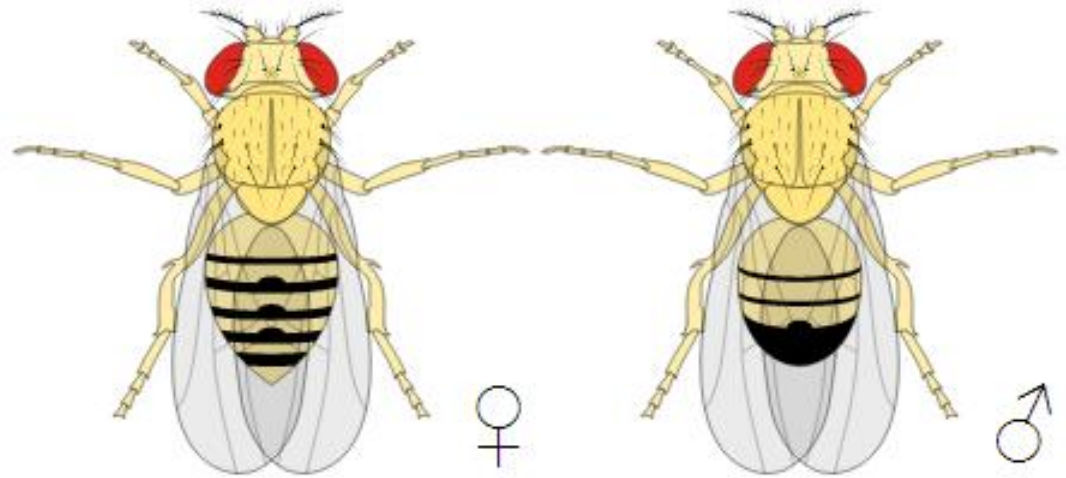
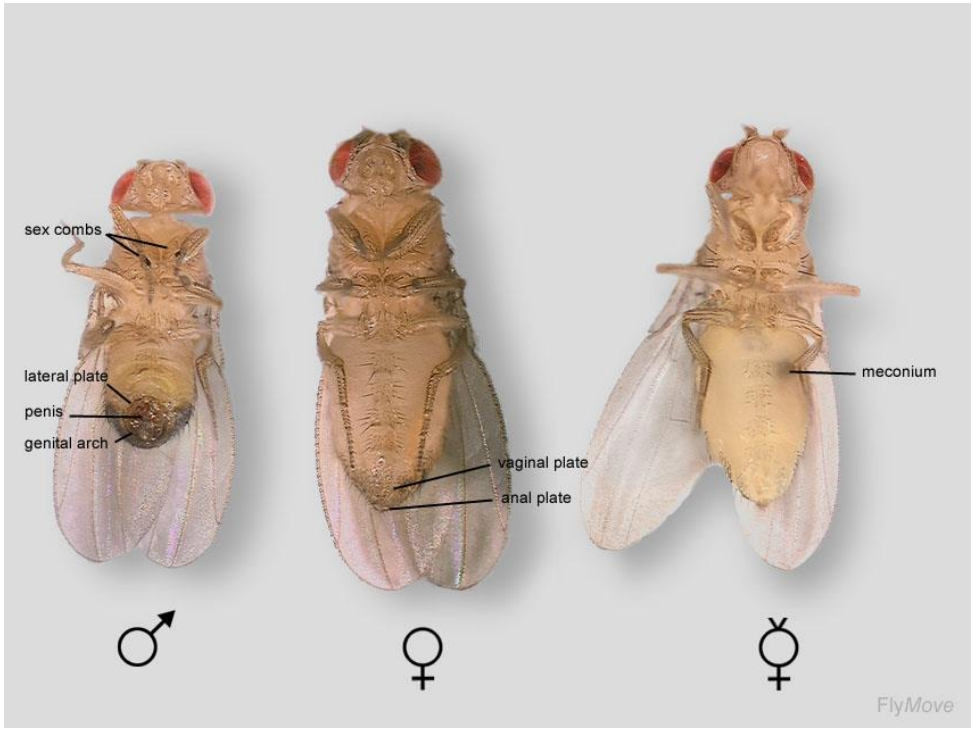
Experimental embryology

