

# **New Theory on Mosquito Larvae Respiration Contradicting the Scheme of Direct Atmospheric Gas Exchange**

**Presentation to:**

**RBM**

**February 2020**

**Geneva Switzerland**

# Contradicting Classical Theory of Mosquito Larval Respiration

**The classical theory of respiration is that the siphon and dorsal tracheal trunks play obligate roles in respiration by exchanging metabolic gas with the atmosphere. Our results indicate the tracheal system at pressure and isolated from the environment.**

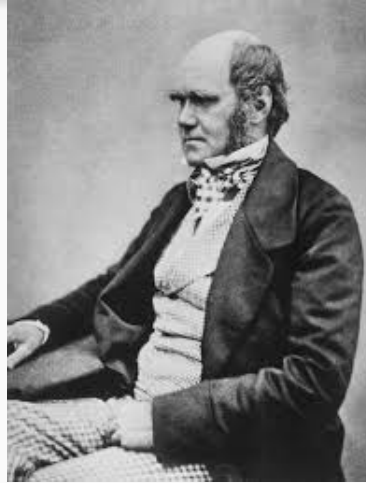


# Evolution of Thought: Marcello Malpighi



**First Identified Spiracles and Trachea are  
Insect Respiratory Organs**

# Evolution of Thought: Charles Darwin



**...when an animal during any part of its embryonic career is active and has to provide for itself... ...the adaptation is just as perfect and beautiful as in the adult animal. From such special adaptation of the larvae or active embryos of allied animals is sometimes much obscured: and cases could be given of the larvae of two species, or of two groups of species, differing quite as much, or even more, from each other than their adult parents.**



