

# Haploid selection driving new genes male germline expression

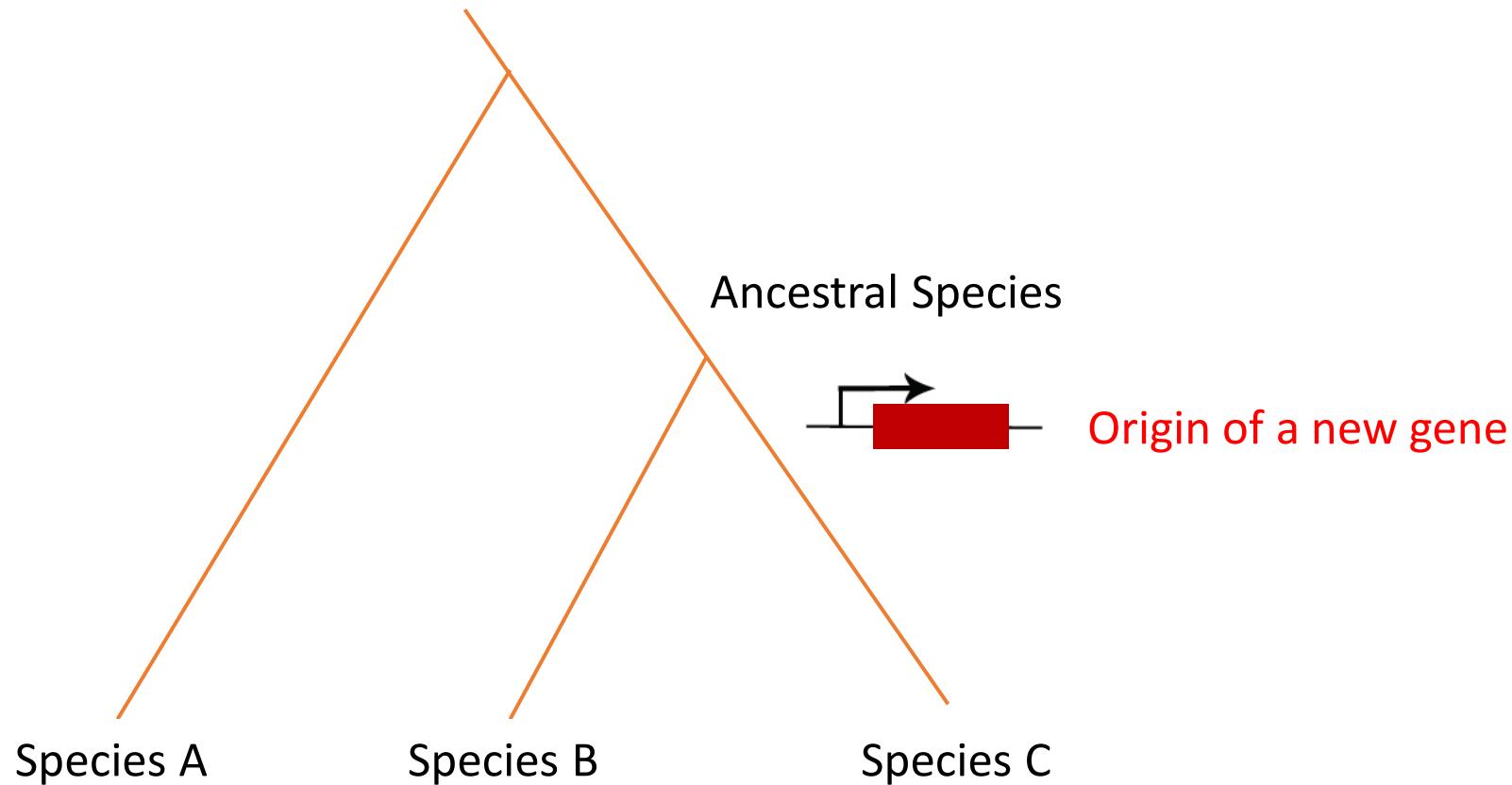


Maria Vibranovski  
University of São Paulo  
ALAG October 7<sup>th</sup> 2019

Photo: Camila Avelino

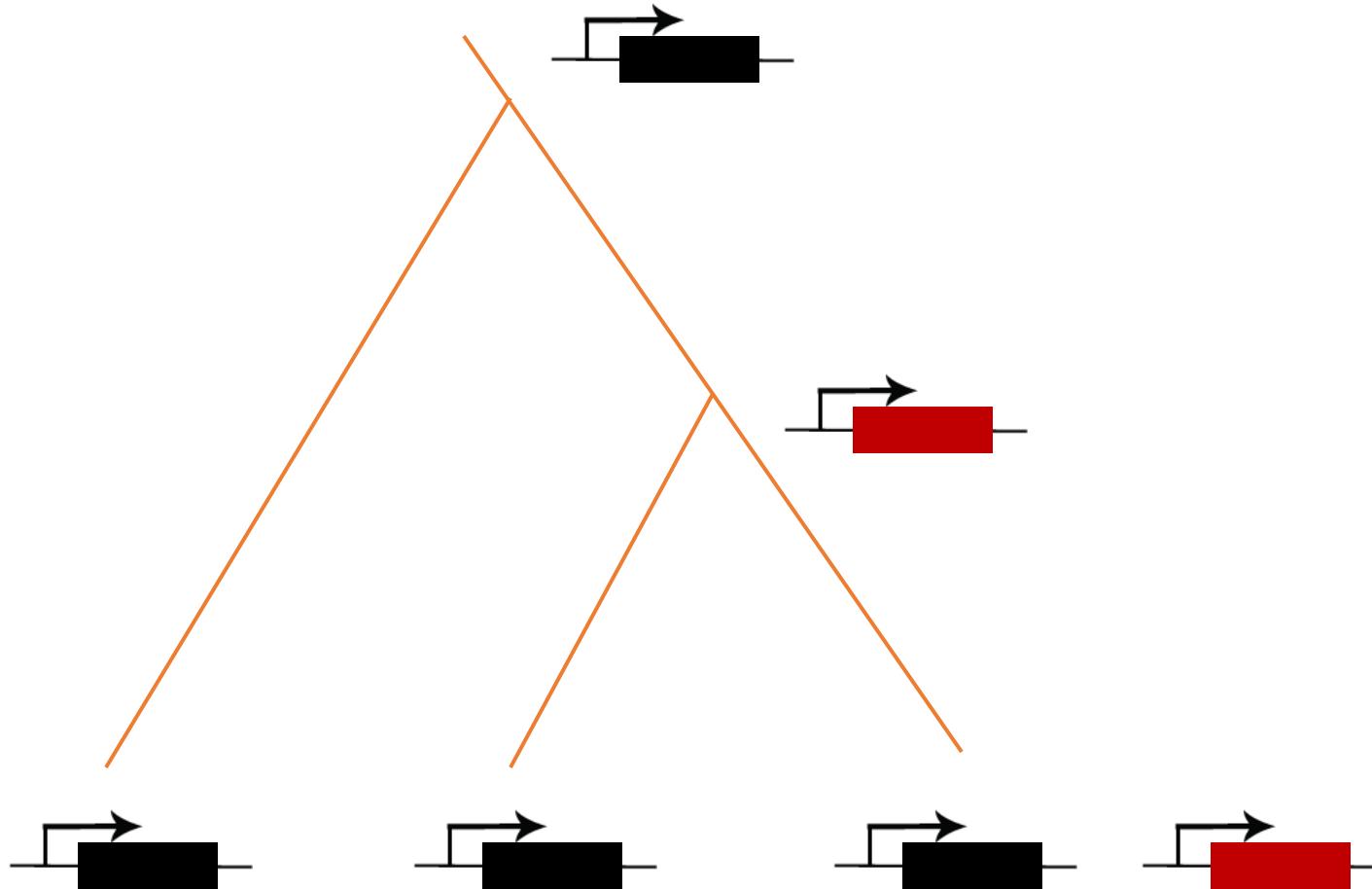
# New genes: origin

---



# Duplicated new genes: pseudogene or new function?

---



# New Gene?

# New Gene?

Innovation

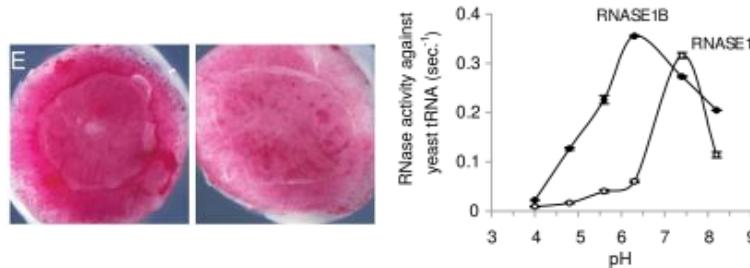
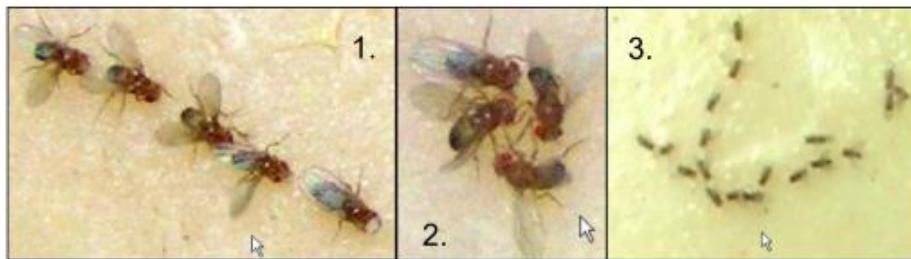
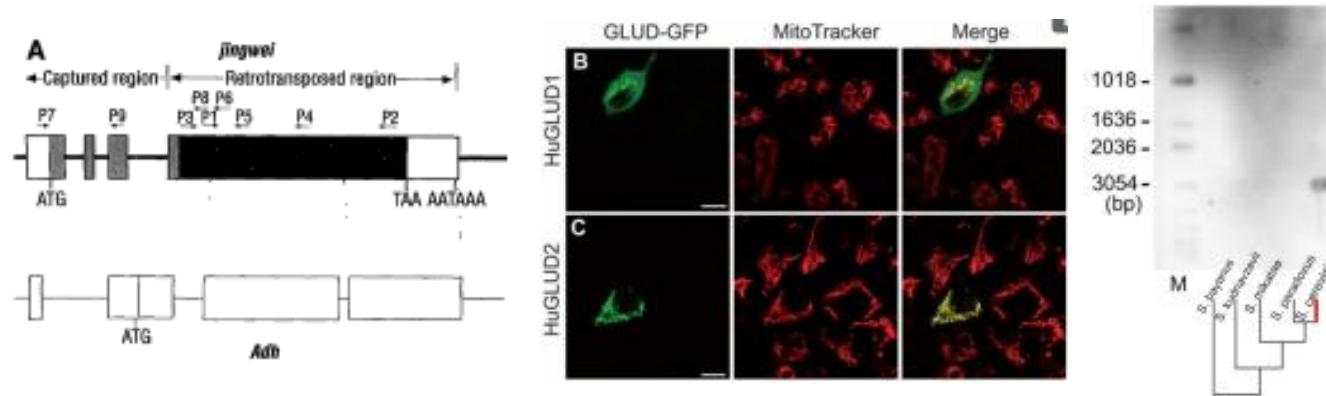
Adaptation

Neo-functionalization

New function

Positive Selection

# New genes: positive selection signature



Long & Langley, 1993. **Science**  
Dupressoir *et al.*, 2009. **PNAS**  
Cai *et al.*, 2008. **Genetics**  
Wang *et al.*, 2002. **PNAS**  
Rosso *et al.*, 2008. **Plos Genetics**  
Zhang *et al.*, 2002. **Nature Genetics**  
Johnson *et al.*, 2001. **Nature**  
Malik & Henikoff, 2005. **Plos Genetics**