Drosophila melanogaster

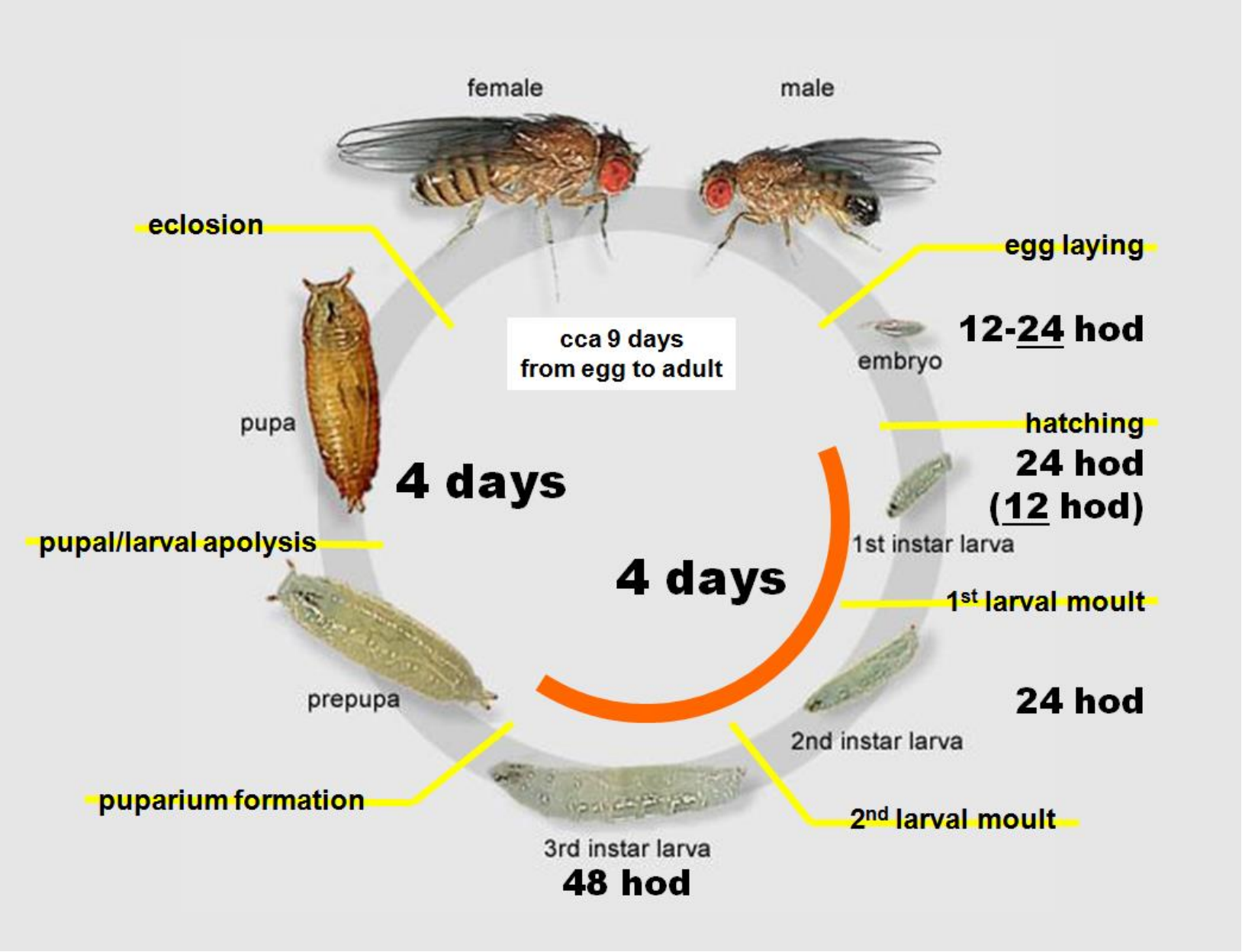
Drosophila melanogaster - common fruit fly (Diptera)





Life cycle

- **short generation time:**
  the shortest, seven days at 28 °C (30 °C: 11 days; 18 °C: 19 days; 12 °C: 50 days)
- lifespan 30 days at 29 °C
- **high number of model animals:**
  females produce about 500 eggs in 10 days
- embryonic development
- embryonic development (comm.)
- Eric Wieschaus talks: Patterning Development in the Embryo