# Evolution of Thought: August Korph



**Corethra Larva Fill Trachea and Inflate Air  
Bladders from Tissue**

# **Acoustic Larvicide**

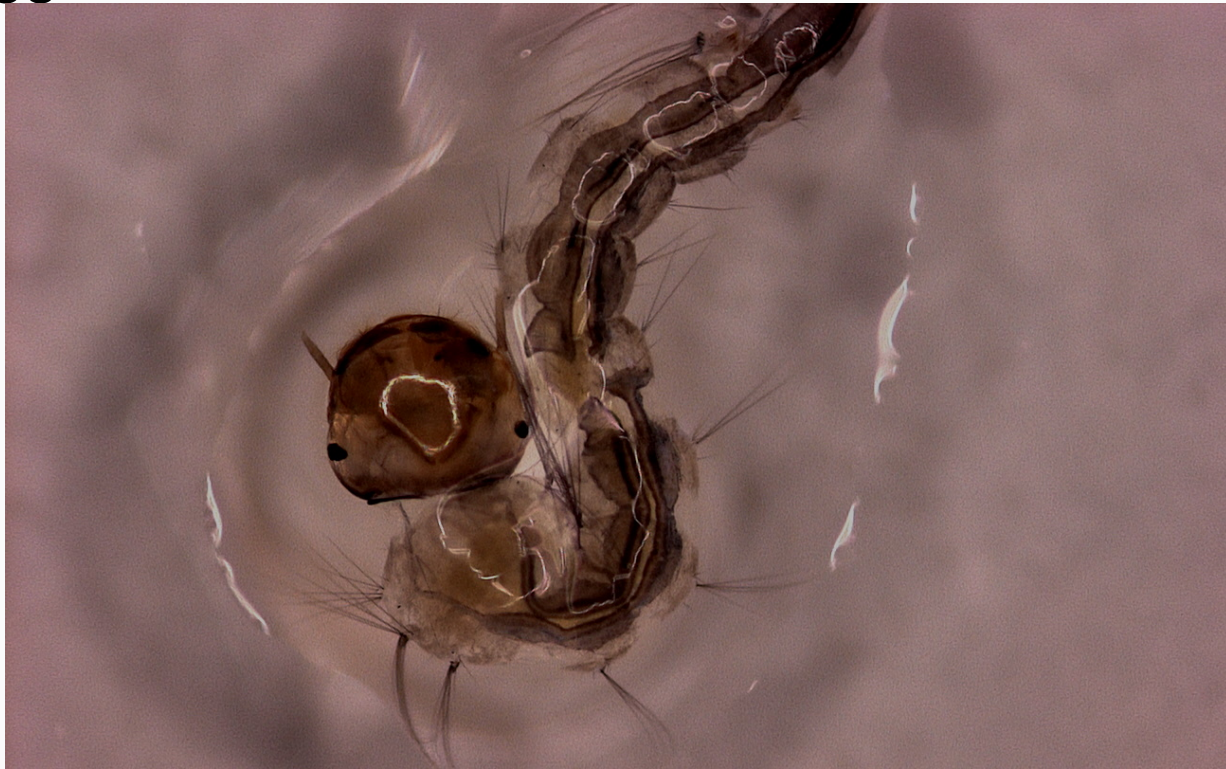
## **A Resonance Phenomenon**

**Sound Energy Transmitted into Water at Resonance