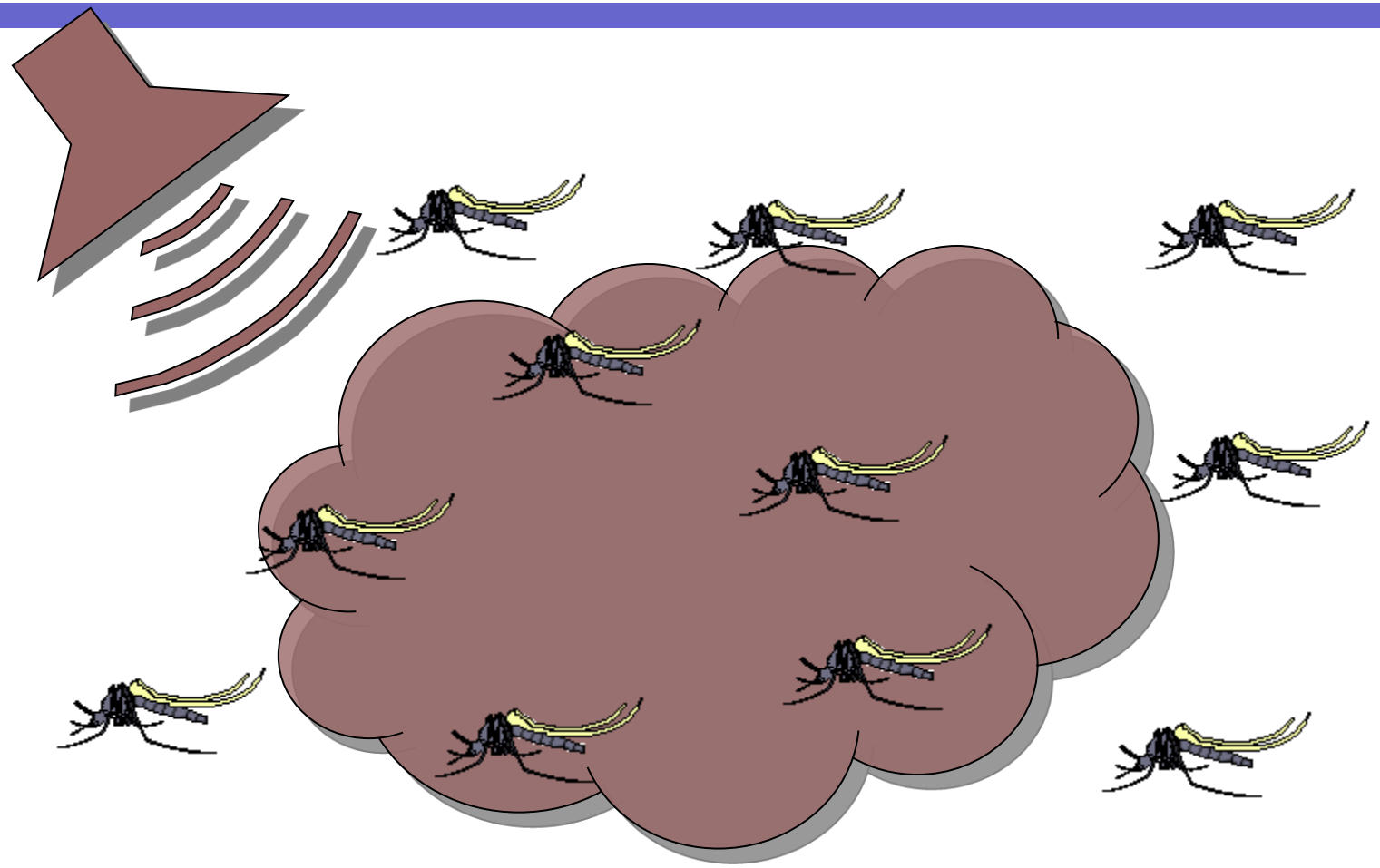
Resistance Bottle Bioassays

Janet McAllister
Division of Vector-Borne Infectious Diseases
Fort Collins, CO
jvm6@cdc.gov

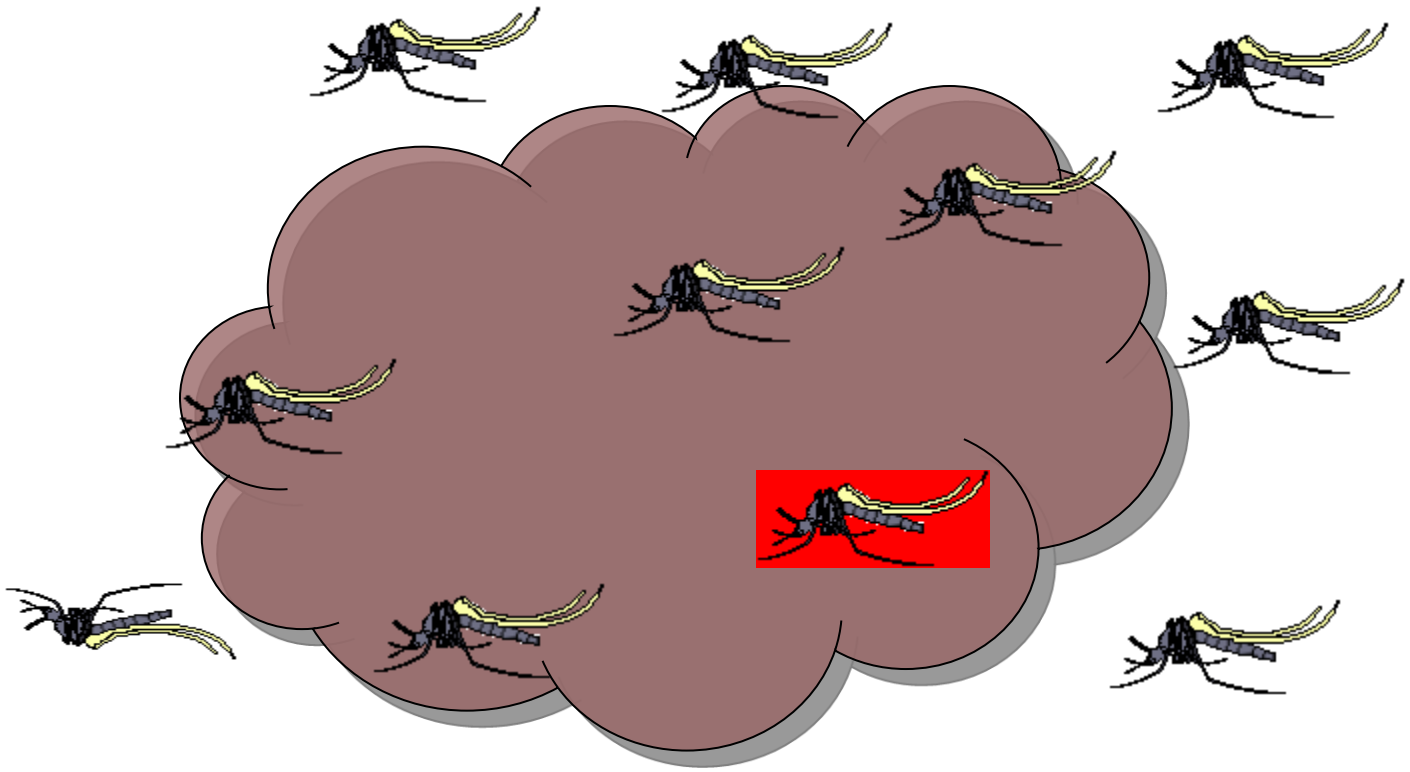
"The findings and conclusions in this presentation are written by authors serving in their capacity as CDC employees and do not necessarily represent an official view of the Centers for Disease Control and Prevention."

