Parasitología, Parásitos, y Vectores y el Sistema Inmune de los Vectores

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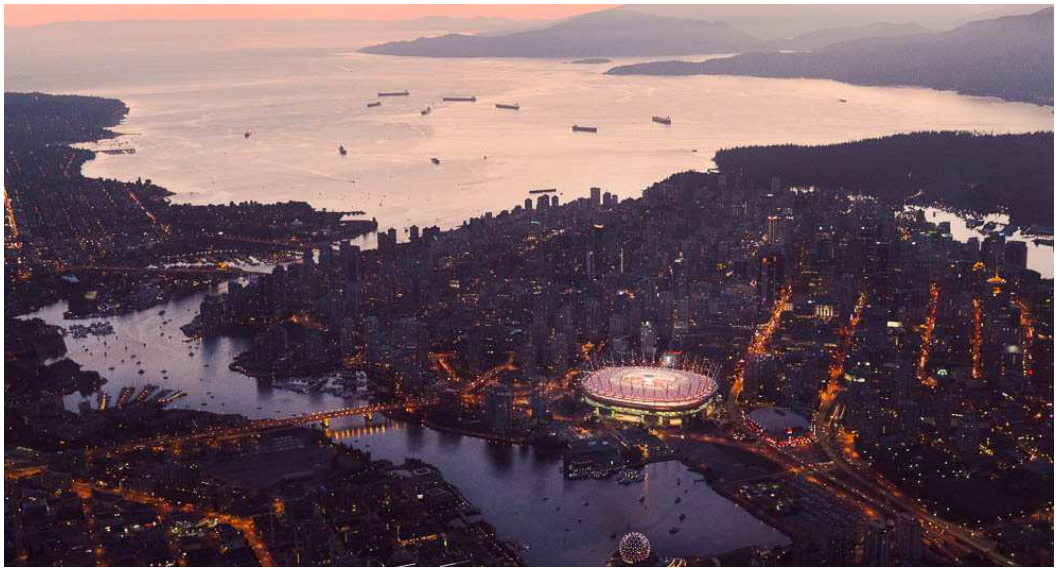


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Types of Ecological Interactions

- Symbiosis are common and span multiple trophic levels

Commensalism

Mutualism

Predation/Parasitism

Competition

Types of Parasitism

- Parasites can be strictly parasitic, free living or a combination

- Original categorization into micro- and macro-parasites
  - Micro-parasites do not display density dependant virulence (ex. viruses)
  - Macro-parasites display density dependant virulence (ex. filarial worms)

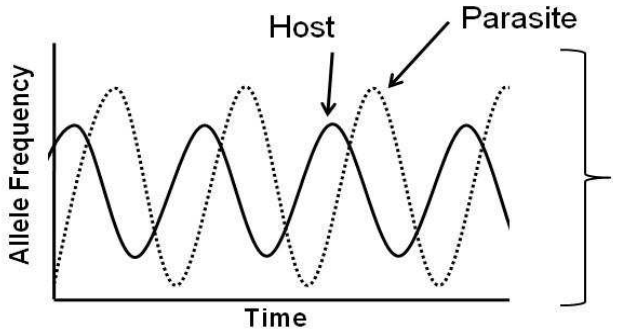
- Simple: one parasite, one host

- Complex: one parasite (or assembly), multiple hosts

- Relationship with hosts differ
  - Developmentally change
  - Replicate
  - Remain infective but do not undergo development

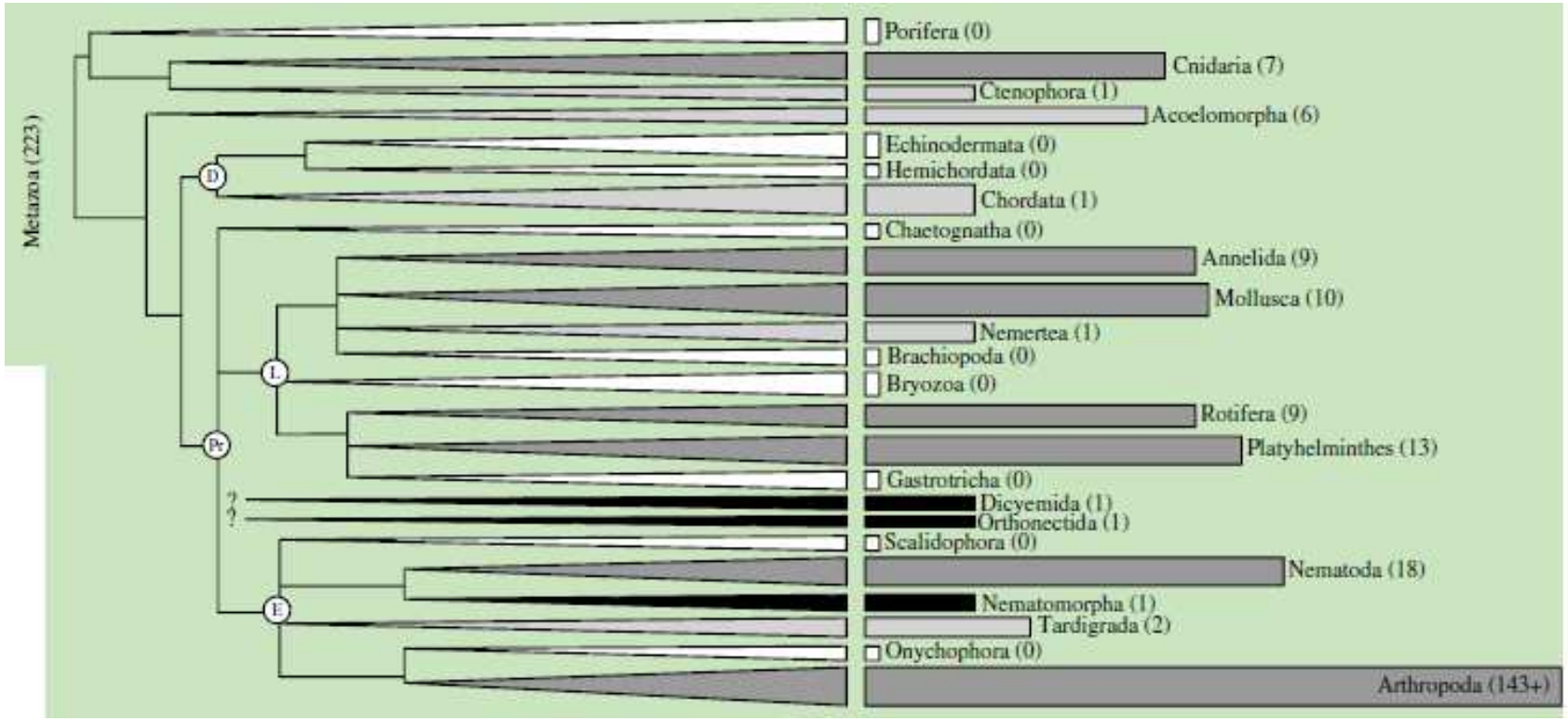
Evolution of Parasitism

- All evolution events restricted to ecological constrains
  - Metabolic capacity, spatial economy and cell multiplication speed
- Shift from free-living to parasitic forms largest evolutionary change in history
- Parasitism evolved through positive selection pressures
- Adaptive radiation to new niches where no competition existed allowed parasites to thrive
- Parasites have evolved due to convergent evolution
  - Patterns of reciprocal adaptation, caused by two species evolving in close association
  - Change in the genetic composition of one species (or group) in response to a genetic change in another (reciprocal evolutionary change)



Evolution of Parasitism

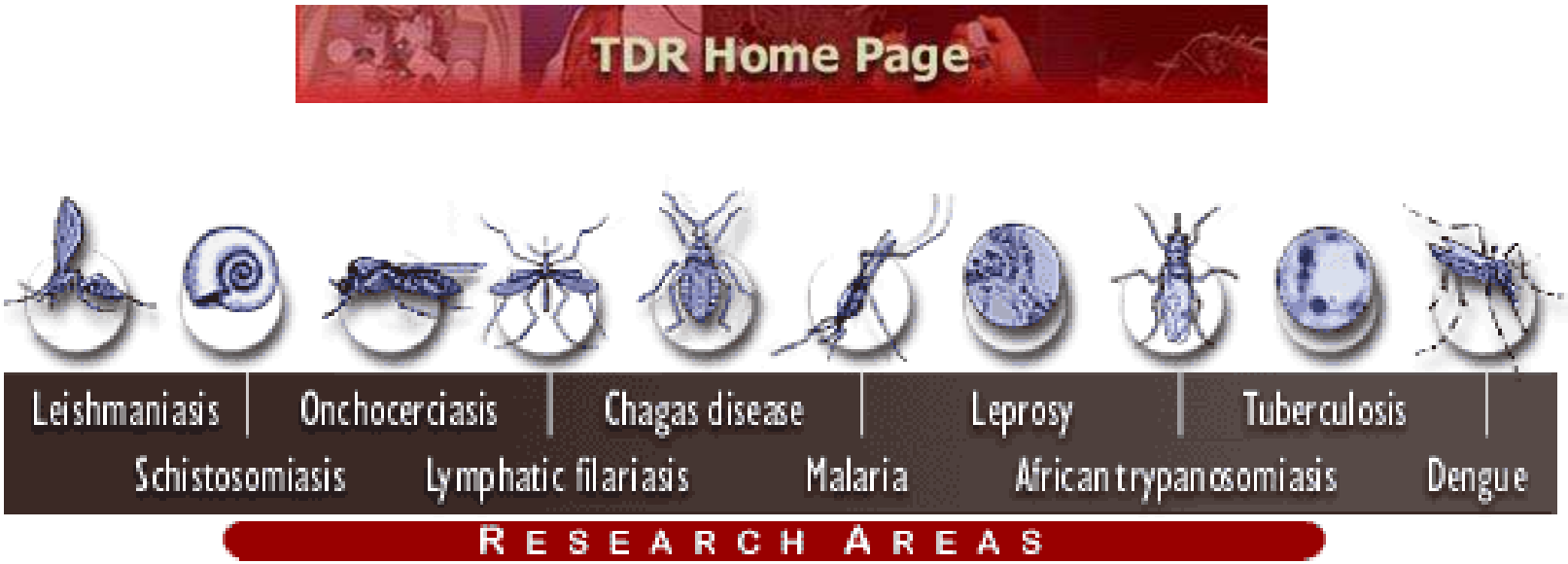
- Evolved independently 223 times
  - Majority are arthropods
  - Many are nematodes, flatworms, molluscs and annelids
- Parasites make up 40% of all animal species
- Some orders do not have any parasitic species



Evolution of Parasitism

- Historical paradigm (Cope's 1896 Law) was that evolution proceeds from unspecialized to specialized forms
    - However parasites tend to be more morphologically simple than their ancestors (although they have more specialized appendages)
- Parasites have evolved quickly due to:
    - Adaption of a host (biotic changing environment) instead of an abiotic comparatively static environment allows for increased rates of adaption
    - Quick generation time and large population size
    - Hijacked and adopted host immune responses
- First parasites evolved mechanisms to avoid/overcome the hosts' immune system, then developed means of neurological manipulation to change host behavior and increase transmission

Are Vector-Borne Disease Important?

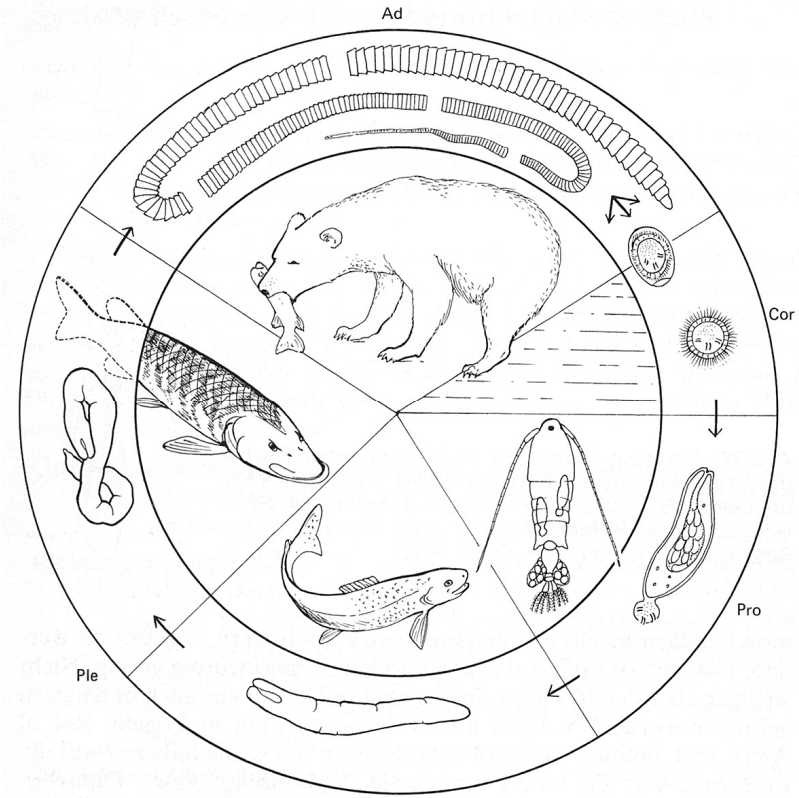


Sept. 15, 2014, 9:04 a.m.

The mosquito borne disease has now spread to Central and South America

Colombia's health ministry has confirmed the first four cases of chikungunya virus in the country.

Hosts and life cycles



- The **definitive host** is by definition the one in which the parasite reproduces sexually
- Additional hosts are then designated **intermediate hosts**
- Host which actively transmit parasites to humans are often called **vectors**
- In **paratenic** or transport hosts no parasite development occurs
- **Reservoir host** are alternate animal host from which the parasite can be transmitted to humans (zoonosis) or domestic animals
- **Accidental host**, not suitable for parasite development, but severe disease might ensue nonetheless

TRANSMISSION OF PARASITES BY VECTORS:

Biological Transmission

I. **A. Cyclopropagative Transmission**

The parasite undergoes cyclical changes and multiplies within the vector, i.e., there are both developmental changes and multiplication of the parasite.

