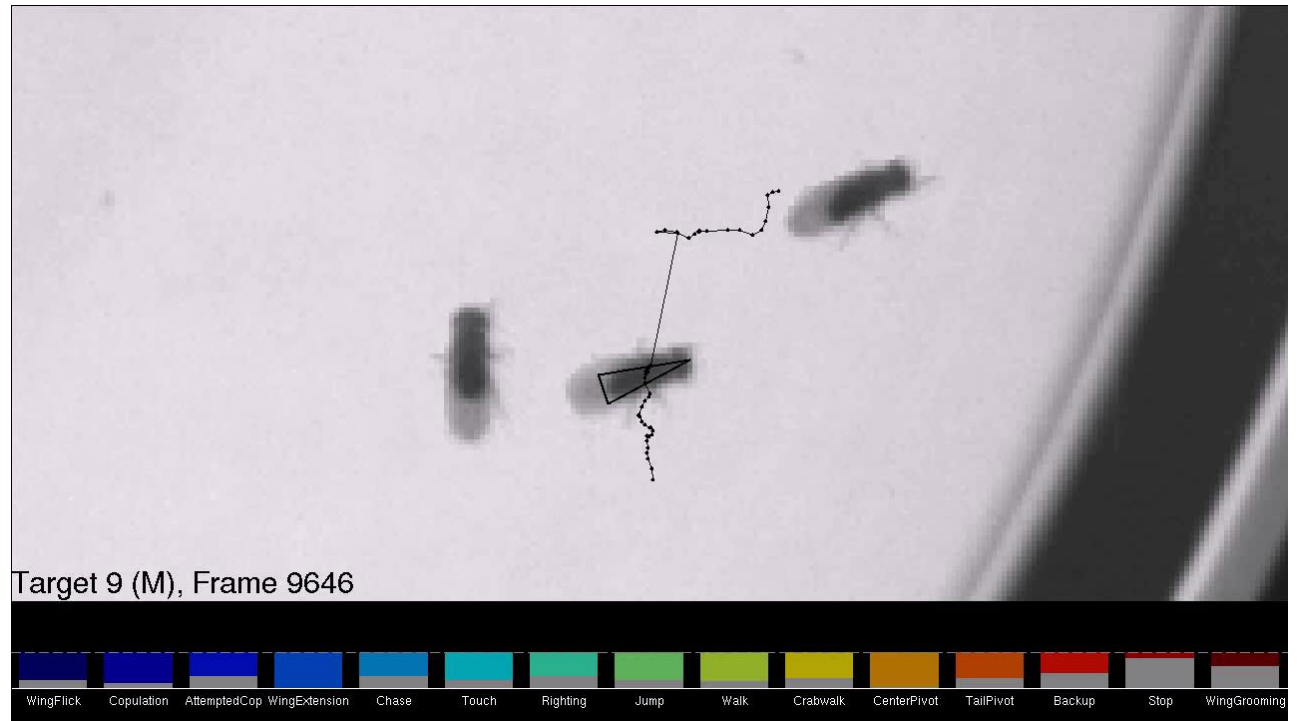
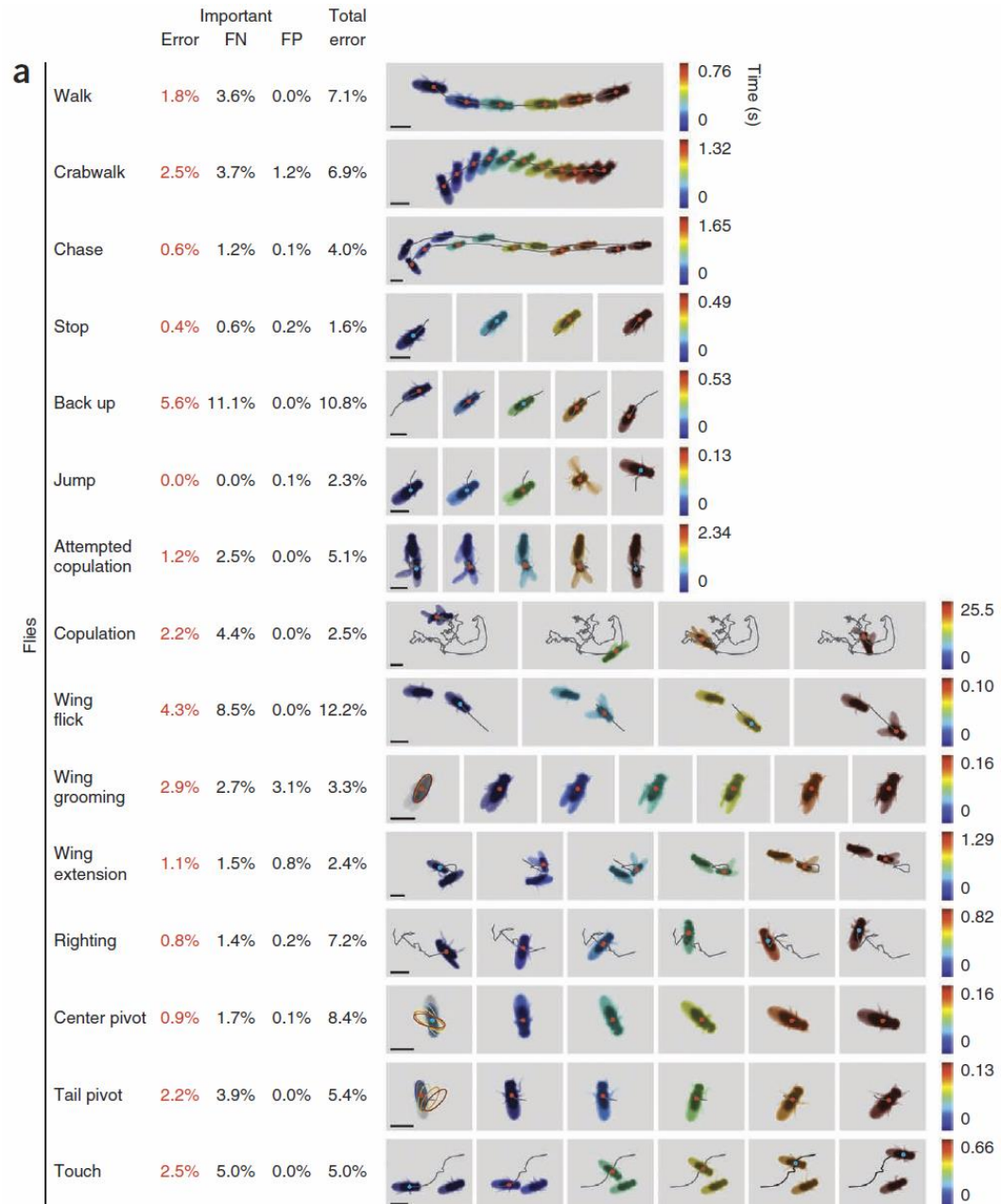
Mayank Kabra^{1,4}, Alice A Robie^{1,4},
Marta Rivera-Alba^{1,2}, Steven Branson^{1,3} &
Kristin Branson¹

GAL4 collection + UAS-*dTrpA1*

Genetic tools: GAL4/UAS and other tools

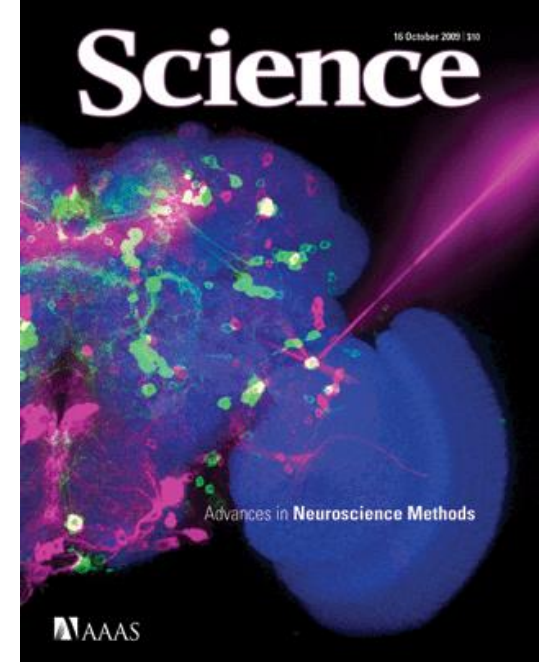
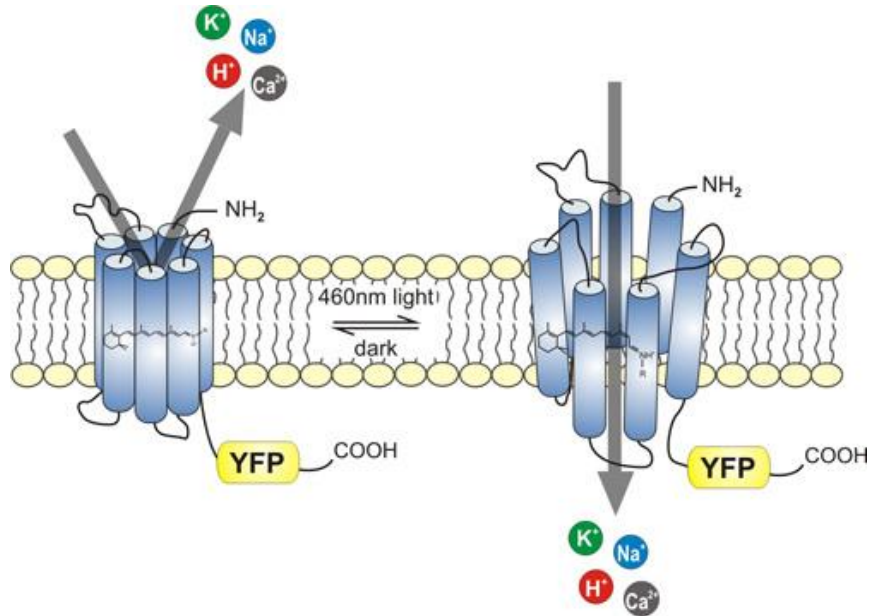


Genetic tools: GAL4/UAS and other tools

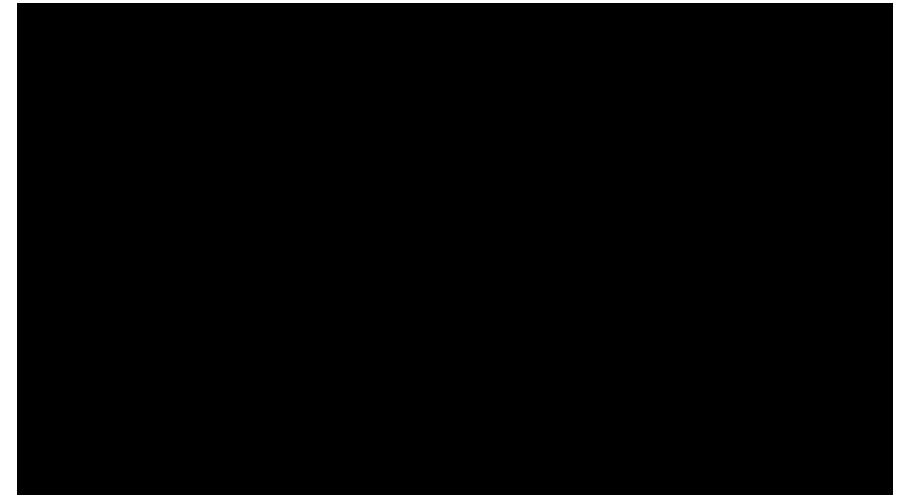


Genetic tools: GAL4/UAS and other tools

Optogenetics: Channelrhodopsin & others

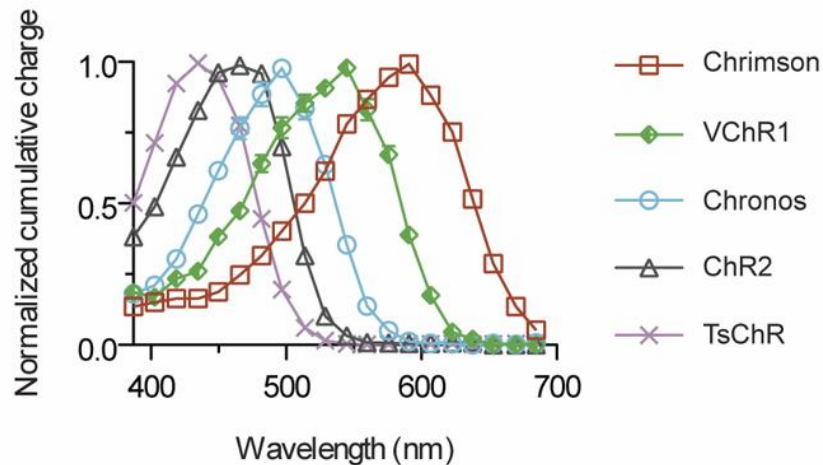


Proboscis extension reflex



sugar-sensing gustatory receptor neurons (GRNs)