CDC

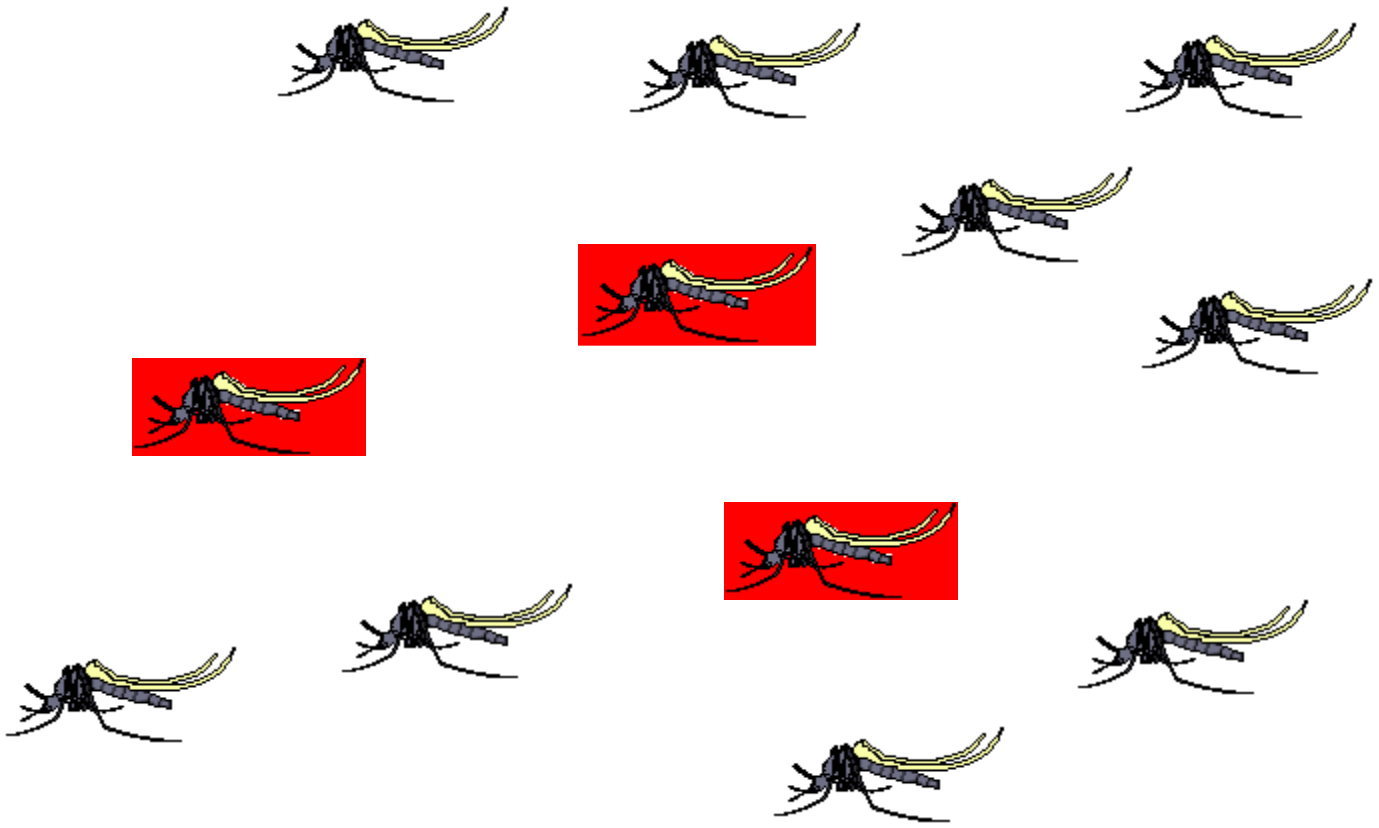
Spray a population