B. Cyclodevelopmental Transmission

The parasite undergoes cyclical changes within the vector but does not multiply, i.e., there are only developmental changes of the parasite without multiplication.

C. Propagative Transmission

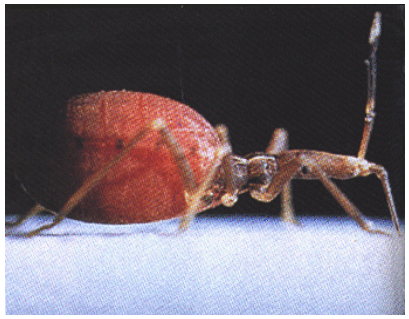
The parasite multiplies within the vector without any cyclical changes, i.e., the parasite increases in number within the vector but does not undergo any developmental changes.

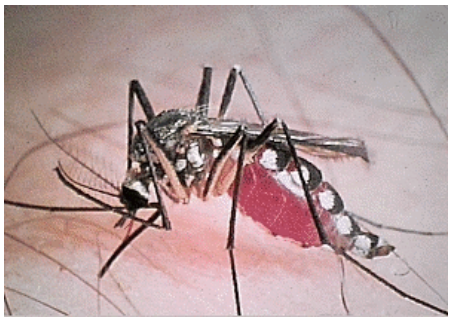
II. **Mechanical Transmission**

This is similar to a "flying syringe" where transmission from one host to another is accomplished because the parasite contaminates the mouthparts of an arthropod and is physically carried to another host.

Why should vectors protect themselves from parasites?

Do human parasites affect their vectors?

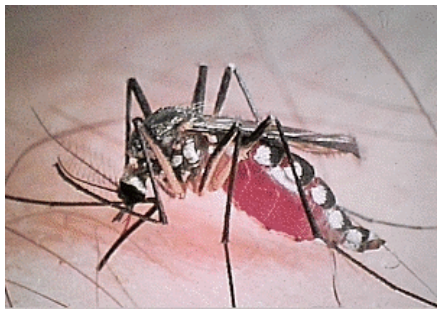


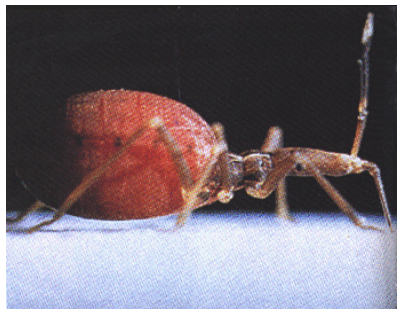


Fitness Costs Associated with Parasite Infection

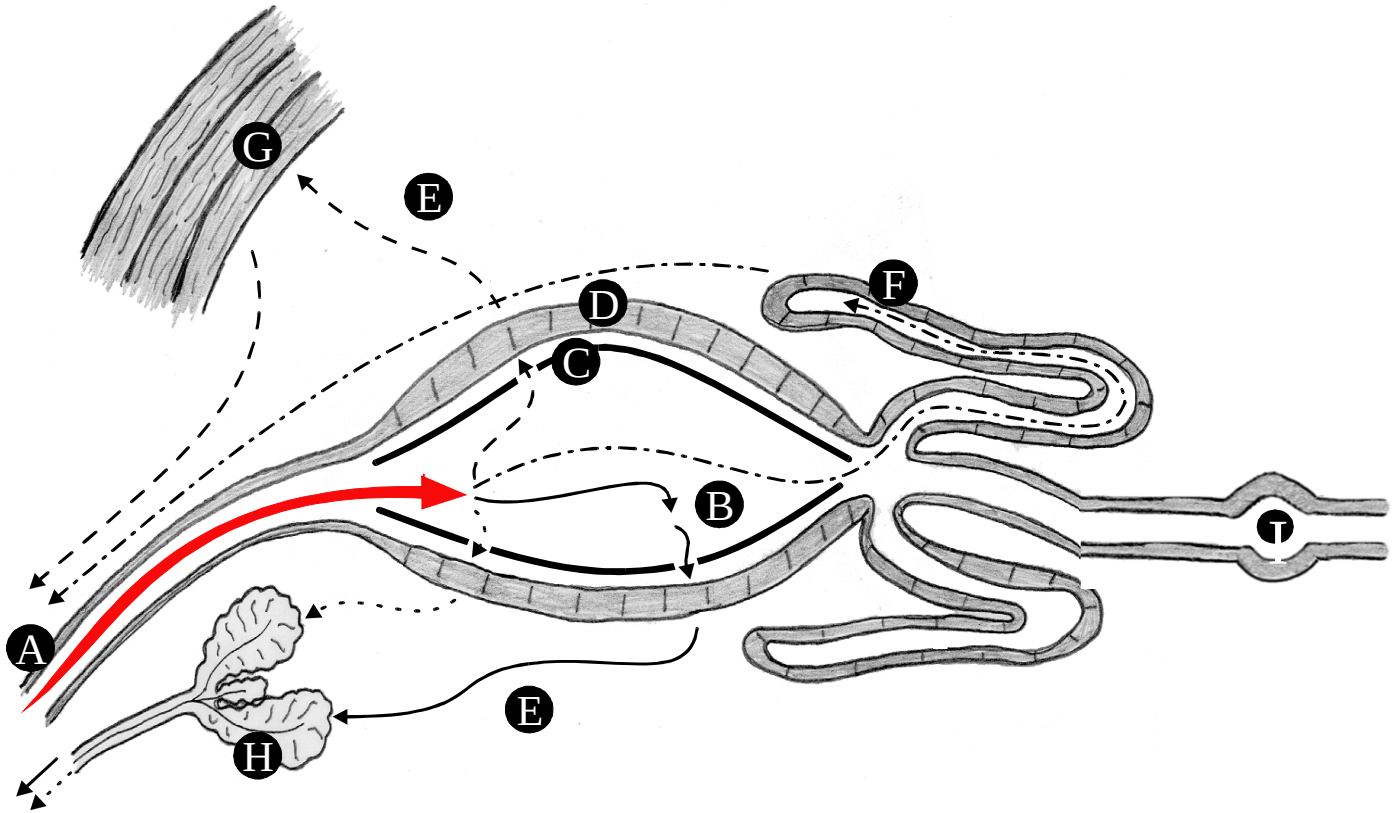
* reduced nutrients available to host
* reduced synthesis of vitellogenin in fat body (Hogg et al. 1997)
* ovary uptake of vitellogenin is impaired (Hogg et al. 1997)
* increase in hemolymph yolk proteins
* resorption of developing follicles (Carwardine and Hurd 1997)
* reduced fecundity (Ahmed et al. 1999)
* reduced fertility (Hacker 1971, Hogg and Hurd 1995)
* bloodfeeding behavior affected (Anderson et al. 1999)

The Vector-Parasite Relationship



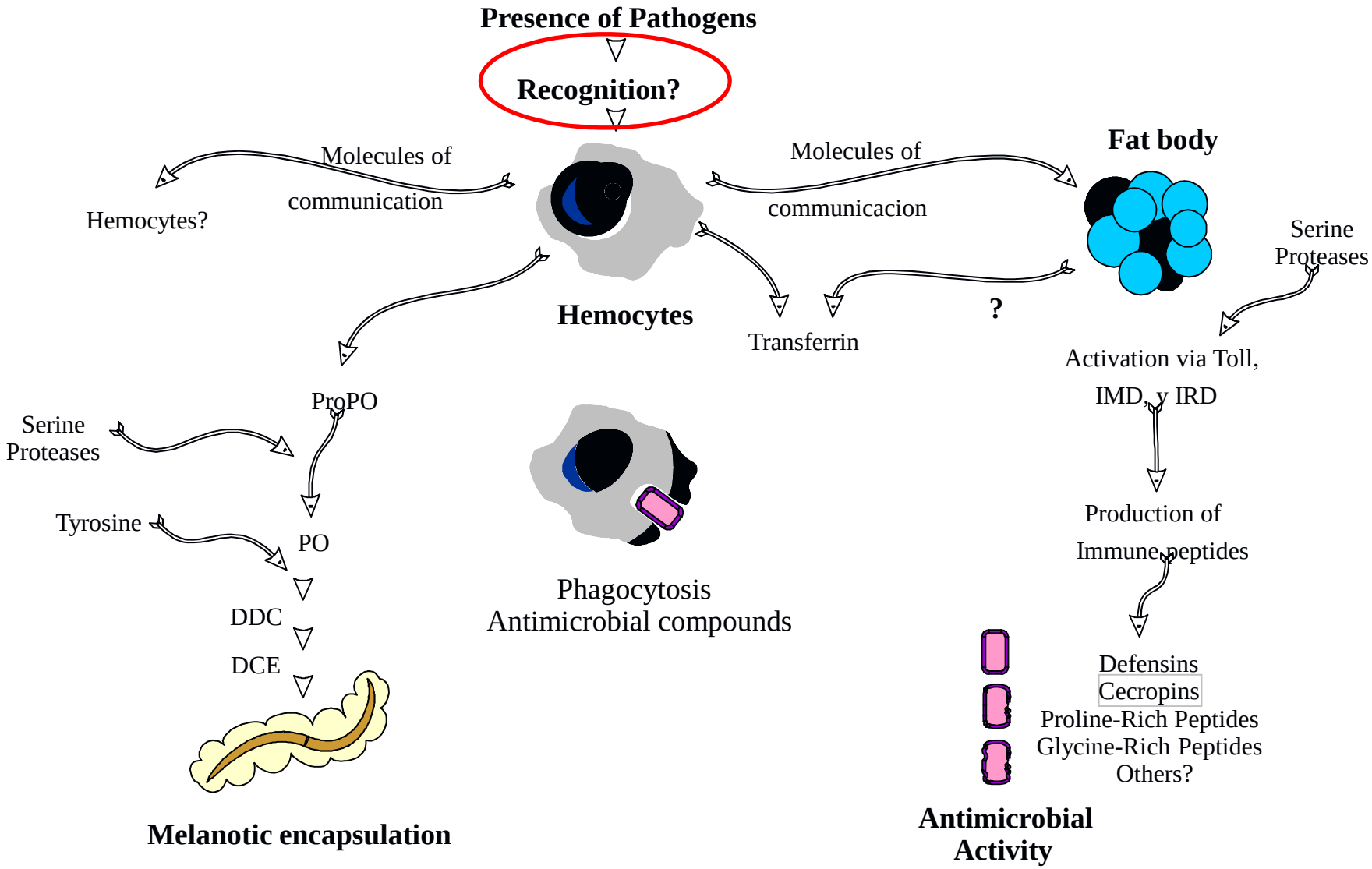
1) Why do insects tolerate parasites and pathogens?
2) How do insects protect themselves against parasites and pathogens?
3) Why do insects not kill all parasites and pathogens?



Why do the insects not kill their parasites???

To answer this we first must understand how insects CAN protect themselves, and then determine why these protective measures are not used or are not effective against the pathogens

Immune Response of Insects



INNATE IMMUNITY OF INSECTS

Innate immunity: nonspecific defense mechanisms used immediately or soon after exposure to a stimulus. This is the immunity one is born with and is the initial response by the body to eliminate microbes and prevent infection.

Innate immunity does not recognize every possible antigen:
It recognizes a few highly conserved structures on different microorganisms.

The structures recognized are called **pathogen-associated molecular patterns (PAMPs**).

The PAMPS are recognized by **pattern-recognition receptors** (**PRRs**).

pathogen-associated molecular patterns (PAMPs) include:

lipopolysaccharide (LPS) from the Gram-negative bacteria cell wall;

peptidoglycans found abundantly in the gram-positive cell wall and to a lesser degree in the gram-negative cell wall

lipoteichoic acids found in the gram-positive cell wall;

mannose-rich glycans (common in microbial glycoproteins and glycolipids);

B-glucans on fungi

To recognize these microbial molecules, various body defense cells have on their surface a variety of receptors called **pattern-recognition receptors** capable of binding specifically to conserved portions of these molecules.

