

# Can RNA interference technology contribute to controlling Asian citrus psyllid in Florida?

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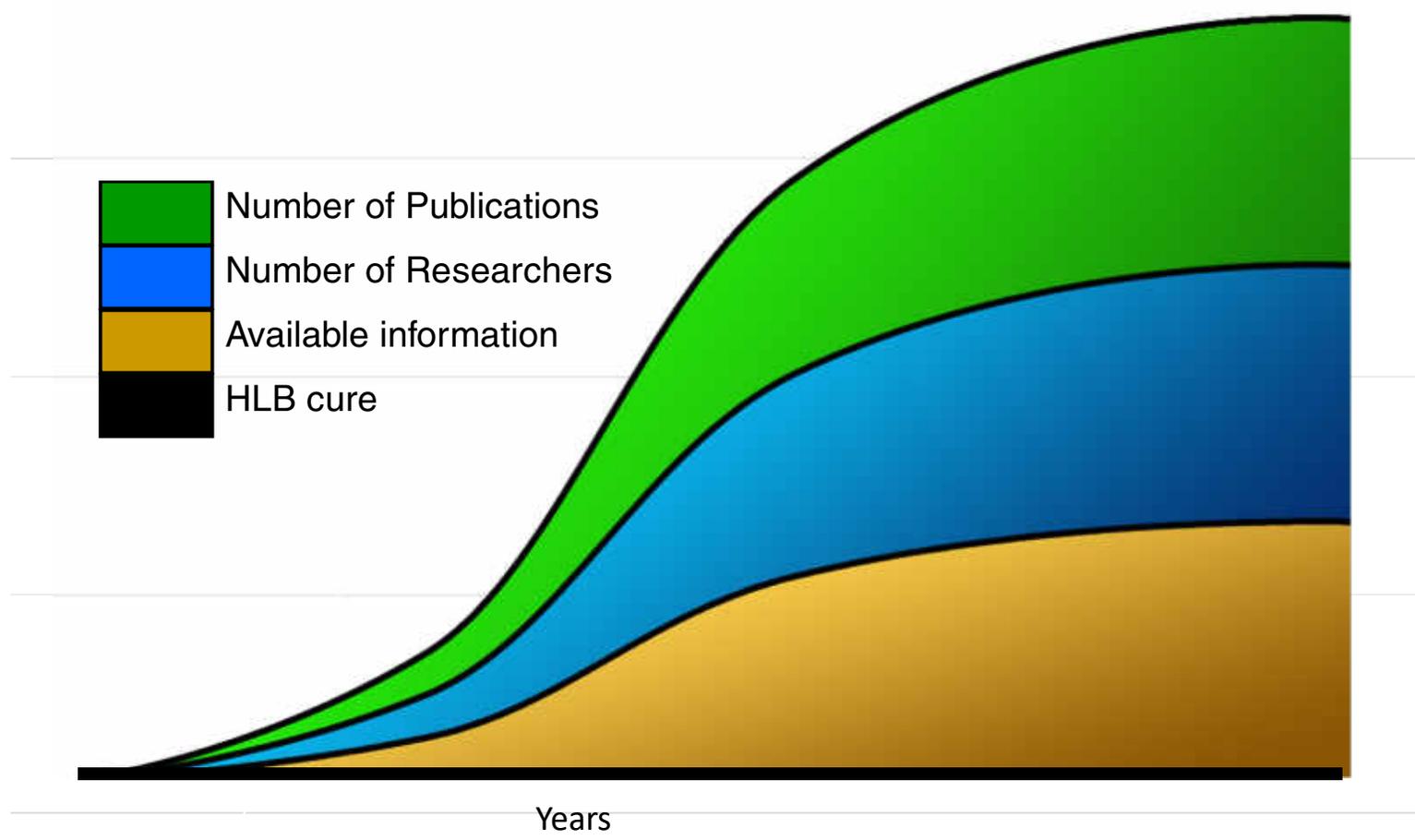
# BEST SOFA FOR UNWANTED GUESTS...



That moment  
when you  
remove an  
unwanted,  
freeloading house  
guest...



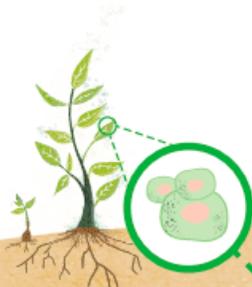
somee cards  
user card.





# Thinking outside the box

ACP is the true enemy



All living things - like this plant - are made up of **cells**, the basic units of life.

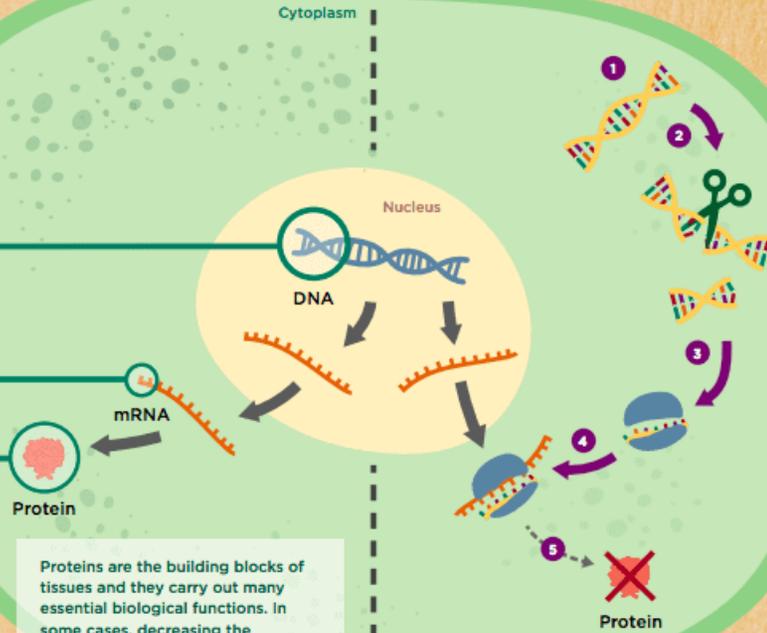
# UNDERSTANDING RNAi

RNAi, or "RNA Interference," is a natural process that occurs in the cells of plants, animals, and people.