# New Gene?

Innovation

Adaptation

Neo-functionalization

New function

Positive Selection

Testis expression

Male germline

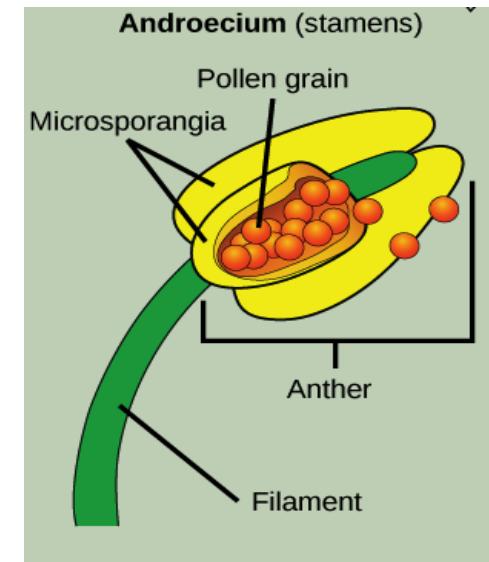
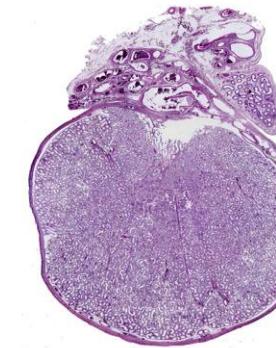
Spermatogenesis

# New genes: testis expression

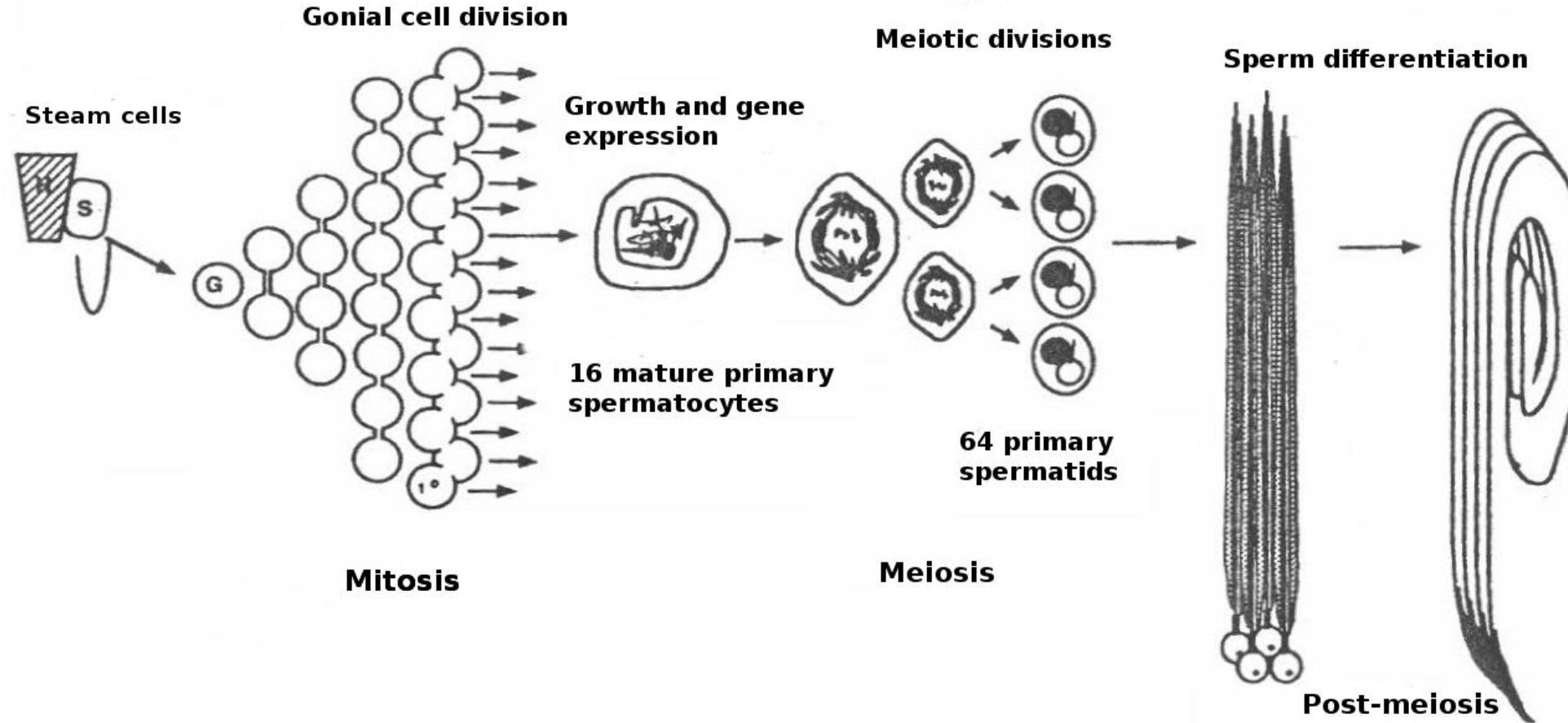
---



Babushok *et al.*, 2007. **Genome Research**  
Levine *et al.*, 2006. **PNAS**  
Betran *et al.*, 2002. **Genome Research**  
Marques *et al.*, 2005. **PLOS Biology**  
Soumillon *et al.*, 2013. **Cell Reports**  
Vibranski *et al.*, 2009. **PLOS Genetics**  
Cui *et al.*, 2015. **Molecular Plant**



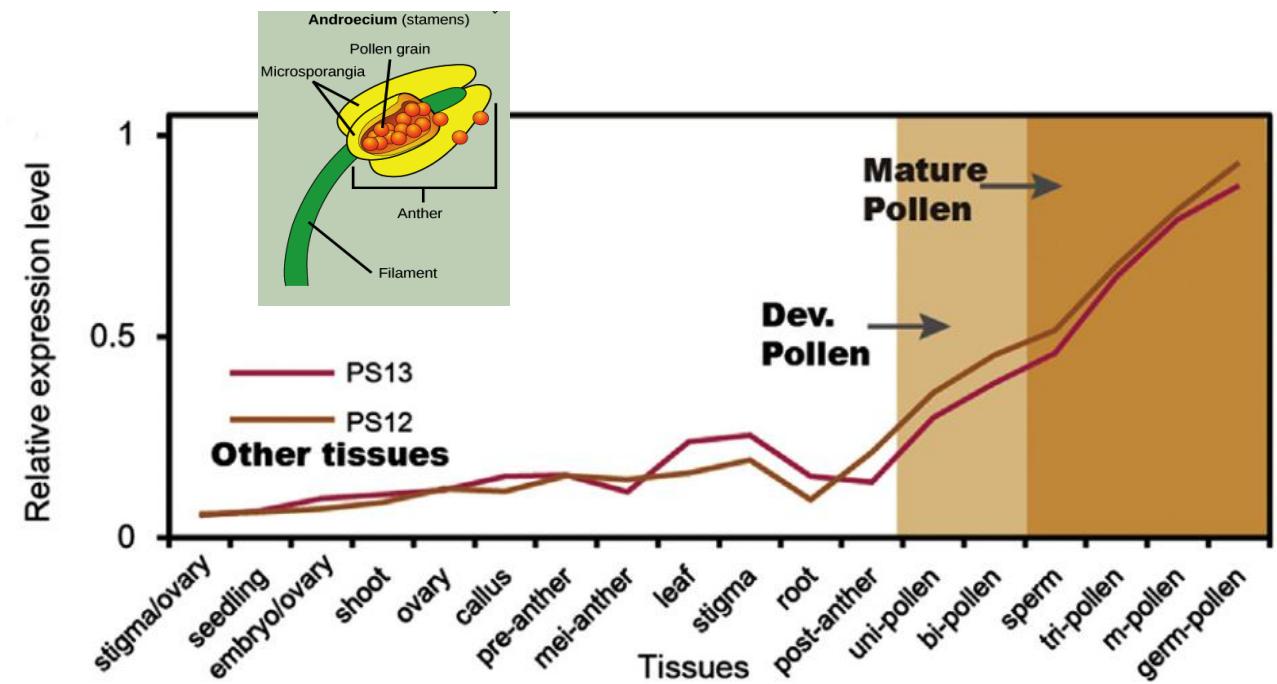
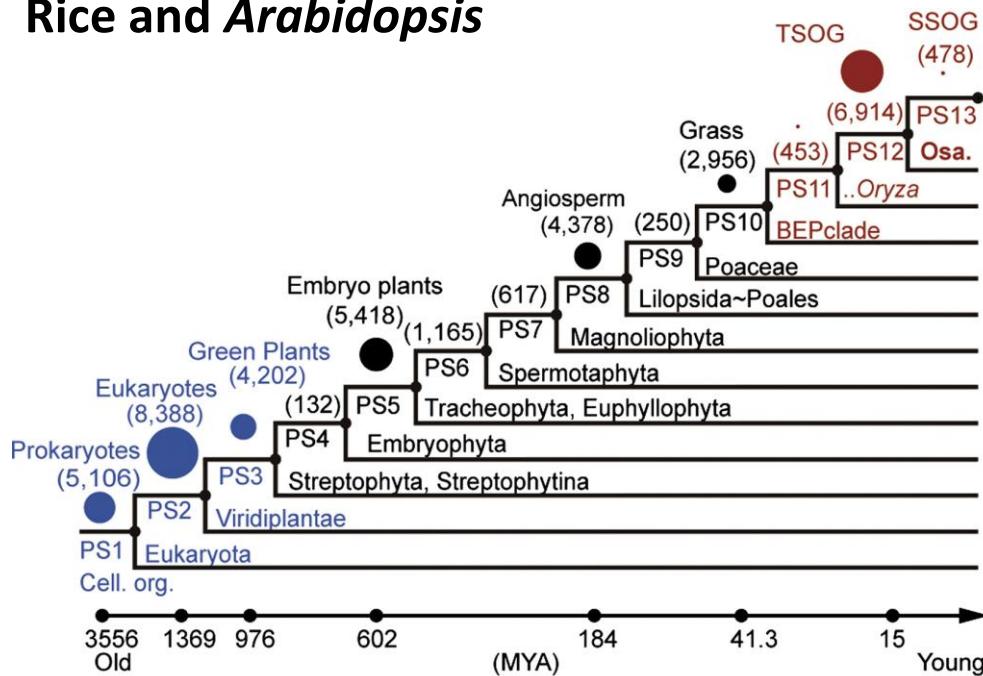
# New genes: spermatogenesis



Fuller, 1993. in *The Development of Drosophila melanogaster*

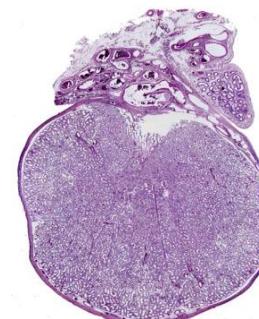
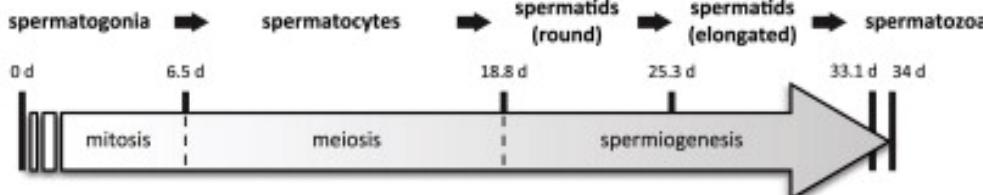
# New genes: later phases of spermatogenesis

## Rice and *Arabidopsis*



Cui et al., 2015. Molecular Plant

## Mouse:



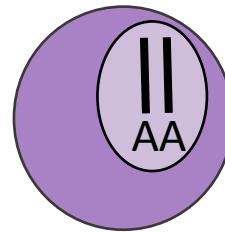
RETROGENES (201)								
	br	lv	ts	sl	sg	sc	sd	sz
Median of expression	0.53	1.7	18.7	1.85	1	29.1	51.3	1.34