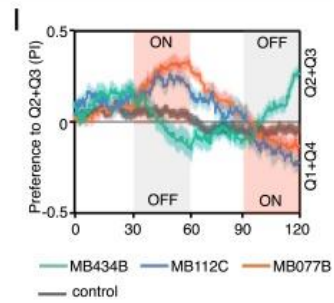
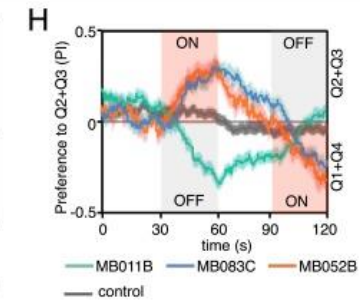
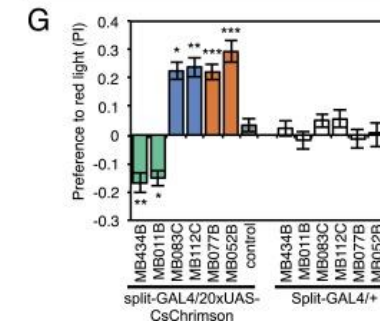
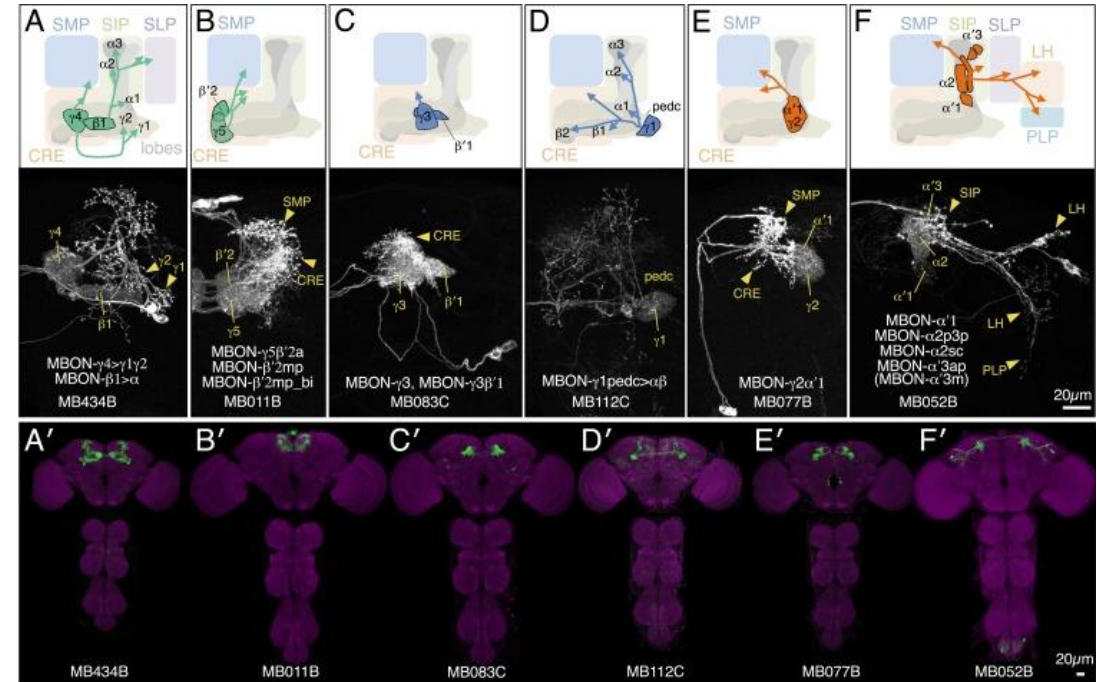
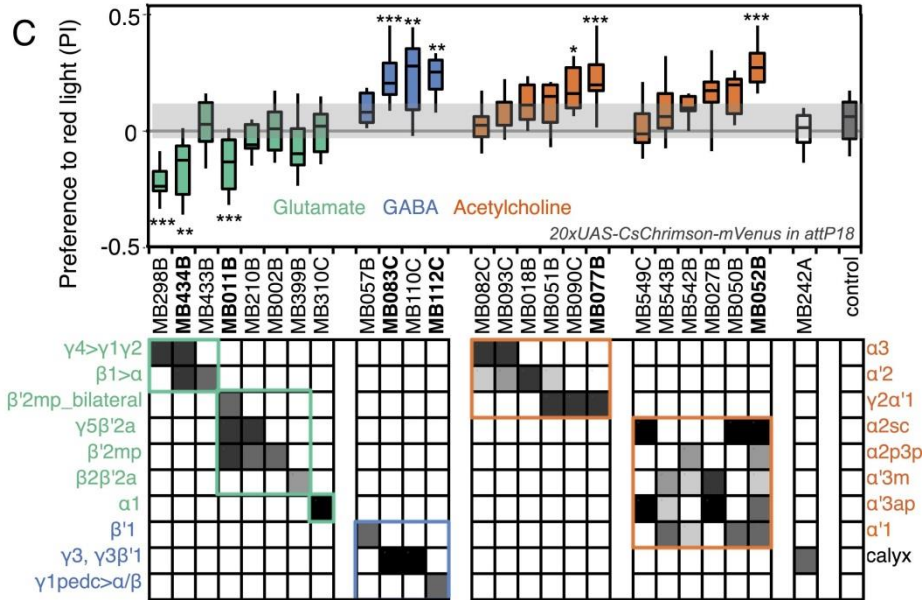
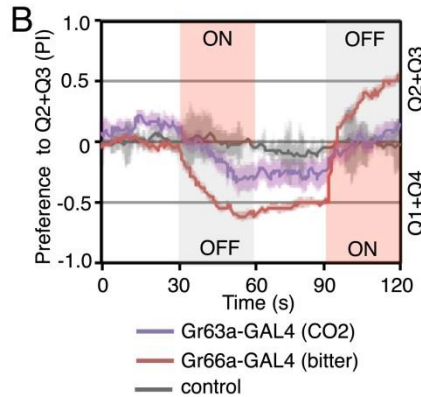
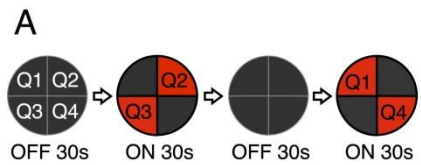
- UAS-ChR2
- UAS-ReaChR
- UAS-Chrimson
- UAS-Chronos



Genetic tools: GAL4/UAS and other tools

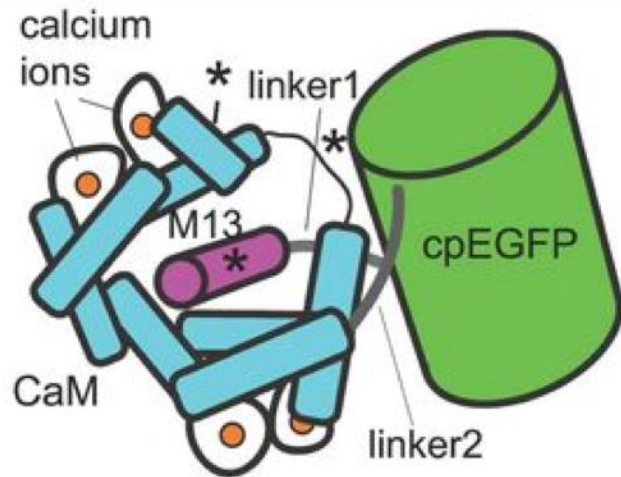
Mushroom body output neurons encode valence and guide memory-based action selection in *Drosophila*

Yoshinori Aso^{1*}, Divya Sitaraman^{1,2,6,7†}, Toshiharu Ichinose^{3,4}, Karla R Kaun^{1‡}, Katrin Vogt³, Ghislain Belliart-Guérin⁵, Pierre-Yves Plaçais⁵, Alice A Robie¹, Nobuhiro Yamagata^{3,4}, Christopher Schnaitmann^{3§}, William J Rowell¹, Rebecca M Johnston¹, Teri-T B Ngo¹, Nan Chen¹, Wyatt Korff¹, Michael N Nitabach^{1,2,6,7}, Ulrike Heberlein¹, Thomas Preat⁵, Kristin M Branson¹, Hiromu Tanimoto^{3,4}, Gerald M Rubin^{1*}