Pattern-Recognition Receptors (Including Toll-Like Receptors)

1. **Pattern Recognition Receptors (PRR)**

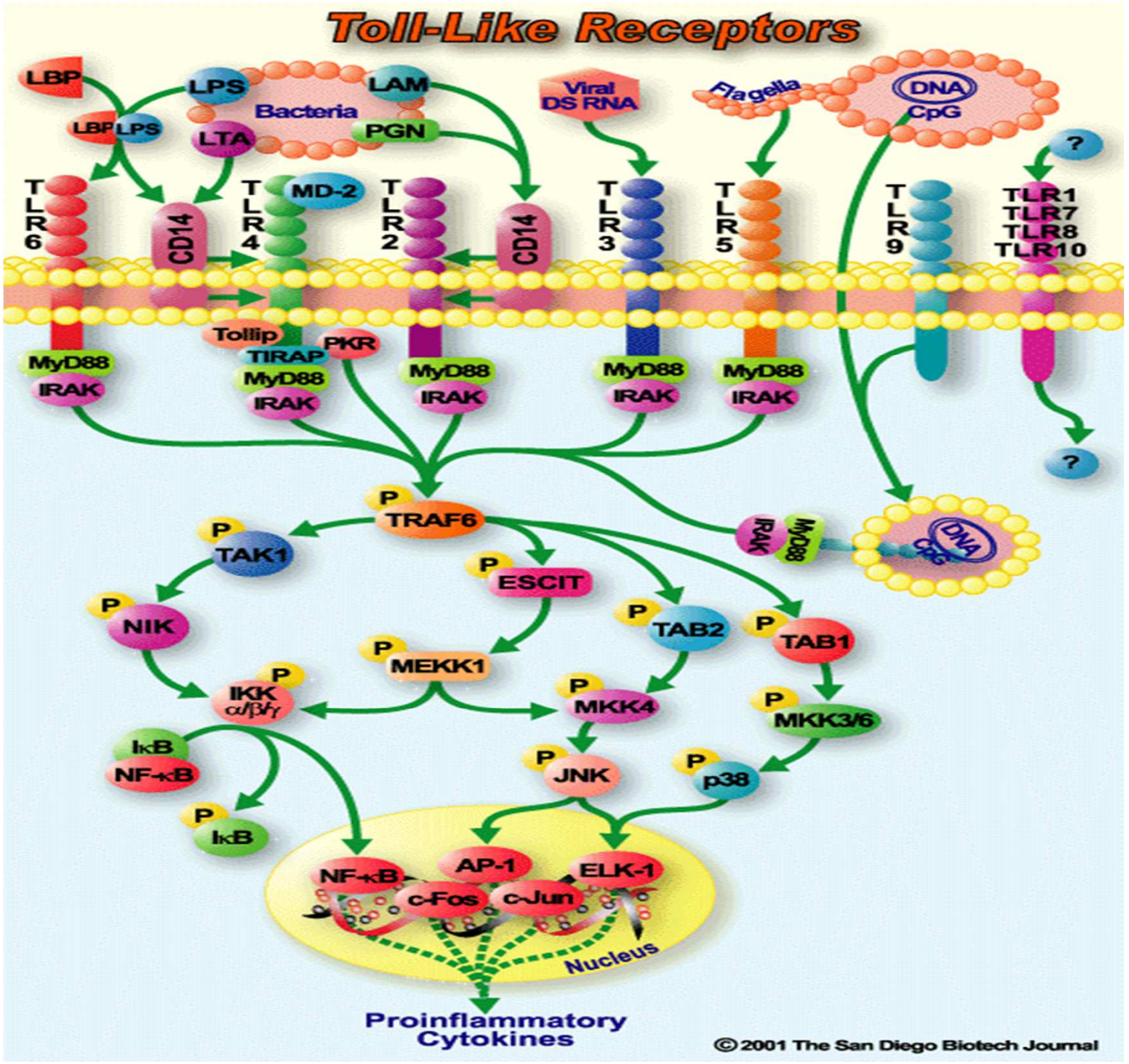
Recognize pathogen associated molecular patterns (PAMP);
conserved molecular patterns on microbes

Toll-Like Receptors (TLR):

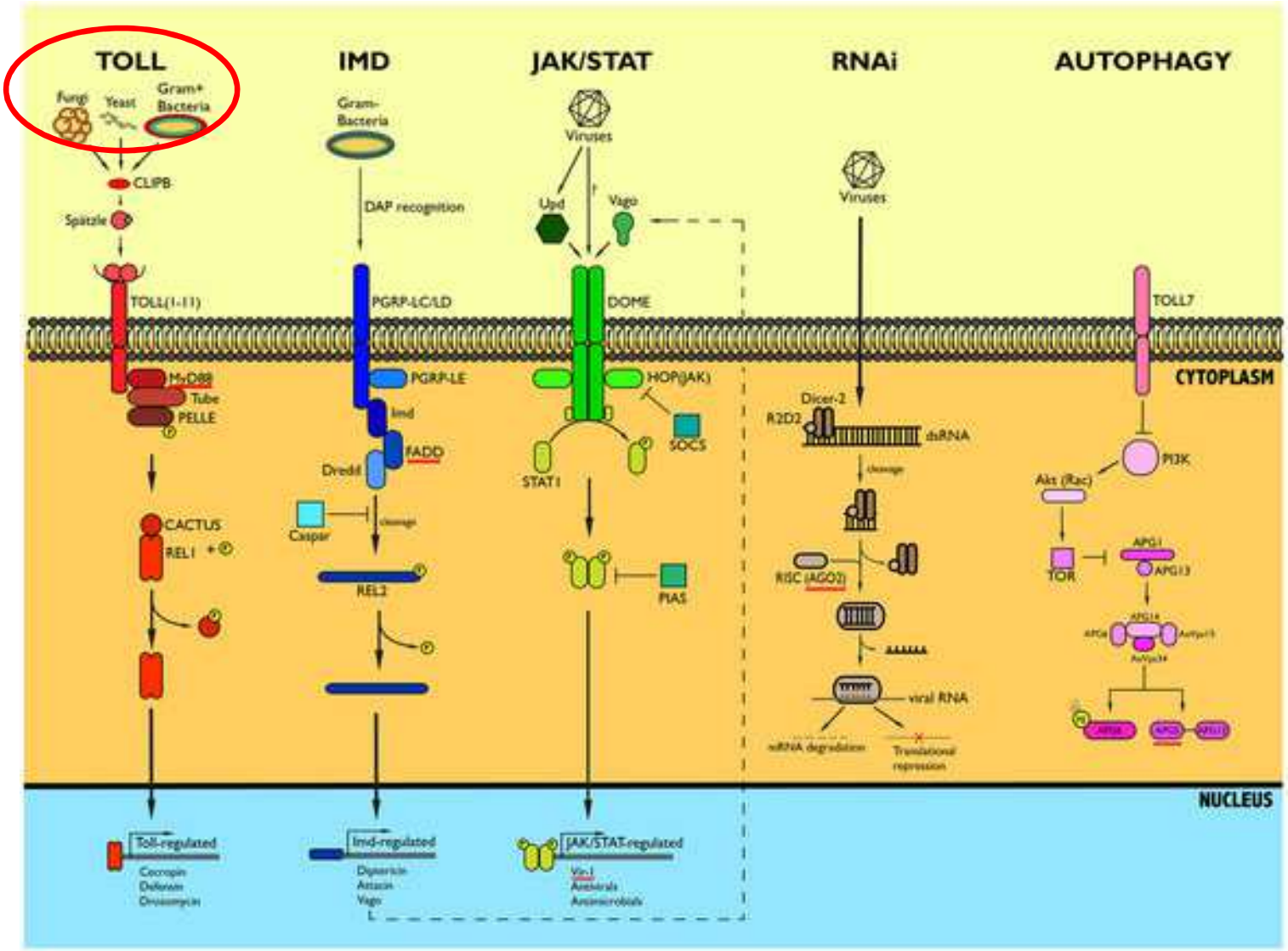
First discovered in Drosophila
Eleven receptors identified in mice and humans

Ligands are **PAMP** (pathogen-associated molecular patterns)

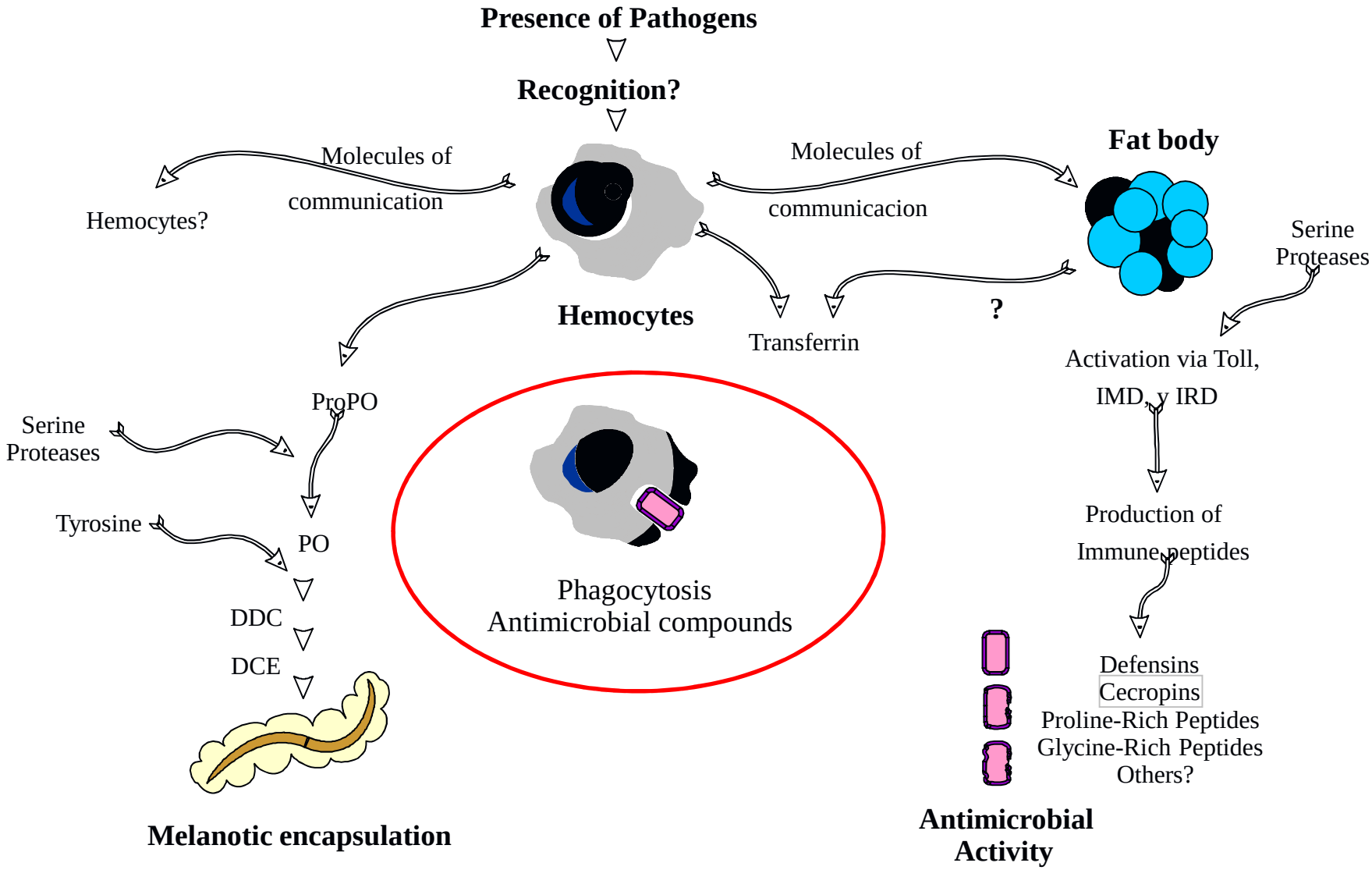
Receptors are **PRR** (pattern-recognition receptors)