Inside the nucleus of each cell is a detailed genetic blueprint, encoded in **DNA**...

... which is transcribed (copied) into **messenger RNA (mRNA)** ...

...which gets translated by the cell's machinery to make a specific **protein**.



Proteins are the building blocks of tissues and they carry out many essential biological functions. In some cases, decreasing the production of specific proteins can be beneficial. RNAi is a natural process that works like a "dimmer switch" to dial down the level of a protein. It likely evolved to protect cells from viruses.

## HOW DOES RNAi WORK?

- 1** It begins when a form of RNA made of two strands (**double-stranded RNA, or dsRNA**) is introduced into the cell, for example by a virus, or produced in the cell.
- 2** When a cell "sees" dsRNA, it activates structures that work like scissors to **cut it up**.
- 3** Next, **other structures** attach to these small pieces of RNA and turn them back into single-stranded RNA.
- 4** These structures then bind to **mRNA with a matching code**.
- 5** As a result, **production of the protein** encoded by that mRNA is prevented.

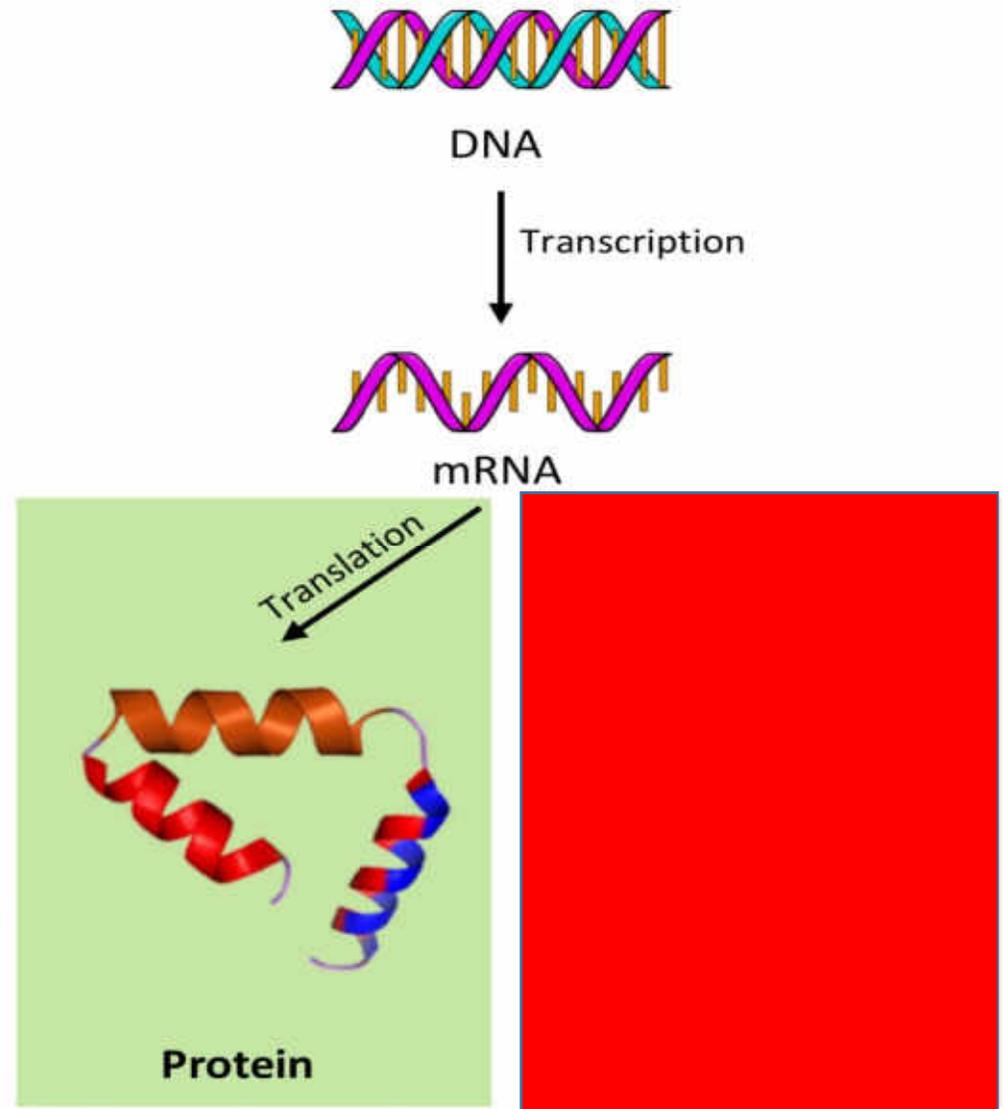
When we know the gene that encodes a certain protein, we can use RNAi to target that protein and dial it down in a highly specific way. In agriculture, for example, this can potentially impact the production of proteins responsible for the development of a disease or essential for a pest's survival, thus protecting plants from such disease or pest infestations.