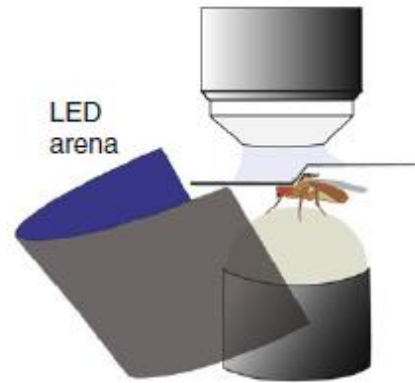


Genetic tools: GAL4/UAS and other tools

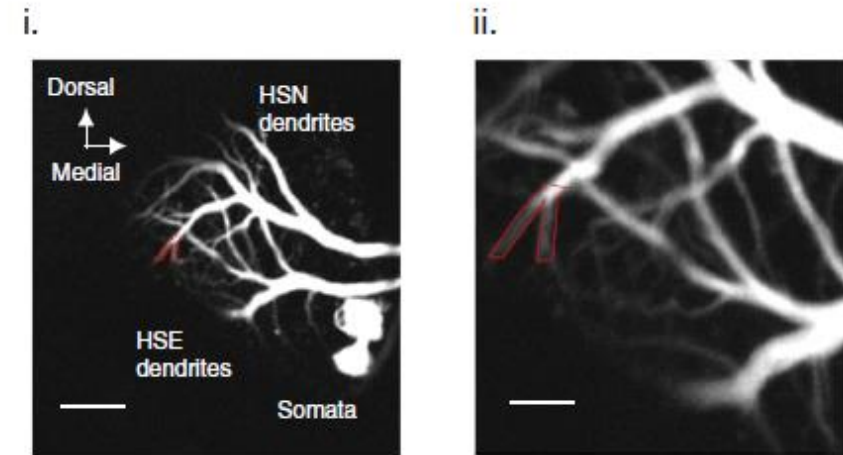
Calcium activity sensors: GCaMP



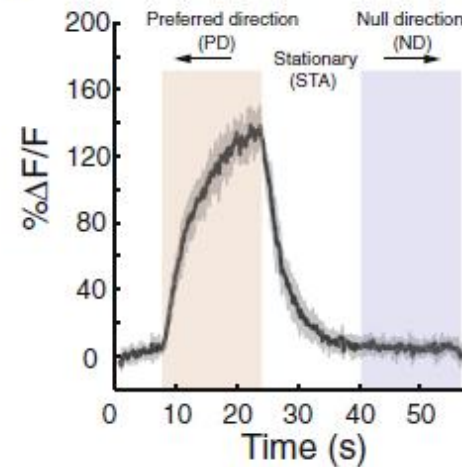
A



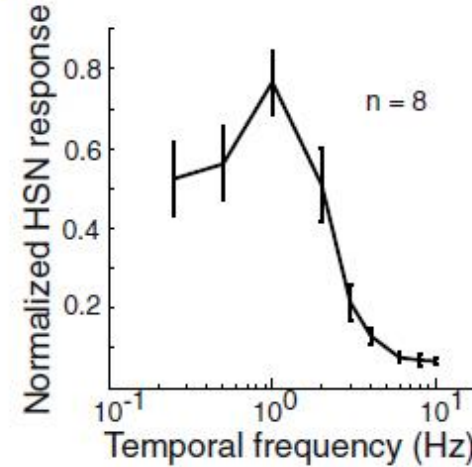
B



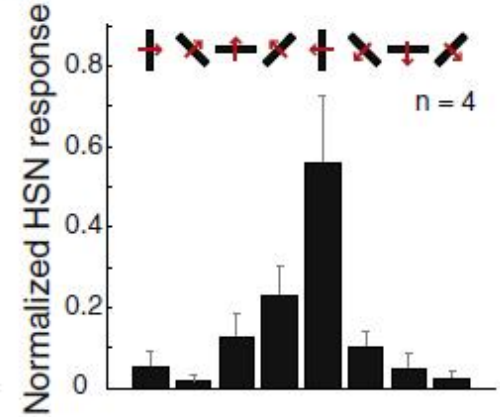
C



D



E



Behavioral paradigms



Behavioral paradigms: Taxis

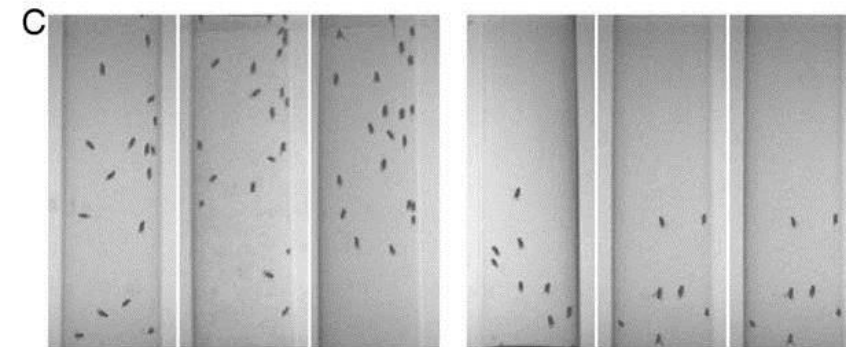
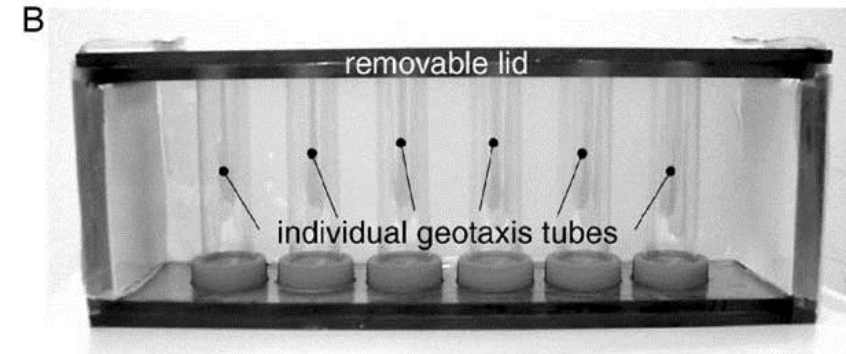
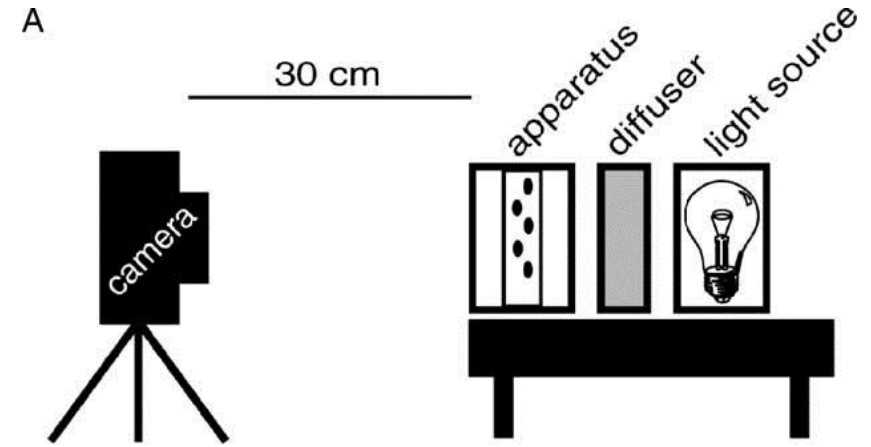
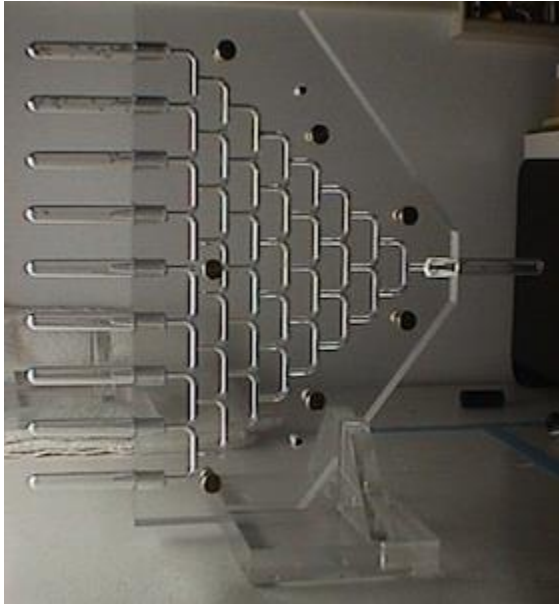
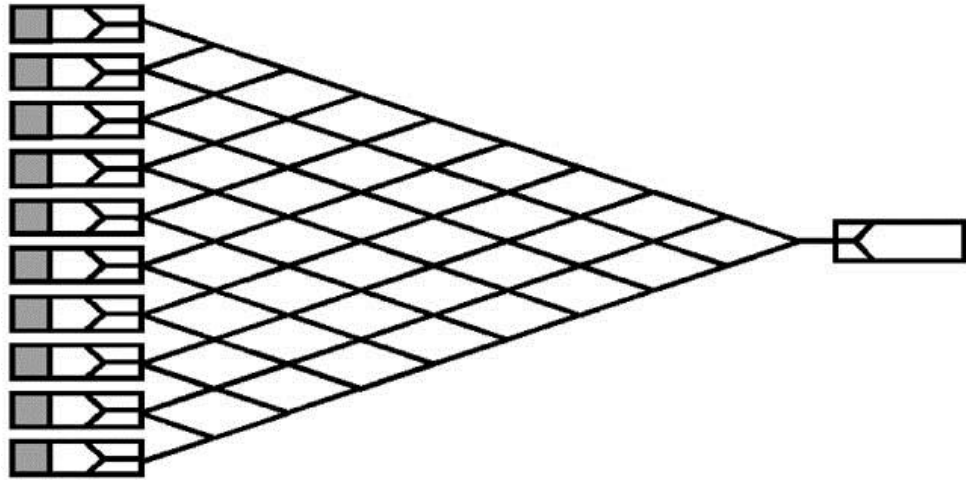
Phototaxis

Geotaxis

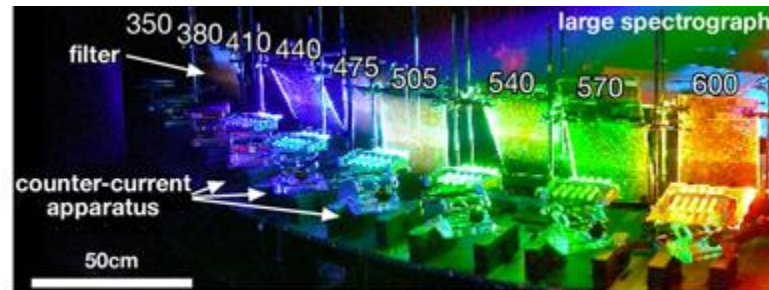
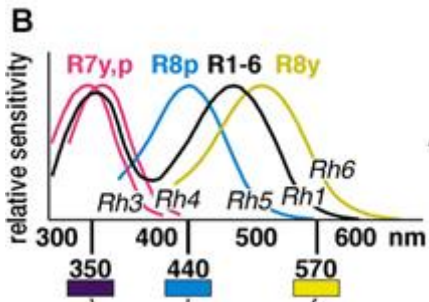
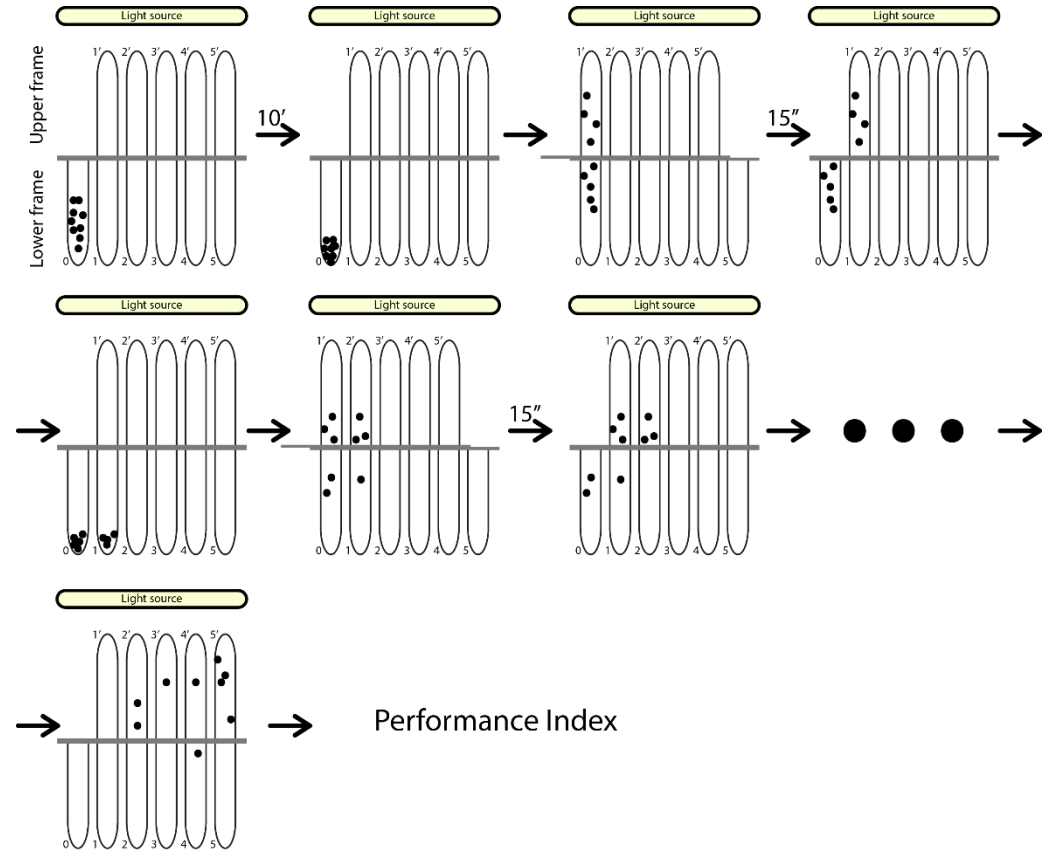


"You know, I'm very attracted to you."

Behavioral paradigms: geotaxis

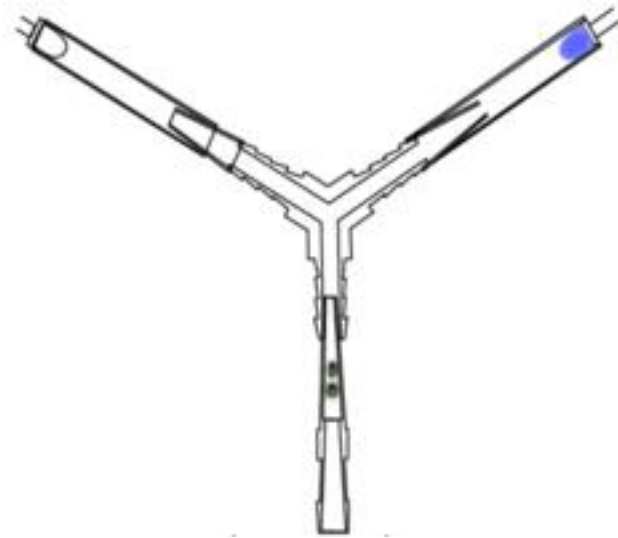
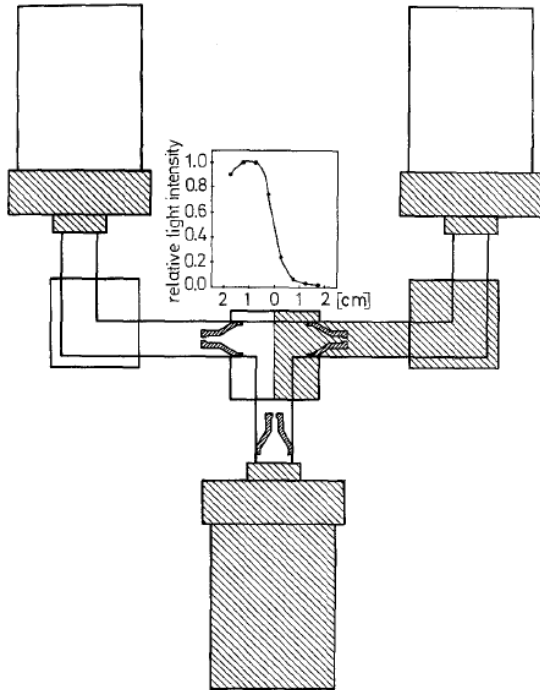