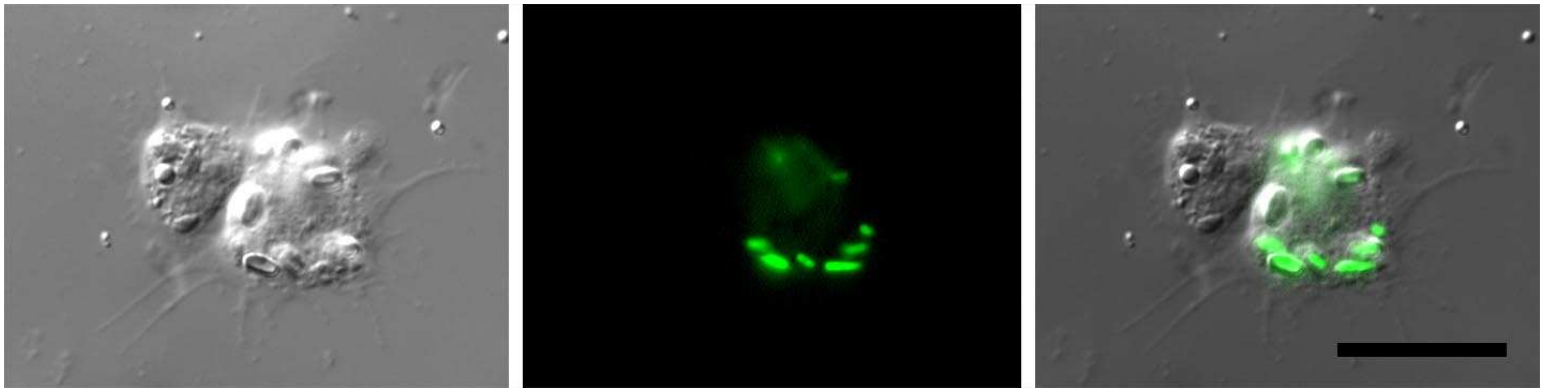
Nobel prize 2011

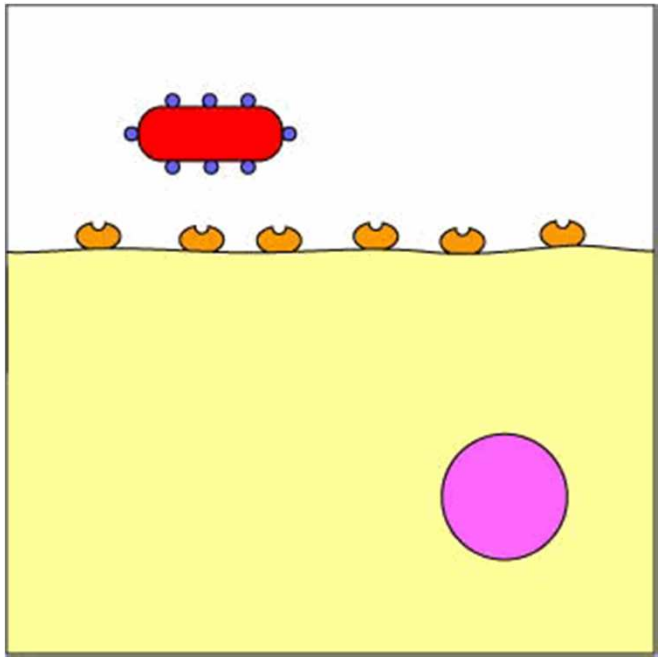


Immune Response of Insects

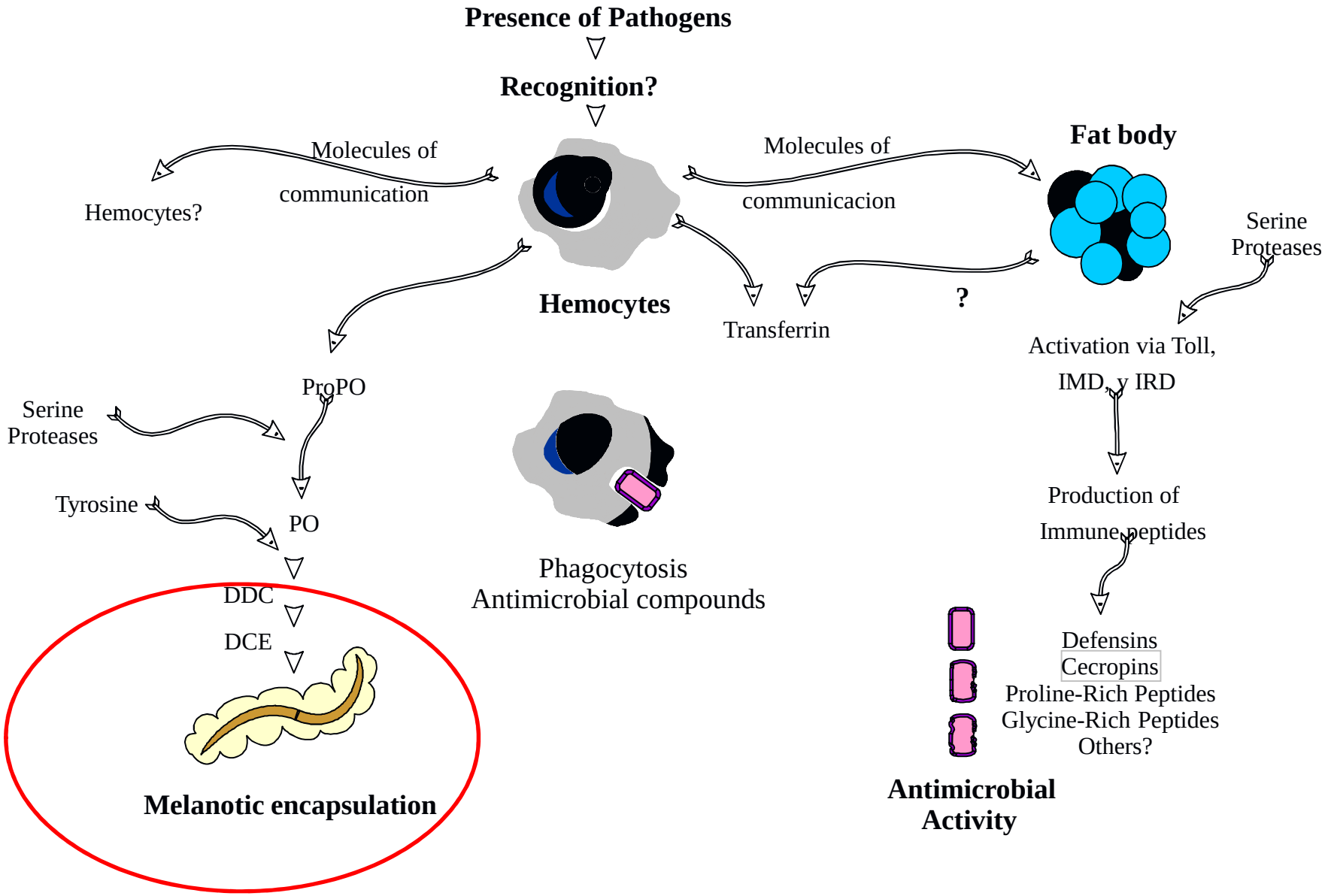


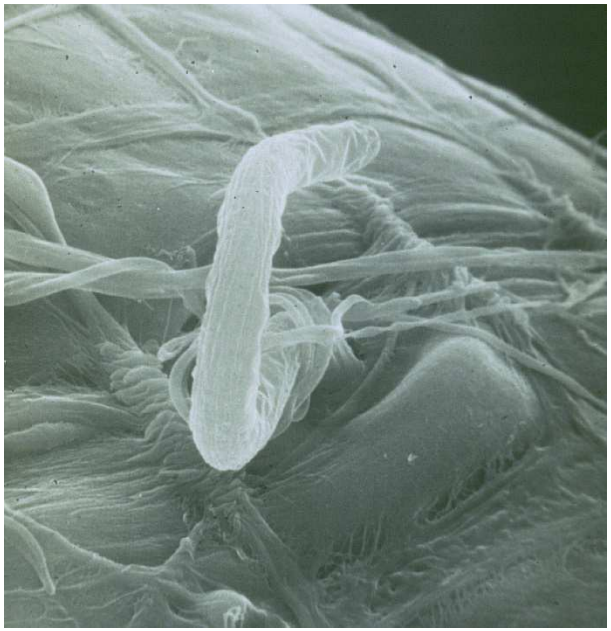
E. coli 3 hr: Phagocytosis





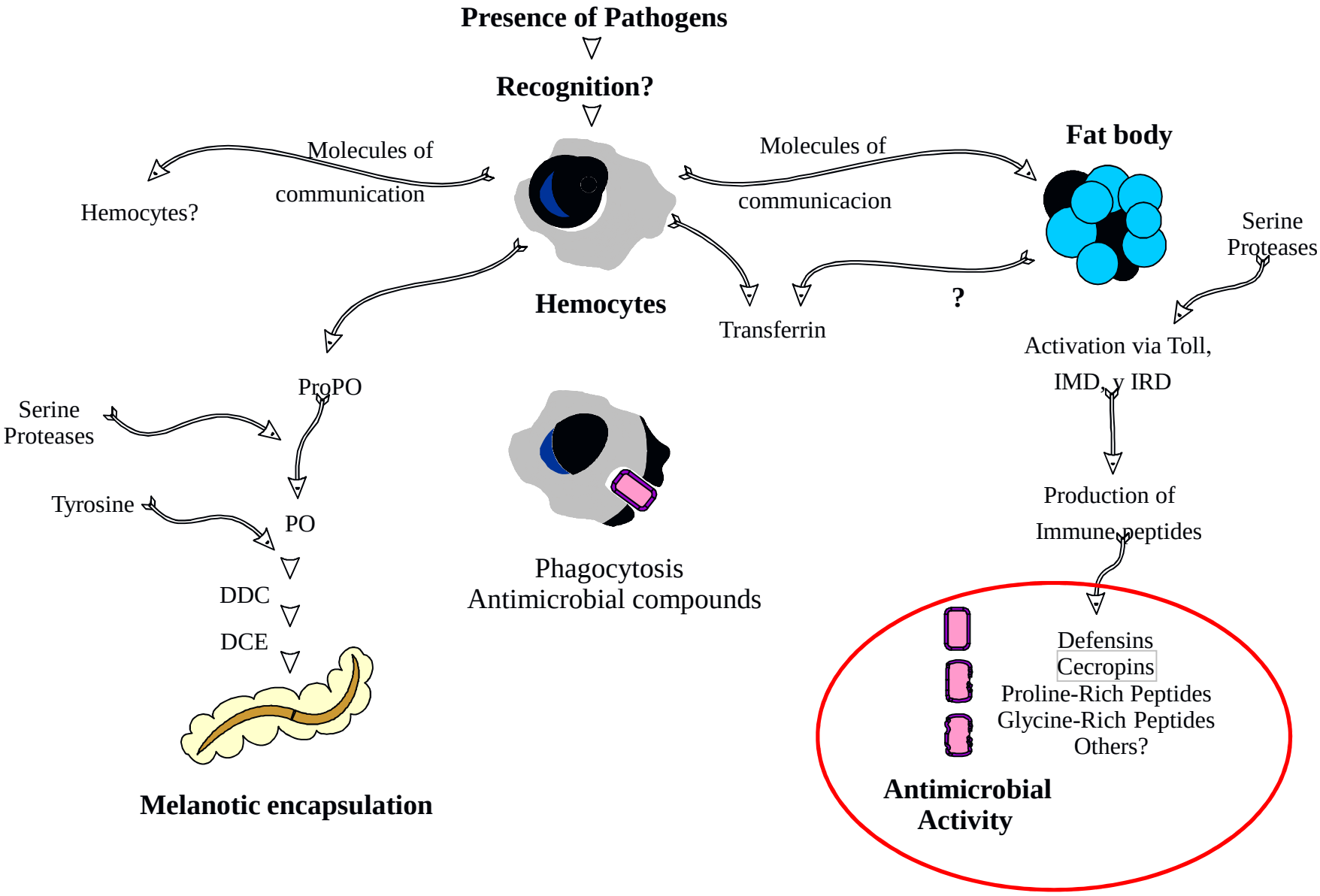
Immune Response of Insects





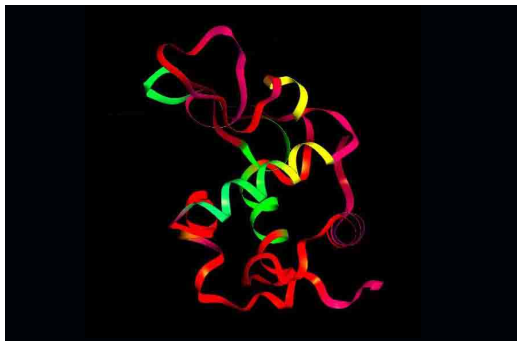
Melanotic encapsulation

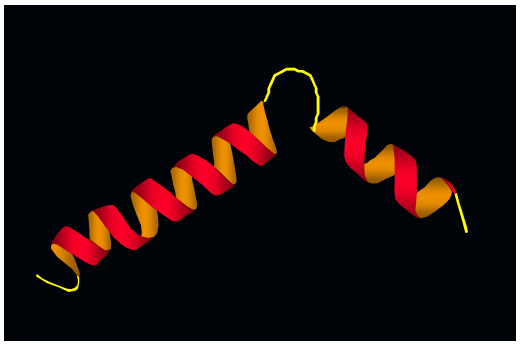
Immune Response of Insects



Invertebrate Antimicrobial Peptides

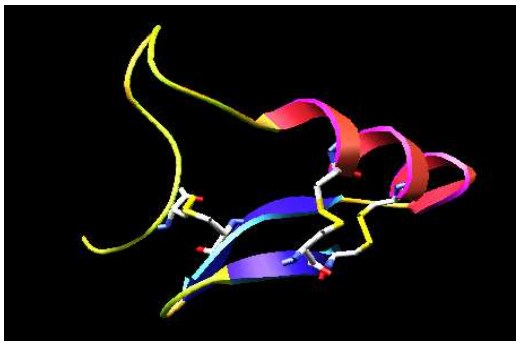
lysozyme





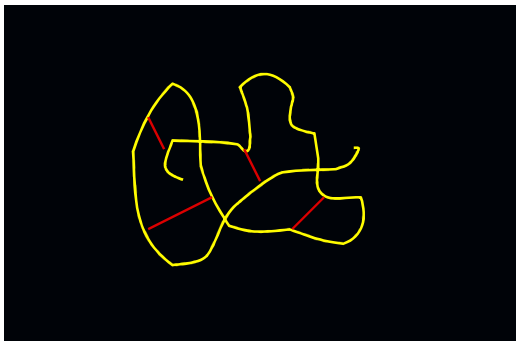
cecropin

Bomanin



defensin

Jacob



gambicin

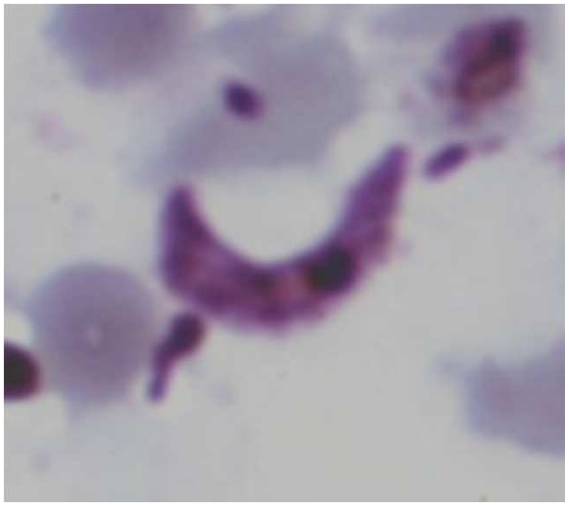
How can parasites survive?