**Andrew Fire,**  
professor of pathology and of  
genetics at the School of Medicine,  
is co-winner of the

**2006 Nobel Prize  
in Physiology  
or Medicine**

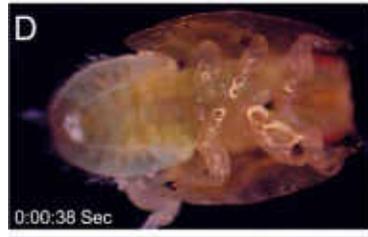
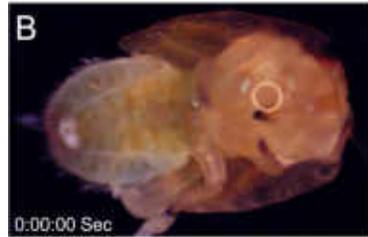
for his discovery of  
RNA Interference

▶ [Read the story](#)  
▶ [Read about RNAi](#)



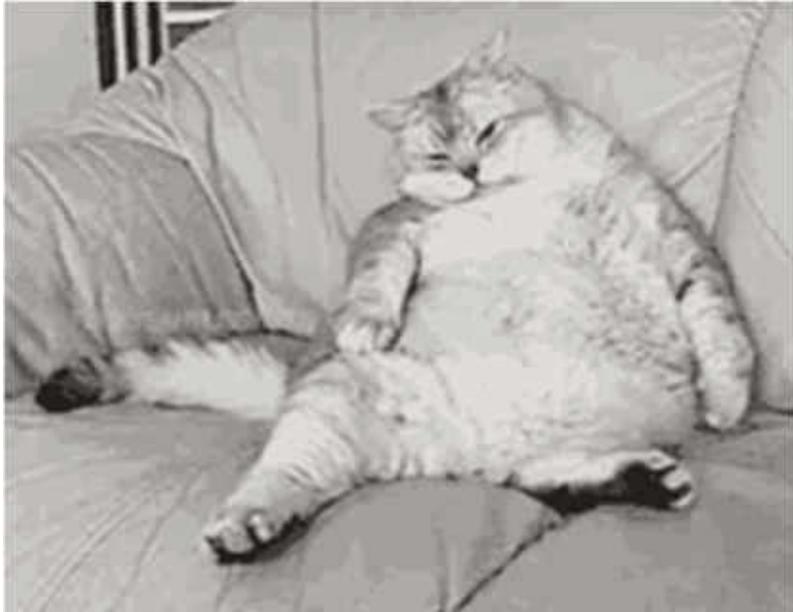
# RNA interference

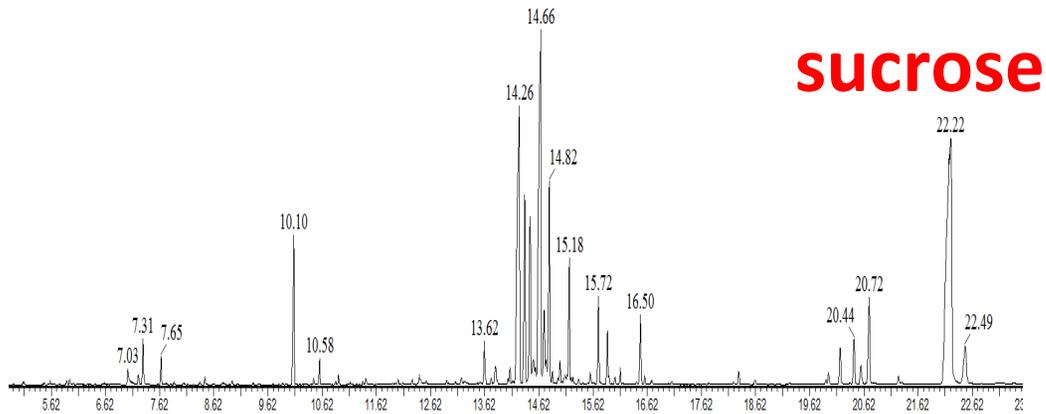
## Reduction of a gene product



# Screening for target genes

## 1: Food coma

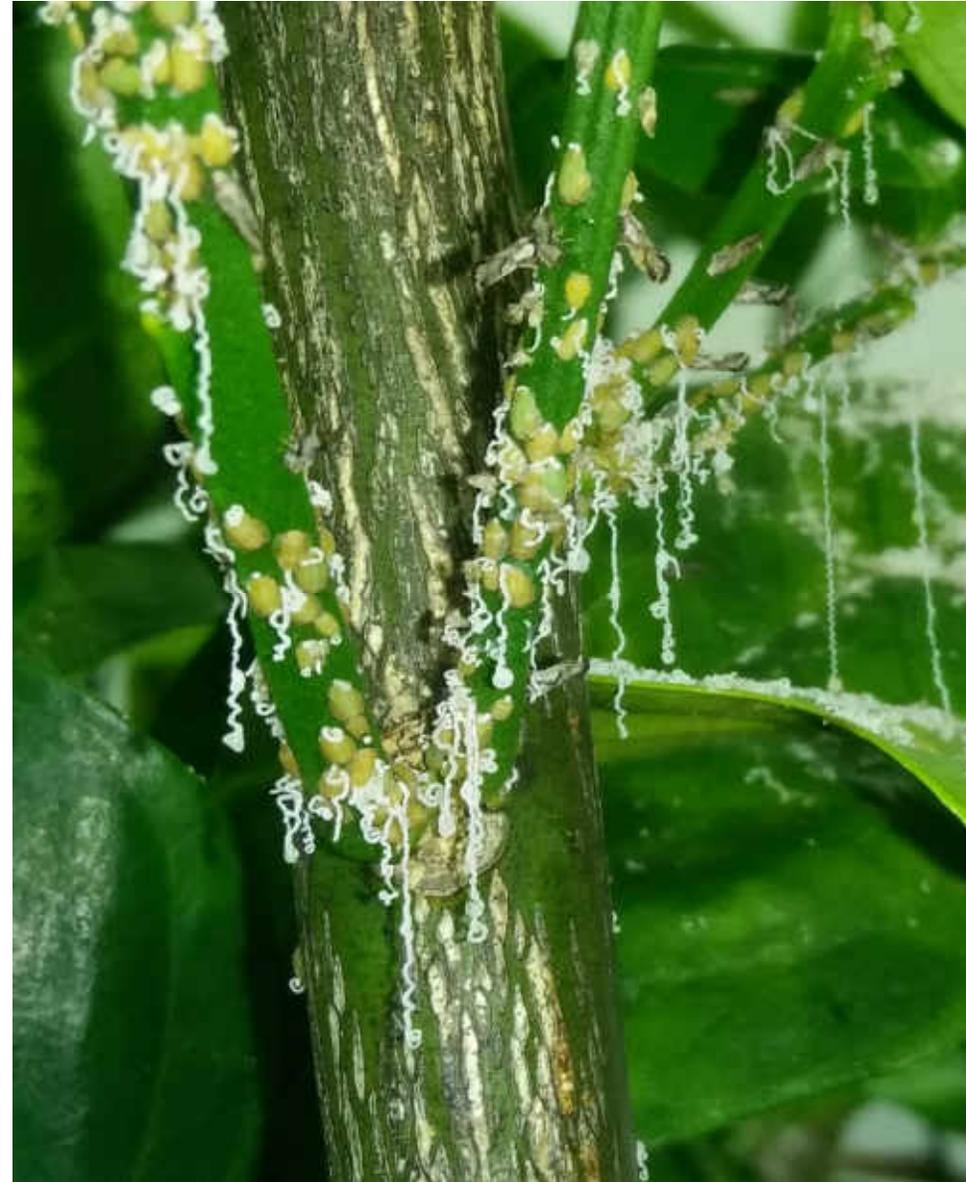


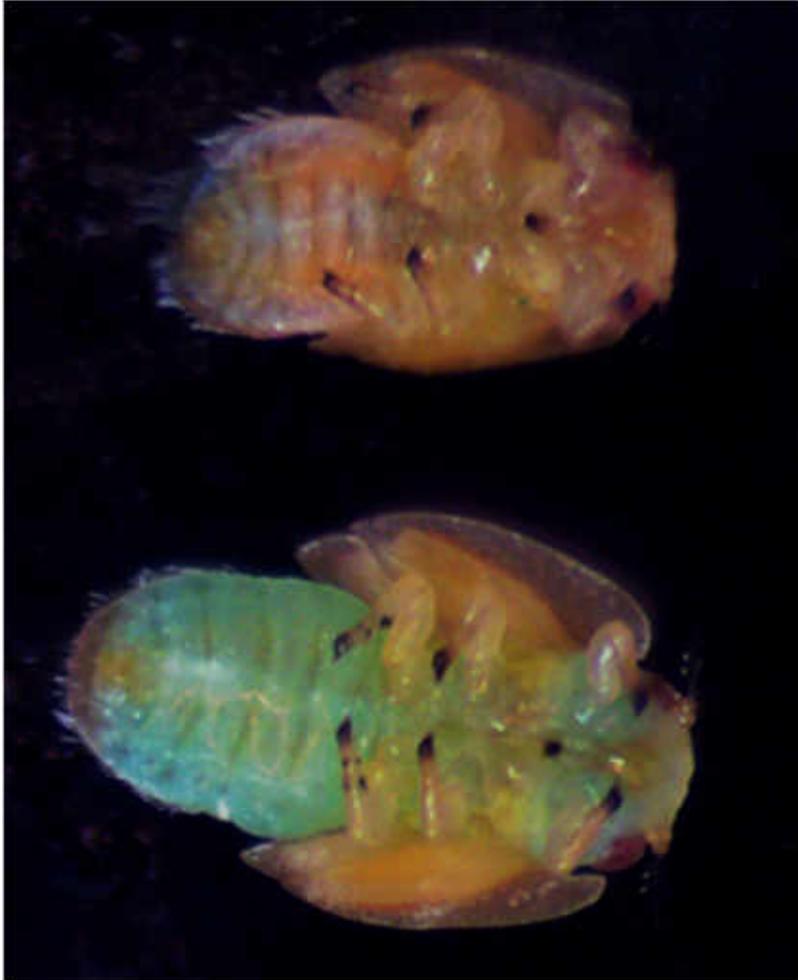


## Osmotic potential:

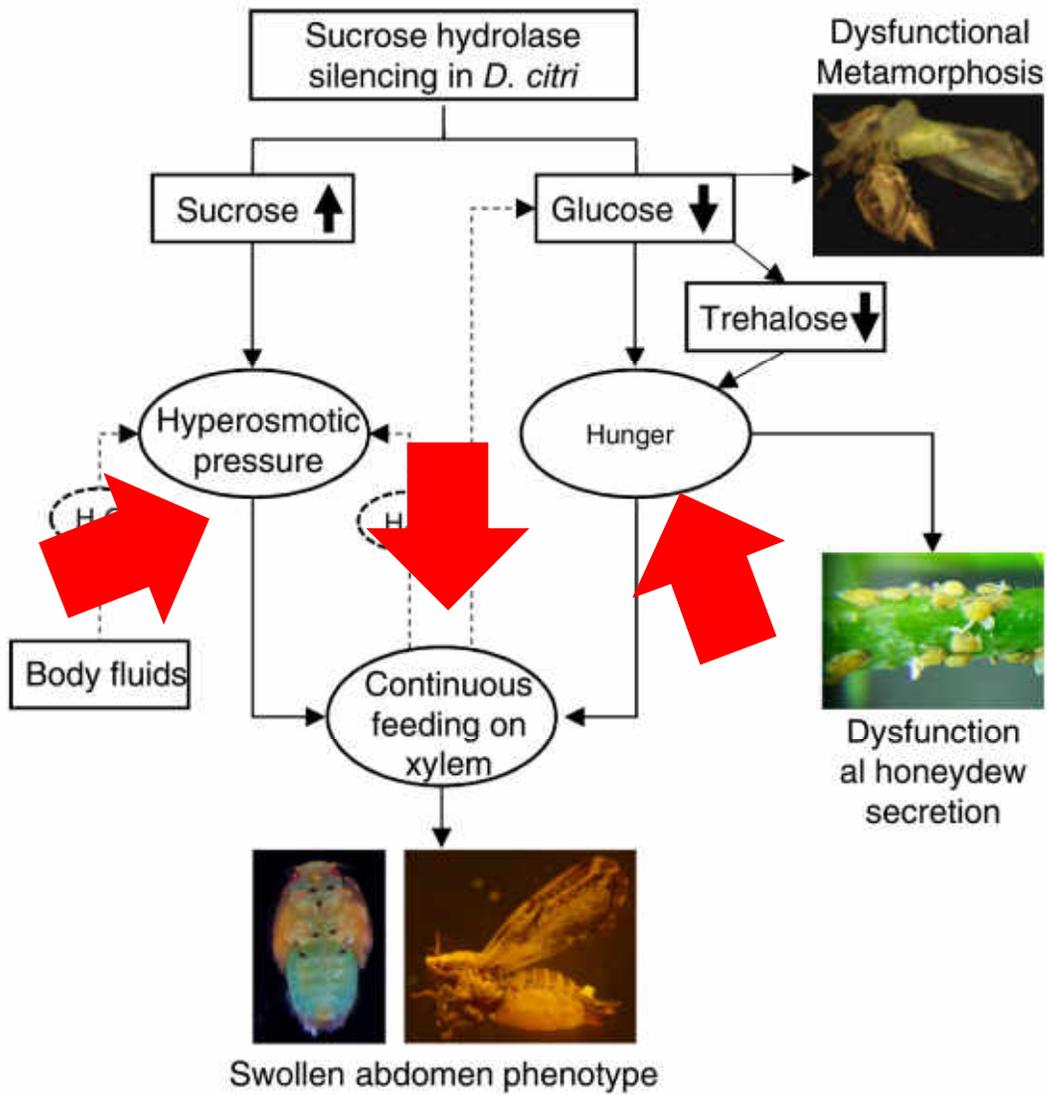
- 1) Hydrolysis of sucrose;