Behavioral paradigms: phototaxis

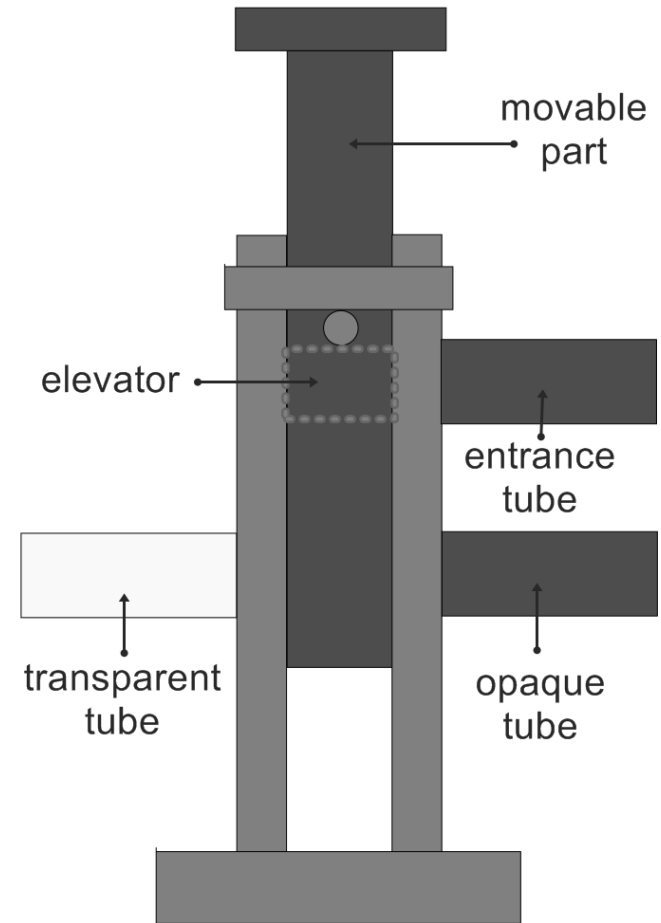


Behavioral paradigms: phototaxis

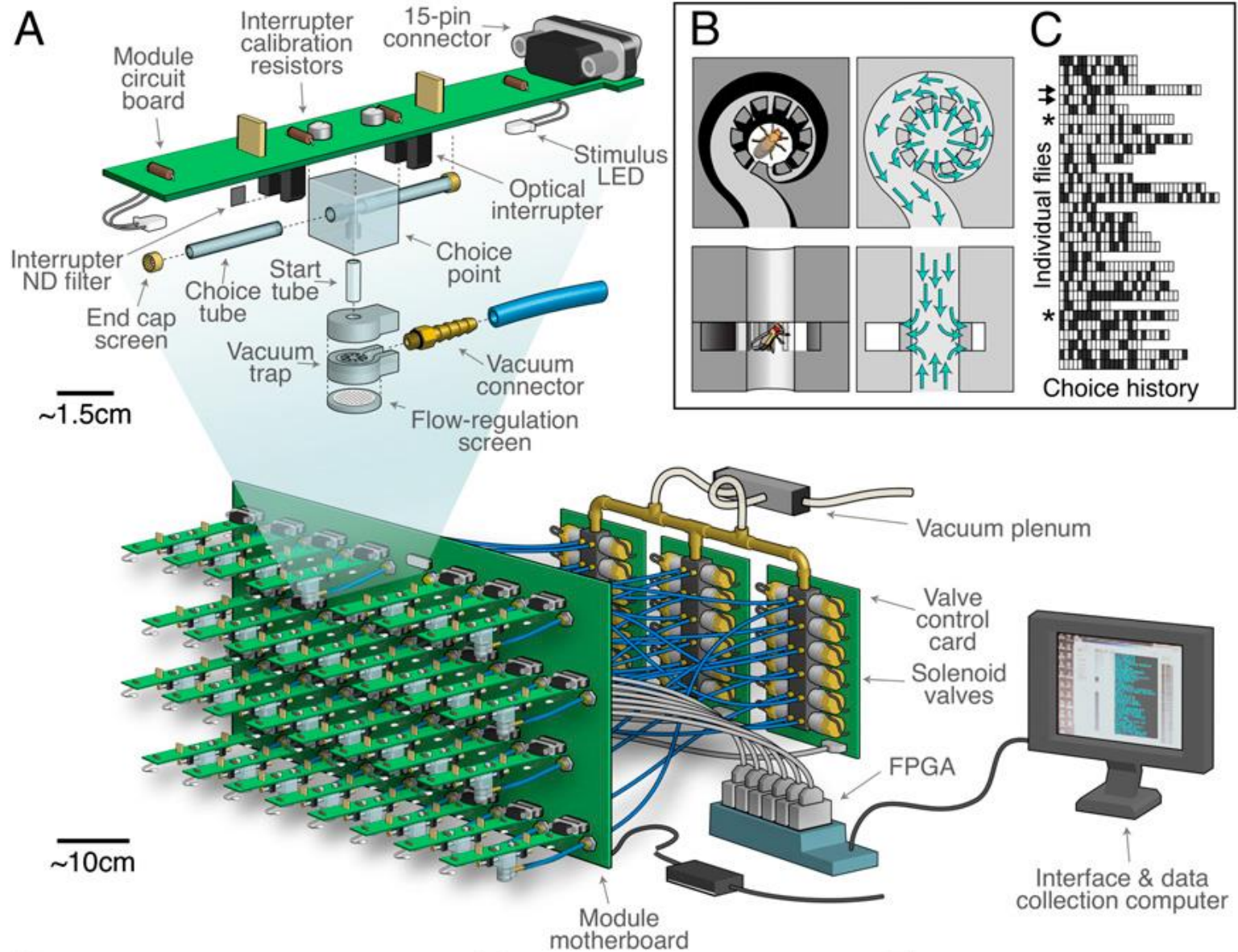
Single fly assay



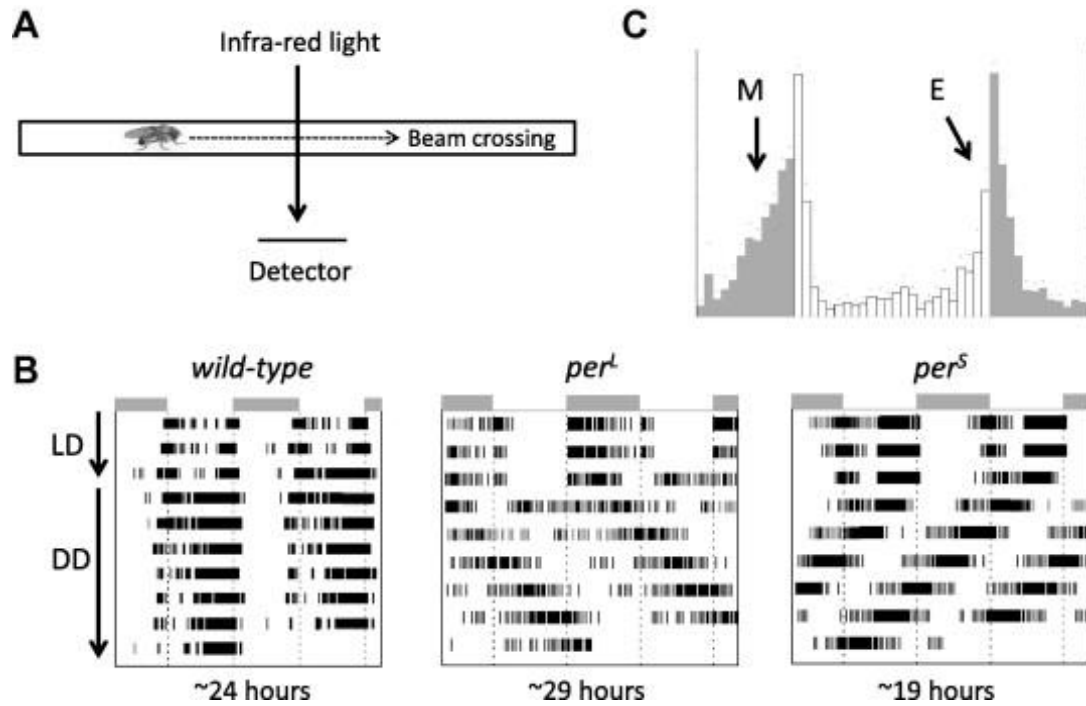
Population assay



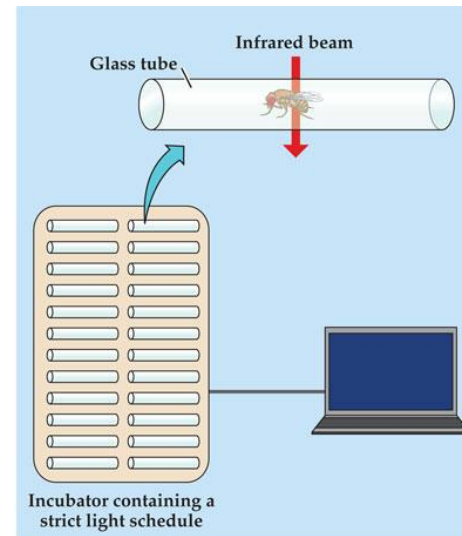
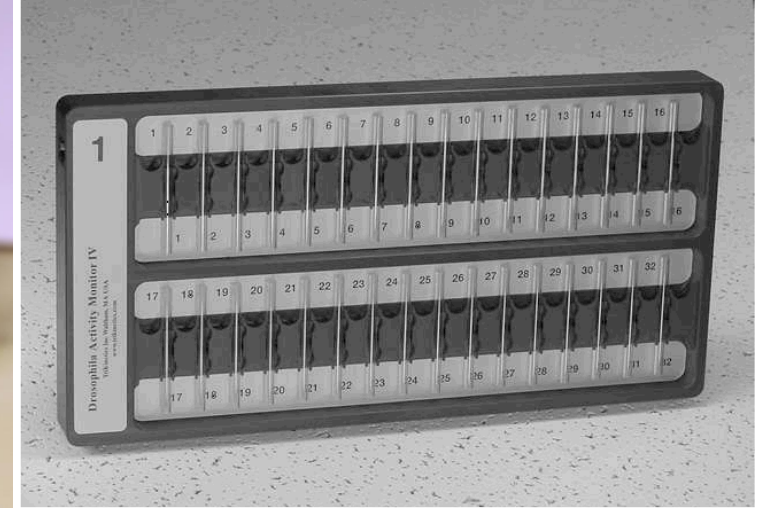
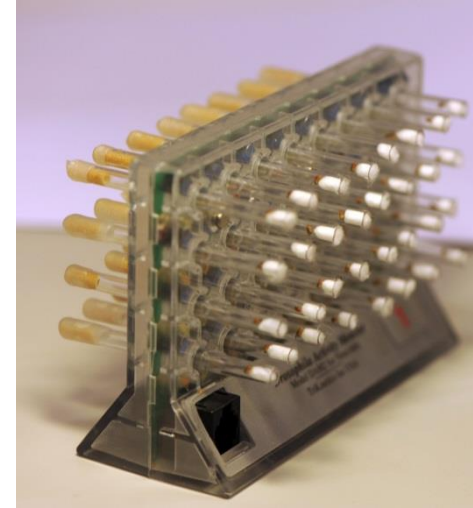
Behavioral paradigms: phototaxis



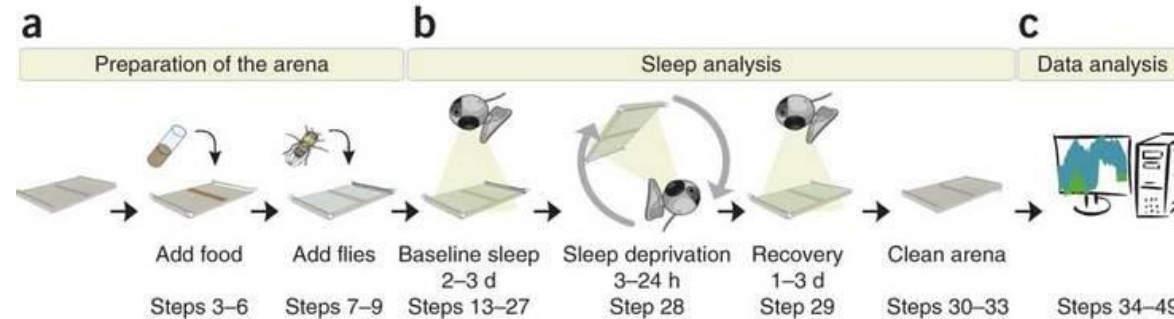
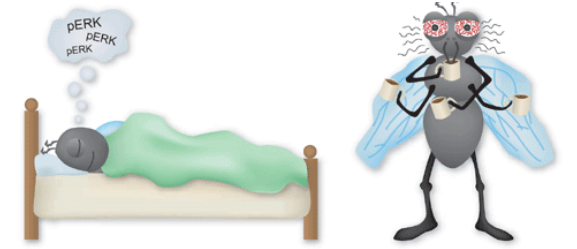
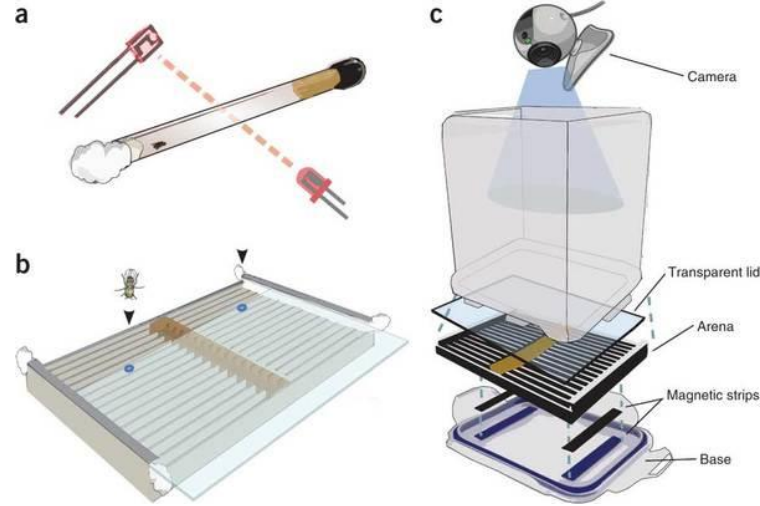
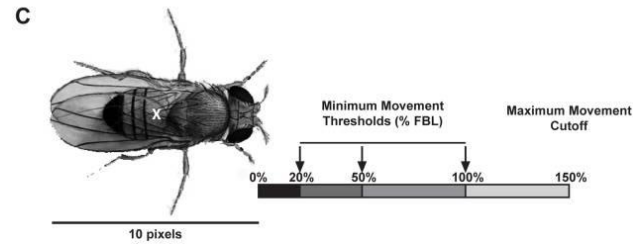
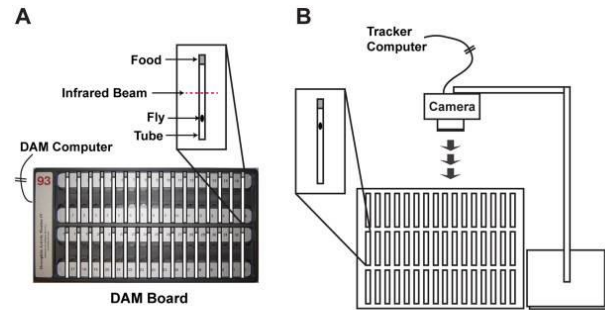
Behavioral paradigms: circadian rhythms & sleep