Soumillon et al., 2013 Cell Rep

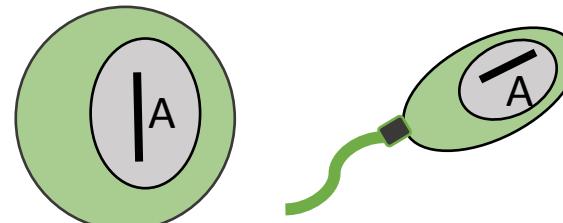
# Haploid Selection and the Origin of New Genes

---

Diploid expression



Haploid expression



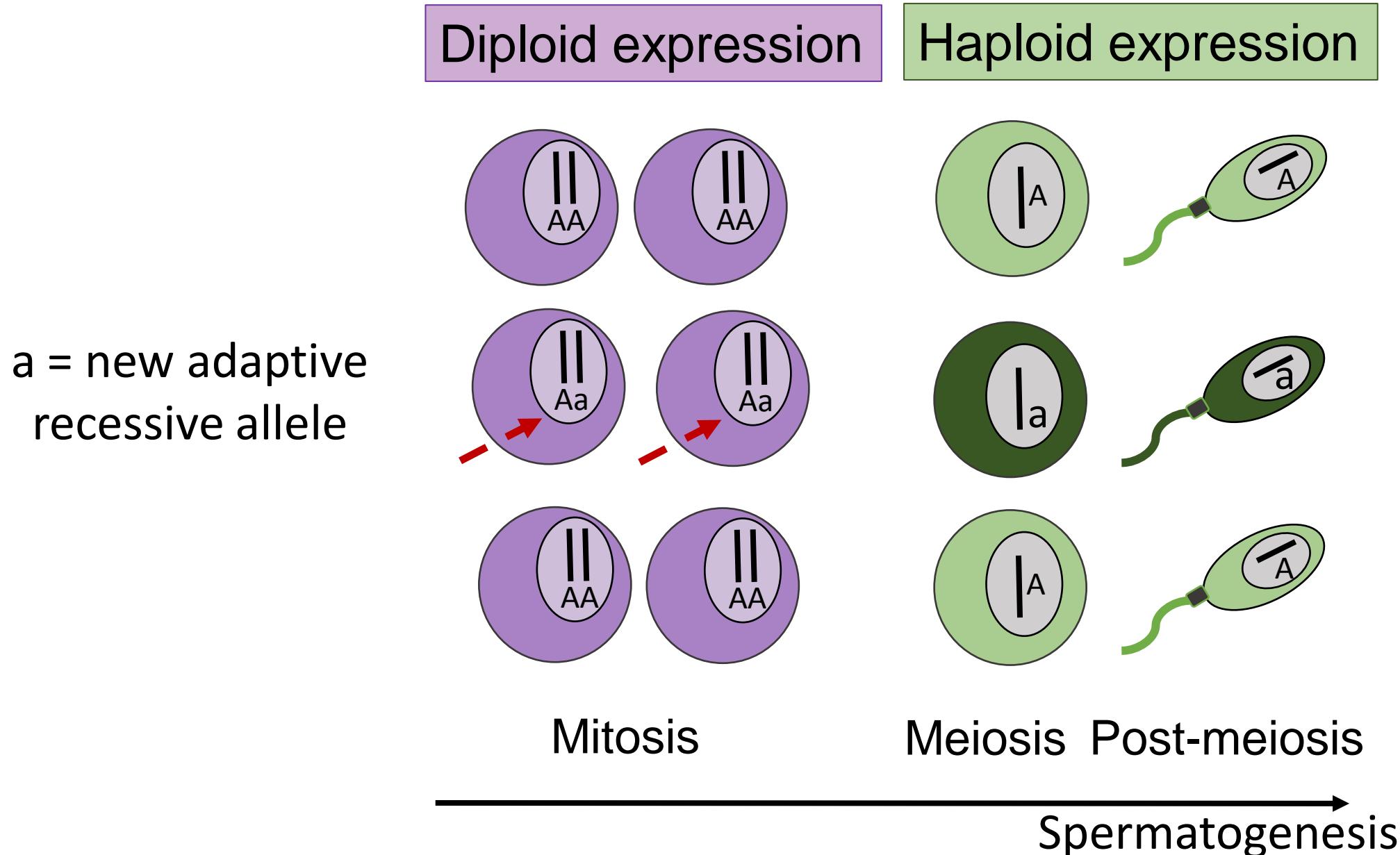
Mitosis

Meiosis Post-meiosis



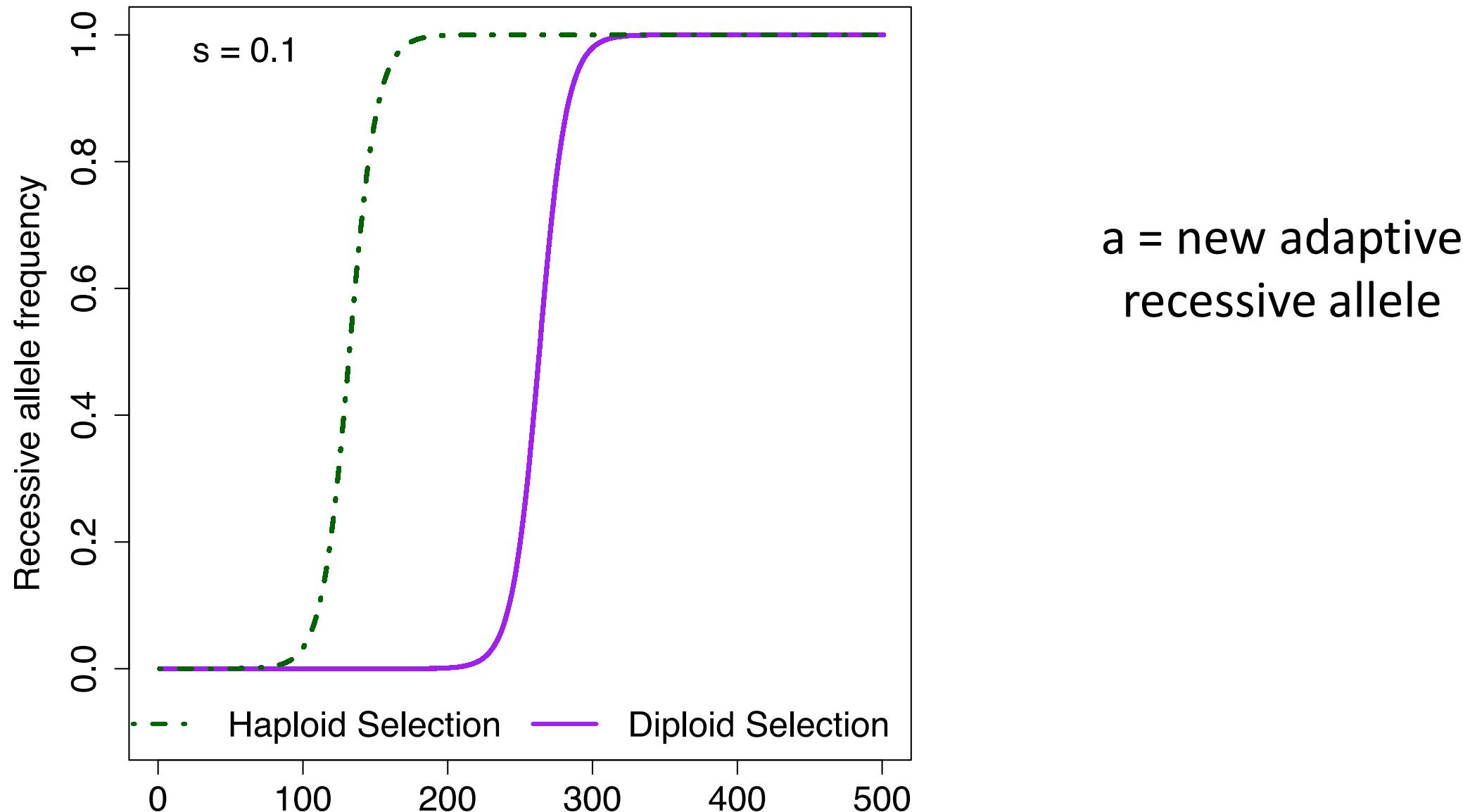
Spermatogenesis

# Haploid Selection and the Origin of New Genes



# Haploid Selection and the Origin of New Genes

---



# Haploid Selection and the Origin of New Genes

---

## Model Predictions:

- 1) New genes should be more expressed in the later phases of spermatogenesis
- 2) Genes expressed in the haploid phases of spermatogenesis should be enriched with positive selection signature
- 3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis

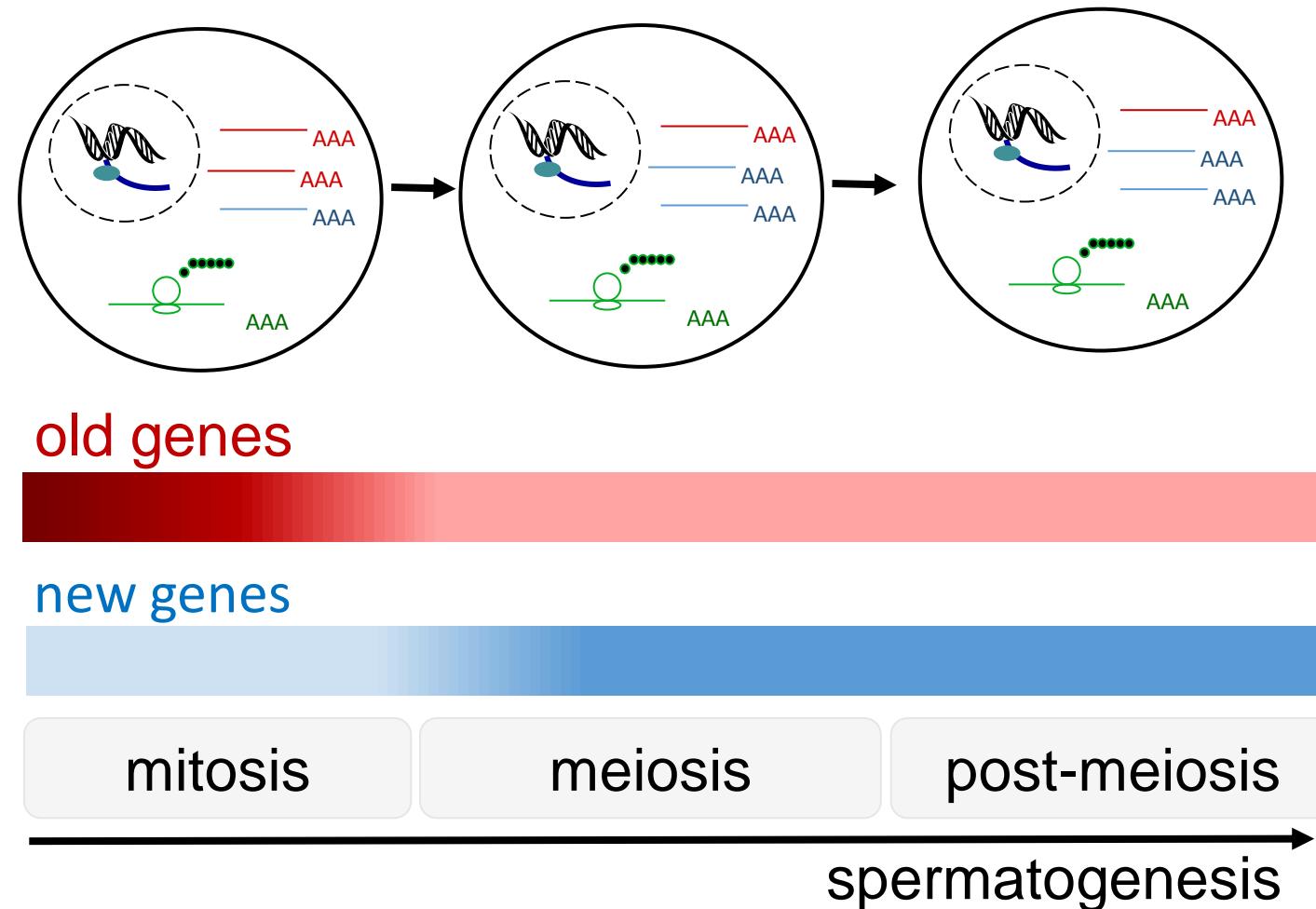
# Haploid Selection and the Origin of New Genes