with Tracheal Gas Causes the Dorsal Tracheal Trunk to Rupture resulting in Mortality or Flightless Mosquitoes**



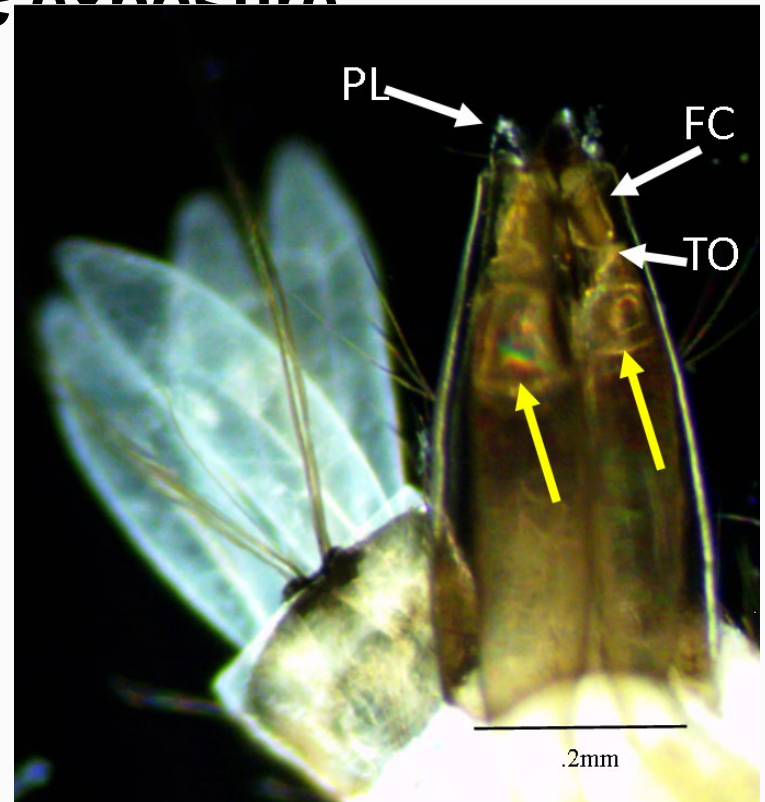
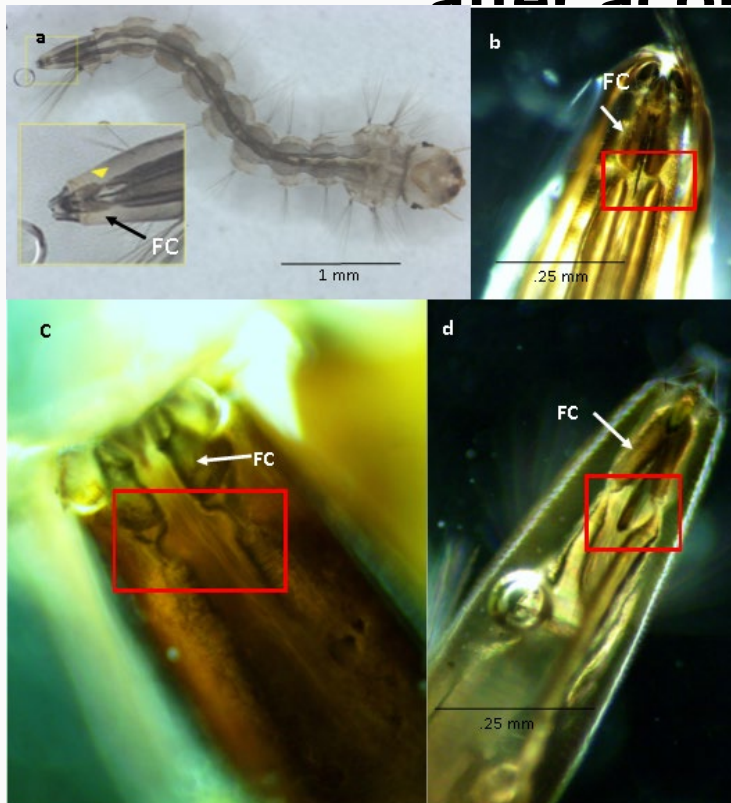
# Precision Acoustic Application Reveals Source and Is Non-Lethal