- 2) transglycosidation of sucrose into oligosaccharides (honeydew)
- 3) dilution by water by feeding on xylem sap

9/12/2019





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# Screening for target genes

## 2: Fly the coop





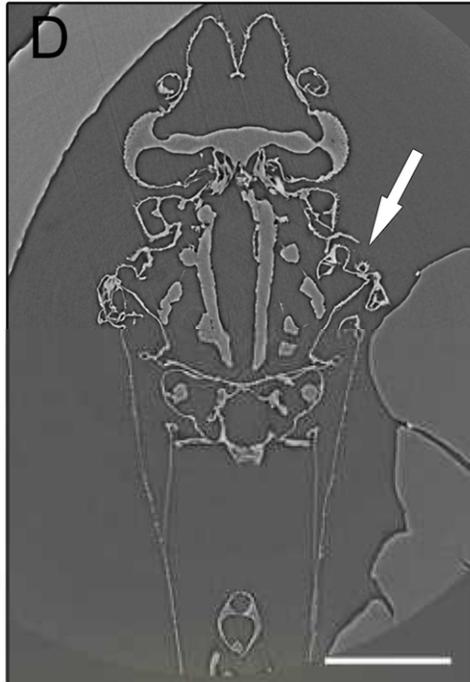
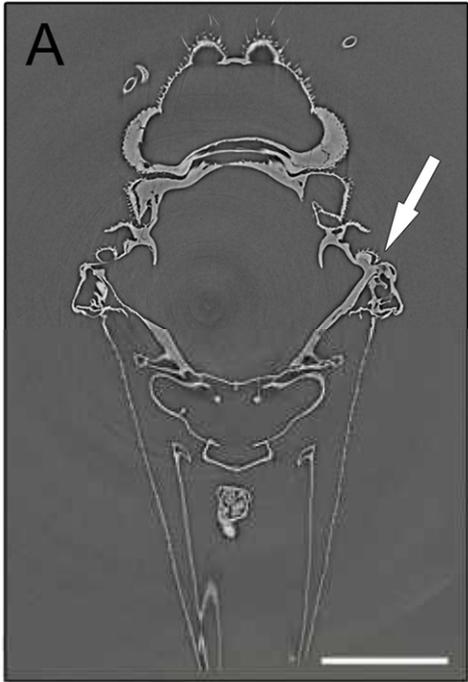
# Screening for target genes