Advantages of *Drosophila* model

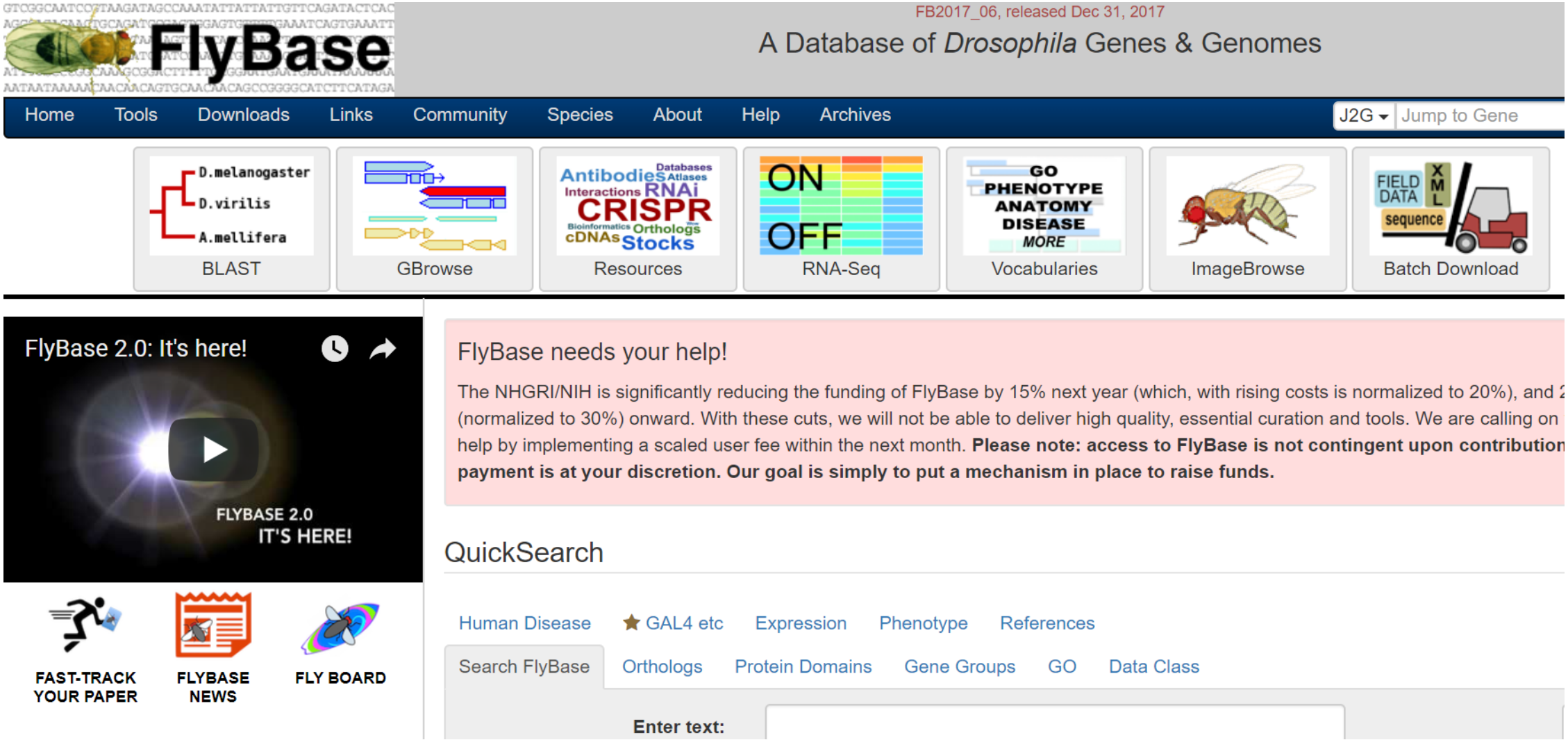
- one of the most studied organisms
- ease of breeding, space-saving, low cost
- well-known fate of individual cell populations
- genetically tractable
- at the molecular and cellular level, many developmental processes are similar to other species

- the basic model of eukaryotic genetics, embryogenesis, studies of maternal and homeotic genes, sex determination
- 4 pairs of chromosomes: female XX (1X:1A), male XY (1X:2A)
- haploid genome: 1.65×10^8 base pairs
- fertilization occurs in the oviduct, the eggs are laid as diploid
- superficial cleavage of eggs
- recombination occurs only in females

What do we need for fly handling?

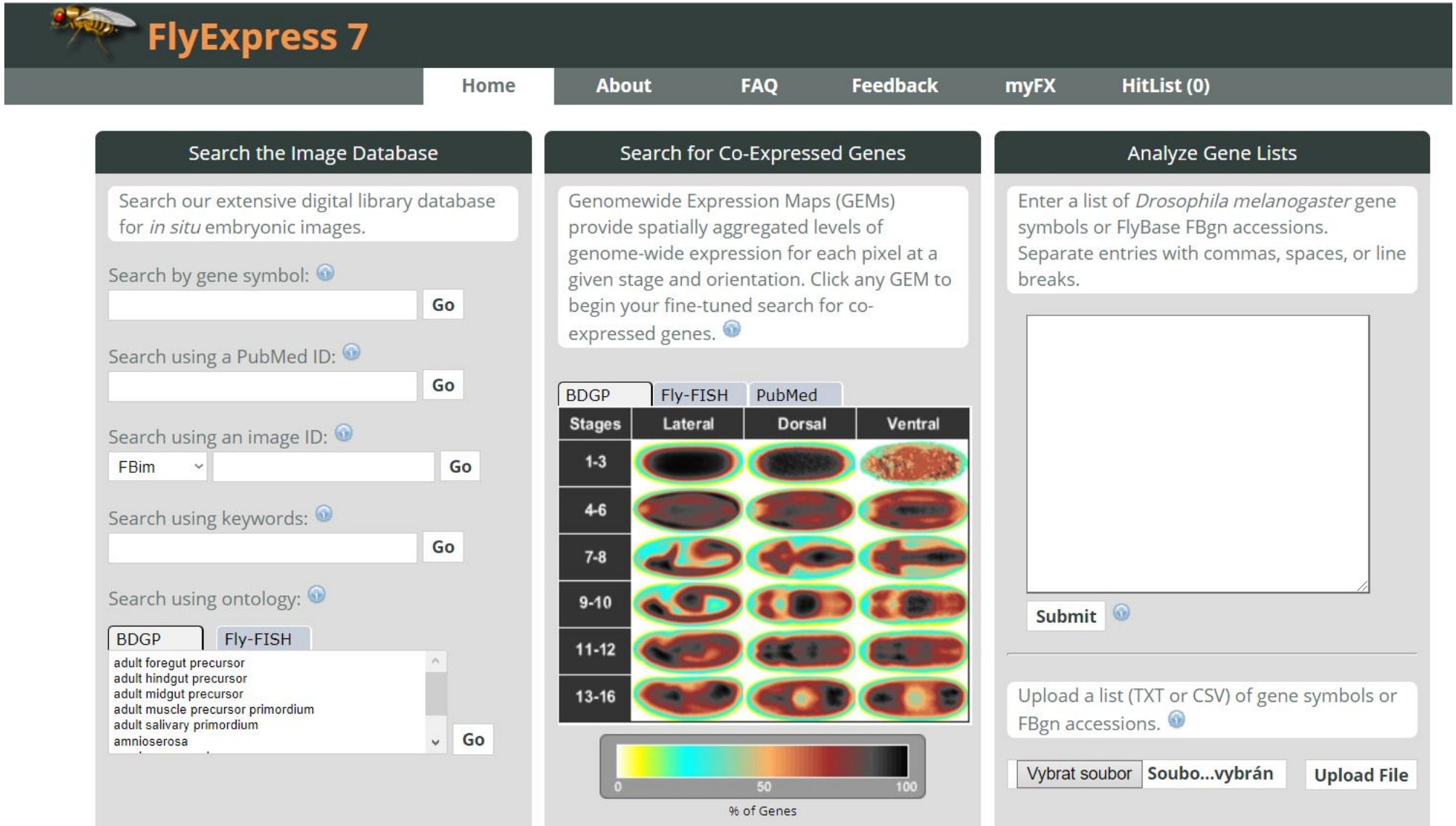
- vials / bottles with caps
- fly food (water, yeast, syrup or sugar, agar, mashed potato powder or soy flour, ascorbic acid, fungicide Nipagin); formerly, crushed banana peel with yeast
- brushes and pad for manipulation
- anesthesia (CO_2, cold, ether, FlyNap® Anesthetic Kit)
- stereomicroscope