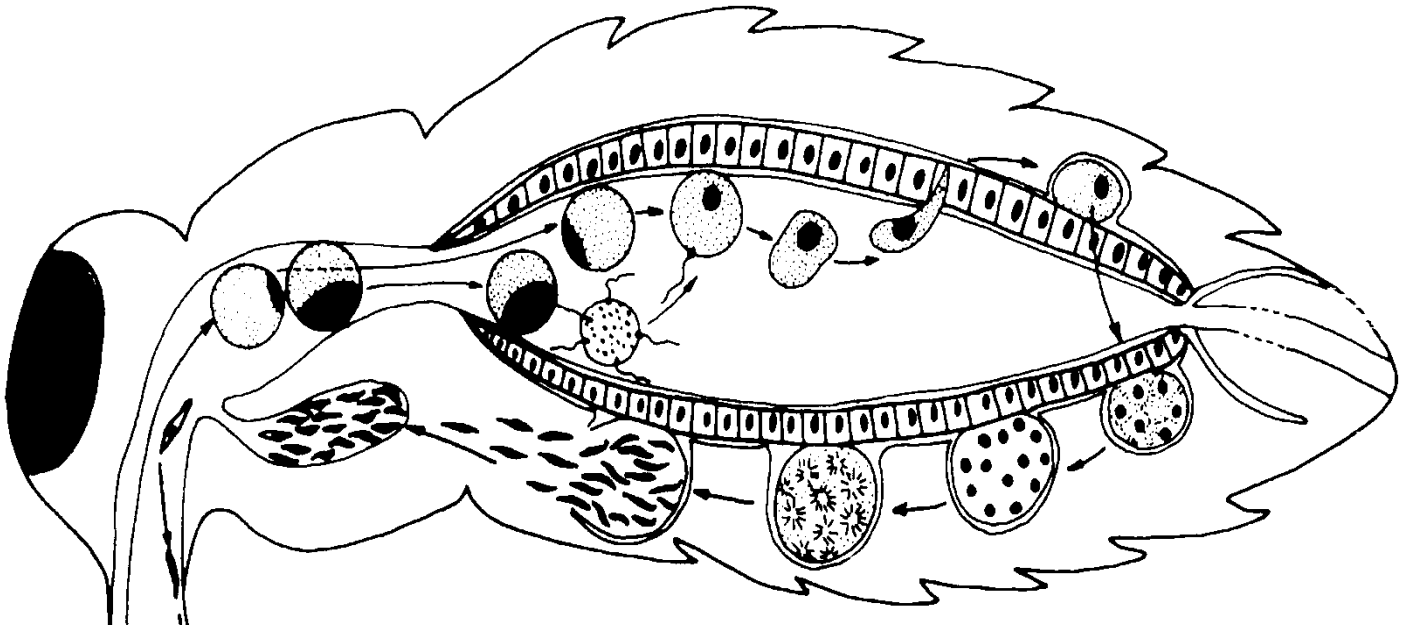
Parasites can:

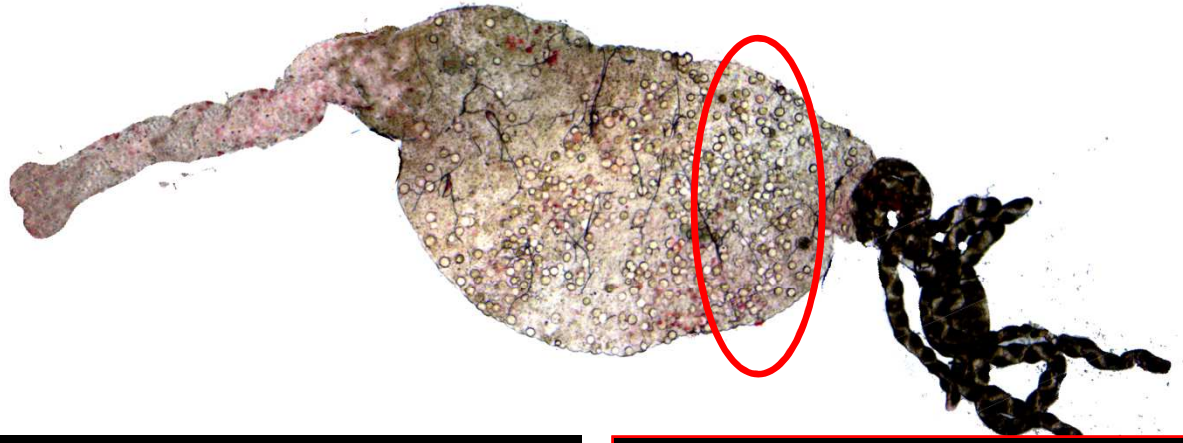
1) Evade immune response
2) Inactivate immune response
3) Avoid contact with immune response

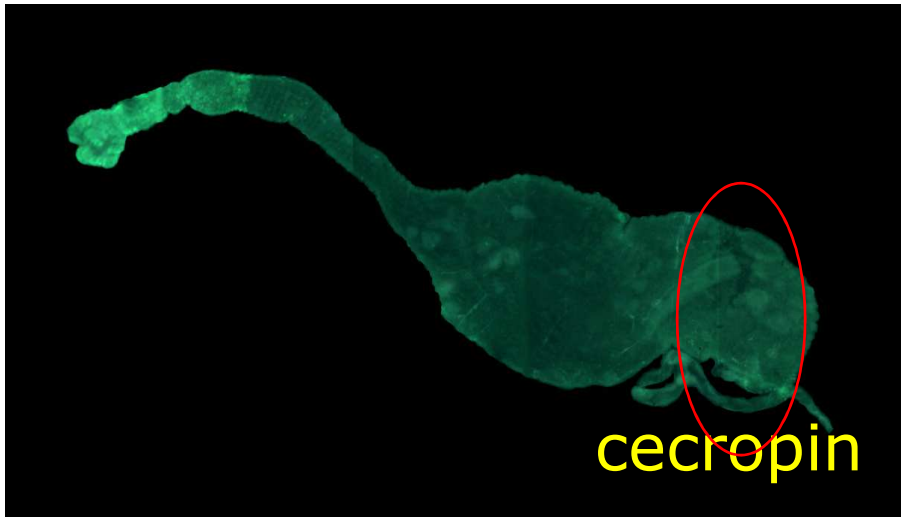
Plasmodium en los mosquitos

- occurs in mosquito (9-21 d)
- fusion of micro- and macrogametes
- zygote → ookinete (~24 hr)
- ookinete transverses gut





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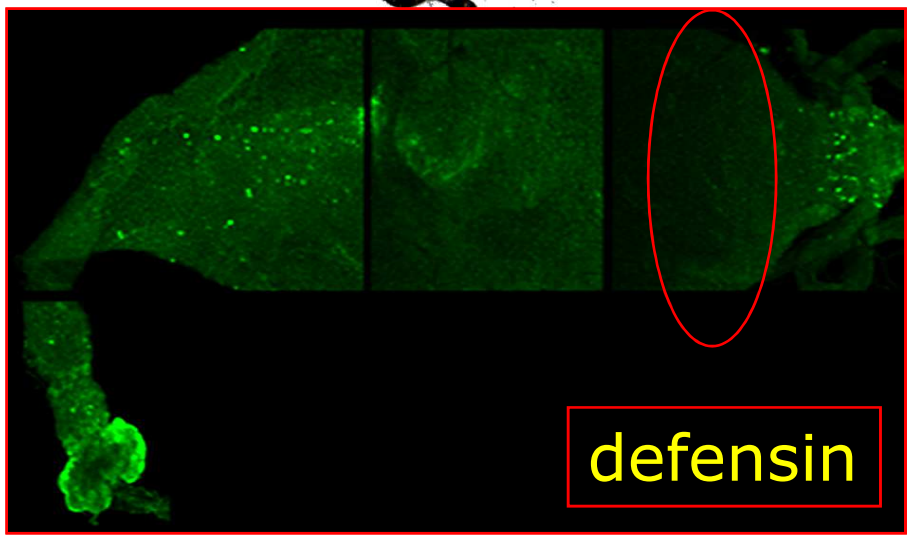
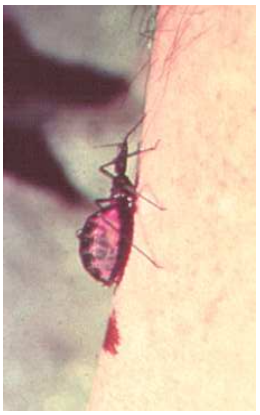
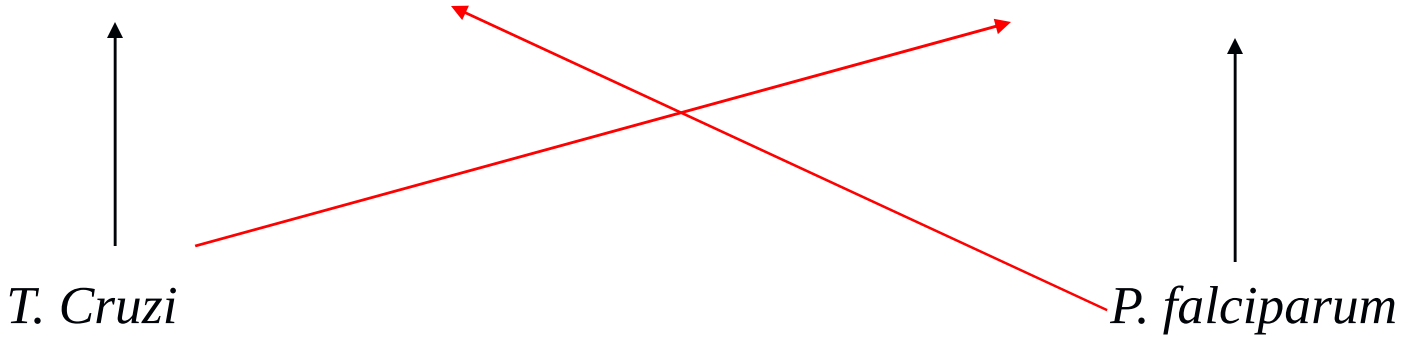

defensin



Rhodnius prolixus

Anopheles gambiae



T. Cruzi

P. falciparum

Fitness Costs Associated with Immune Response

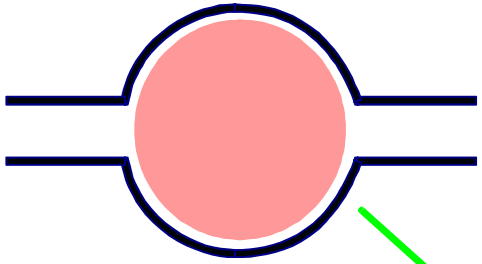
Melanization

* reduced fertility
* increased time to oviposition
* reduced longevity
* competition for resources needed for egg
* development and melanin synthesis

Immune peptides and Phagocytosis

* No apparent reduction in fertility
* No significant reduction in longevity
* Competition for resources?

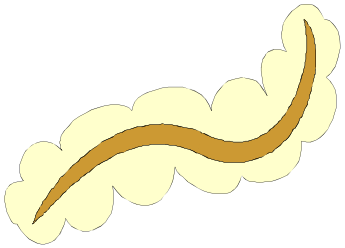
TRADE OFFS



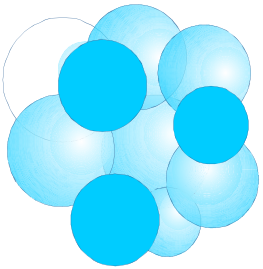
Phenylalanine

PAH

Tyrosine

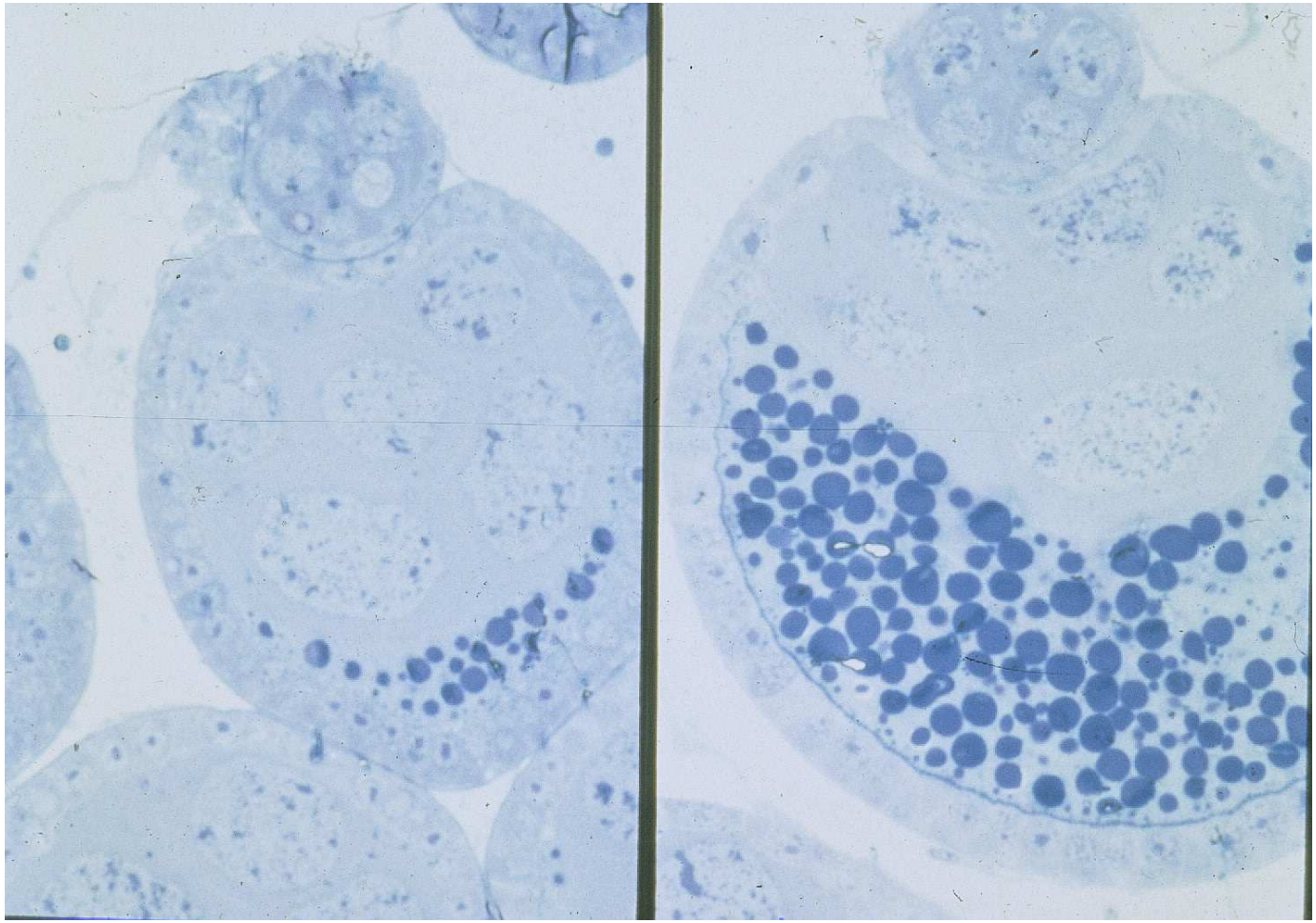


Competition



Defense

Egg Development

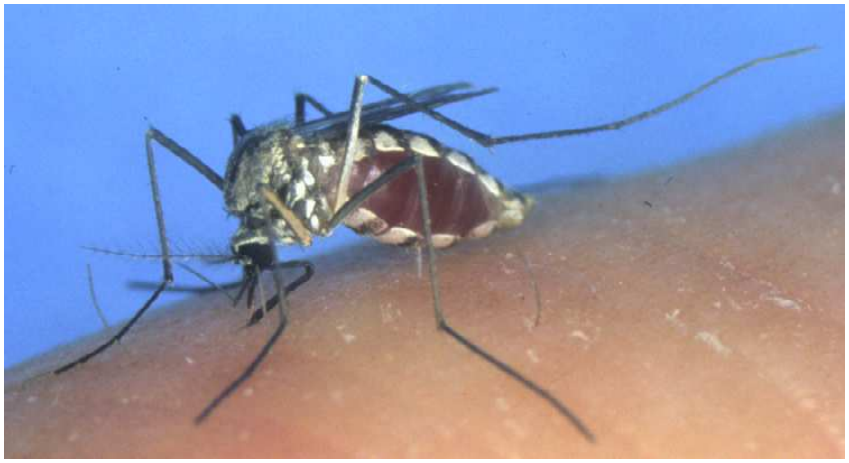


Undergoing immune response

Controls

(Ferdig et al. 1993)