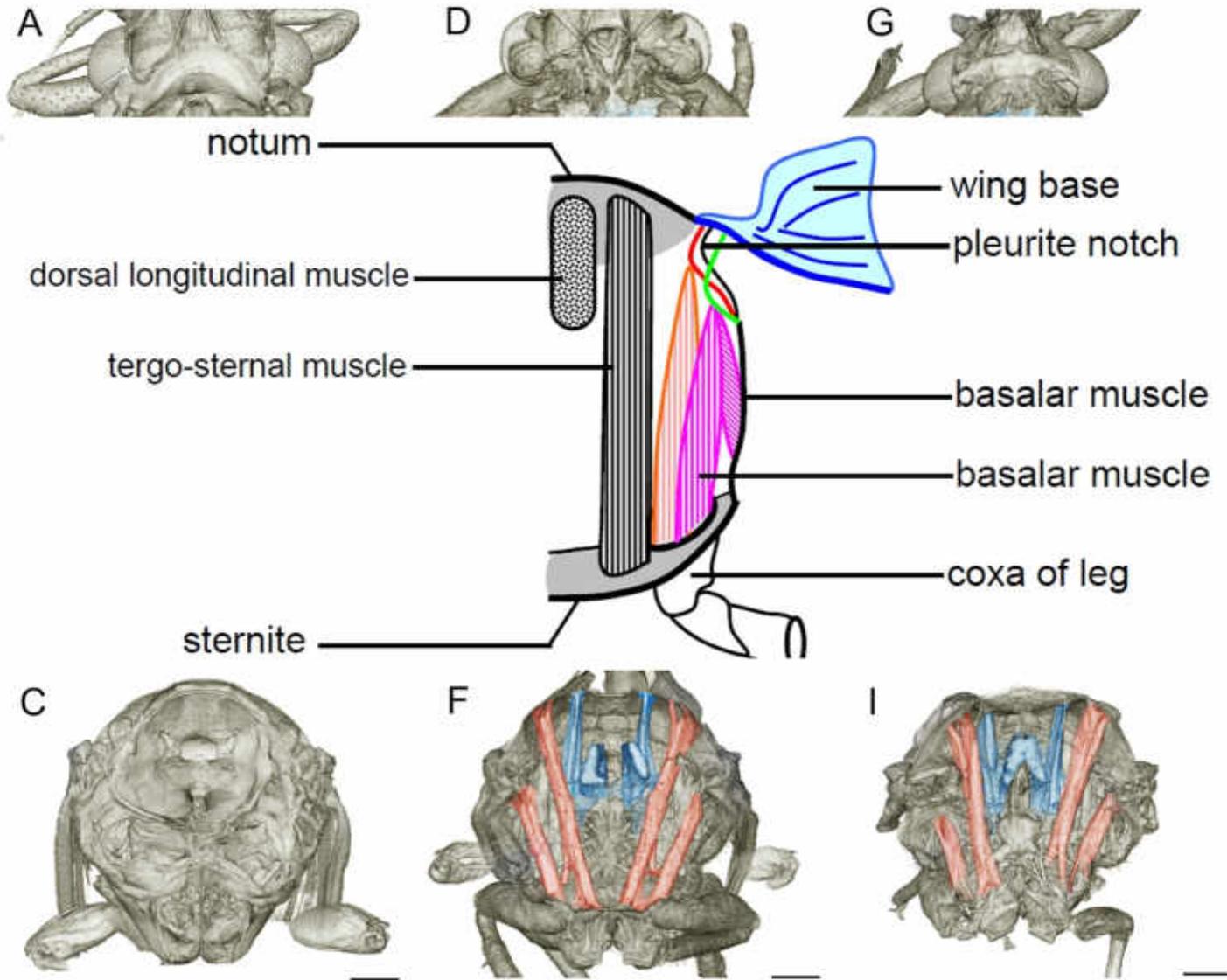
## 3: Lazy-bones



Hot plate (60 °C)