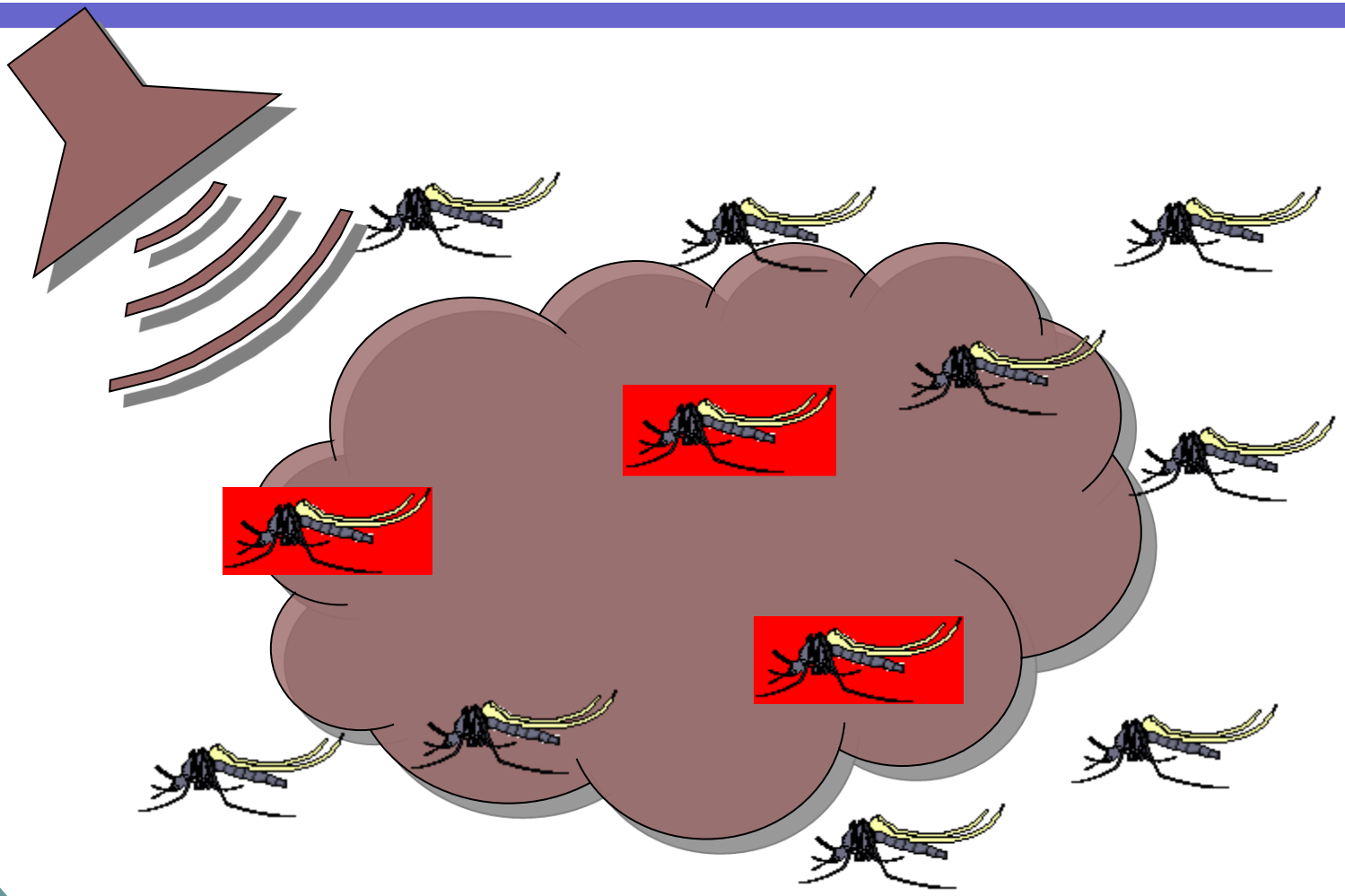
Survivor with “something special”