Armigeres subalbatus



Brugia pahangi

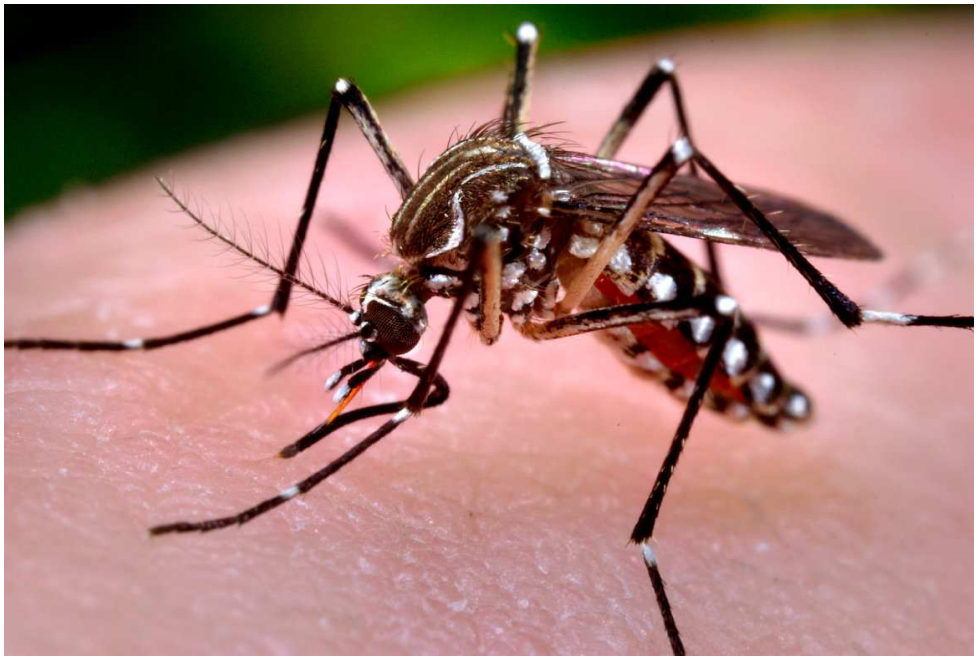
develops

Brugia malayi

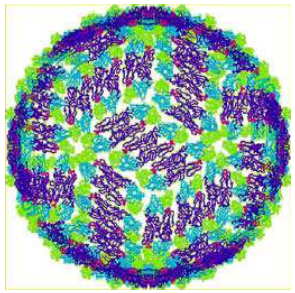
killed

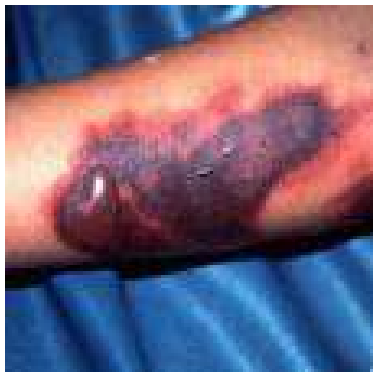
Aedes aegypti

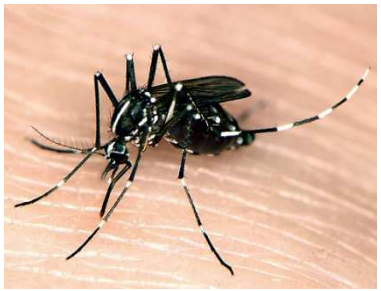
- Vector of Dengue, Zika, Yellow Fever, and Chikungunya
- Present in most tropical and subtropical environments
- Anthropophilic, bites during the day



What happens with Intracellular Parasites?







DENv comprise 4 antigenically distinct serotypes: DENv-1, -2, -3, -4

- 2.5 billion people at risk
- 50-100 million new infections/year
- ~500,000 cases of DHF, DSS
- No vaccine, no drugs

Transmitted by mosquitoes

- *Aedes aegypti, Aedes albopictus, Aedes polynesiensis*

Intracellular viruses are not freely exposed to classical components of the vector immune response

Chikungunya Outbreak 2014

Chikungunya is a viral disease first found in Tanzania in 1952

Symptoms similar to dengue virus

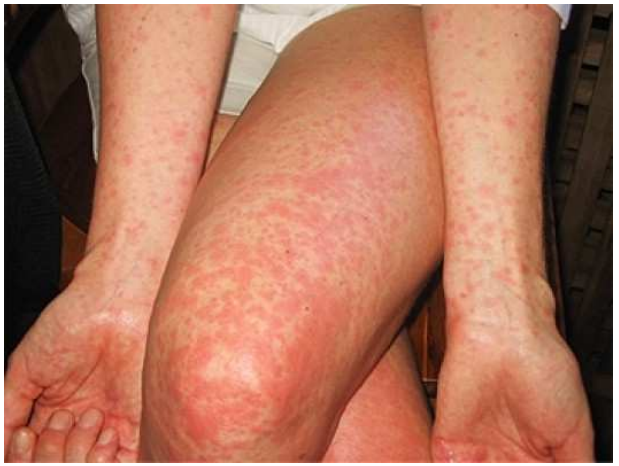
As of January 2015: 34 countries infected in the Americas mostly in the Caribbean, now moving to South America

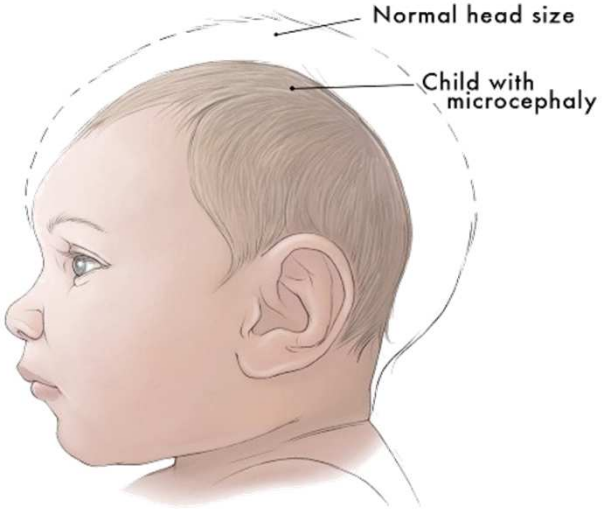
>1,200,000 suspected and 24,000 confirmed chikungunya cases were reported

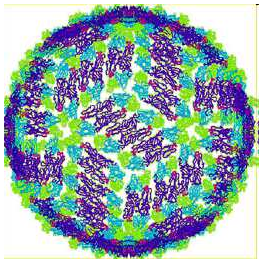
How did it get to the Americas?

Zika: The Disease

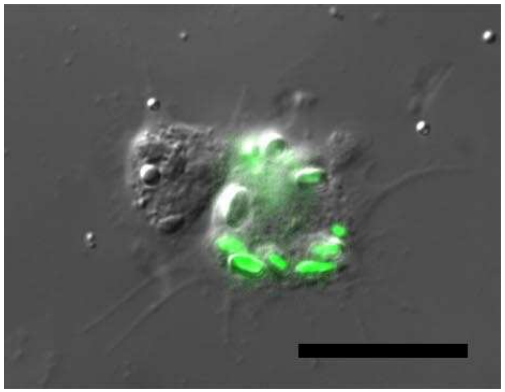
- 1 in 4 people generally develop symptoms
- No vaccine or drugs for treatment
- Symptoms: Fever, joint pain, rash and red eyes
  - Death and severe disease are extremely rare
- Linked to cases of Guillain-Barré syndrome and microcephaly

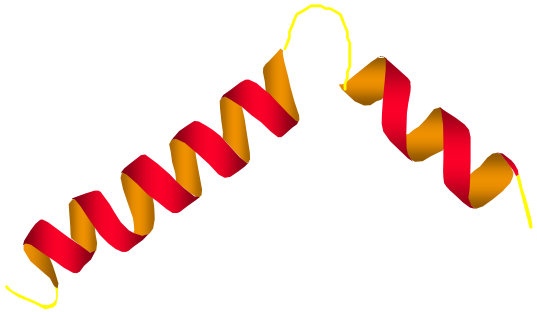






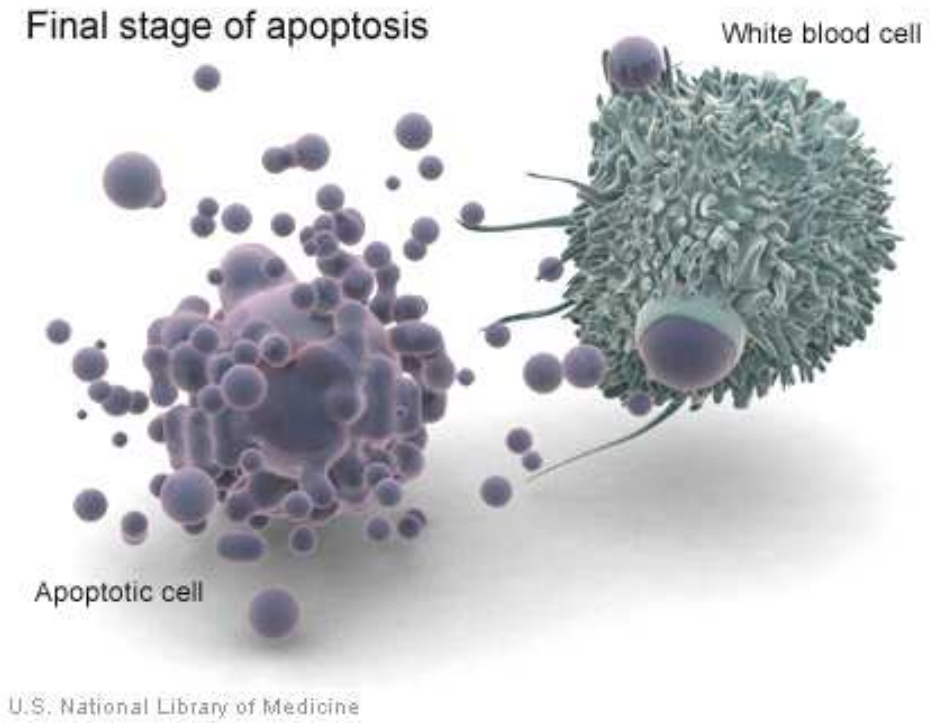
Intracellular viruses are not freely exposed to classical components of the vector immune response





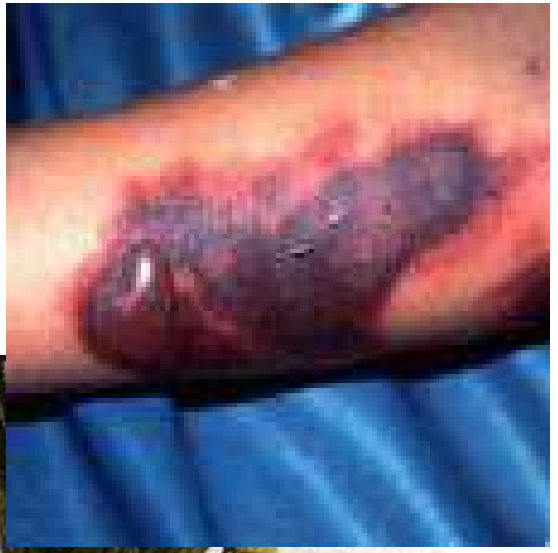
Apoptosis: Programmed Cell Death

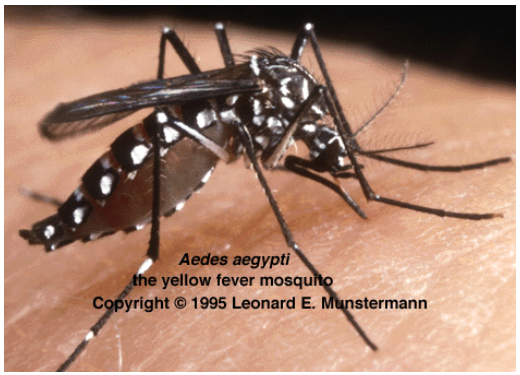
- Cellular response to damage, age, and stress
  - Intracellular infection
- Cells respond to viral infection by initiating apoptotic cell death
- Powerful immune response
  - severely limit virus production
  - reduce or eliminate the spread of progeny virus