---

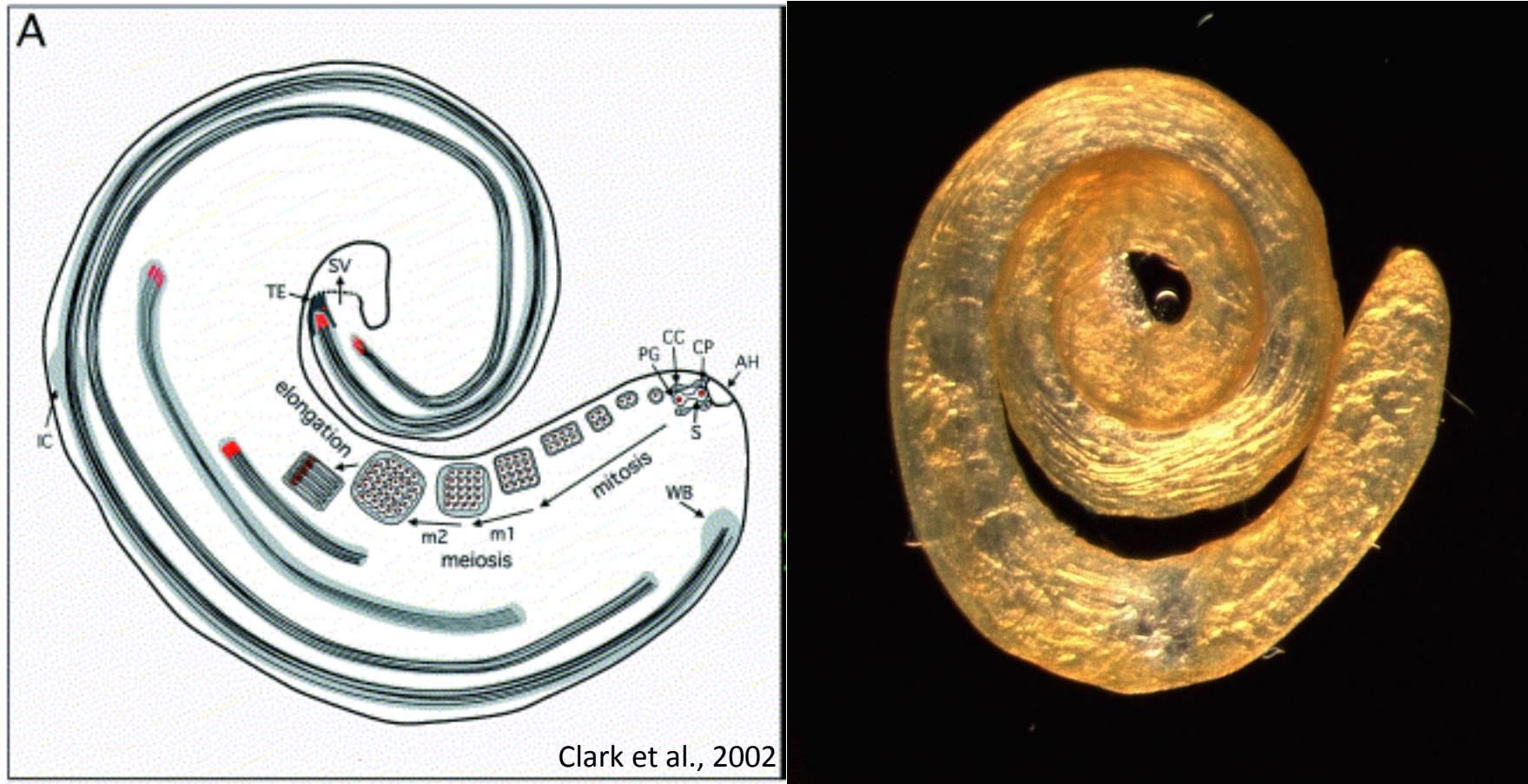
## Model Predictions:

- 1) New genes should be more expressed in the later phases of spermatogenesis
- 2) Genes expressed in the haploid phases of spermatogenesis should be enriched with positive selection signature
- 3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis

# 1) New Genes should be more expressed in later phase of spermatogenesis

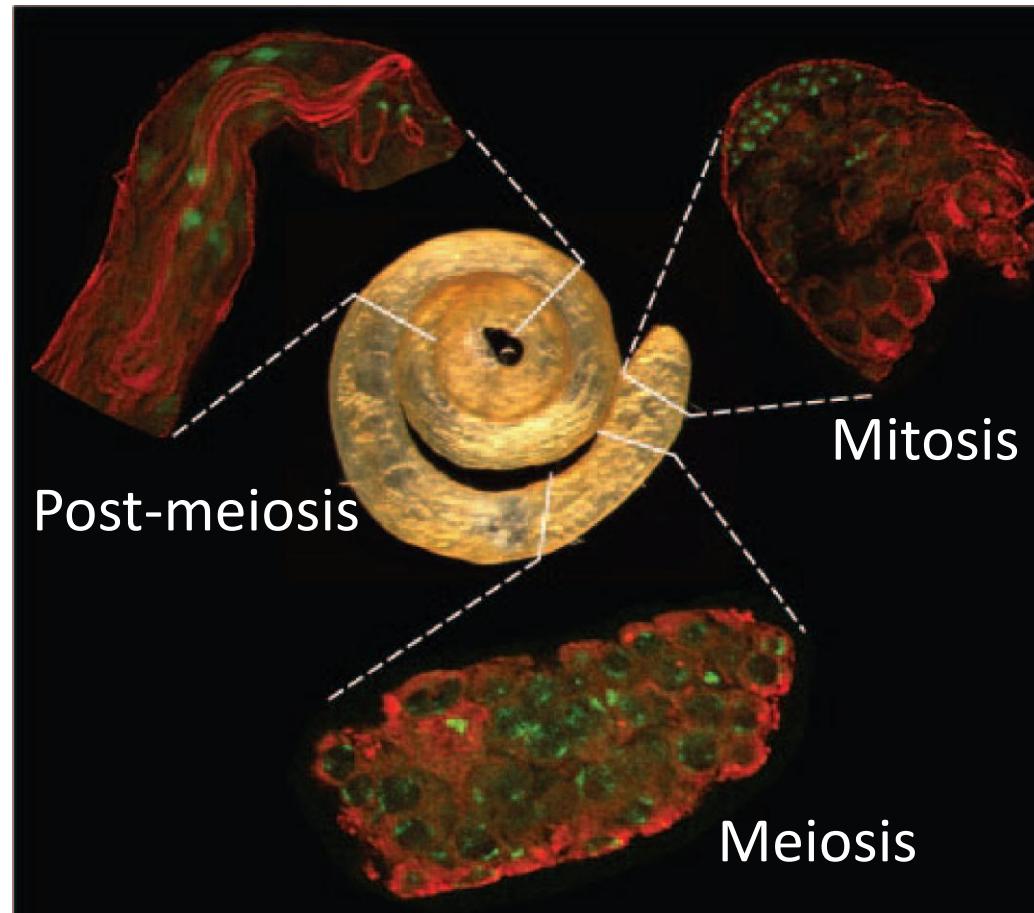


# *Drosophila* Spermatogenesis expression: Vibranovski et al. 2009



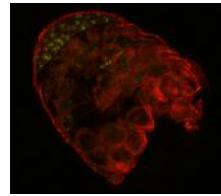
# *Drosophila* Spermatogenesis expression: Vibranovski et al. 2009

---



# *Drosophila* Spermatogenesis expression: Vibranovski et al. 2009

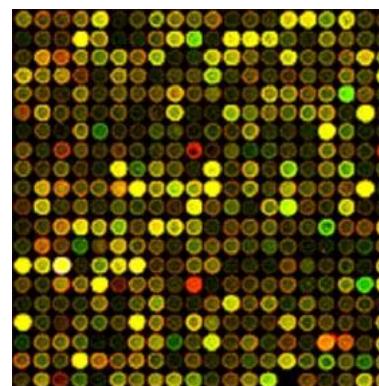
Tissue Isolation (n=3)



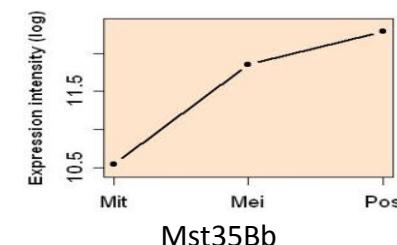
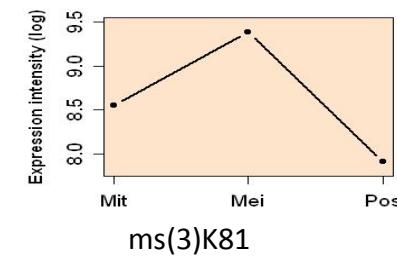
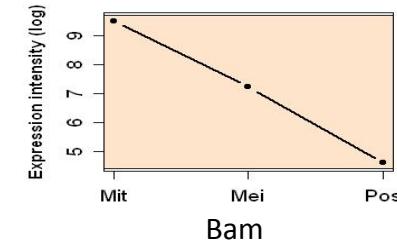
RNA extraction



Microarray Hybridization



Gene profile



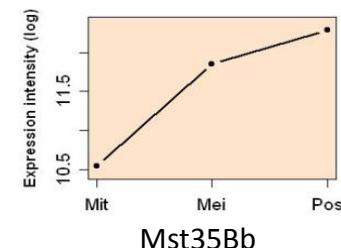
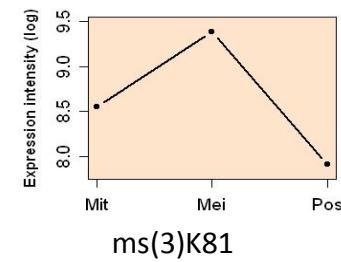
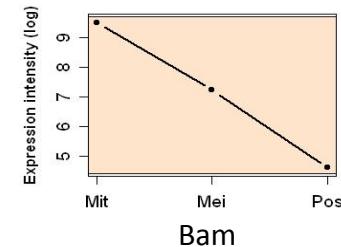
# *Drosophila* Spermatogenesis expression: Vibranovski et al. 2009