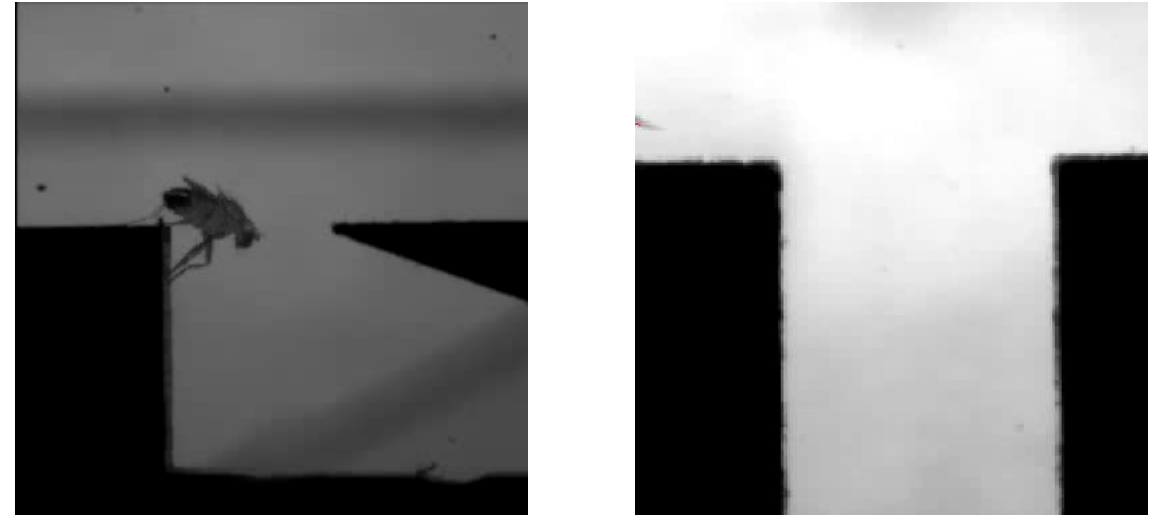
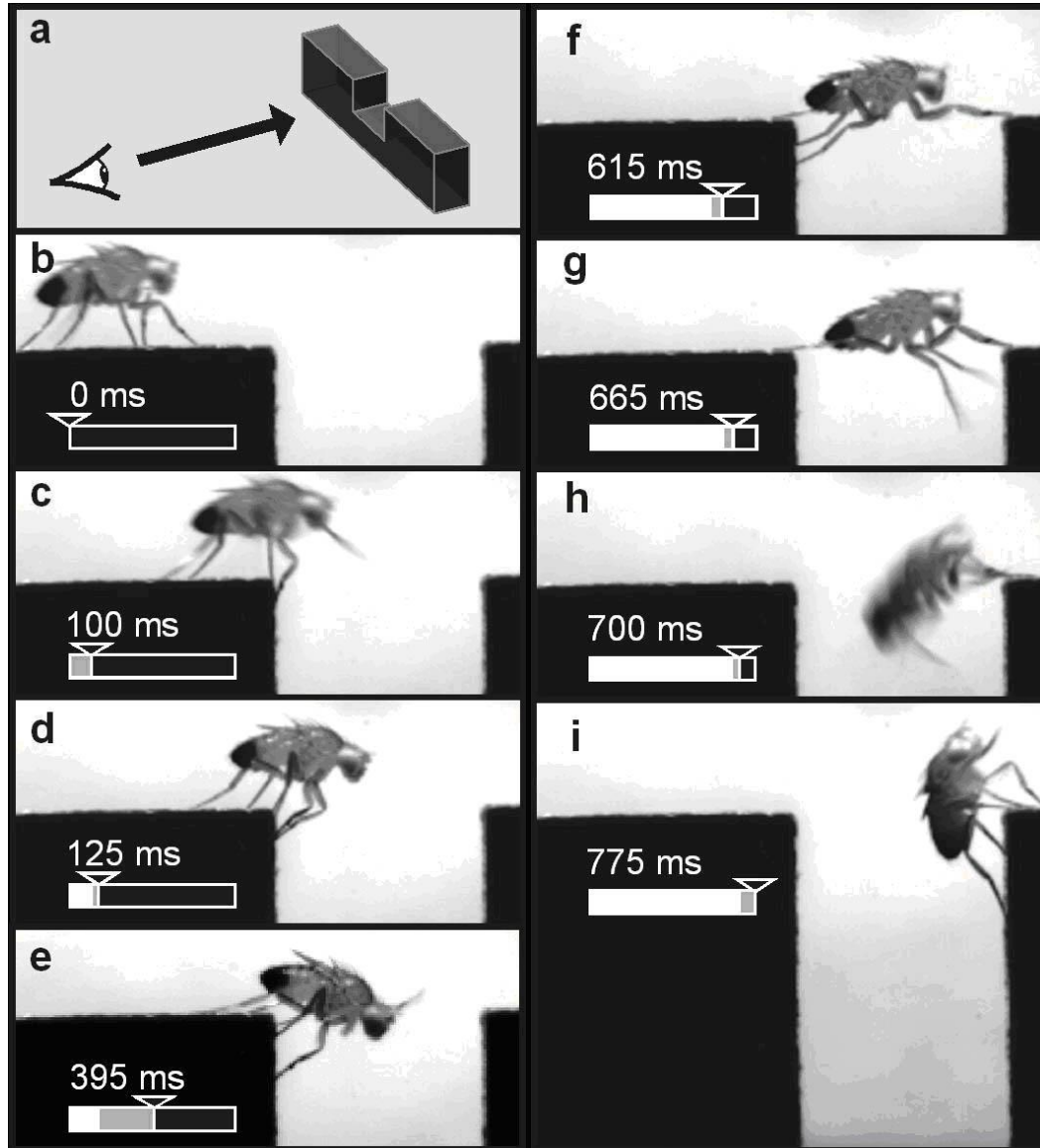
Ozgur Tataroglu & Patrick Emery, *Methods*, 2014, 140 - 150



Behavioral paradigms: circadian rhythms & sleep



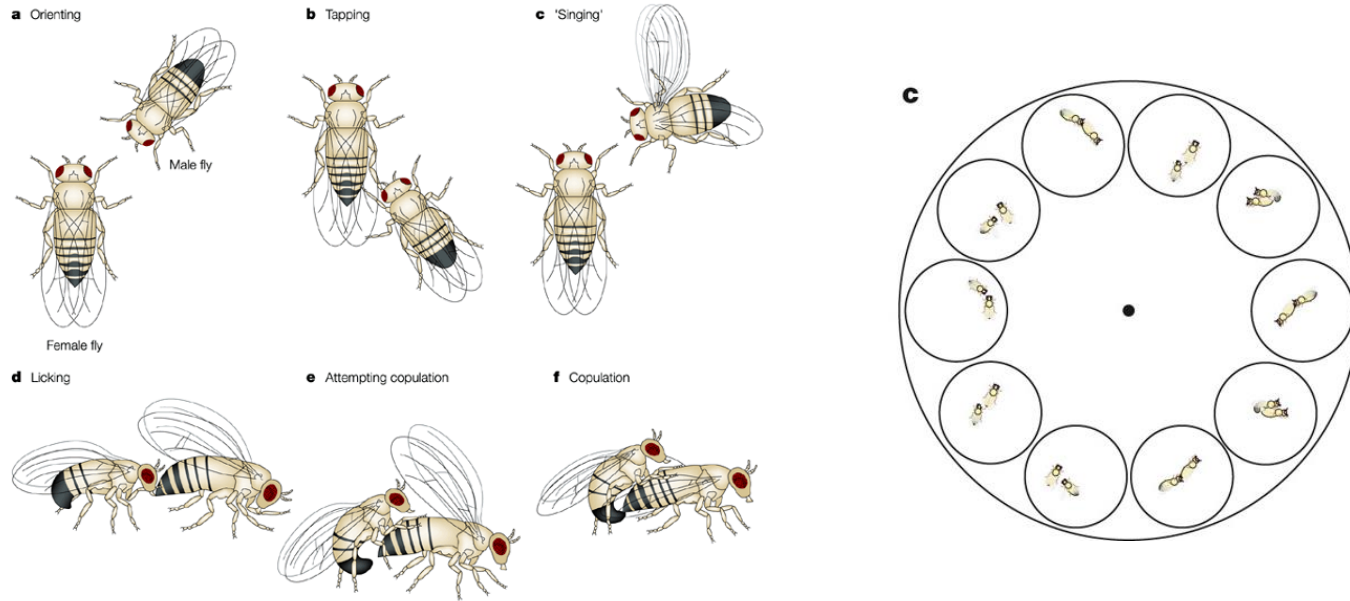
Behavioral paradigms: gap



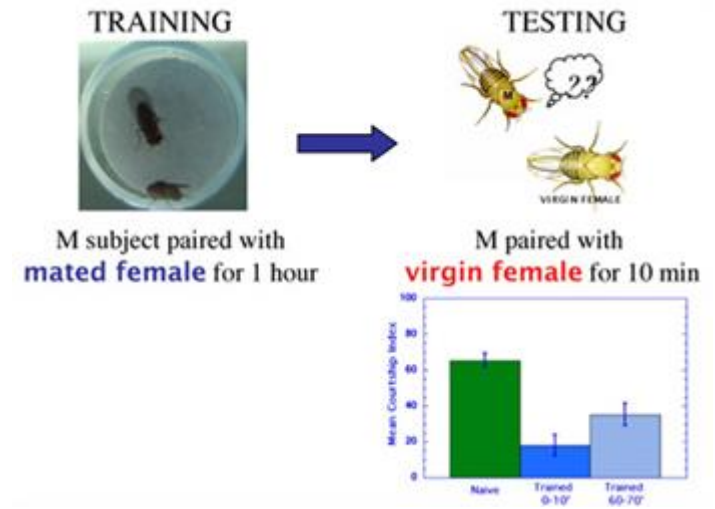
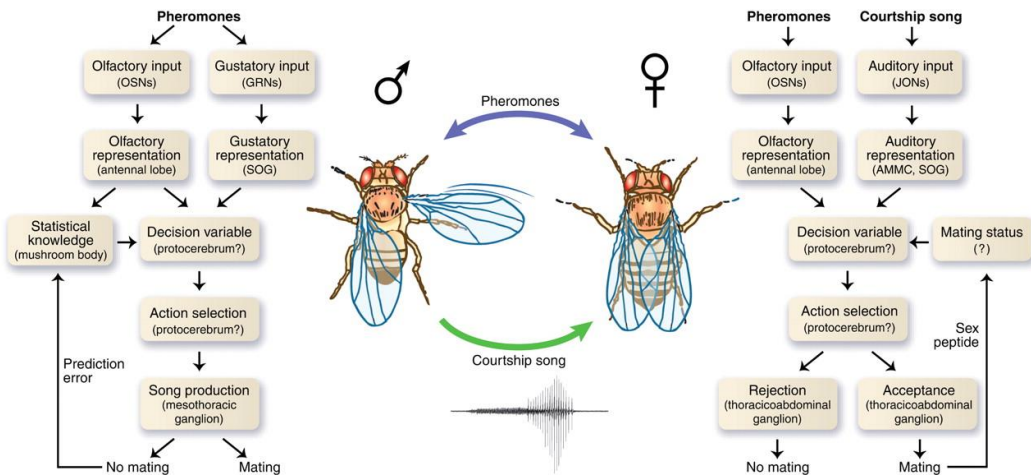
Motor coordination

Goal-directed actions

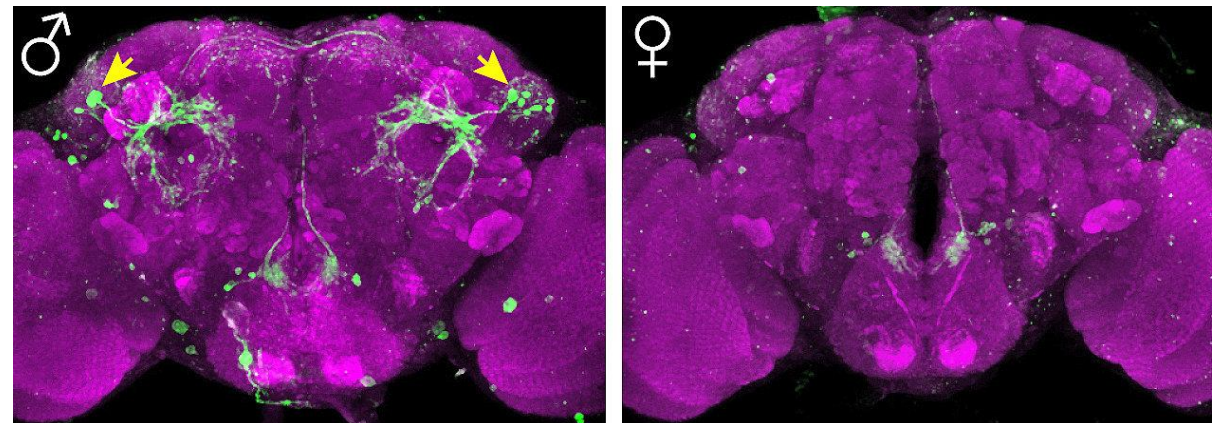
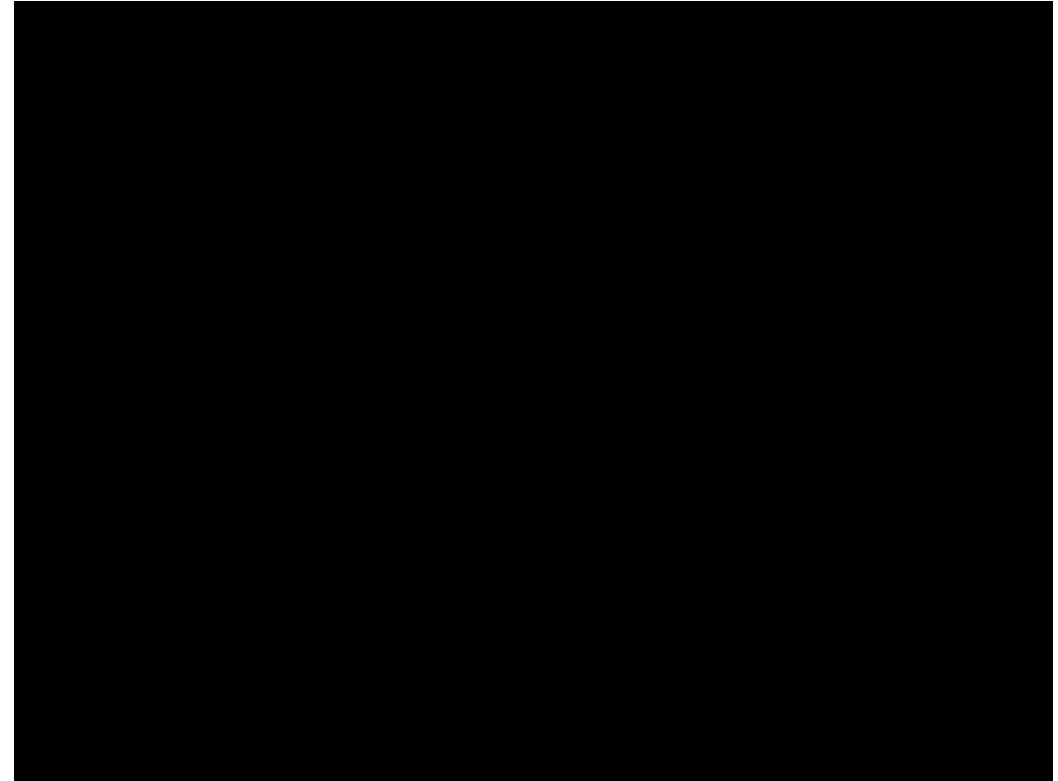
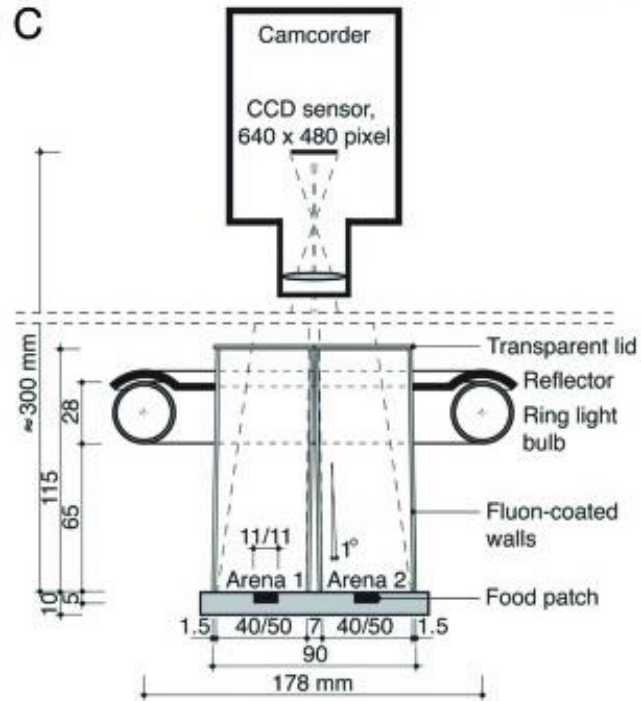
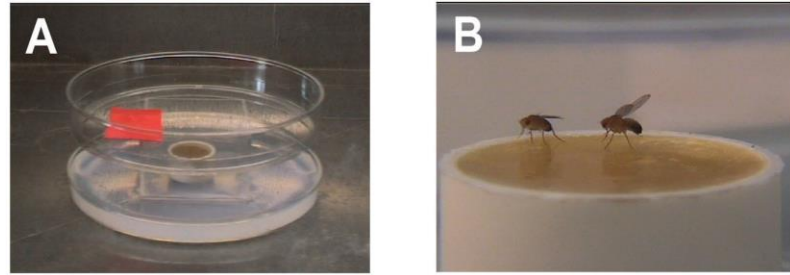
Behavioral paradigms: courtship