Arboviruses: Dengue virus

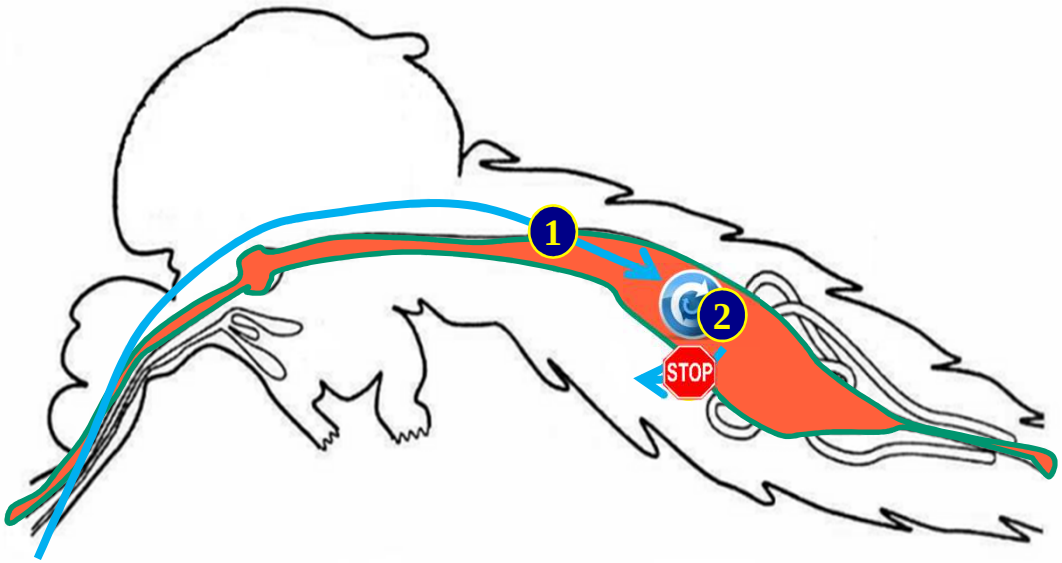
- Geographically wide spread arbovirus
- 2.5 billion people at risk
- 50-100 million new infections annually
- ~500,000 cases of DHF
- No vaccine, no drugs

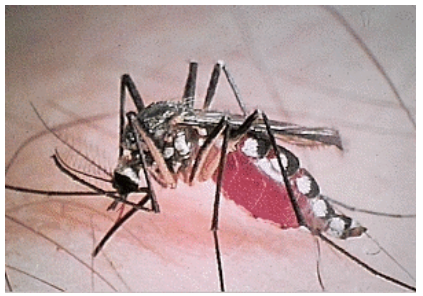




Dengue Refractory *Aedes aegypti*

- **Naturally** DENv resistant populations of mosquitoes found in Cali, Colombia
  - 70% are Susceptible (S) while 30% are Refractory (R)





We believe:

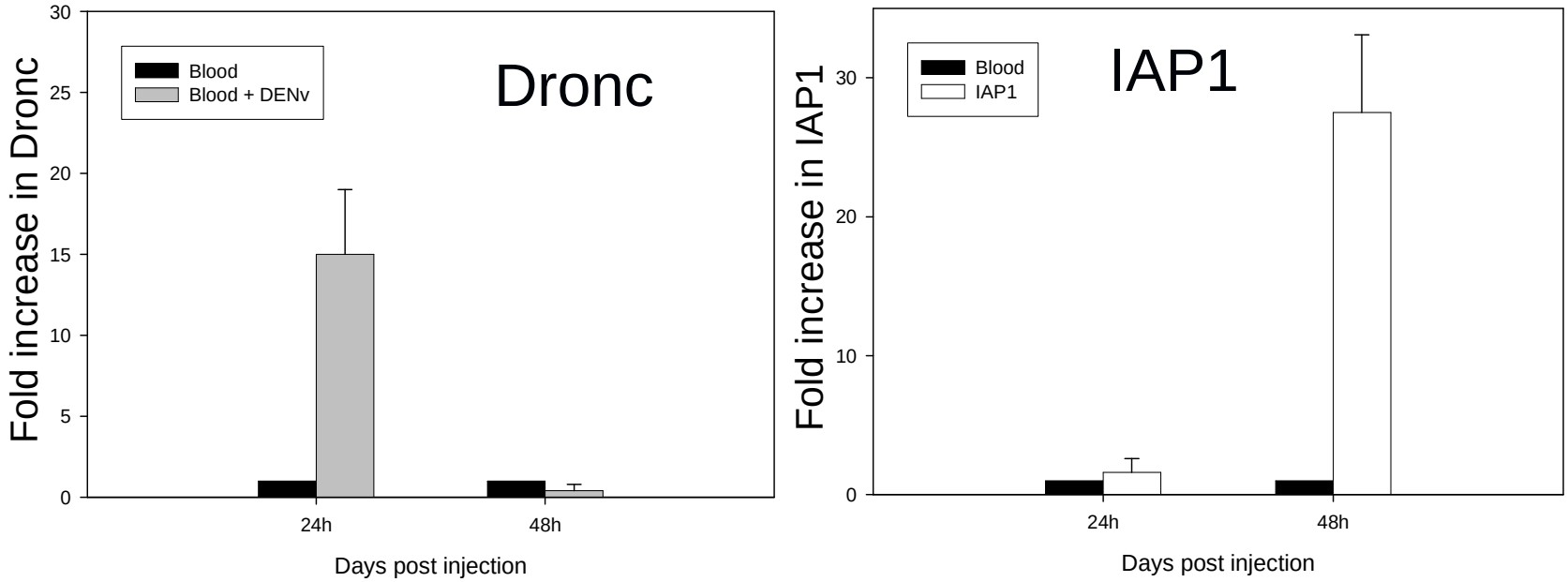
Dengue enters cells-

Mosquito activates apoptosis

virus over expresses IAP1

Apoptosis inhibited until virus has replicated

Cells allowed to burst- releasing virions



Relative expression of *Aedronc* and *Aelap1* during DENv-2 challenge.

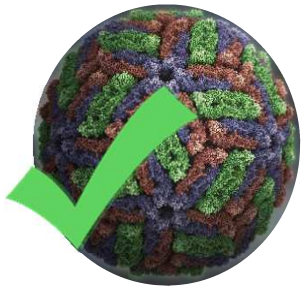
- **Aim 1:** Identify differentially expressed genes that contribute to the refractory (R) or susceptible (S) phenotype

- Tissues from 12 treatments of interest (biological variation within each treatment)

Condition	Strain	Time Period (hours post feeding)		
Blood	S	24	36	48
Blood + DENv	S	24	36	48
Blood	R	24	36	48
Blood + DENv	R	24	36	48

- Aim 1: Identify differentially expressed genes that contribute to the refractory (R) or susceptible (S) phenotype
- We compared genes expressed under different treatments in the different strains
  - Putatively assigned these genes as ‘anti-viral’ or ‘pro-viral’

Pro-Viral (PV) Genes



Highly expressed in S mosquitoes
May help dengue enter & replicate

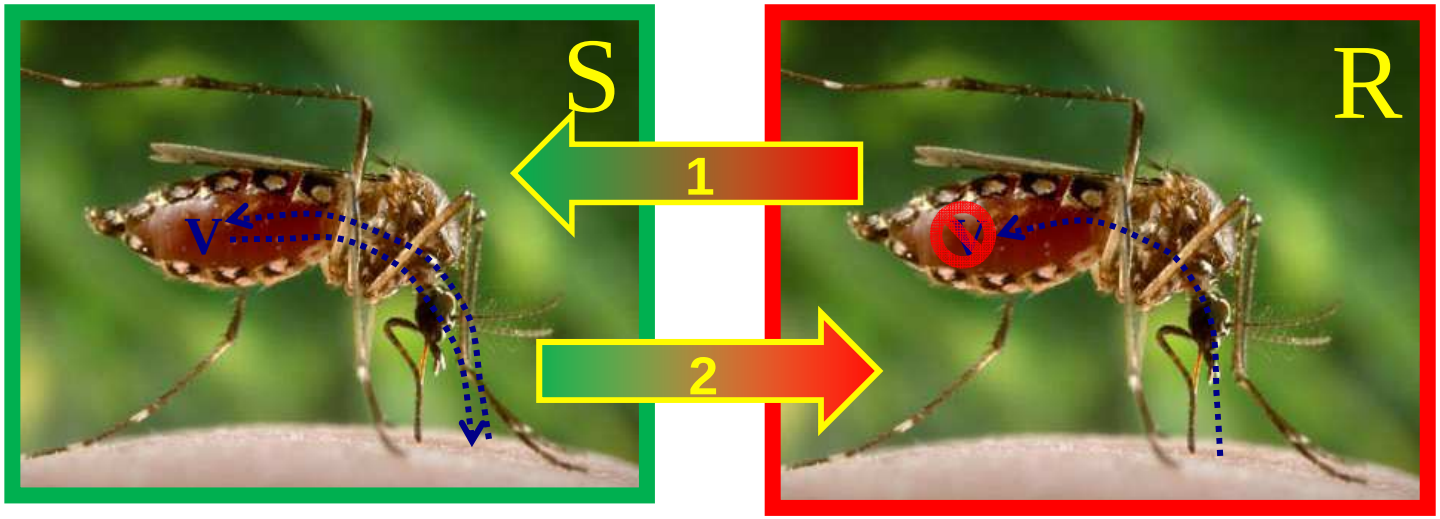
Anti-Viral (AV) Genes

Highly expressed in R mosquitoes
May stop dengue replication

SFU CIDEIM

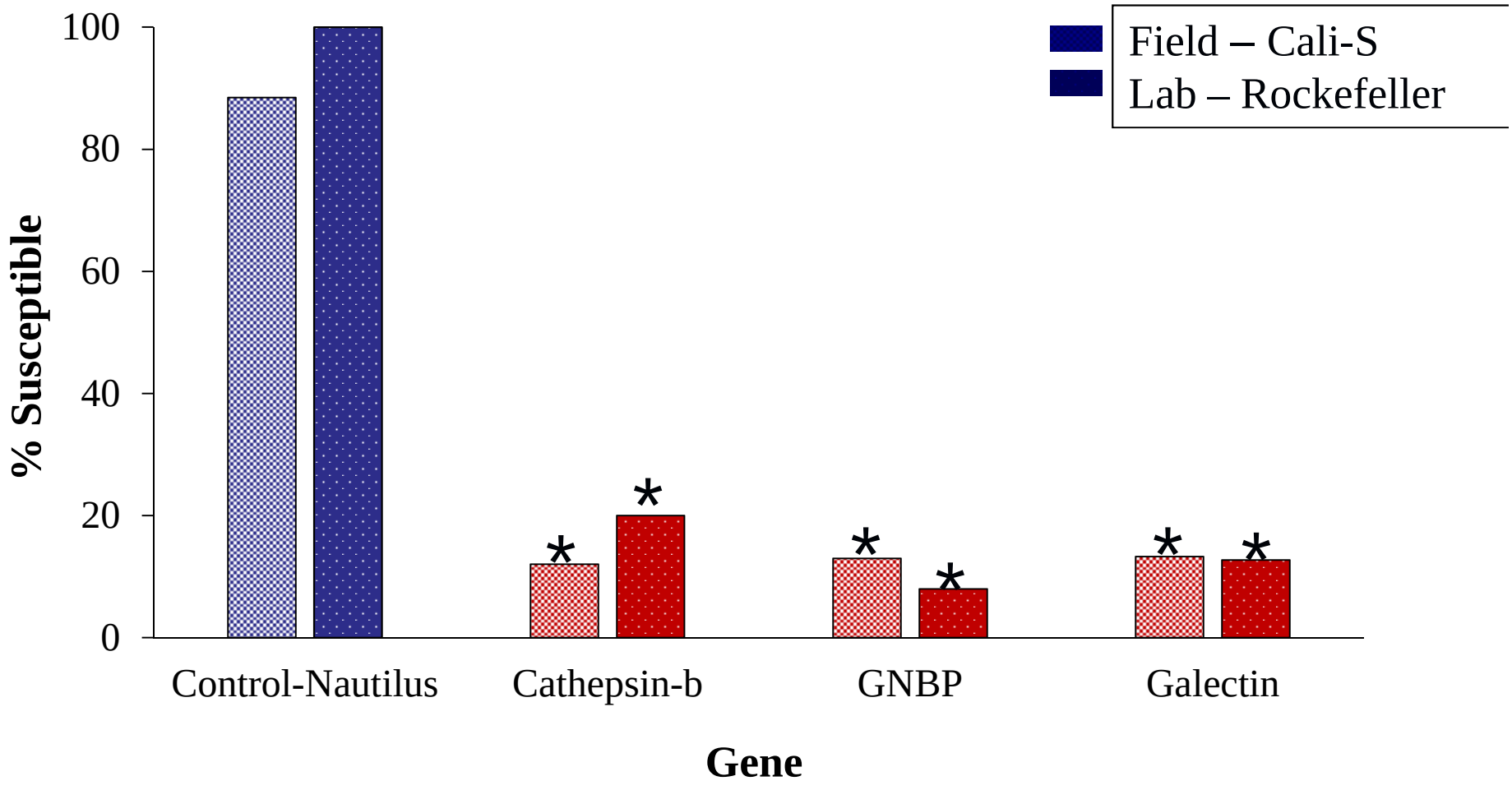
Flipping Phenotypes

- Knock down selected genes of interest
  - **1** - Knocking down **Anti-Viral** genes
    - **Help** dengue to enter cells, aid in replication
  - **2** - Knocking down **Pro-Viral** genes
    - **Stop** dengue from entering cells, stop replication



Flipping Phenotype (S → R)