## Statistical Analyses

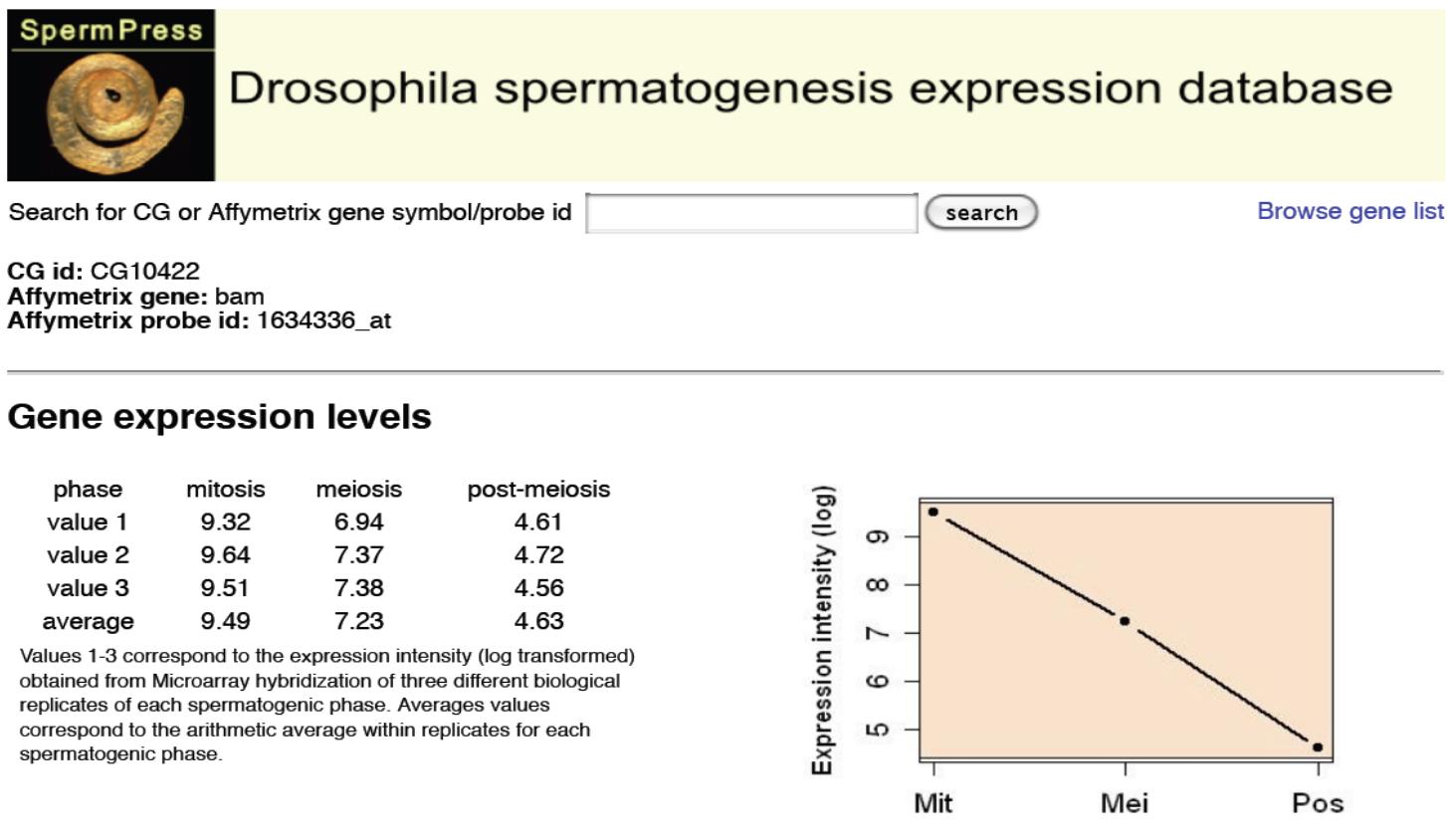
### Bayesian Model

- Avoids the arbitrariness of fold-thresholds
- Avoids multiple testing distortions

Gene profile



# *Drosophila* Spermatogenesis expression: Vibranovski *et al.* 2009



### Comparison of expression levels

phase	mitosis	meiosis	post-meiosis
mitosis	-	Over	Over
meiosis	-	-	Over
post-meiosis	-	-	-

The classifications in this table (Equal, Over or Under) were obtained by our Bayesian Statistical Model (Methods). Each classification is given to a pair-wise comparison between two spermatogenic phases. For example, "Over" mitotic vs. meiotic classification means that, for this gene, the mitotic expression is significantly higher than the meiotic expression.

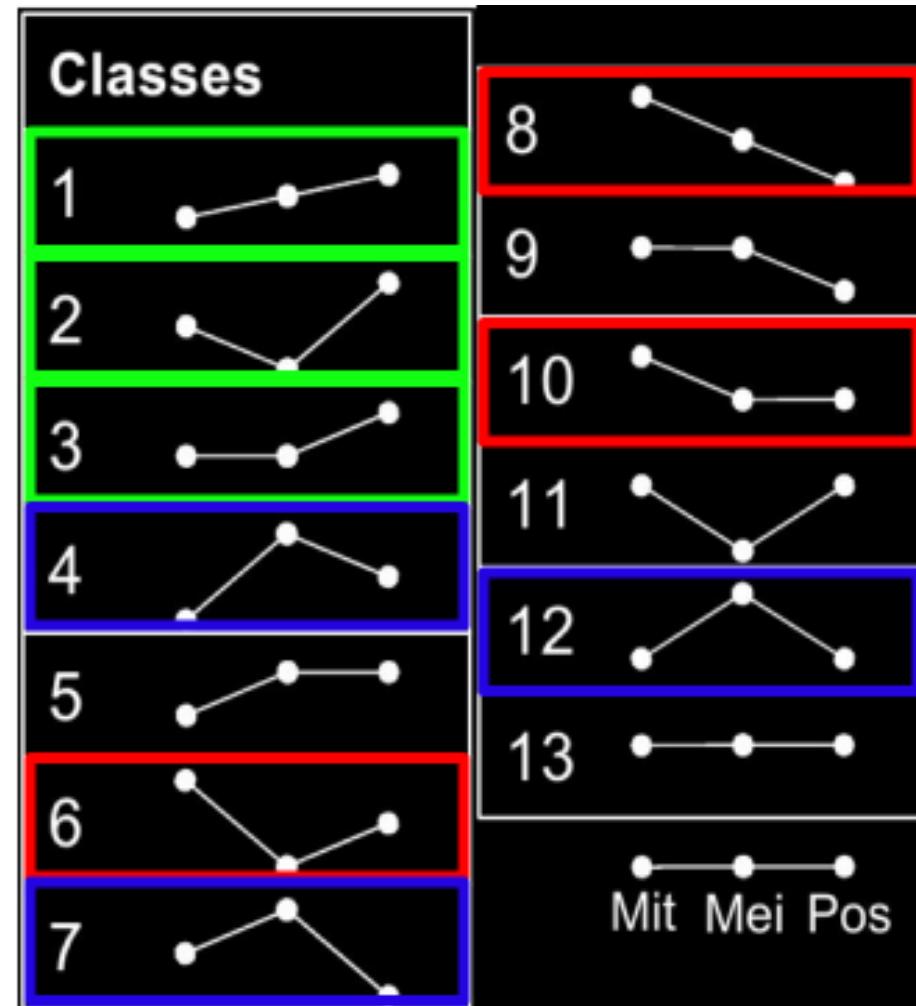
# *Drosophila* Spermatogenesis expression: Vibranovski et al. 2009

Gene classification according to their expression pattern:

