Blood
Immunity

Insects are Different

Blood, circulation and immunity

- Anatomy
- Blood flow
- Thermoregulation
- Immunity
- Other functions

Functions of insect circulatory systems

- thermoregulation
- immunity
- reservoir of substances of maintain homeostasis (eg. water)
- transport of macronutrients, hormones
- hydraulic fluid/skeleton

Is there a major function missing?

Hemolymph does NOT transport respiratory gases!

What is in hemolymph?

- Water – 90%
- Ions and molecules in solution
- Hemocytes (blood cells)

In insects, the tracheal system takes gases (oxygen and CO₂) to and from tissues
Anatomy of circulatory systems

- pumps and tubes (muscles, vessels)
- devices that control direction of flow (diaphragms, ostia)
- interaction with nervous and endocrine systems

Vessels: aorta, heart
Supported by dorsal diaphragm
Muscle: alary muscles drive flow

structures that control the direction of flow (sheets and holes)
- Diaphragms, dorsal and ventral
- Ostia (s. ostium)

dorsal and ventral diaphragms
Ostia can let flow

- In only
- Out only
- Both ways

how do they get blood into antenna, legs, wings?

accessory vessels and pulsatile organs

Insect Leg APO

- leg space divided by septum
- valves and heart muscle work to push and pull blood along

In some insects, the story is more interesting: flow can reverse

- found in adult, winged Holometabola
- probably related to flight

Standard story: Cyclical contraction, relaxation of heart draws blood in and contraction moves it forward
the case of *Manduca sexta*

- the larval heart has forward flow only

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the case of *Manduca sexta*

- in the adults, flow can alternate forwards and backwards
- this means that there should be two pacemakers, one at each end

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Cardiograms in *Manduca*

- slow backward (retrograde)
- fast forward (anterograde)
- pause

*Manduca* heart physiology – factors affecting contractions

- muscle contractions/stretch (myogenic)
- neural input

- Do experiments manipulating each of these.

How can neural input be manipulated?

1. Chemically
2. Cut/stimulate nerve

Tetradotoxin is a neurotoxin that blocks nerve output
Effect of TTX

- How do you interpret this result?

Which nerve?

Is this nerve the rear pacemaker?

- Cut the dorsal 8 nerve. What happens?

Forward phase stopped
Only backward phase remains

What happens when you stimulate the nerve?

Stimulation cause change to
**Results**

- neurotoxin stops forward flow, only backward flow remains
- transecting the nerve to the rear heart chamber stops forward flow, only backward flow remains
- stimulating of the nerve during the backward flow cycle causes immediate shift to forward flow

**Interpretation**

- posterior pacemaker is neural
- posterior pacemaker can override anterior one
- anterior pacemaker is probably myogenic and requires no neural input

**The Role of Hemolymph in Thermoregulation**

- muscles must be >32 deg. to support flight
- muscles generate heat when contracting
- how can this heat be kept in the thorax?

**Thermoregulation**

- muscles must be >32 deg. to support flight
- muscles generate heat when contracting
- how can this heat be kept in the thorax?
Countercurrent exchanger

- Waist is counter current heat exchanger
- Heat of backward blood is transferred to forward blood

Honey bees use another wrinkle

- Aorta is coiled
- Adds extra length to heat exchanger

The Case of the Winter Moths

- Winter Noctuid moths emerge as adults in fall or late winter.
- They feed, mate, and lay eggs before dying in the spring.
- Insects need to have warm flight muscles to fly (35º C).
- How do winter moths do this?

Attract moths with bait

- At rest, same temperature as air
- Ready for flight, temperature is 30º C, even when air is at freezing (0º).

Producing Heat

- After landing, thorax temperature drops to ambient
- To heat up muscles, shiver
- Must be 30º C to takeoff
Huge loss of heat to environment

- they don’t heat up to 30º unless they have to fly
- fly, land, shiver, fly, land, shiver...
- How to conserve heat?

How do they conserve heat?

- furry thorax
- ears between thorax and abdomen enclosed by air sacs - excellent insulators
- air sacs also channel returning blood flow around heart

restricting heat to the thorax

- counter current heat exchangers in BOTH abdomen and thorax
- moths get heat stroke if it is >20º outside

how winter moths do it

- behavior: shiver, fly, land.....
- insulation: air and hairs
- anatomy of circulatory system: 2 counter current heat exchangers, air sacs channel return flow

contrast tropical moths

- no special exchangers
- heat shunted directly to head and return flow
Immunity

defenses against disease and parasites

Immunity

• a trait of organism not to become diseased when exposed to potential parasites and pathogens

Some types of immunity

• Behavioral defense
• Biochemical defense – proteins or other biomolecules
• Innate immunity – hereditary protection against infection with little capacity for memory
• Acquired immunity – enhanced protection against re-infections with the same kind of pathogen

Innate Immunity

• “non-specific” mechanisms that protect an organism from intruders
• can be cellular, chemical, or structural (cuticle)

Cellular responses

• Phagocytosis
• Nodule formation, encapsulation
• Release of defensive proteins

HEMOCYTES are the primary agents of immunity
Where do hemocytes come from?

- hemopoietic organs
- usually near the heart
- in larval Lepidoptera, in wing discs

Prohemocyte

- may be stem cell for other hemocytes
- mostly nucleus
- ~5% of all hemocytes

Granular hemocytes

- Granular hemocytes phagocytose small objects
Plasmatocyte

- encapsulation of objects too large to be phagocytosed.

Encapsulation

larger multicellular invaders (parasites)

Encapsulation

oenocytoids

Oenocytoids lyse within 3 minutes – release phenoloxidase which crosslink proteins

P, G, +OE cooperate in nodule formation

- Attack on large numbers of bacteria
Defensive proteins - the next level

- Immunity proteins are generally antimicrobial-
- Over 25 families have been isolated and sequenced

Antibiotic proteins

- **Hemolin** in moths is an immunoglobulin induced by pathogen
  It in turn induces 15-25 proteins including cecropins and attacins
- **Cecropins** and **Attacins** disrupt bacterial membranes

Innate immunity can have some specificity and memory

The Experiment

- Innoculate some flies with yeast and some with bacteria
- Later inoculate pretreated flies and untreated flies

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Drosophila infected with bacteria or yeast

- Green spots are proteins from blood of untreated flies and orange spots are from inoculated flies
- Treated flies produce different proteins than untreated ones
- Bacteria treatment produces different response than yeast treatment

Yes there is some specificity and memory
Significance of the different patterns?

- Different classes of pathogens stimulate different pathways of immune responses

2 pathways for different pathogens

- Fungi and gram+ bacteria activate toll pathway and result in expression of one set of immunity genes
- Gram- bacteria activate IMD pathway and result in expression of another set of immunity genes

Mosquitos take immunoglobulins to an even higher level of specificity

- AgDscam is an alternatively spliced hypervariable immunoglobulin domain-encoding gene
- Down syndrome cell adhesion molecule!
- 101 exons that can make over 31,000 forms with different combinations of adhesive domains and interaction specificities

Mosquitos take immunoglobulins to a higher level

- Just looked at one region of the gene, different sections were expressed in response to different pathogens
- Tested bacteria, surface proteins from bacteria, plasmodium (malaria bug)

Do insects have acquired immunity?

- Response to specific antigens that results in increased future responses to that same antigen
Can arthropod immune systems remember?

- Yes
- But how long do they need to remember anyway?

Some families of flies have greater resistance to parasitoid wasps

- **COST:** Larvae that are genetically more immune to wasps grow more slowly
- **COST:** Immune larvae grow up to be adults with reduced fecundity

*A Cost of Bearing Arms*