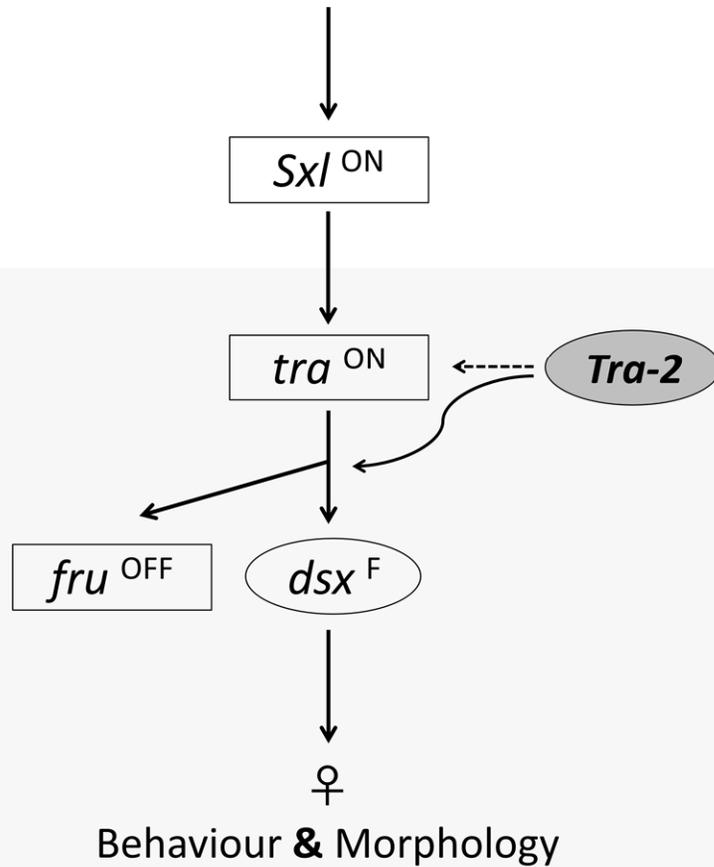
# Screening for target genes

## 4: Island of lost men

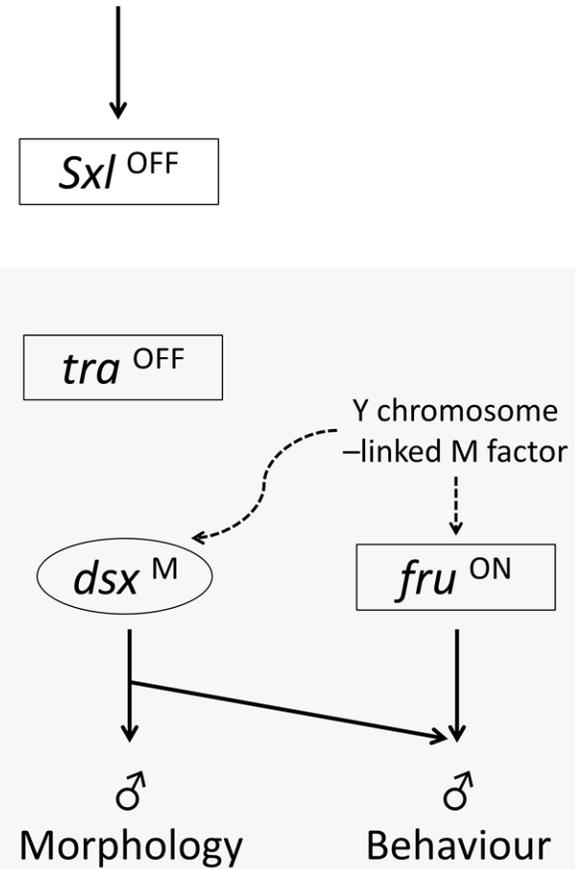


**A****XX embryo**

(high X chromosome dose)

**B****XY embryo**

(low X chromosome dose)



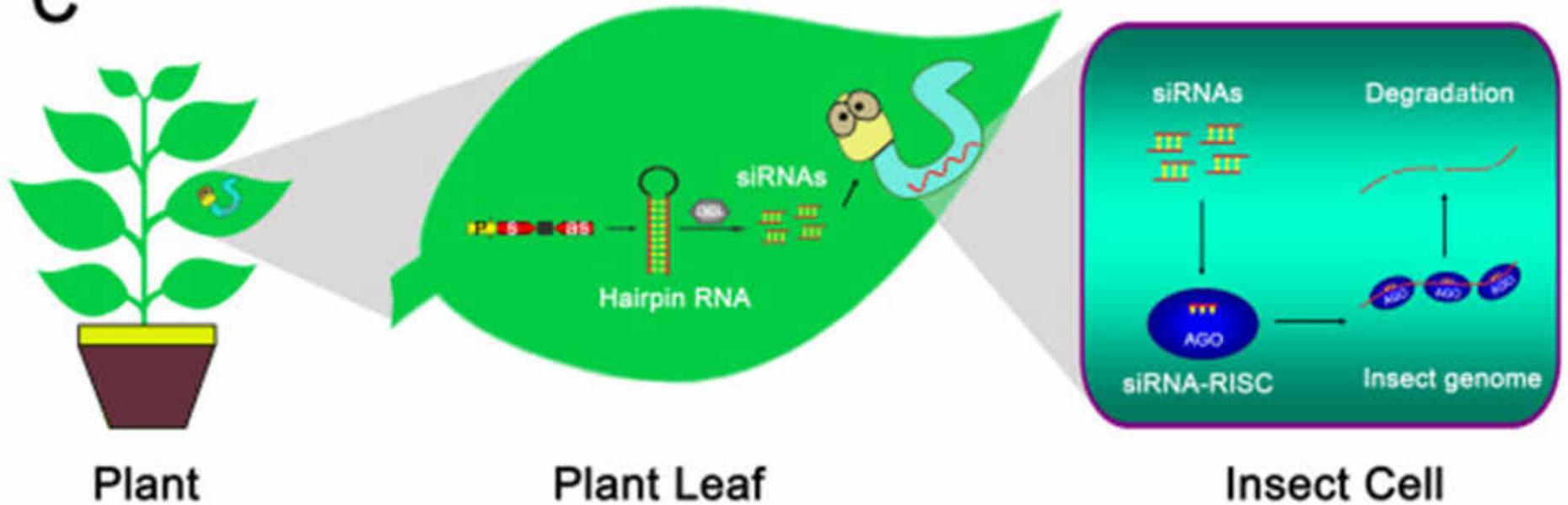
## Best gene candidates

- Significantly affect survival, fertility, transmission of HLB bacteria,.....
- Asian Citrus Psyllid specific
- RNAi of a gene does not require its entire sequence



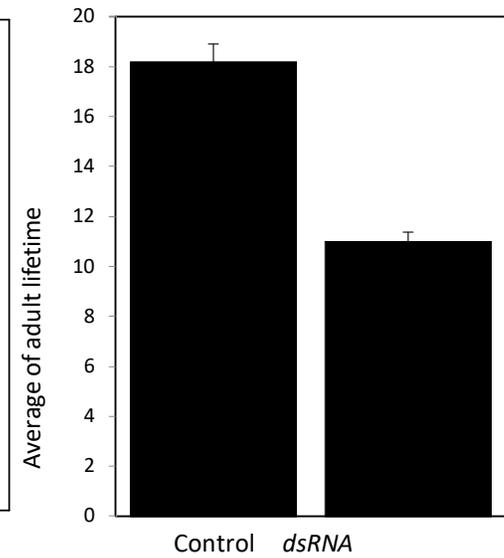
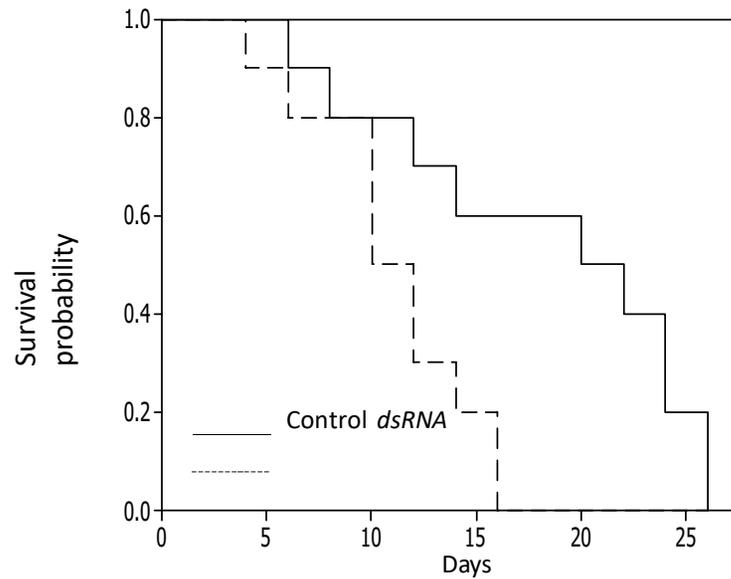
# 1-Virus induced gene silencing using CTV