FlyBase – gene database and availability of fly lines

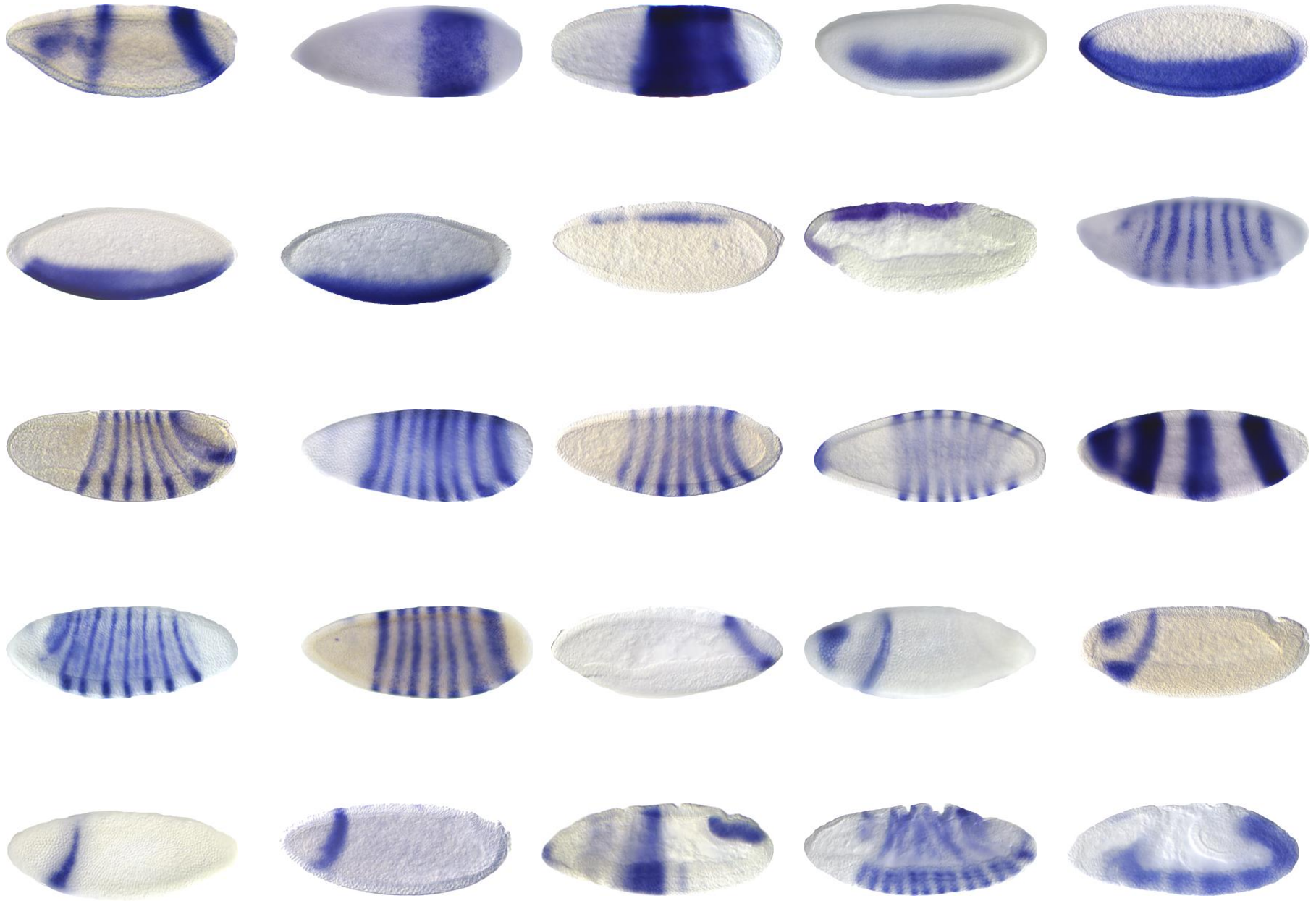


http://flybase.org/

FlyExpress – gene expression maps



http://www.flyexpress.net



FlyMove – developmental processes

FlyMove

Stages | Processes | Organogenesis | Genetics | Methods

- Amnioserosa
- Dorsal vessel
- Epidermis
- Fat body
- Gonads
- Gut
- Hemocytes
- Imaginal discs
- Lymph gland
- Malpighian tubules
- Muscles
- Nervous system
- Ring gland
- Salivary glands
- Trachea