**“During Insect Development the Gas Filled Trachea of a Given Instar Becomes Enclosed in Larger, Liquid Filled Coaxial Tube” Keister & Buck 1955**  
**Acoustic Rupture Outward Flare of “Active DTT” Revealed Gas Source**



# An Isolating Occlusion at the DTT - Felt Chamber Transition

Newly Identified Tracheal Occlusion, Indicates Complete Isolation, hemolymph, tissue or gas does not pass through siphon after acoustic exposure

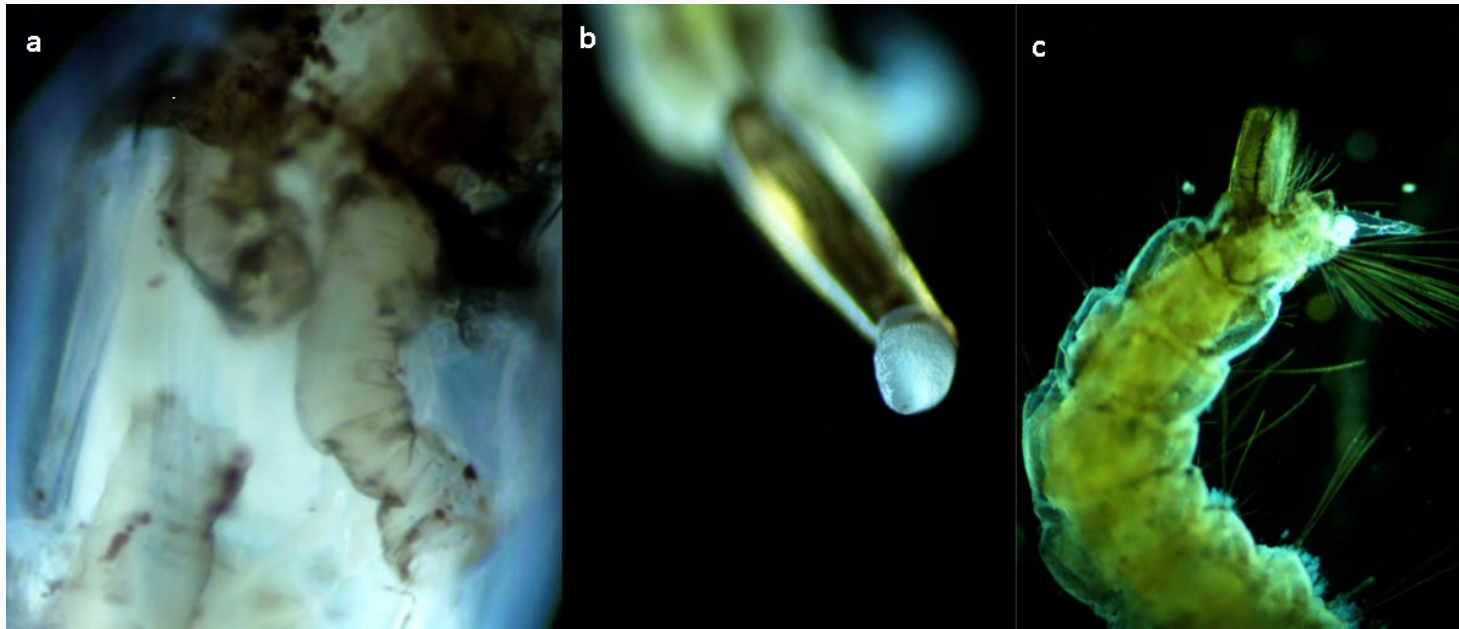




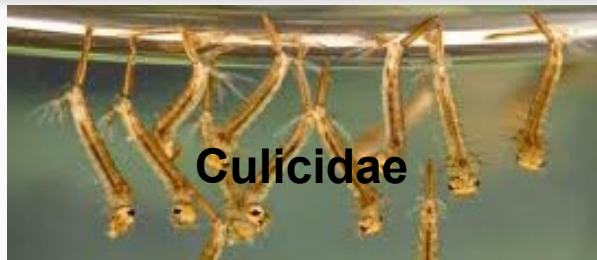
# Impairing the Siphon Reveals That Atmospheric Respiration is Facultative

- DTT not Necessary for Survival,
- Siphon Blocked with Paraffin Not Lethal
- Severed Siphon at Tracheal Occlusion and FC Renders Acoustic Larvicide Ineffective

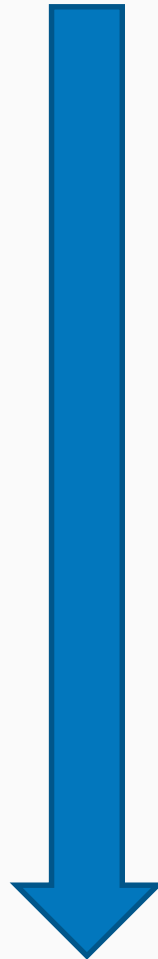
**Kroph – Corethra DTT Connecting Air Bladders Have No Respiratory Function**



# Members of Culicomorpha Infraorder Adapting for Rest and Respiration and Survival



Depth



- **Culicidae (mosquito)** hang motionless in the highest stratum of dissolved oxygen
- **Chaoboridae (phantom midge, glass worm)** remains neutrally buoyant mid water and rests. Its Ventral Fan is more developed than mosquito larvae. Note vestigial siphon
- **Simuliidae (Black Fly)** Remain attached to rocks in flowing high oxygenated streams filtering food.
- **Chironomidae (non-biting midge)** Rest protected in sediment, low  $O_2$  forced development of efficient Hemoglobin

# More Questions than Answers

- **Exactly what is the mechanism of respiration**
- **Internal Gas Transport**
- **Morphology and Molting Sequence of DTT**
- **Tracheal Filling**
- **To Do –**
  - **More details on Tracheal Occlusion and nomenclature**
  - **Rename the Anal Gills to something else**
  - **Rename the Ventral Brush and its function**
  - **Clear up misconceptions on DTT**
    - **Large size to store air for deep dive?**  
**Wrong**
    - **Mode of action of Petroleum Surfactants**

# Impacts and Opportunities

- **Review the Mechanism of Petroleum Surfactants**
- **Acoustic Larvicide may be Applied to Other Pests**
- **New Acoustic Larvicide Interventions**
  - **More Automated**
  - **Less labor**
  - **Solar Powered**
- **Fixed Cistern Float/ Solar or Domestic power**
- **Solar Powered Lethal Ovitrap**
- **Totally autonomous ROV (University of**