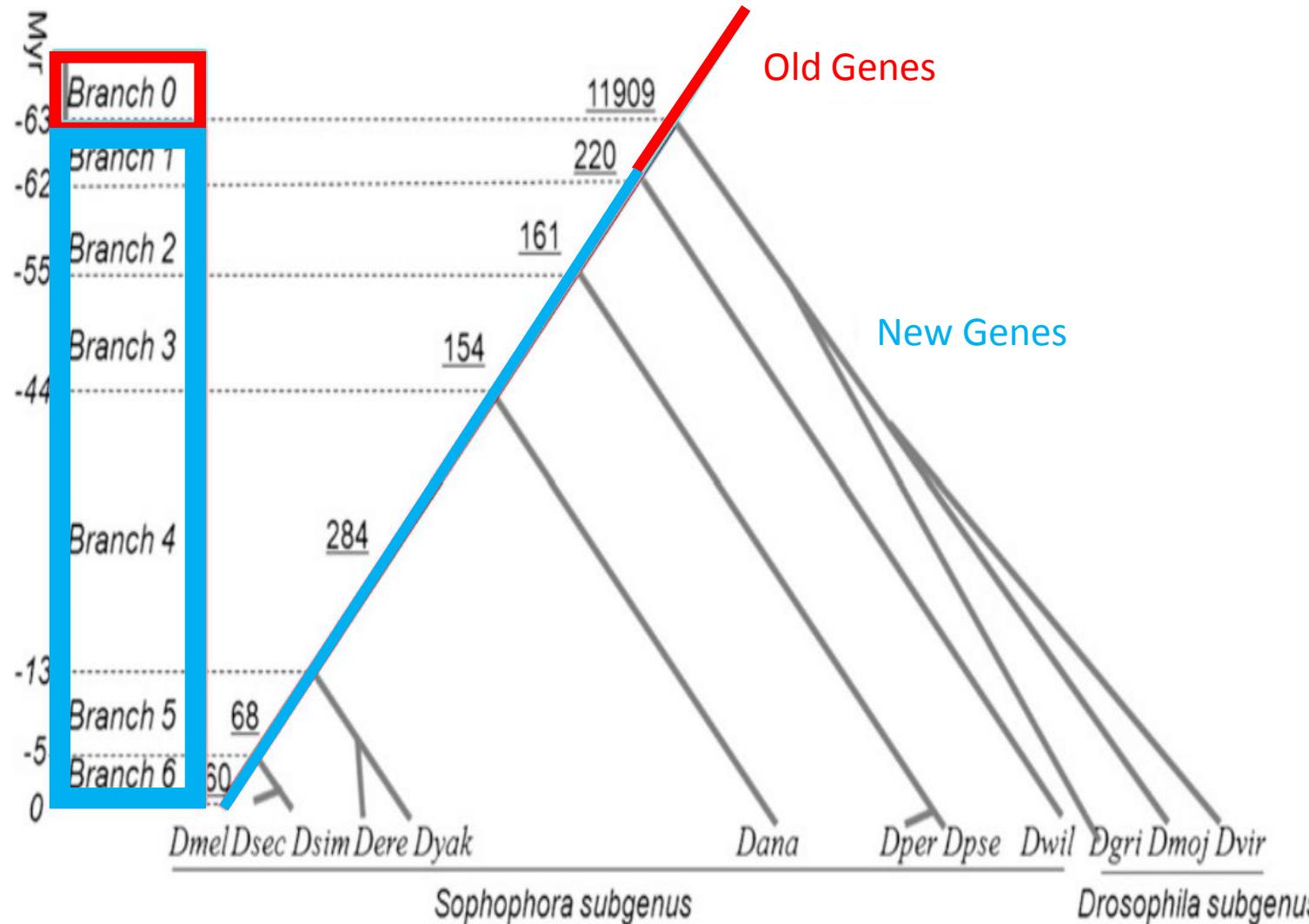
Post-meiotic Genes

Meiotic Genes

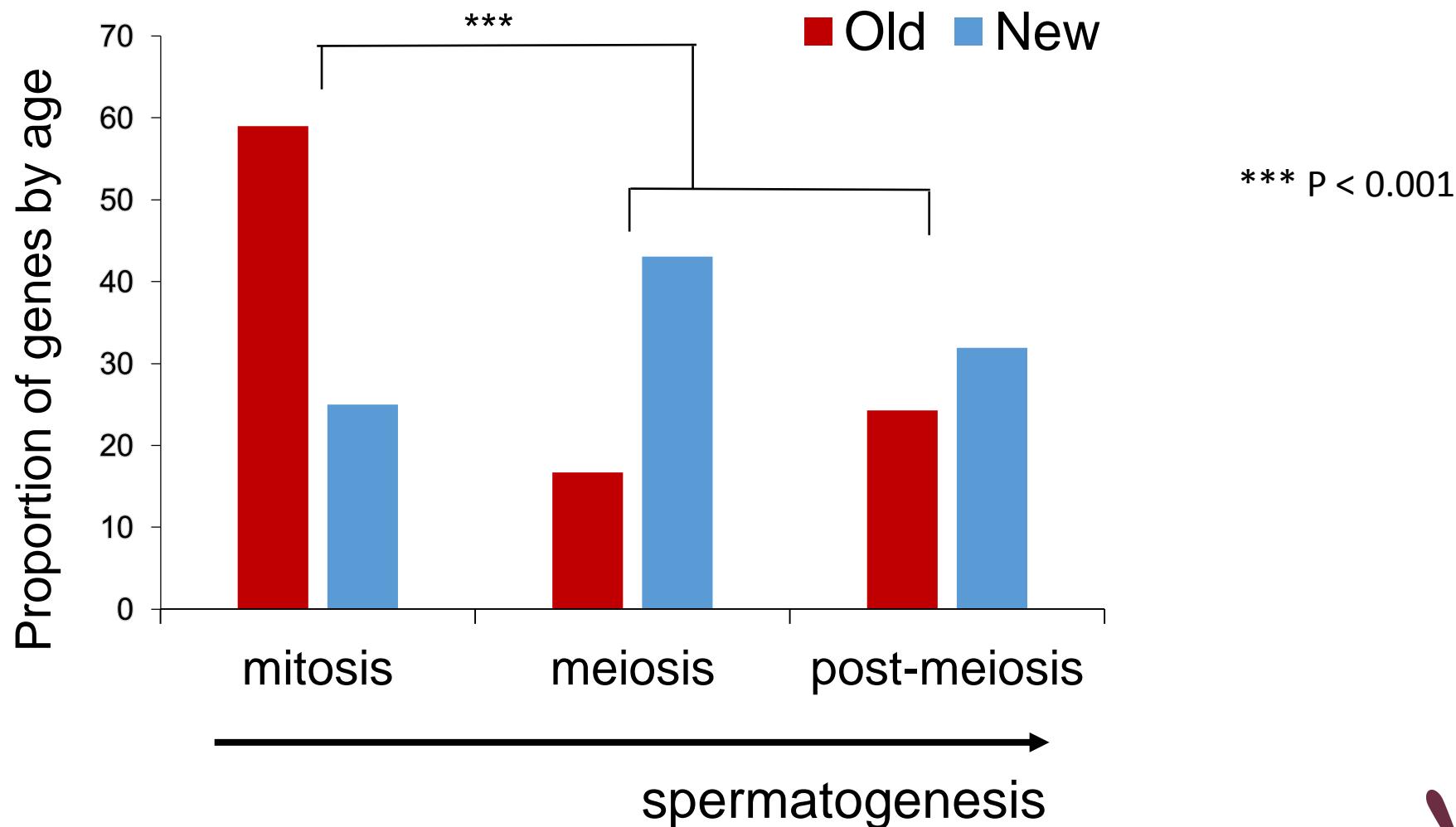
Mitotic Genes



# *Drosophila* Gene's age: Zhang et al. 2010



# 1) *Drosophila*: New Genes are more expressed in later phase of spermatogenesis



# Haploid Selection and the Origin of New Genes

---

## Model Predictions:

- 1) New genes should be more expressed in the later phases of spermatogenesis ✓
- 2) Genes expressed in the haploid phases of spermatogenesis should be enriched with positive selection signature
- 3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis

# New Gene?

Innovation

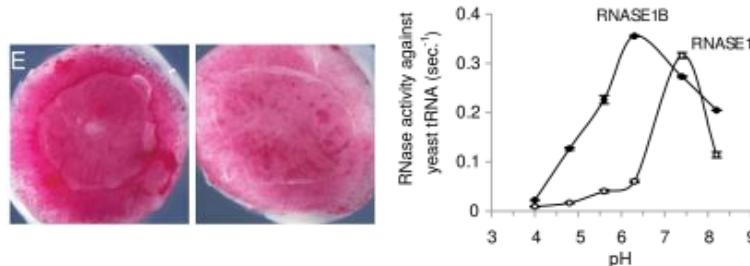
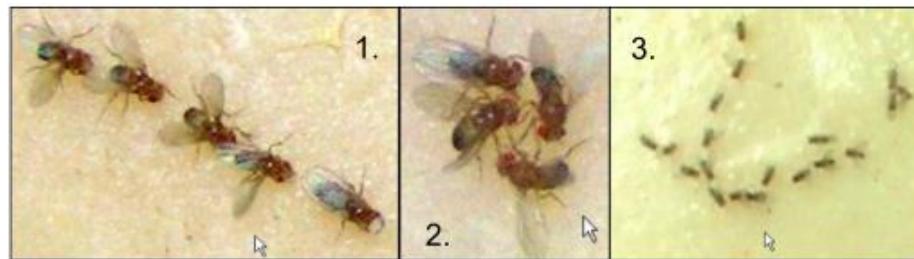
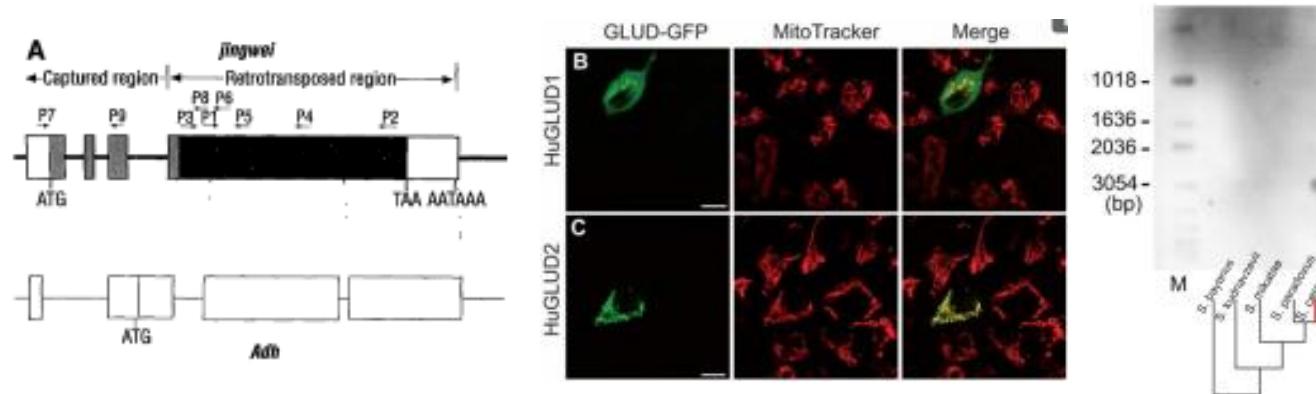
Adaptation

Neo-functionalization

New function

Positive Selection

# New genes: positive selection signature



Long & Langley, 1993. **Science**  
Dupressoir *et al.*, 2009. **PNAS**  
Cai *et al.*, 2008. **Genetics**  
Wang *et al.*, 2002. **PNAS**  
Rosso *et al.*, 2008. **Plos Genetics**  
Zhang *et al.*, 2002. **Nature Genetics**  
Johnson *et al.*, 2001. **Nature**  
Malik & Henikoff, 2005. **Plos Genetics**

# Haploid Expression and Positive Selection

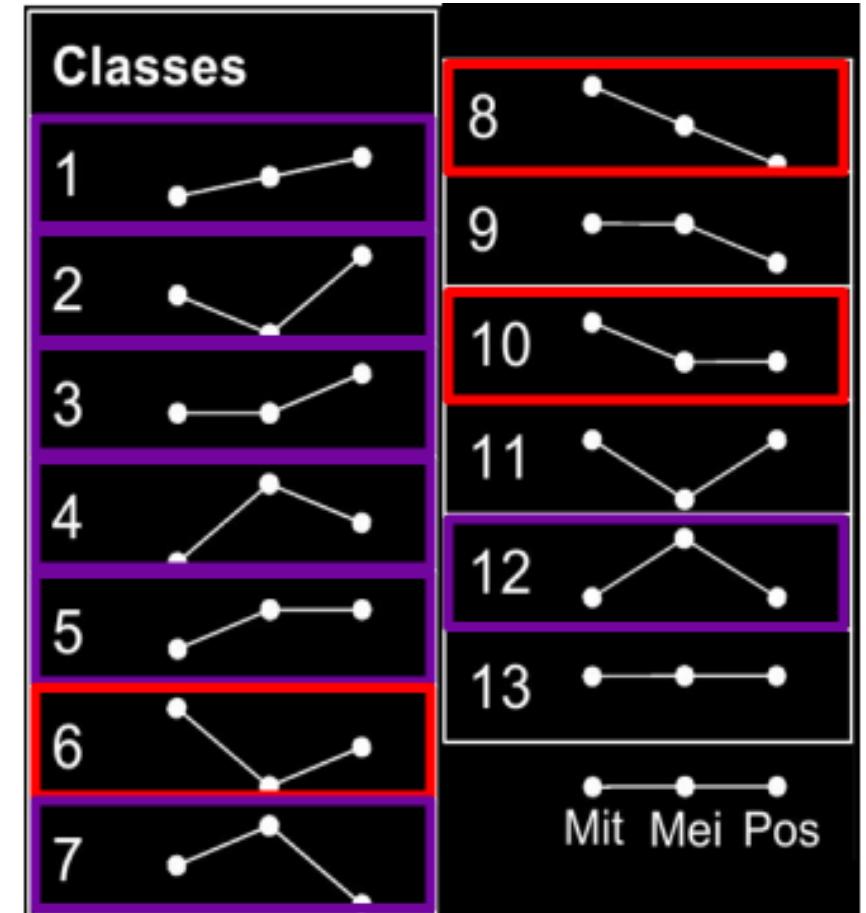
Gene classes according to their expression pattern to test for positive selection signature

Mitotic Genes:

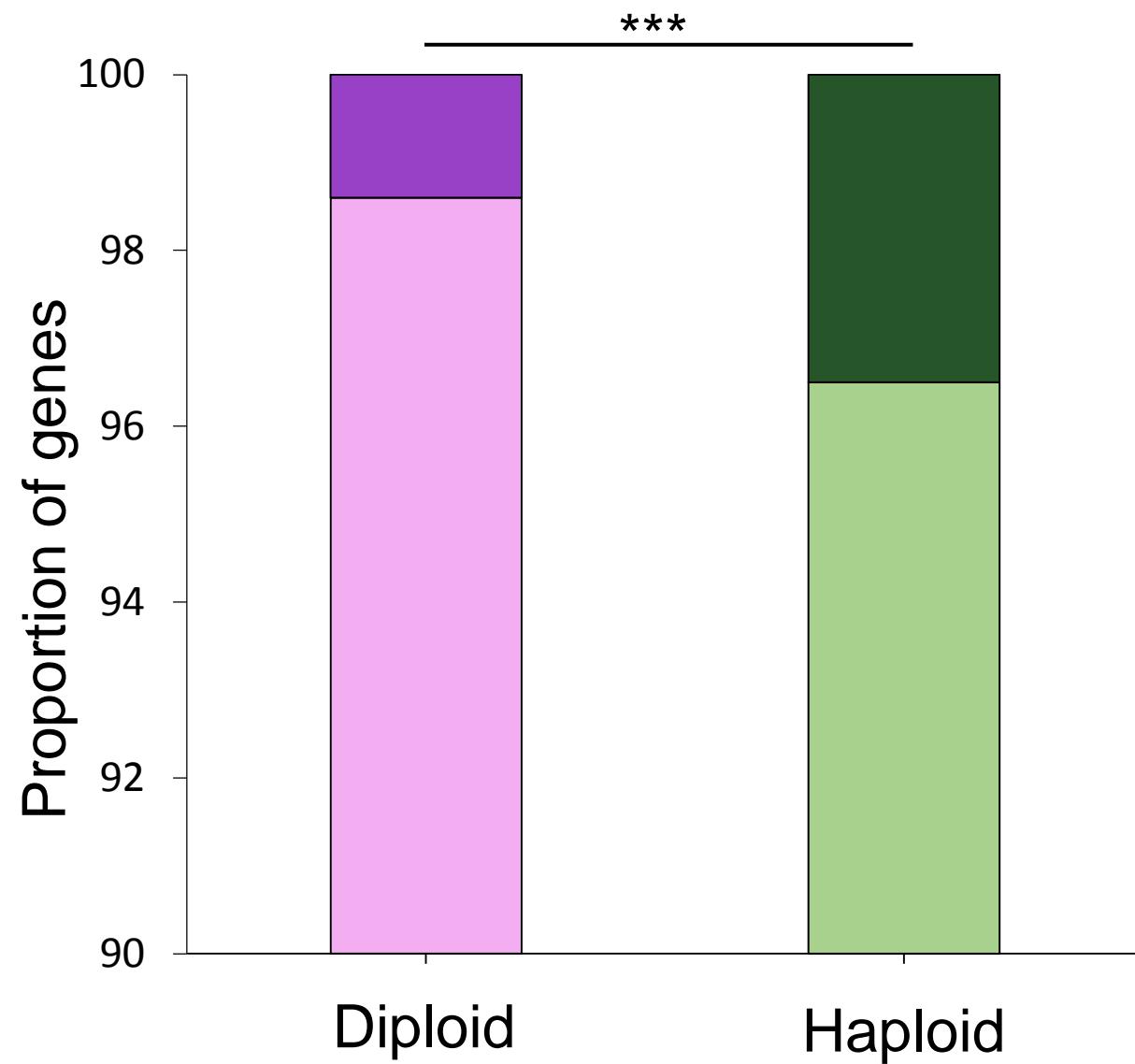
higher expressed in Mitosis (diploid/control)

Haploid Genes:

higher expressed in Meiosis and/or Post-meiosis



## 2) Genes expressed in the haploid phases of spermatogenesis should be enriched with positive selection signature



Stanley and Kulathinal, 2016.