Quiz

Organogenesis

Media

Within just one day the fertilized egg develops from a single cell into a complete and autonomous organism consisting of many different organs.

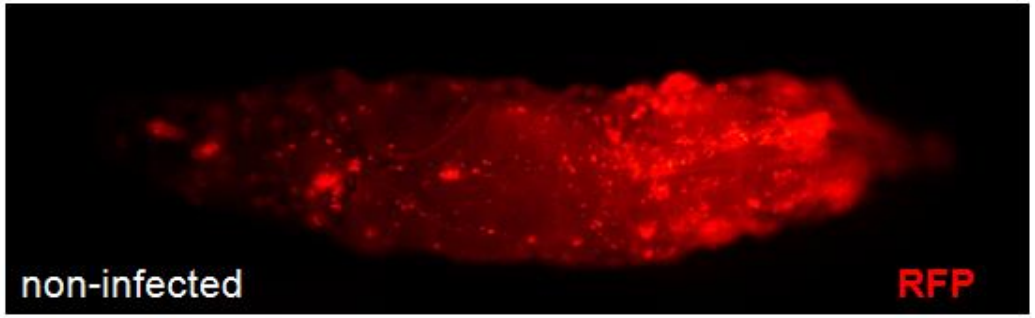
This section deals with the formation of the major organ systems from the cellular blastoderm stage onward.

At the blastoderm stage, the 6000 cells giving rise to all somatic tissues are located as a single cell wide epithelium at the surface of the embryo. Cell lineage analyses have shown, that the three germ layers – ectoderm, endoderm and mesoderm – are already determined at this stage. Thus, a so-called "anlagenplan" or fate map can be pojected onto the embryo.

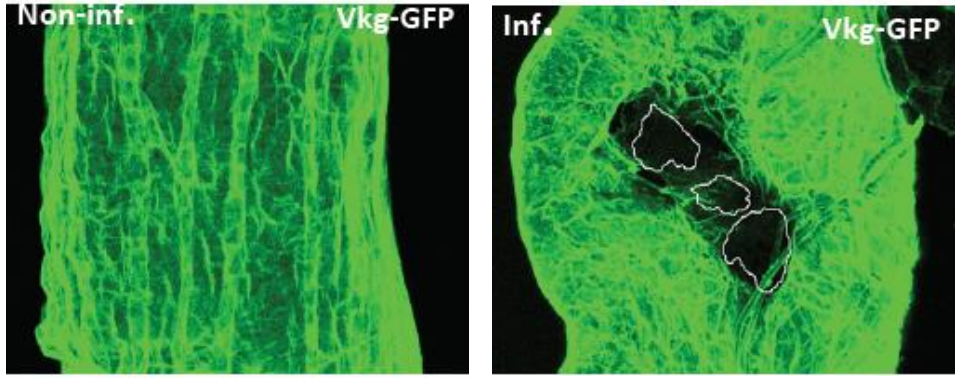
- The ectoderm differentiates the nervous system, the epidermis, the fore- and hindgut, the salivary glands and malpighian tubules as well as the trachea and the ring gland.
- The two endoderm fractions represent the anlagen of the midgut.
- The mesoderm gives rise to all types of muscles, the fat body, the dorsal vessel, the