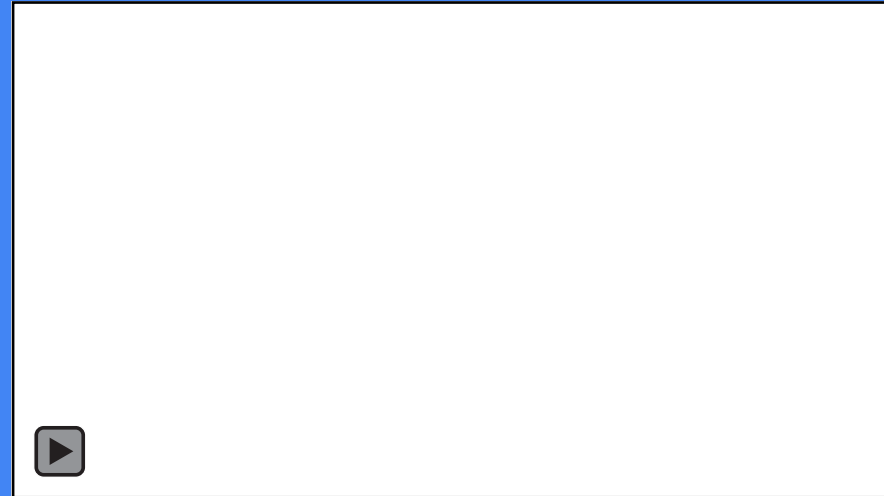
# Acoustic Lethal Ovitrap Set & Forget Daylight & Twilight Fliers

- Low Cost – Recycled Tires!
- Will not become breeding site
- Solar Power
- No Toxic Pesticide Handling
- Multi-Mode Attractant
  - Water, Blue LED with Motion, Dark Interior
- Internal Radius Shade (tire)
- Black-White Variegated Exterior
- No Consumables (Sticky Paper/Pesticides)
- No Moving Parts to Fail
- Bottom Slope Prevents Breeding
- Optional Attractants
  - Octanol Strips, Organic Brew, Sugar Bait Applique

Large water view

Shade Space

Solar Powered



Aedes Oviposition Sites

Draining Port

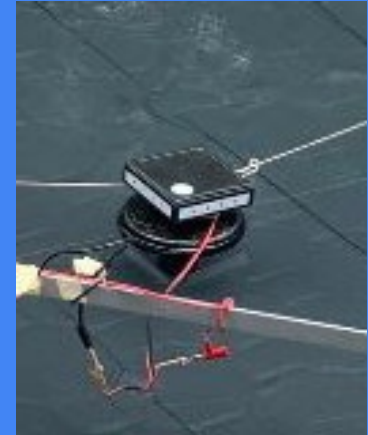
“Studies have shown that population densities can be reduced with sufficiently large numbers of frequently-serviced traps.”  
(<http://www.who.int/denguecontrol/research/en/>)

# Set and Forget Low Cost Cistern Solution

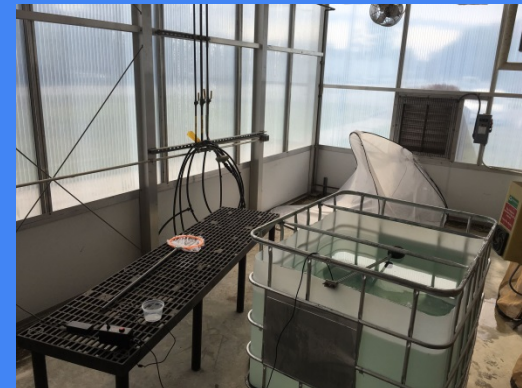
Cisterns, wells are increasingly important for survival providing excellent breeding habitats



Low-cost, low-power (solar) fixed set and forget system proved effective



Laboratory (USDA) and Field Trials Conducted Currently under Evaluation at University Sains Malaysia