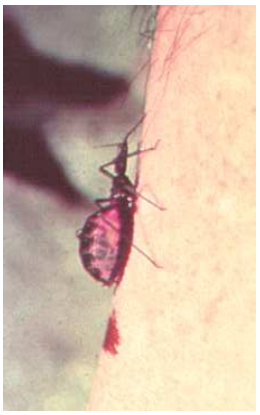
- Knock down of **pro-viral** genes

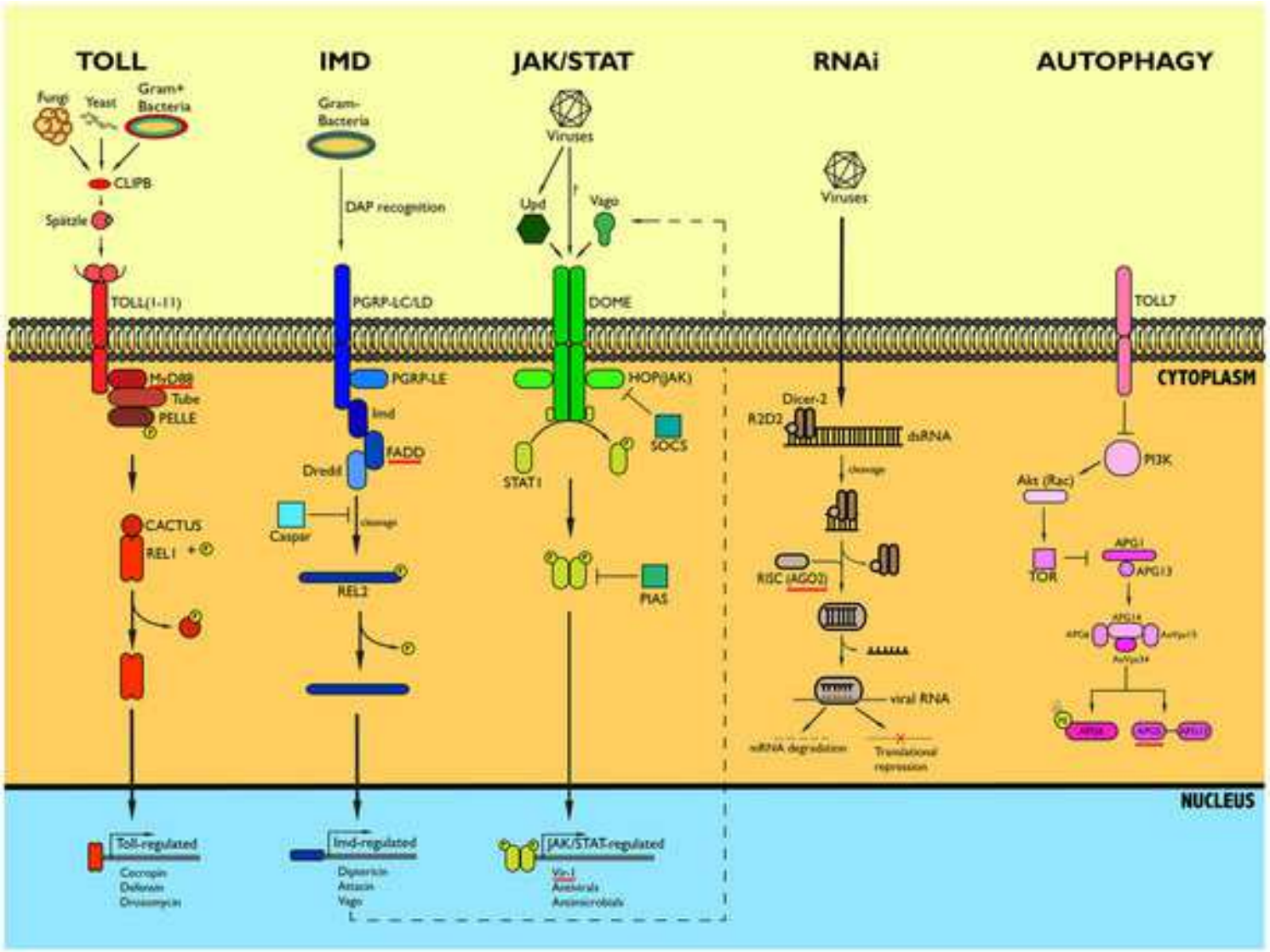


In Summary:

1) Cali-S seems dominant
2) Selection for Cali-MIB and Cali-MEB only reached ~50%
3) Longevity of adults not significantly different
4) Delays in development, time to emergence and bloodfeeding
    - therefore oviposition, and hence fewer gonotrophic cycles
5) Egg hatching reduced

COSTS TO BEING REFRACTORY




TOLL
IMD
JAK/STAT
RNAi
AUTOPHAGY
Fungi
Yeast
Gram+ Bacteria
CLIPB
Spätzle
TOLL(1-11)
MyD88
Tube
PELLE
CACTUS
REL1
Gram- Bacteria
DAP recognition
PGRP-LC/LD
PGRP-LE
Imd
FADD
Dredd
Caspar
REL2
Viruses
Upd
Vago
DOME
HOP(JAK)
SOCS
STAT
PIAS
Viruses
Dicer-2
R2D2
dsRNA
RISC (AGO2)
viral RNA
TOLL7
CYTOPLASM
PI3K
Akt (Rac)
TOR
NUCLEUS
Toll-regulated
Cecropin
Diptericin
Drosomycin
Imd-regulated
Diptericin
Attacin
Vago
JAK/STAT-regulated
Vir-1
Antiviral
Antimicrobials

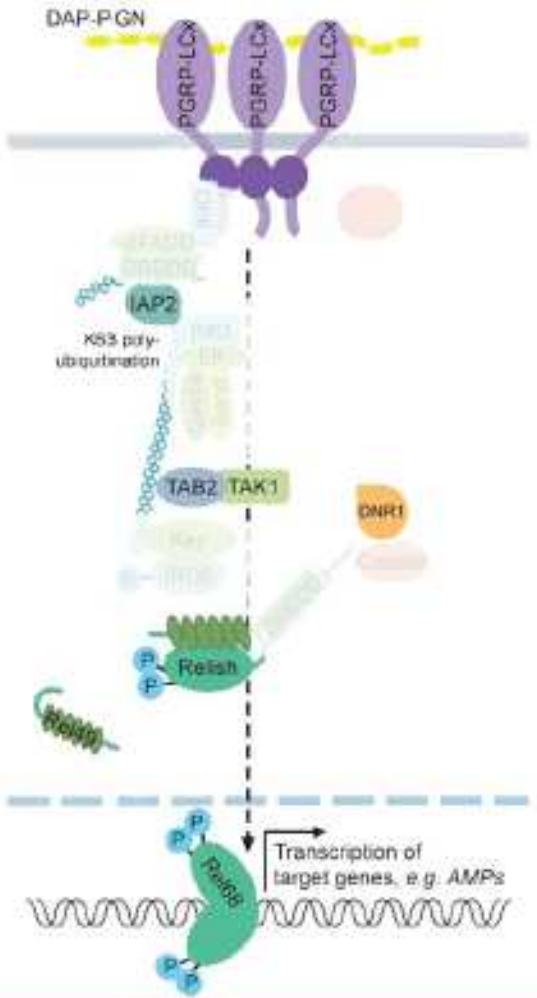
R. prolixus – IMD pathway

Hypothesis 1.

Novel proteins link the existing IMD pathway components.

Hypothesis 2.

Non-canonical pathway



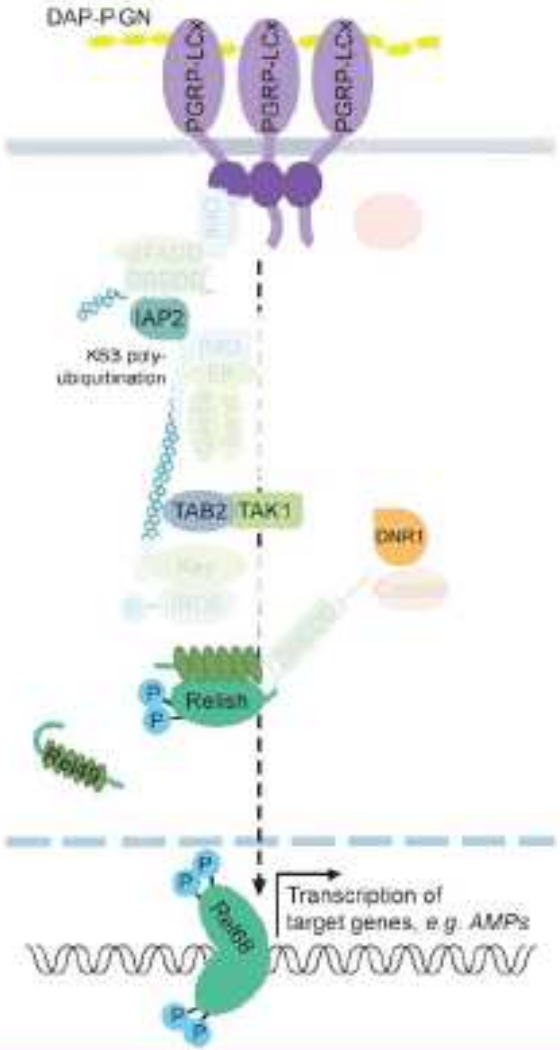
R. prolixus – IMD pathway

Hemimetabolous

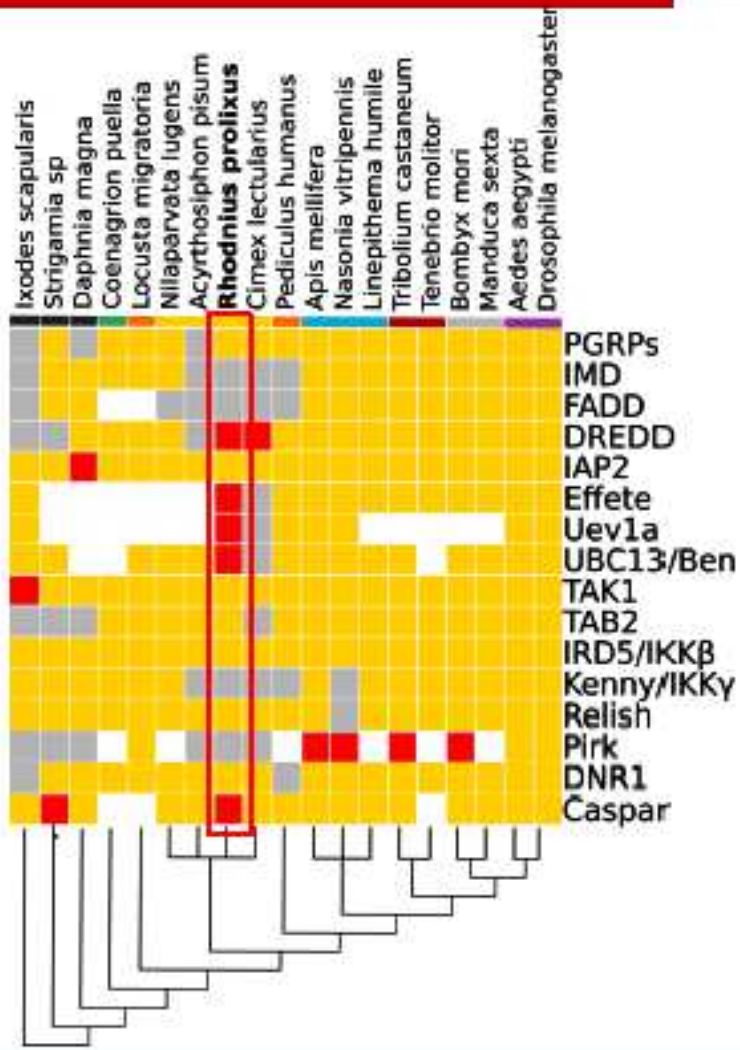
Pea aphid *Acyrthosiphon pisum*

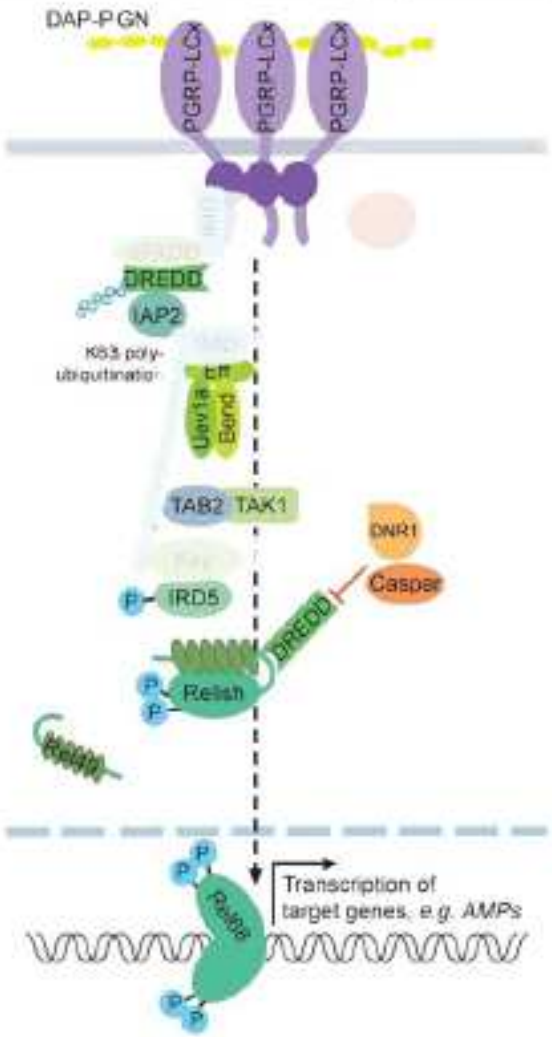
Bedbug *Cimex lectularius*

Head louse *Pediculus humanus*



Objective 1 - Results






CHAGAS

Questions