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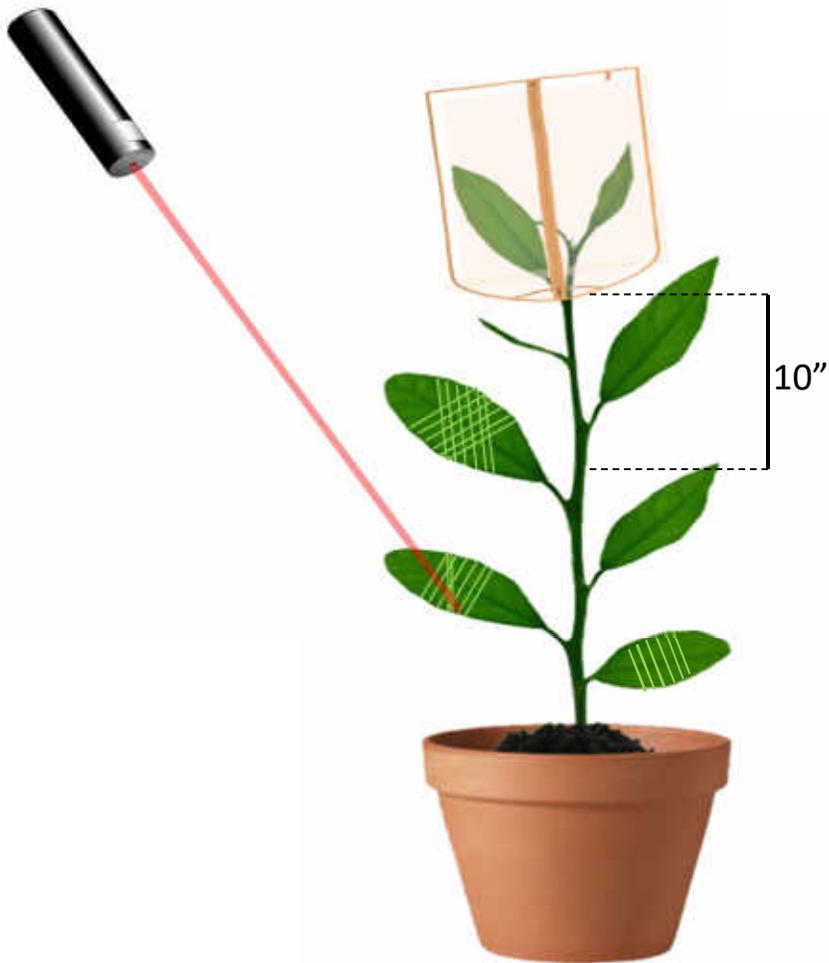


# Virus induced gene silencing

## CTV

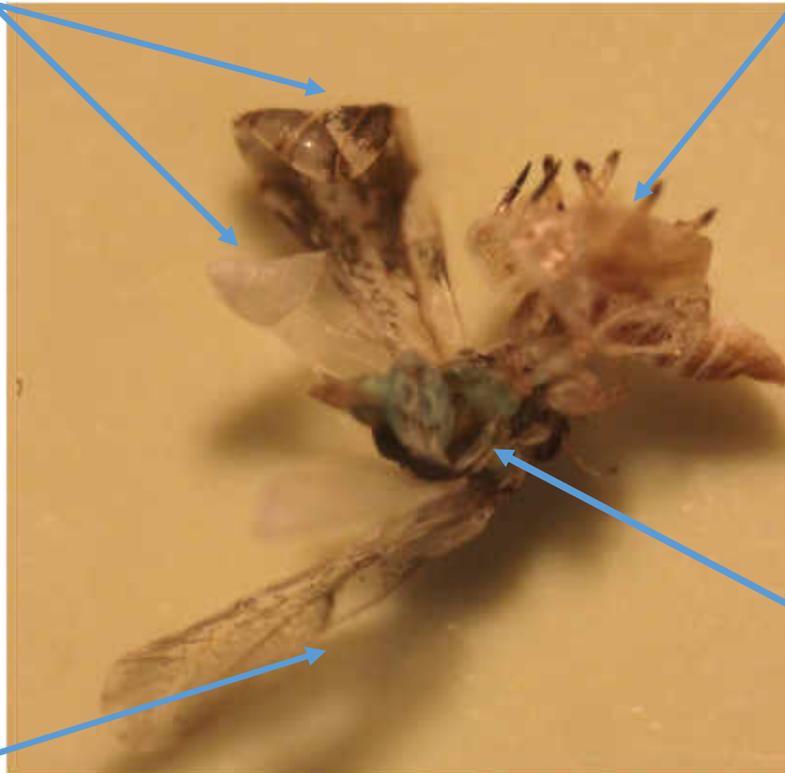


## 2- Laser delivery of dsRNA



**Curled wings  
(cup shaped)**

**Incomplete  
molting**



**Stretched wing**

**Stunted  
malformed body**

### 3- Gene-editing using CRISPR-Cas9





# Conclusion

- RNA interference is new technology where we can silence important genes in Asian citrus psyllid causing mortality or inability to fly, and/or increase pesticide susceptibility.
- RNAi is a promising and potential tool to control Asian citrus psyllid but is not ready yet for application in the field.
- Combinations of RNAi and other methods such as pesticide application or Bt-toxin could offer a very efficient control strategy.

**THANK YOU**