# Thank you

**Kunihiro Moto**

**University of Notre Dame**

**Samual Rund, Laura Labb, Paul Hickner**

**The Connecticut Agricultural Experiment Station**

**Theodore Andreadis, Phillip Armstrong**

**John Shepard**

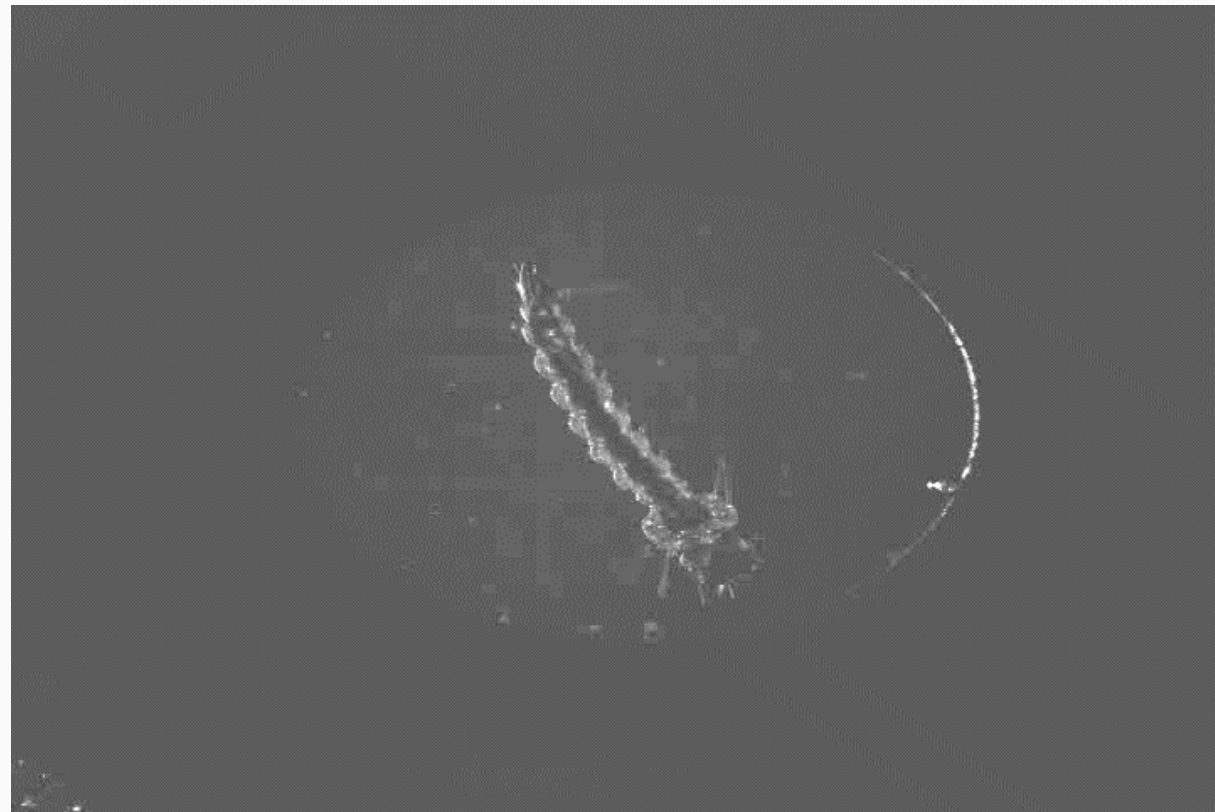
# Surfactants

## Century of Evidence Indicate Petroleum Surfactants are Neurotoxins

- **Glenn Richards Jr. 1941. “Differentiation Between Toxic and Suffocating Effects of Petroleum Oils on Larvae of the House Mosquito (*Culex Pipiens L*)”**
- **“Considerable Concrete Evidence has Accumulated Showing that the more Volatile Petroleum Oils have a Direct Toxic Effect”.**
- **Toxic Activity Very Rapid**
- **Asphyxiation Due to Forced Submergence Takes Many Hours**
- **Larval Oral Ingestion of Contaminated Nutrients Similar Toxic Results**