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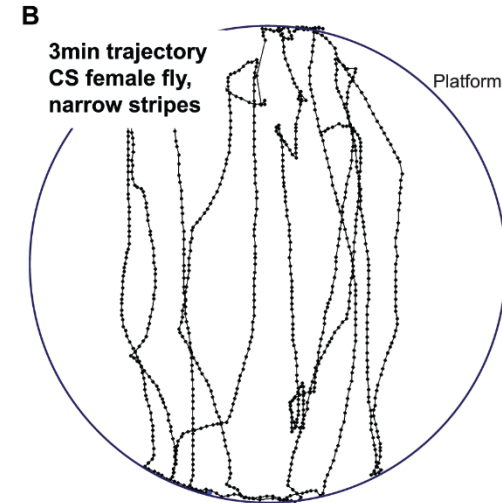
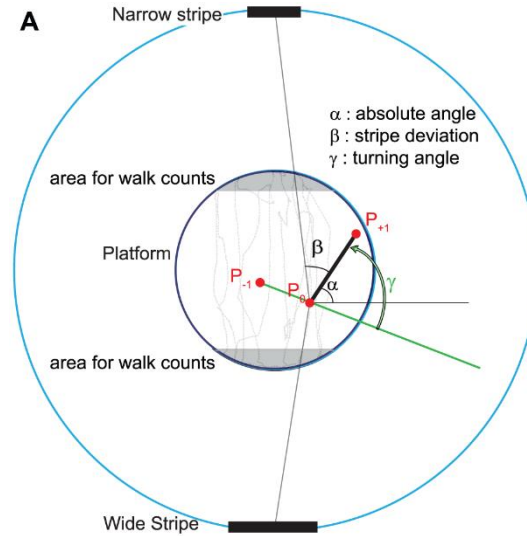
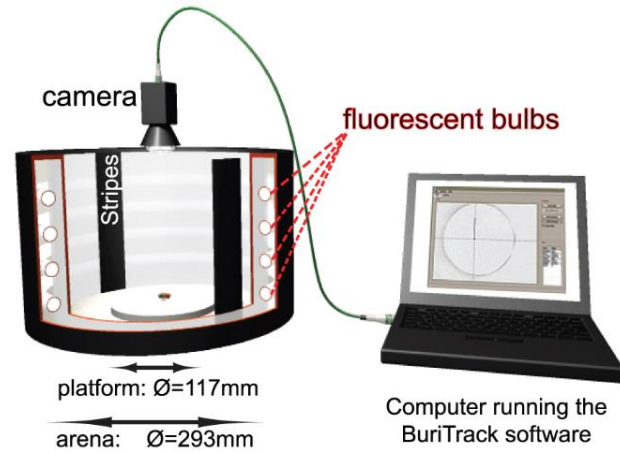


Behavioral paradigms: aggression

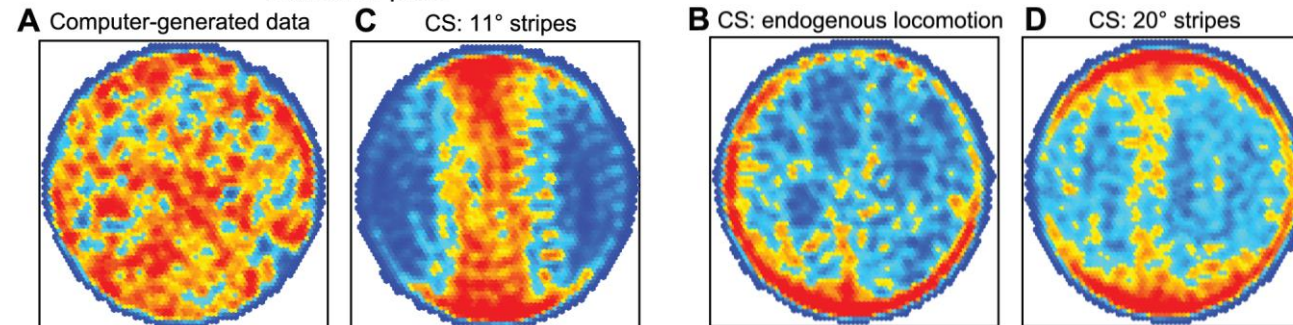


Tachykinin-expressing neurons

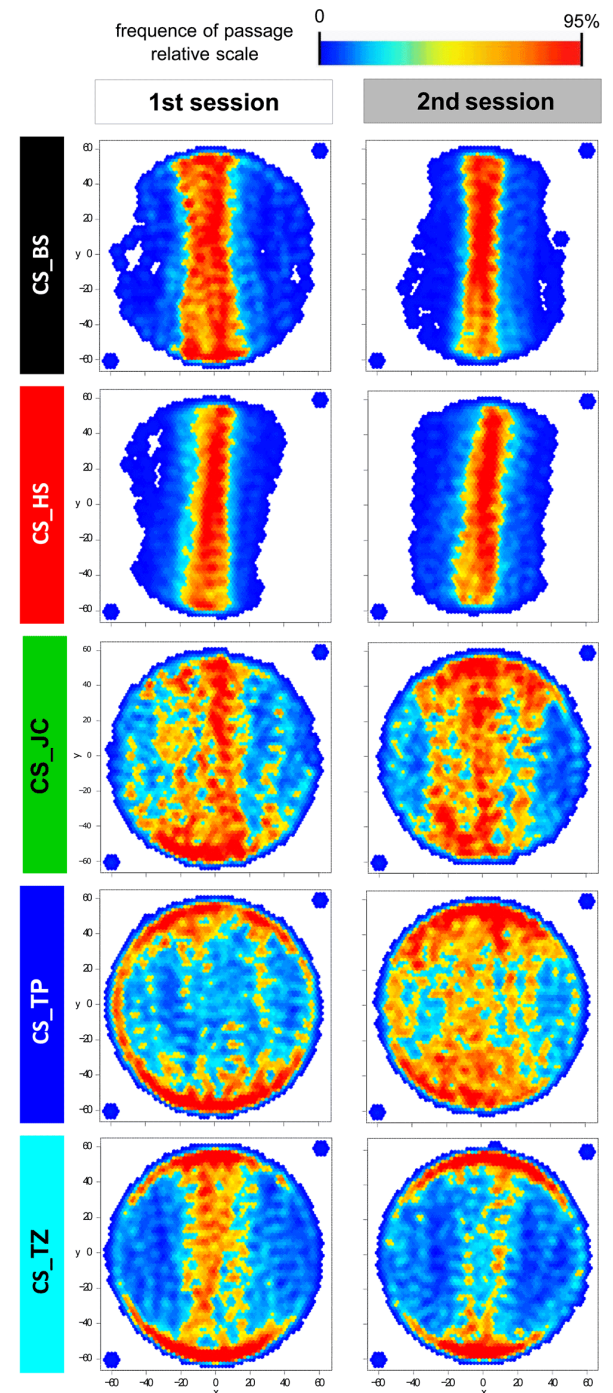
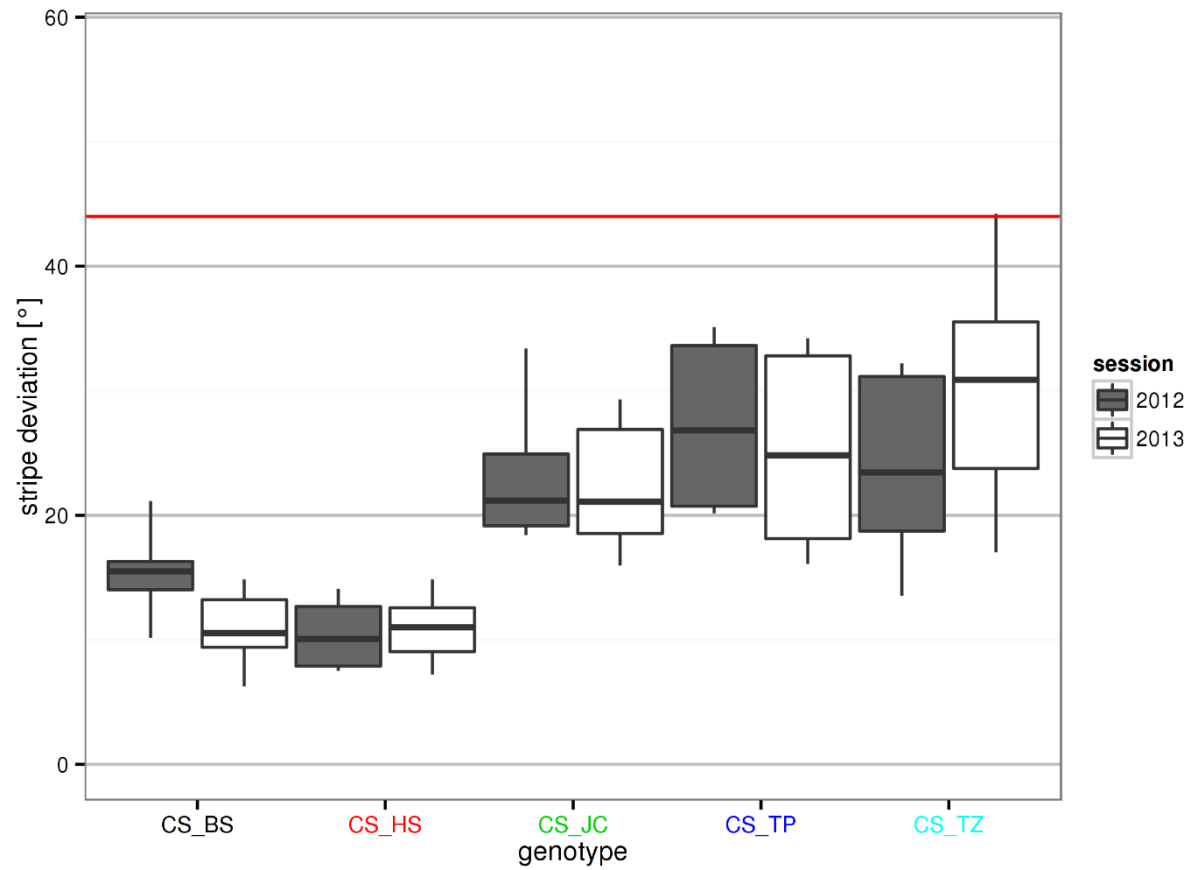
Behavioral paradigms: Buridan