Reporter lines

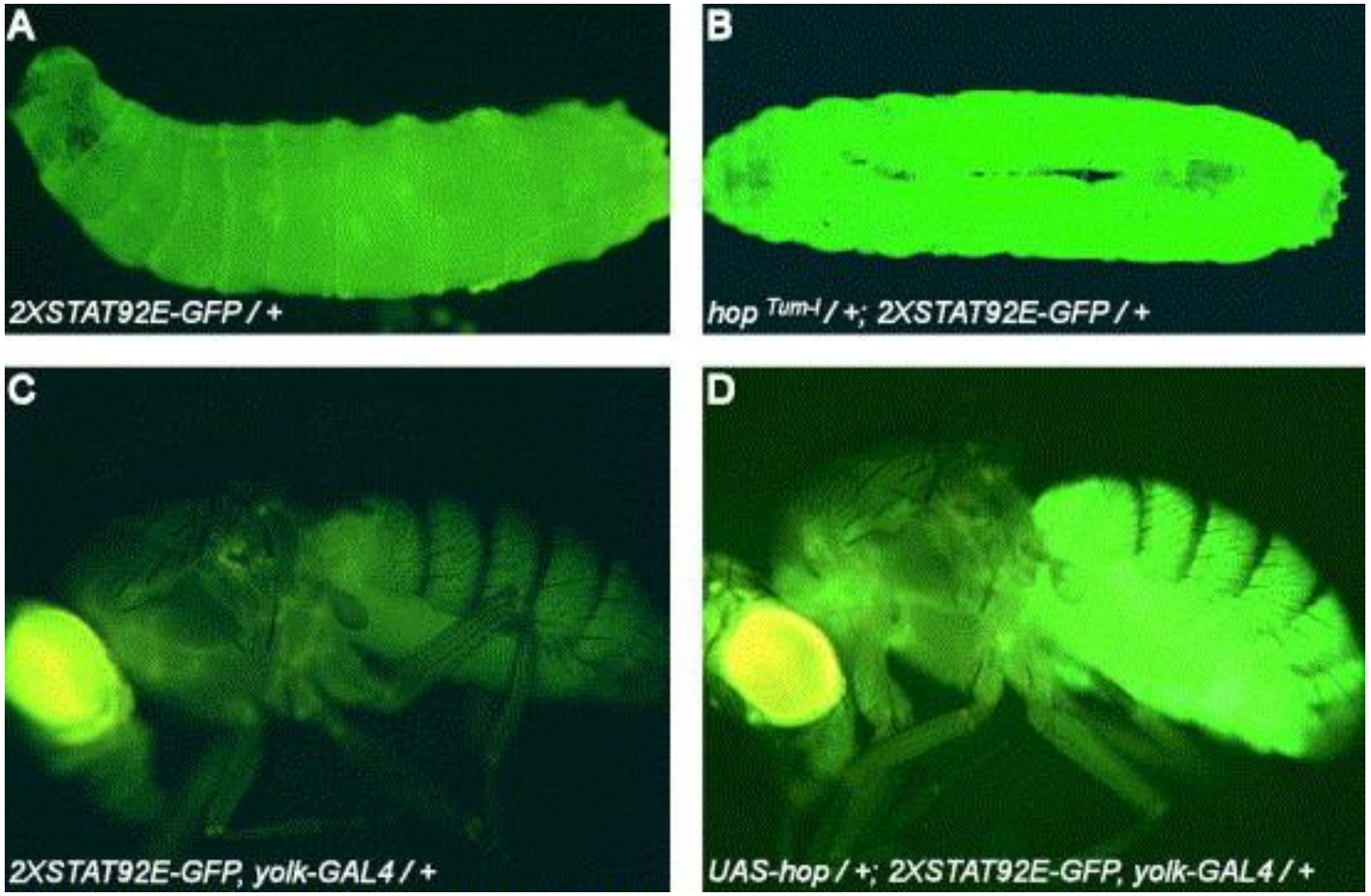
Haemocyte reporter line (hml-Gal4 UAS-RFP)



Collagen visualisation (Vkg-GFP)



STAT pathway reporter line (STAT92E-GFP)