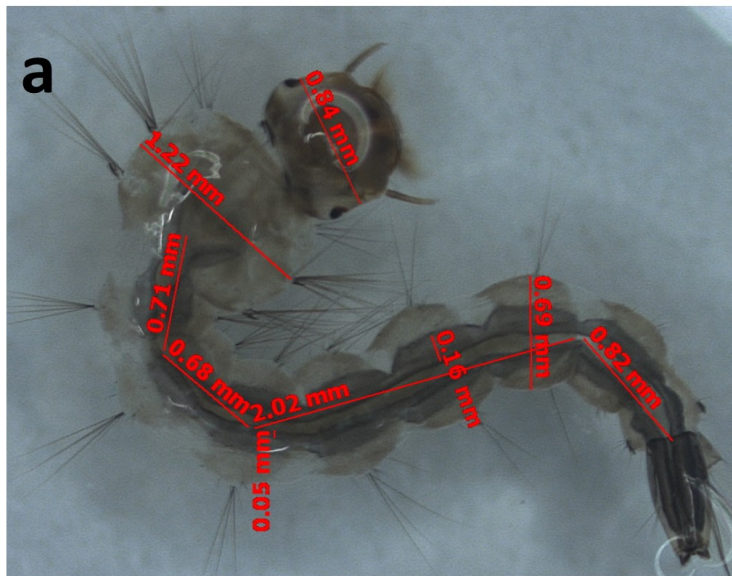
# Preliminary Indications

**Bubble Size and High Speed Video  
Indicated Elevated Pressure**

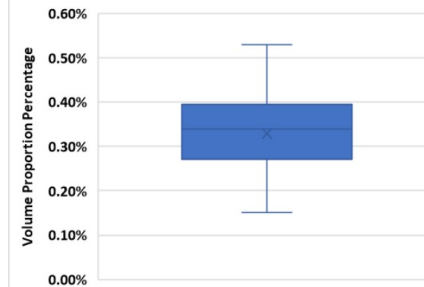




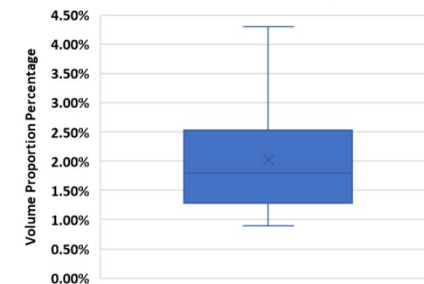
# Natural Gas Law and Relative Expansion - Two Proofs of Pressurization



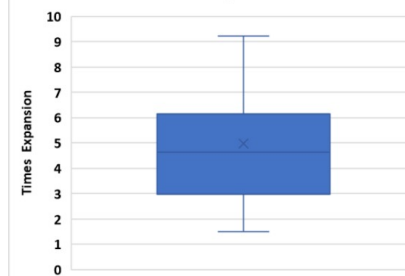
**c** Volume Proportions of *Ae. aegypti* Gas Filled Tracheal System to Body Volume



**d** Volume Proportion of *Ae. aegypti* Released Gas Bubbles to Body Volume



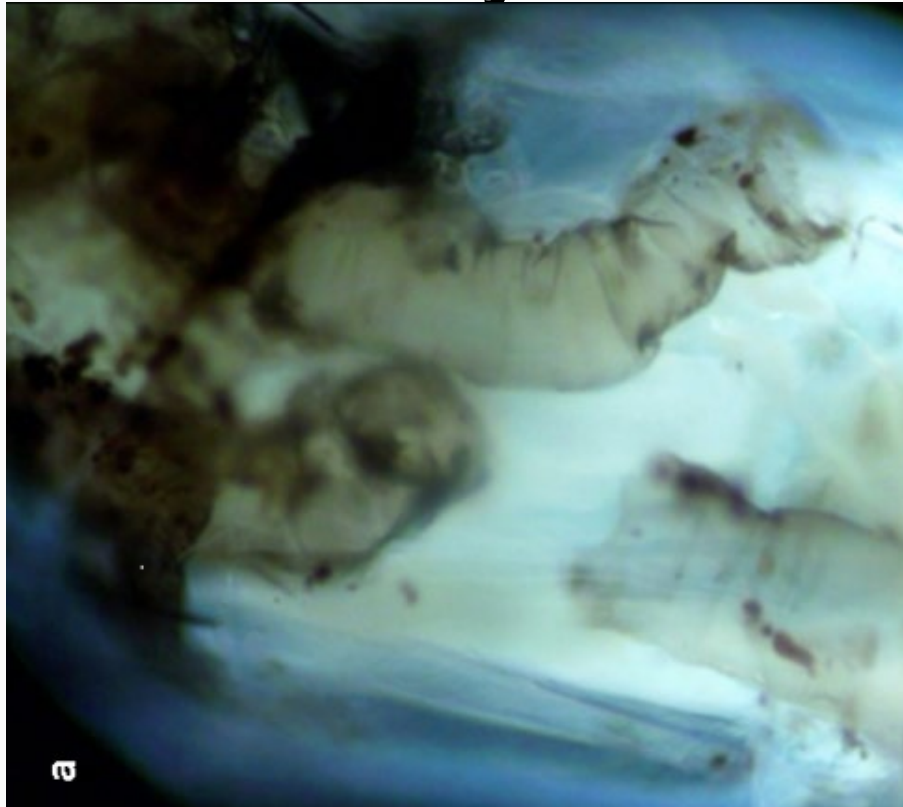
**e** Gas Expansion Over *Ae. aegypti* Gas Filled Tracheal System Volume