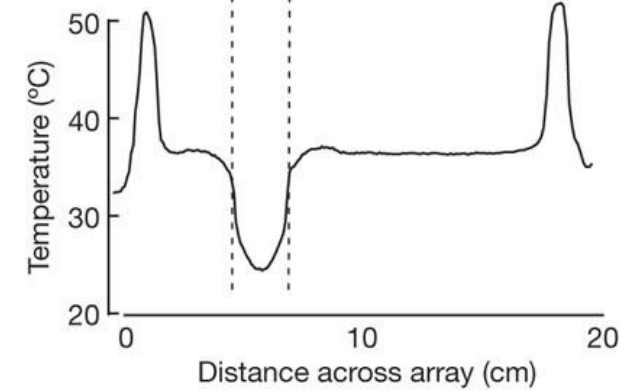
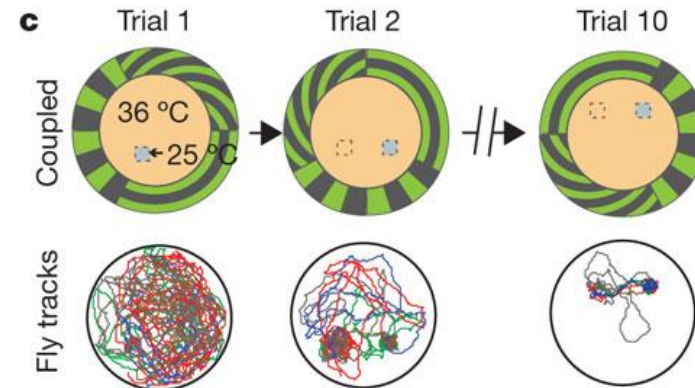
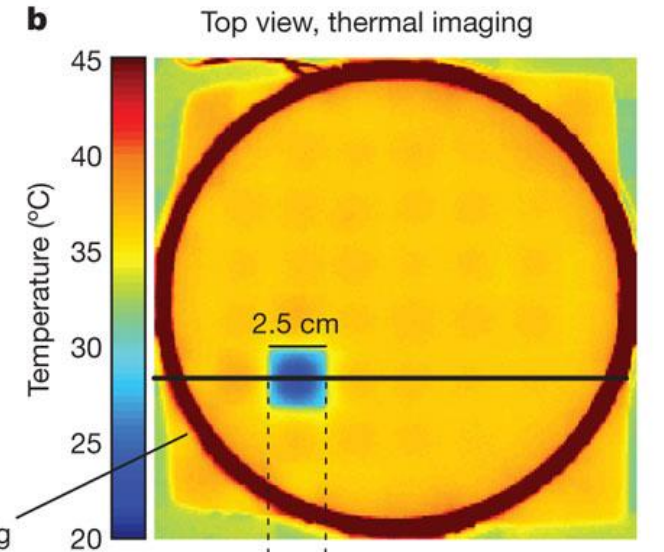
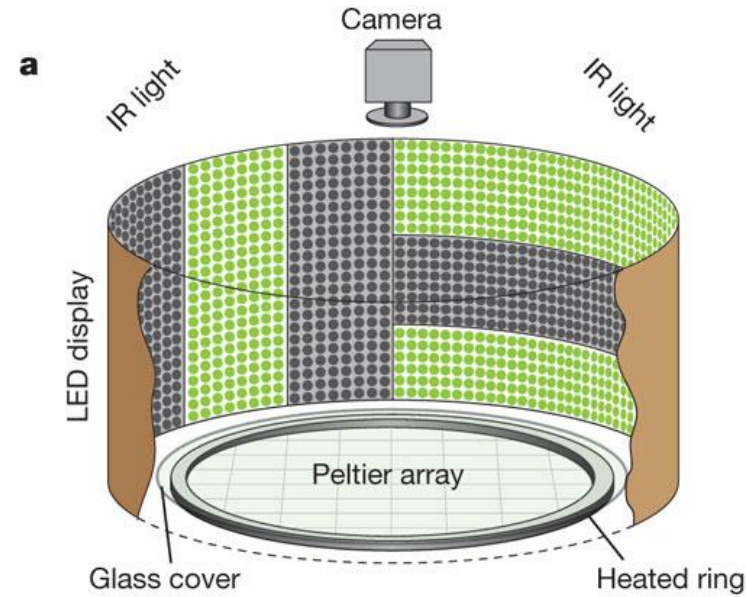
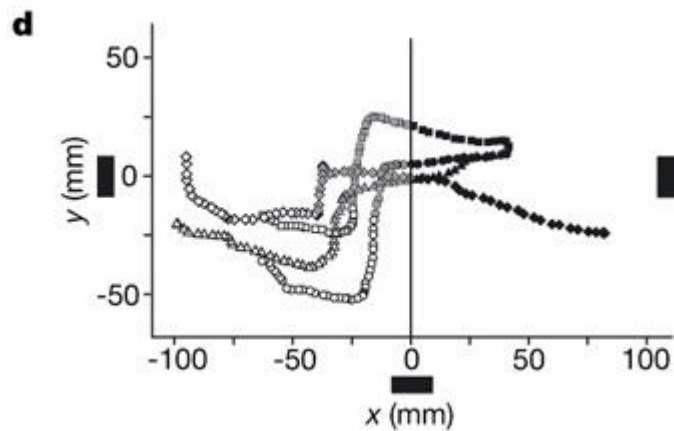
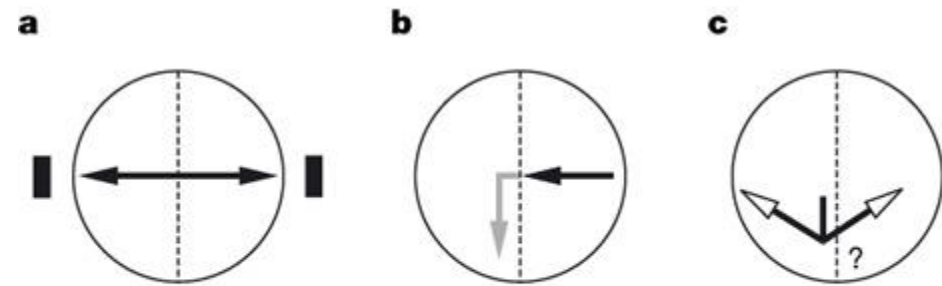
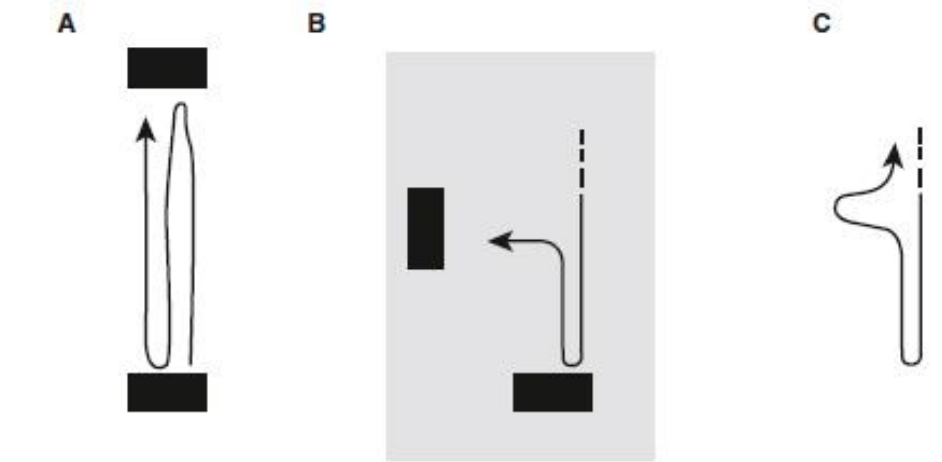
Transition plots



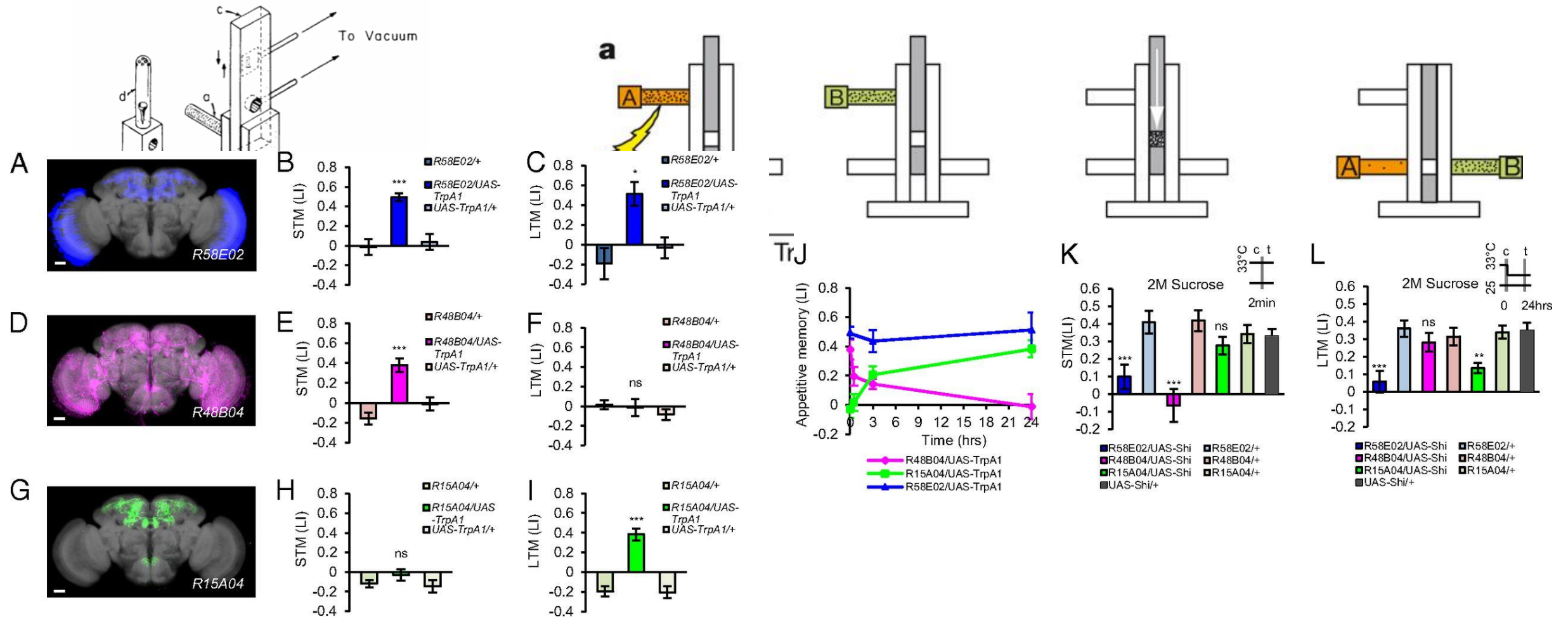
Behavioral paradigms: Buridan



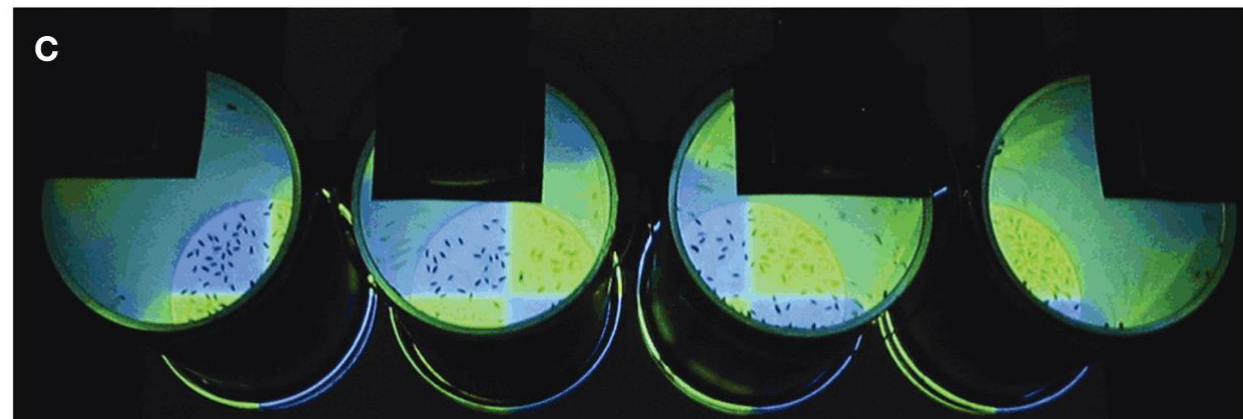
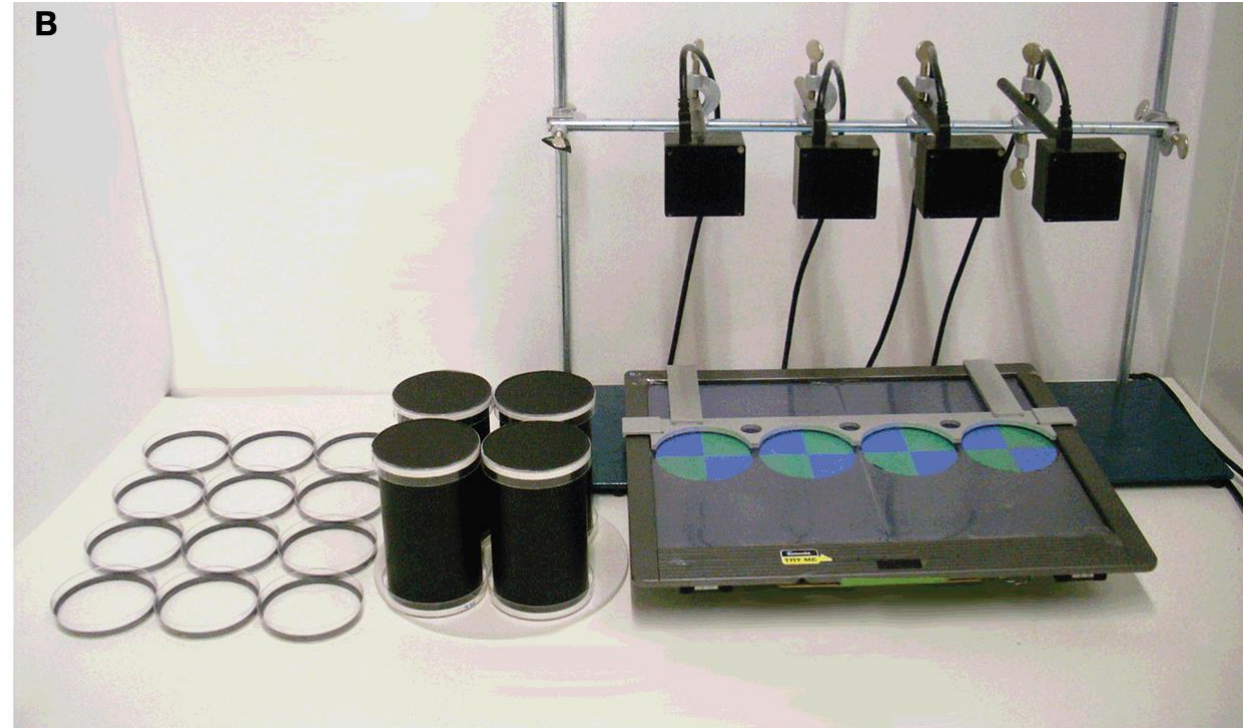
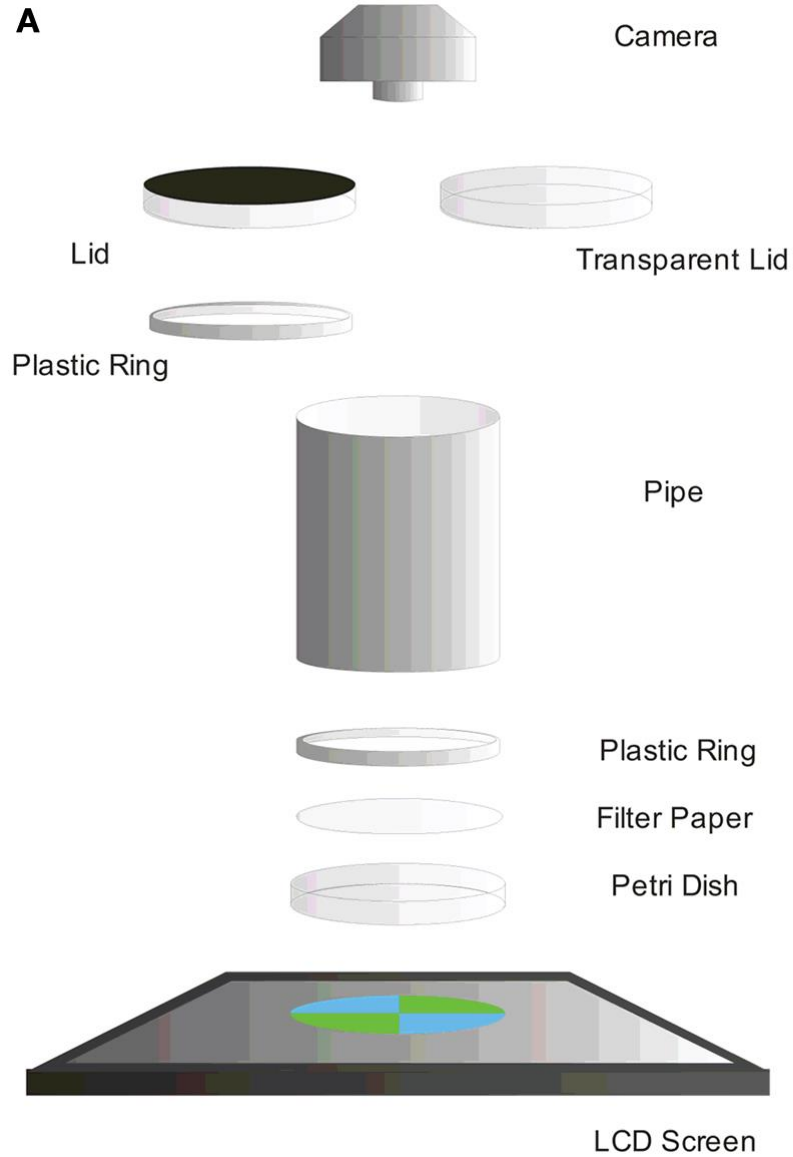
Behavioral paradigms: detour & visual place learning