\*\*\* P < 0.001

Comparisons using  
PALM models (Yang, 1997):

- 1) M1a vs. M2a
- 2) M7 vs. M8
- 3) M8 vs. M8a

# Haploid Selection and the Origin of New Genes

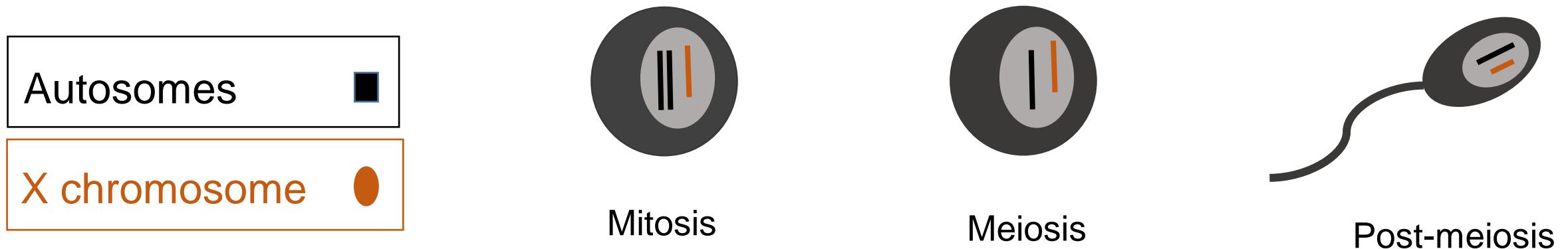
---

## Model Predictions:

- 1) New genes should be more expressed in the later phases of spermatogenesis ✓
- 2) Genes expressed in the haploid phases of spermatogenesis should be enriched with positive selection signature ✓
- 3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis

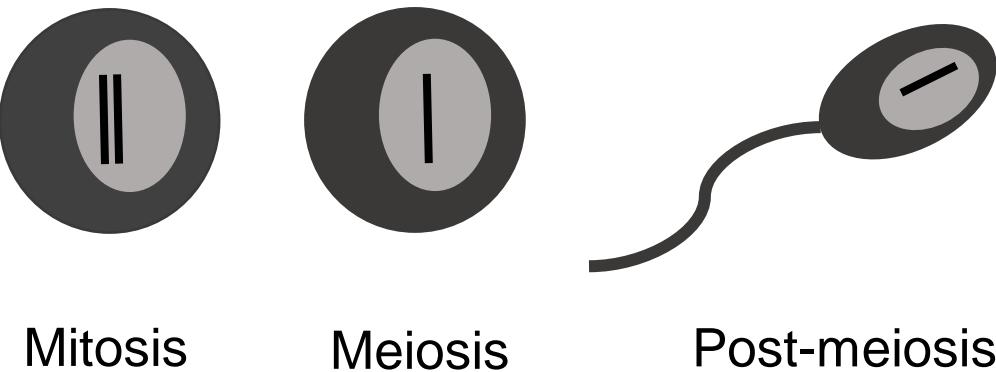
3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis

---



## 2) Autosomal new genes more expressed in haploid phases spermatogenesis (not X-linked genes)

---

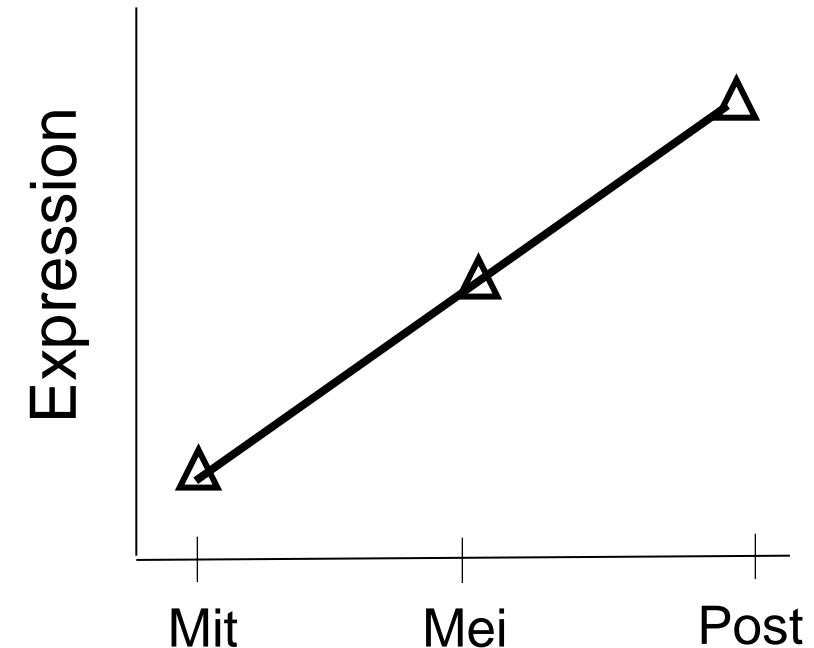


Mitosis

Meiosis

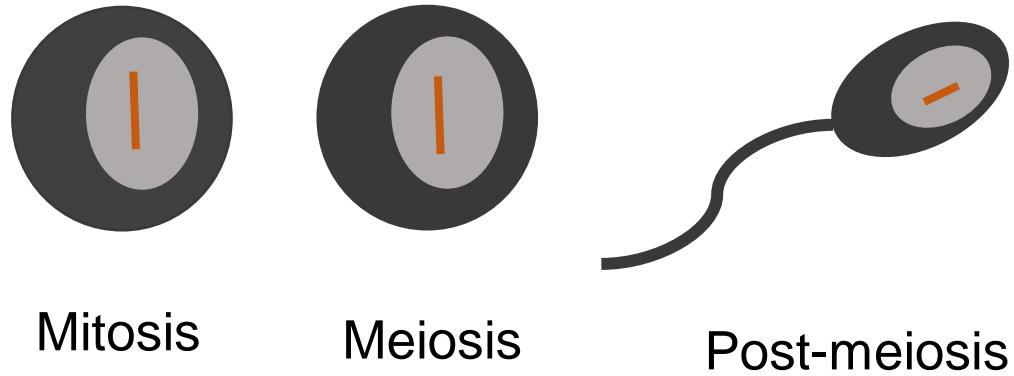
Post-meiosis

Autosomes       $\Delta$



## 2) Autosomal new genes more expressed in haploid phases spermatogenesis (not X-linked genes)

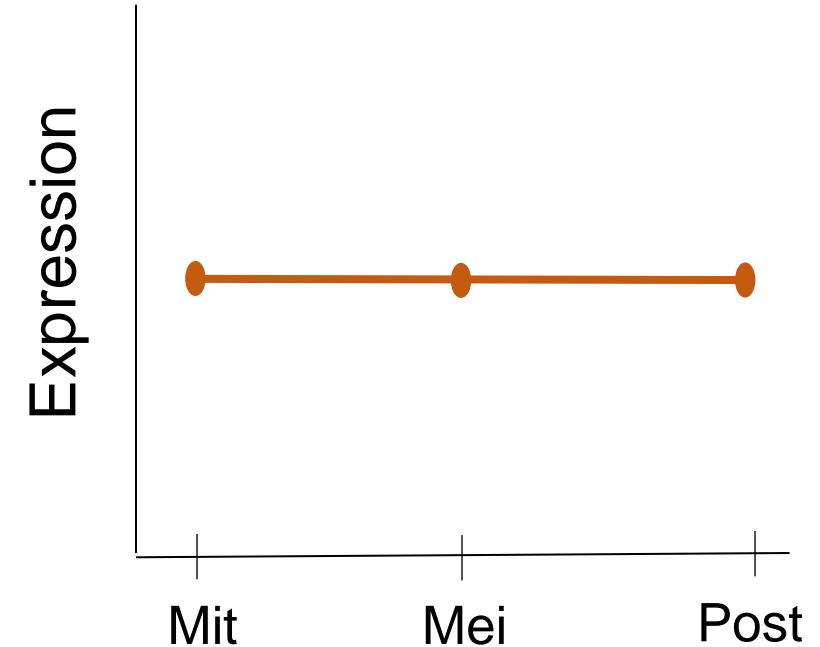
---



X chromosome

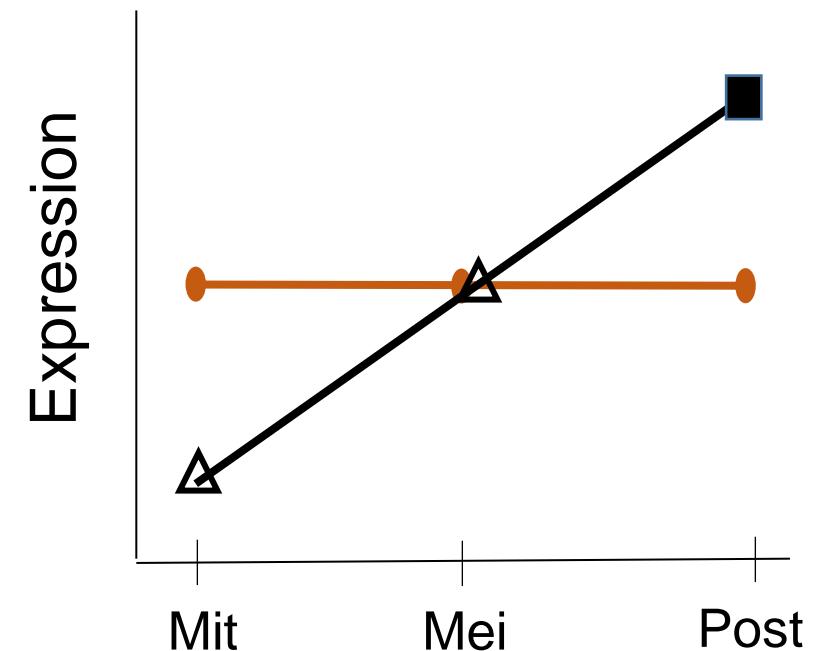
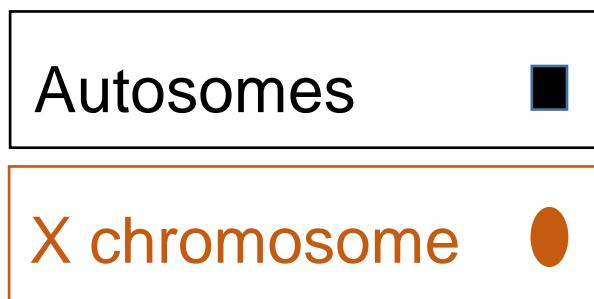
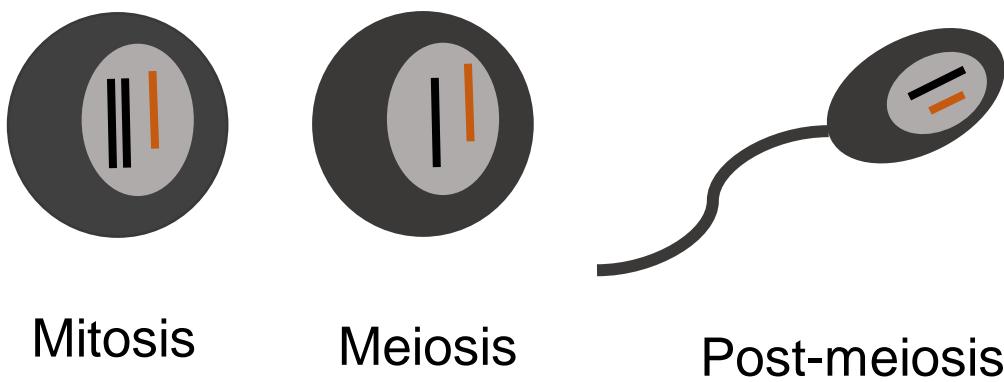


Always hemizygous in male cells!