# Severed Dorsal Tracheal Trunk Shows No Obligate Need for Atmospheric Respiration

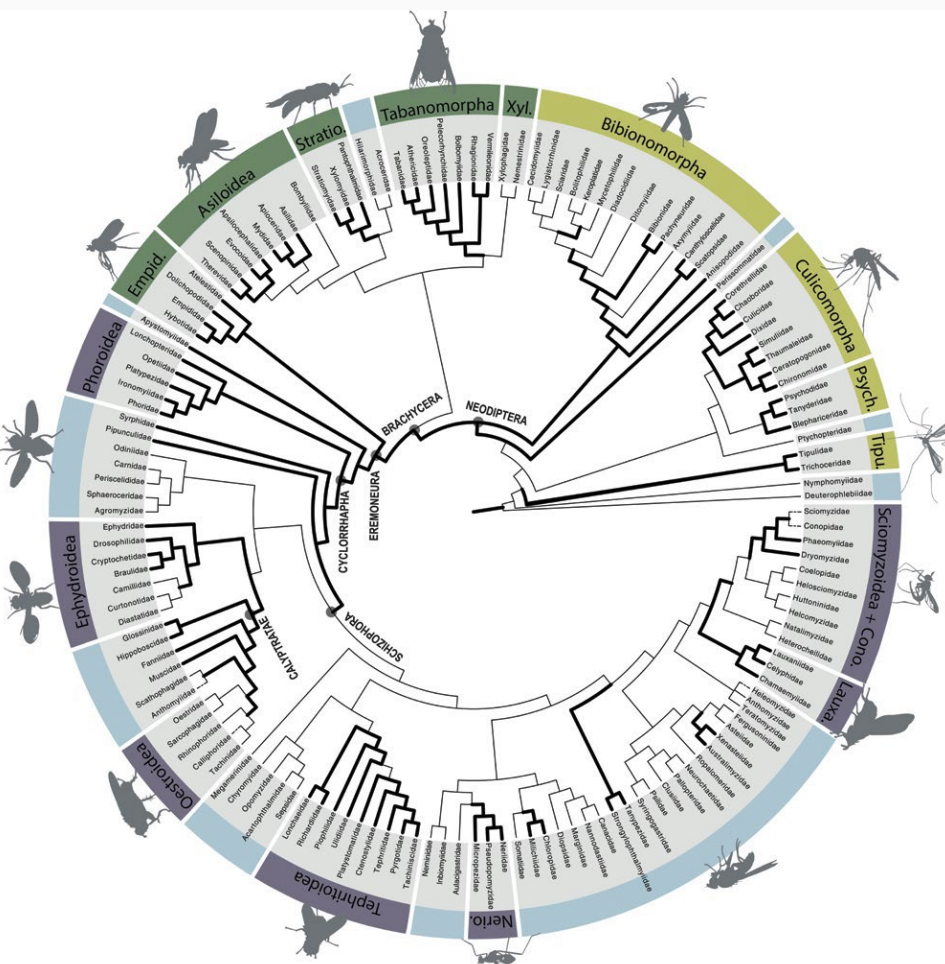
**Kroph – Corethra DTT Connecting Air Bladders  
Have No Respiratory Function**

**Culex Survive Over 20 Days Without An intact DTT**



# Infraorder Culicomorpha is a Large Clade, Larvae of All These Families are Reported to be Aquarius Respirators Except Culicidae

## Phylogenetic tree of Diptera<sup>3</sup>



Ceratopogonidae  
Chironomidae

Chaoboridae  
Corethrellidae  
Culicidae  
Dixidae

monophyletic

Simuliidae  
Thaumaleidae