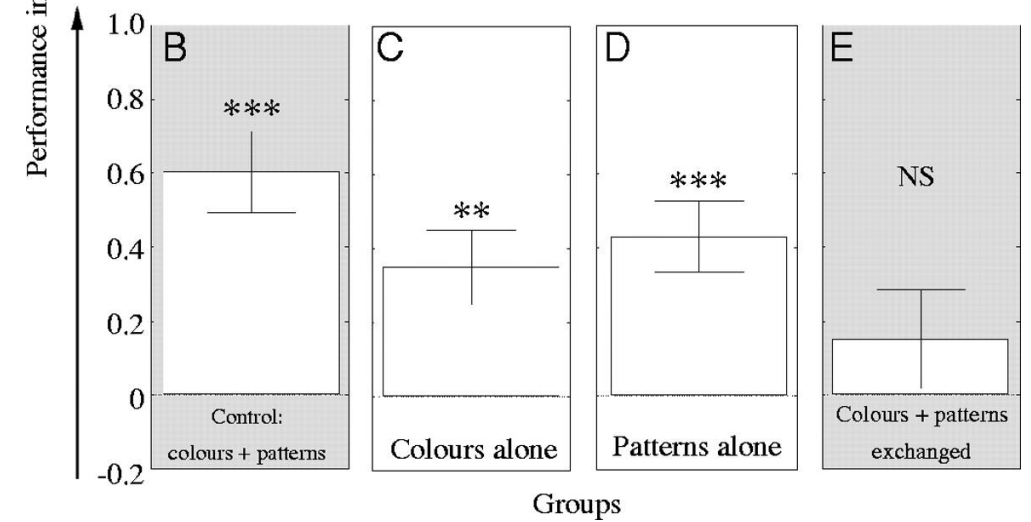
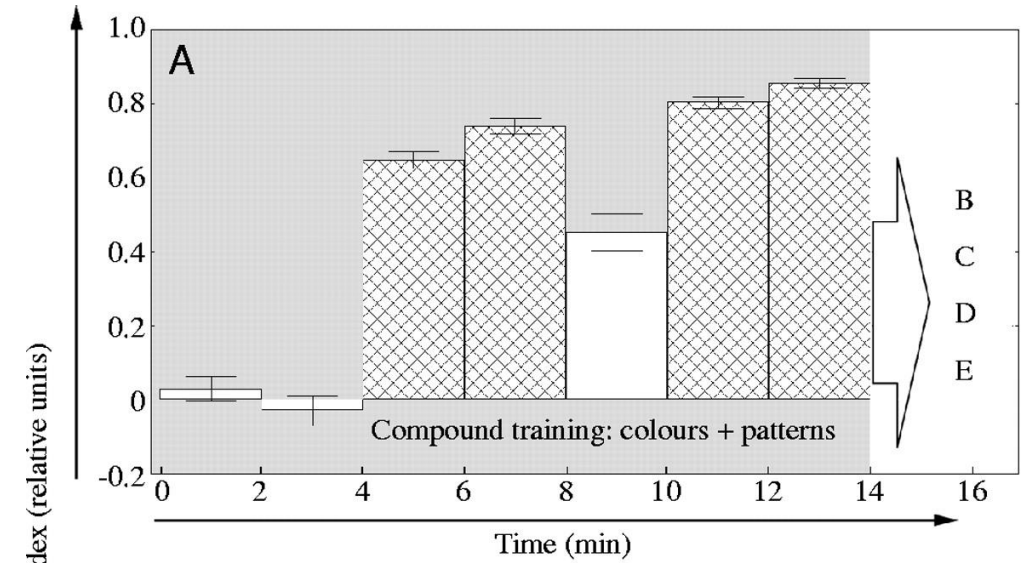
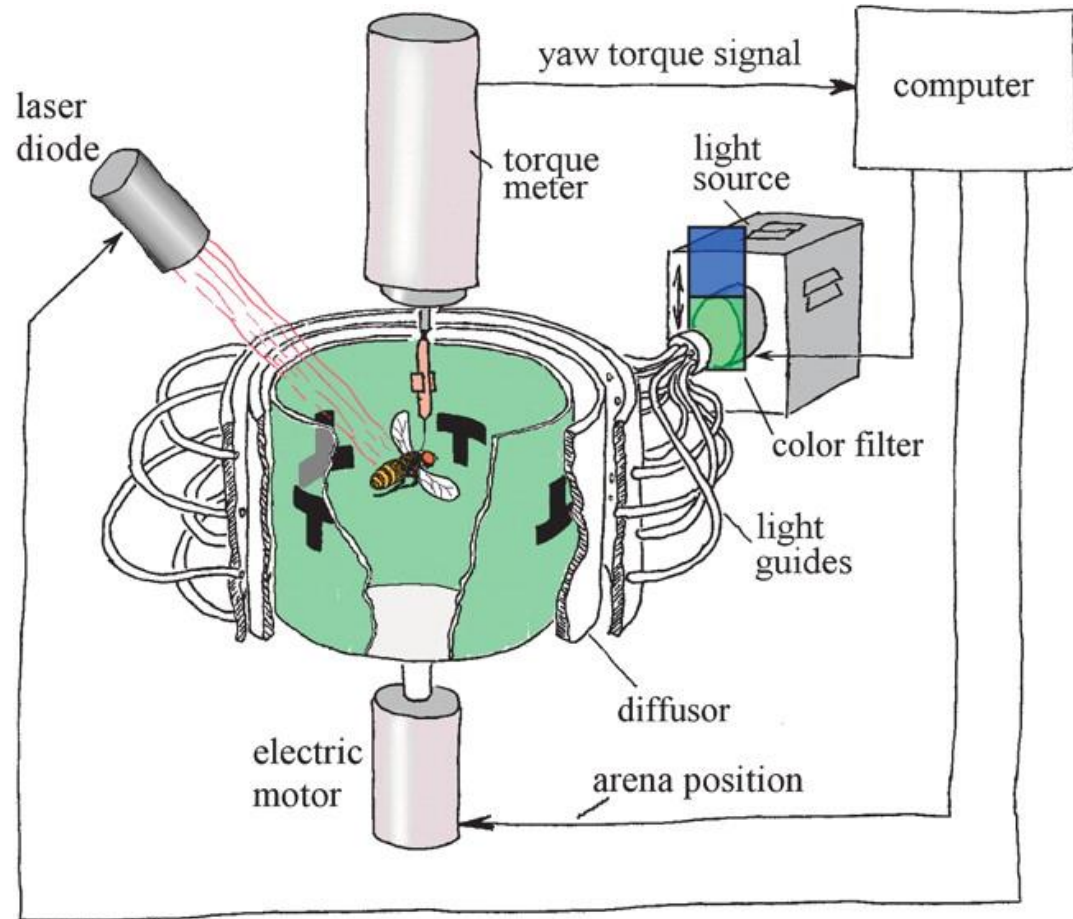
Behavioral paradigms: olfactory learning

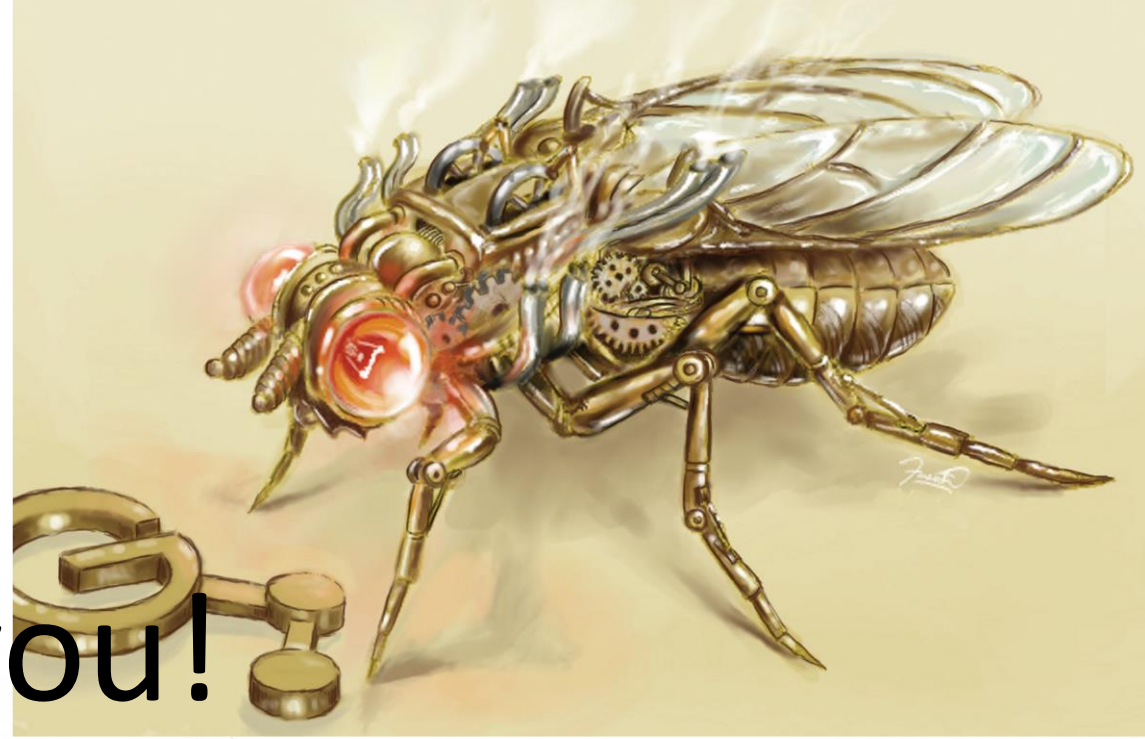
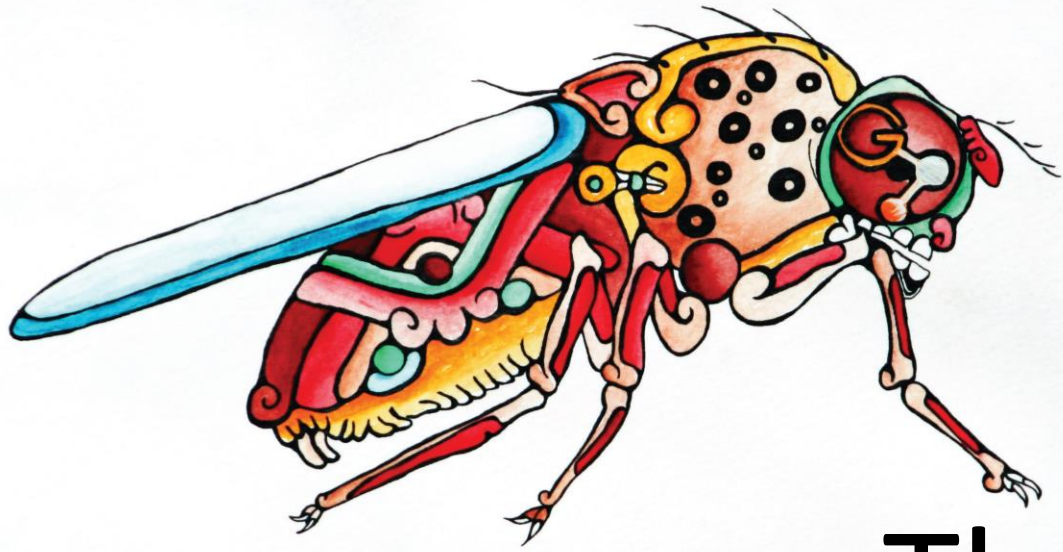


Behavioral paradigms: visual learning



Behavioral paradigms: flight simulator





Thank you!