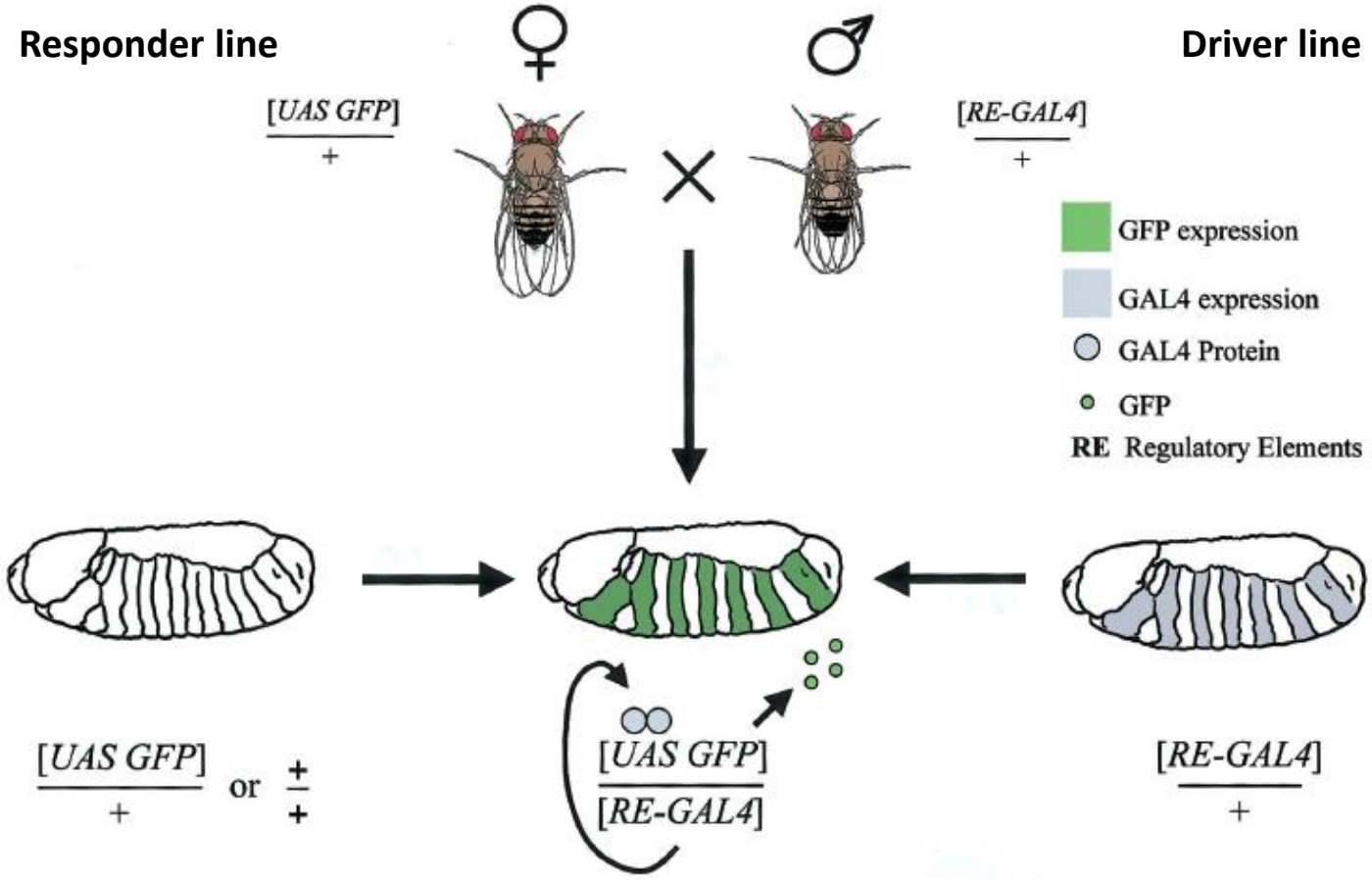


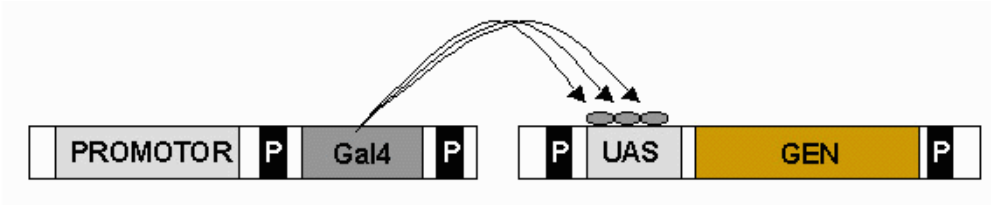
Bach et al. (2007) GFP reporters detect the activation of the *Drosophila* JAK/STAT pathway *in vivo*. https://doi.org/10.1016/j.modgep.2006.08.003

UAS/Gal4 system for targeted gene expression

„driver line“ – specific promotor mediates spatial and temporal synthesis of Gal4
– Gal4 binds to DNA and activates transcription

„responder line“ – **U**pstream **A**ctivating **S**equences bind Gal4 protein
– RNAi construct / GFP / copy of the target gene – its transcription is regulated by UAS which require Gal4 binding

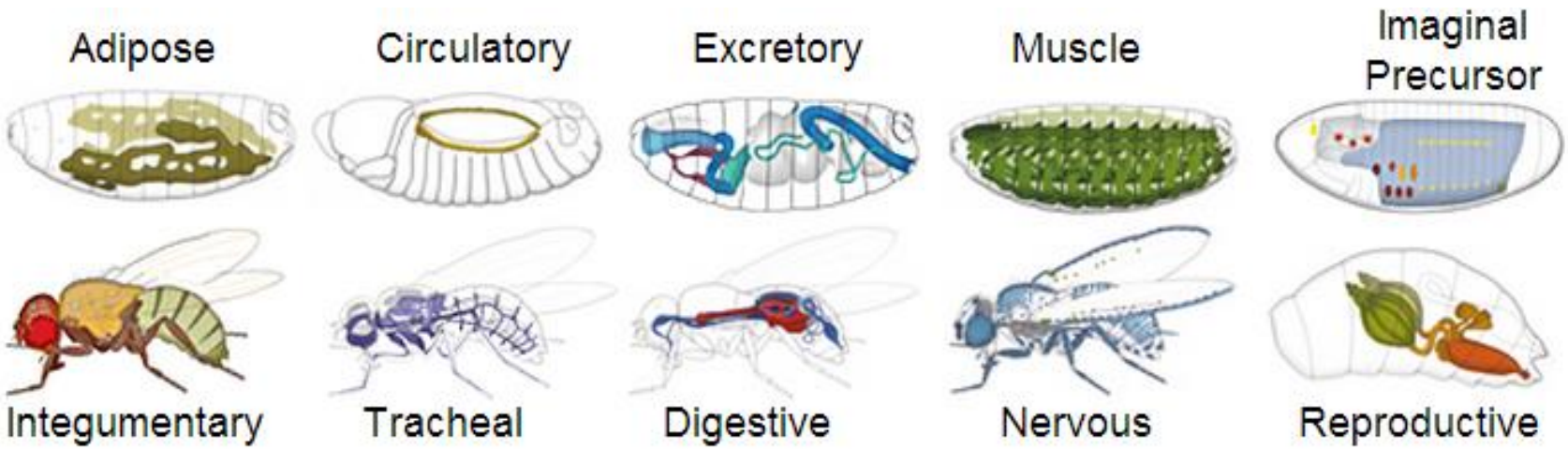




Duffy. *Genesis*, 34(1-2): 1-15, 2002.