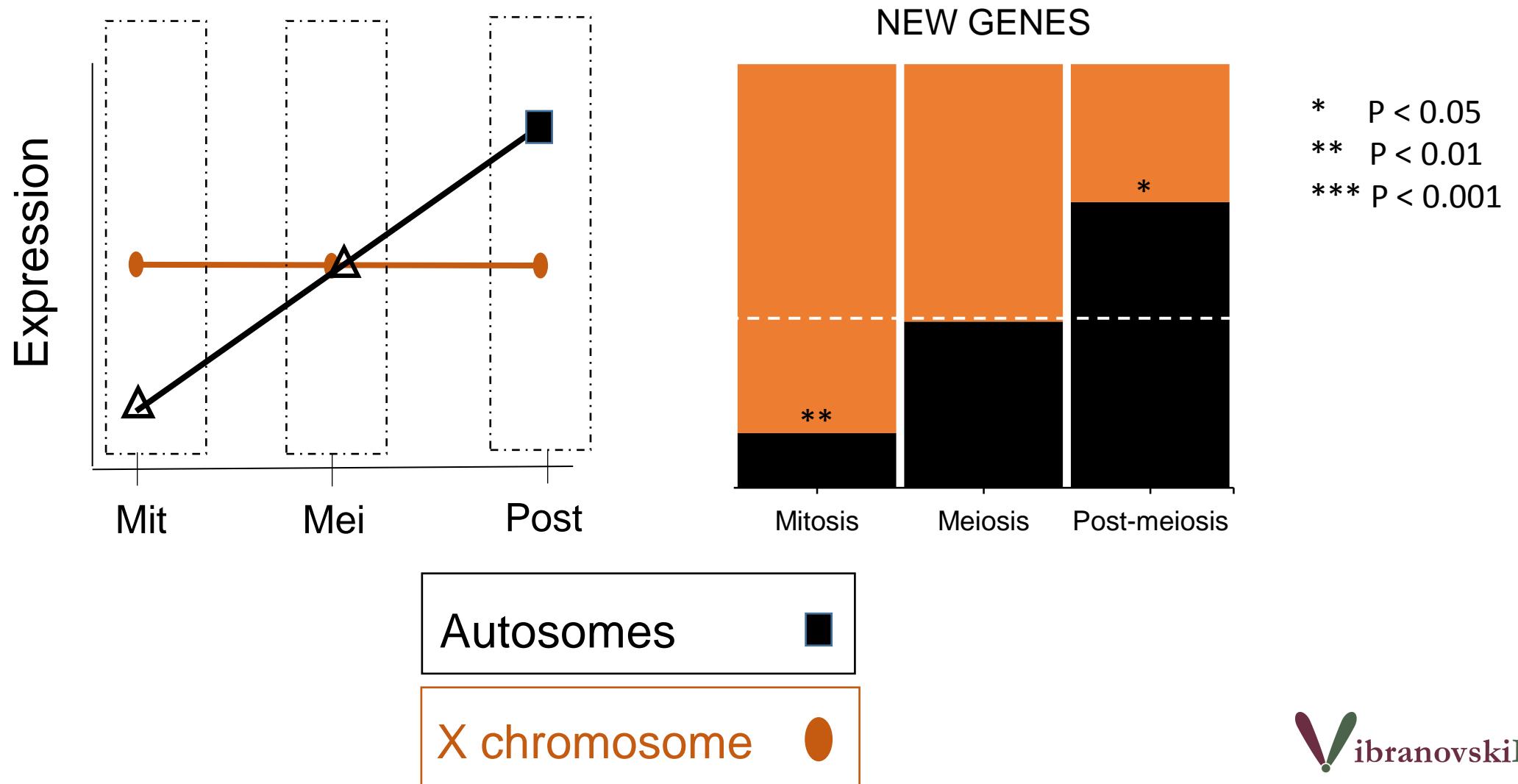


### 3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis

---



### 3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis



# Haploid Selection and the Origin of New Genes

---

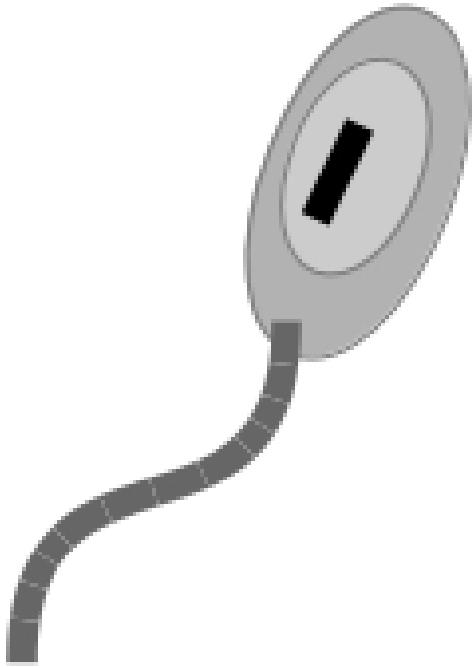
## Model Predictions:

- 1) New genes should be more expressed in the later phases of spermatogenesis ✓
- 2) Genes expressed in the haploid phases of spermatogenesis should be enriched with positive selection signature ✓
- 3) Autosomal new genes (not X-linked genes) should be more expressed in haploid phases of spermatogenesis ✓

# Haploid Selection and the Origin of New Genes

---

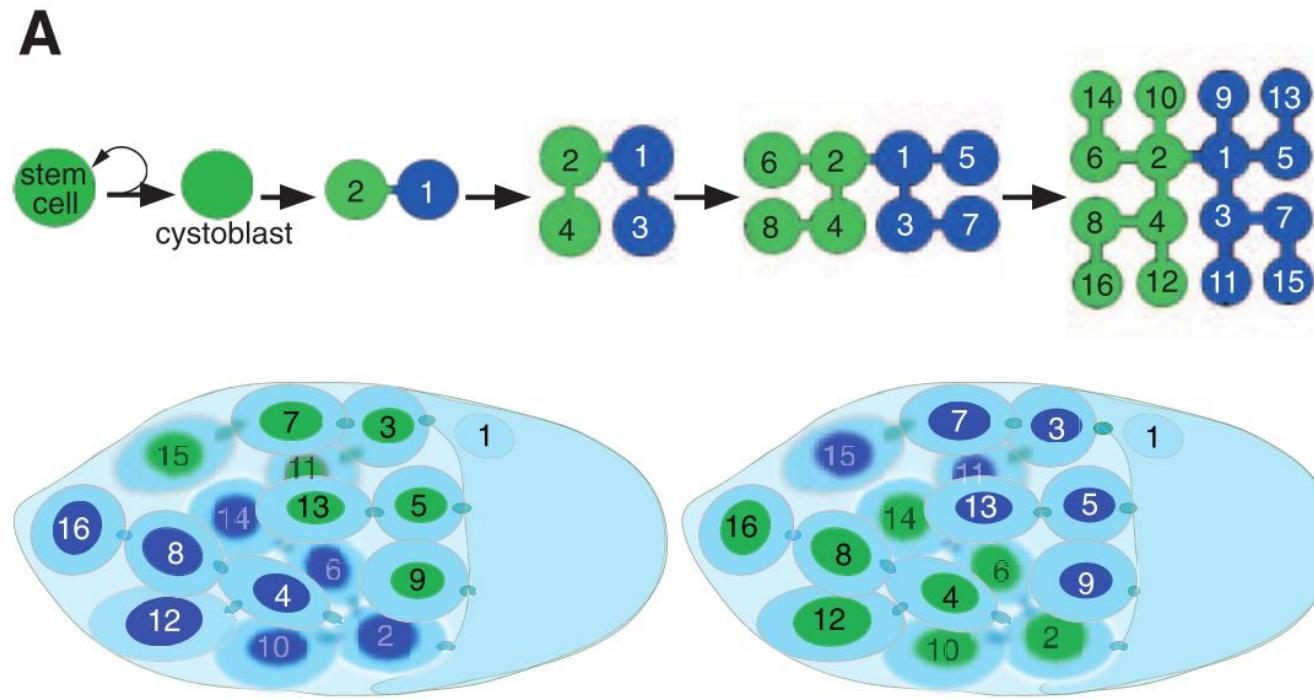
## *Discussion*



# Haploid Selection and the Origin of New Genes



## 1. How about female haploid cells?

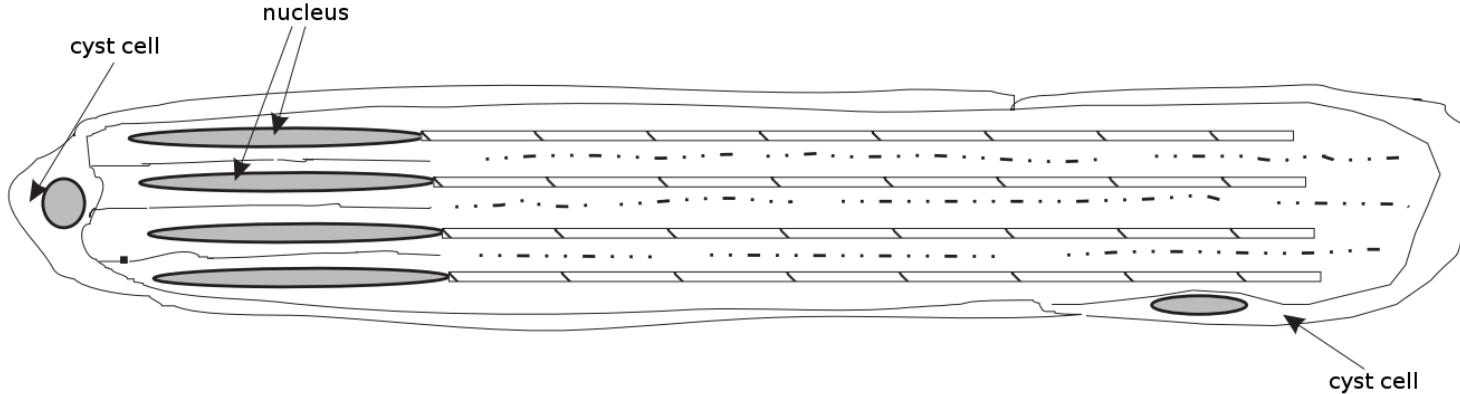


Cáceres and Nilson, 2005 Development

# Haploid Selection and the Origin of New Genes



## 2. How about RNA interchange between haploid male cells?



Fabrizio *et al.*, 1998 Development

Genotype:	A <sub>1</sub> A <sub>1</sub>	A <sub>1</sub> A <sub>2</sub>	A <sub>2</sub> A <sub>2</sub>
Relative fitness:	1	1-hs	1-s

Complete dominance:  $h=1$   
vs.  
incomplete dominance:  $1 \geq h > 0.5$

# Haploid Selection and the Origin and fixation of New Genes

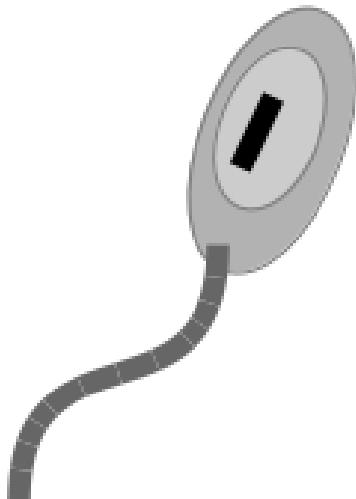
GENOME  
RESEARCH

Research

## Haploid selection drives new gene male germline expression

Julia B. Raices,<sup>1,2</sup> Paulo A. Otto,<sup>1</sup> and Maria D. Vibranovski<sup>1</sup>

<sup>1</sup>*Department of Genetics and Evolutionary Biology, Institute of Biosciences, University of São Paulo, São Paulo, Brazil, 05508-090*



Júlia Raíces



Paulo Otto



Universidade de São Paulo



Lab Retreat 2018:  
National Park of Itatiaia, Brazil