UAS/Gal4 system for targeted gene expression – driver lines

- Actin-Gal4 (ubiquitous expression)
- hemolectin-Gal4 (embryonic/larval haemocytes; no lamellocyte expression observed)
- Hemese-Gal4 (haemocytes, embryonic/larval lymph gland)
- Lz-Gal4 (crystal cells)
- PPL-Gal4 (embryonic and larval fat body)
- NP1-Gal4 (gut)



UAS/Gal4 system for targeted gene expression

- **knock-down** (UAS-RNAi, UAS-rpr), **over-expression** (UAS-gene sequence) or **visualisation** of target gene expression
- allows spacial and temporal control of gene expression (Gal4 drivers available both tissue and time-specific; eg. Bloomington)
- allows the maintenance of lethal mutations and mutations causing sterility (genotype is split into two fly lines and the harmful effect will not appear until both lines are crossed at 29 °C)

Critical steps of the use:

- the sequence of regulated gene must be known
- off-target effects
- rearing the flies at 29 °C
- non-complete knock-down (leaky expression)

The Nobel Prize in Physiology or Medicine 1995

The Nobel Assembly at the Karolinska Institute in Stockholm, Sweden, has awarded the Nobel Prize in Physiology or Medicine for 1995 to **Edward B. Lewis, Christiane Nüsslein-Volhard and Eric Wieschaus** for their discoveries concerning **“the genetic control of early embryonic development”**.

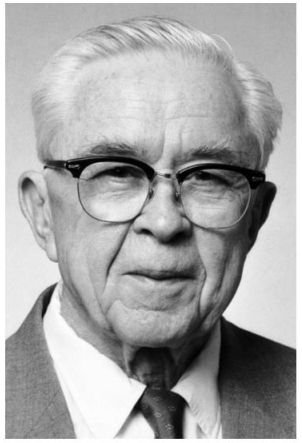


Photo from the Nobel Foundation archive.
Edward B. Lewis
Prize share: 1/3

Photo from the Nobel Foundation archive.
Christiane Nüsslein-Volhard
Prize share: 1/3

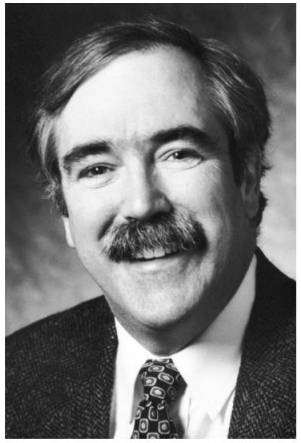
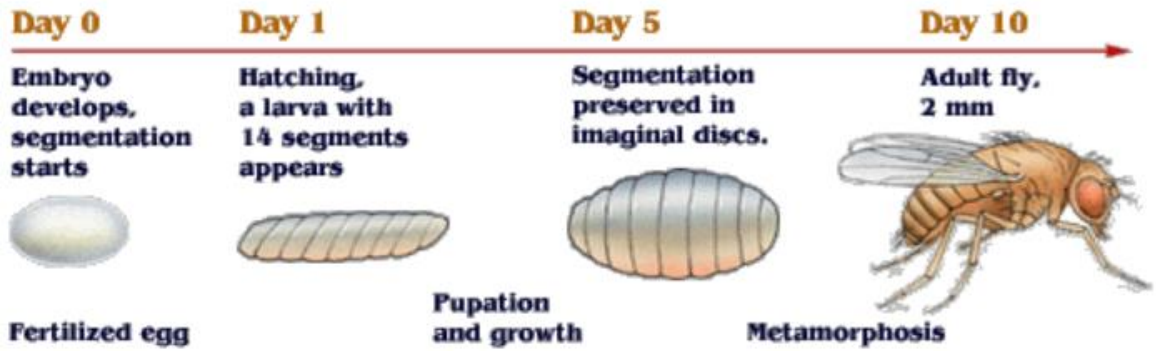


Photo from the Nobel Foundation archive.
Eric F. Wieschaus
Prize share: 1/3



- the mechanisms of Wnt action have emerged from several systems: genetics in *Drosophila melanogaster* and *Caenorhabditis elegans*
- the Wnt homepage

materialstoday
Volume 14, Issue 5, May 2011, Pages 190-195

Review

Drosophila – a versatile model in biology & medicine

Barbara H. Jennings ✉

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https://doi.org/10.1016/S1369-7021(11)70113-4

J. Dev. Biol. **2017**, *5*(3), 9; **https://doi.org/10.3390/jdb5030009**

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Editorial

Introduction: *Drosophila*—A Model System for Developmental Biology

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Minireview

Posterior gut development in *Drosophila*: a model system for identifying genes controlling epithelial morphogenesis

Judith A Lengyel ✉ & Xue Jun Liu

Cell Research **8**, 273–284(1998) | Cite this article

The *Drosophila* lymph gland as a developmental model of hematopoiesis

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Practical part

1) fly handling
   collection of virgins
   visualisation of fat body using UAS/Gal4 system

2) preparing of *D. melanogaster* larvae for nanoinjection
   microscopy of reporter *D. melanogaster* lines

3) nanoinjection of *D. melanogaster* larvae and adults