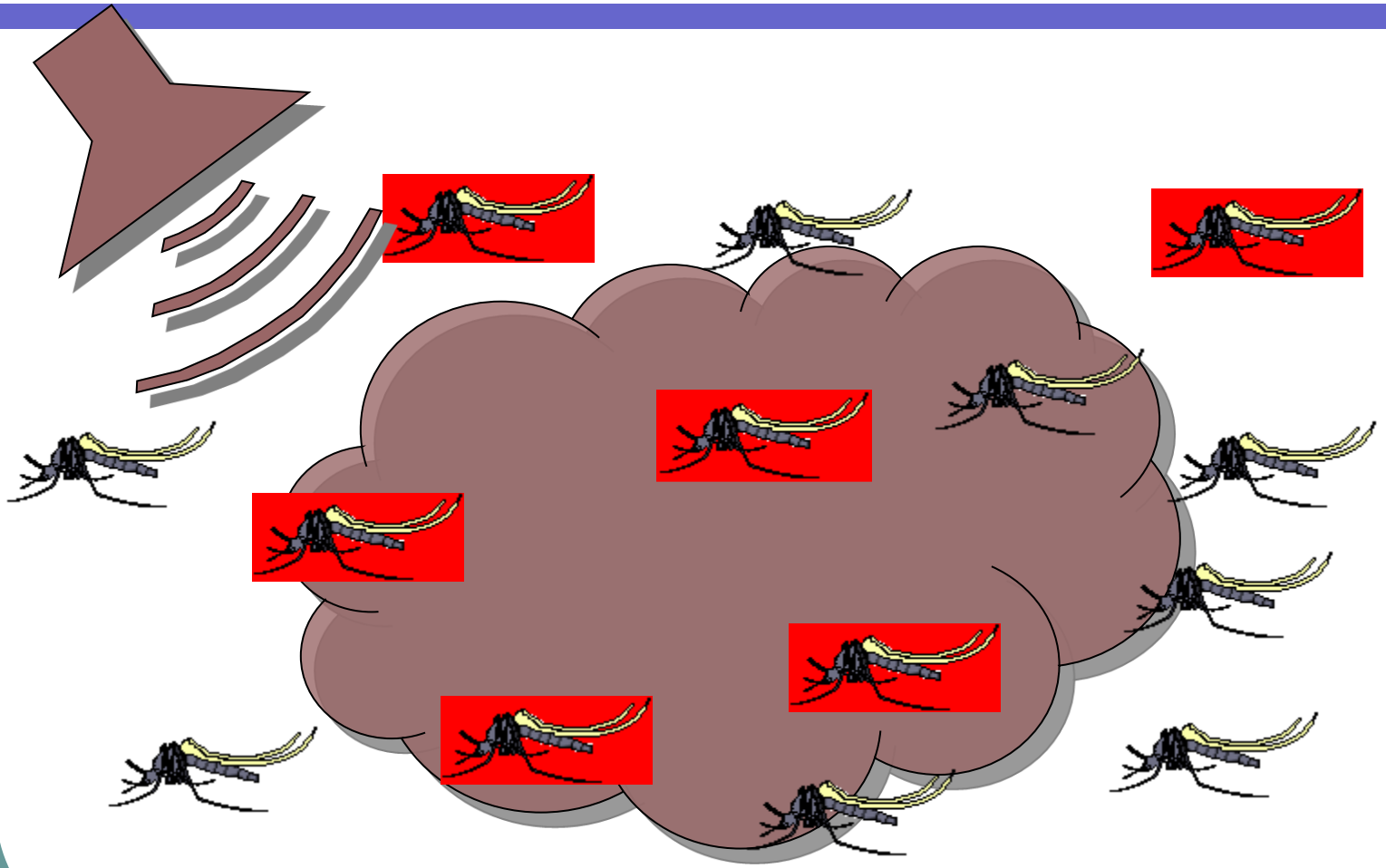
Offspring of the survivor



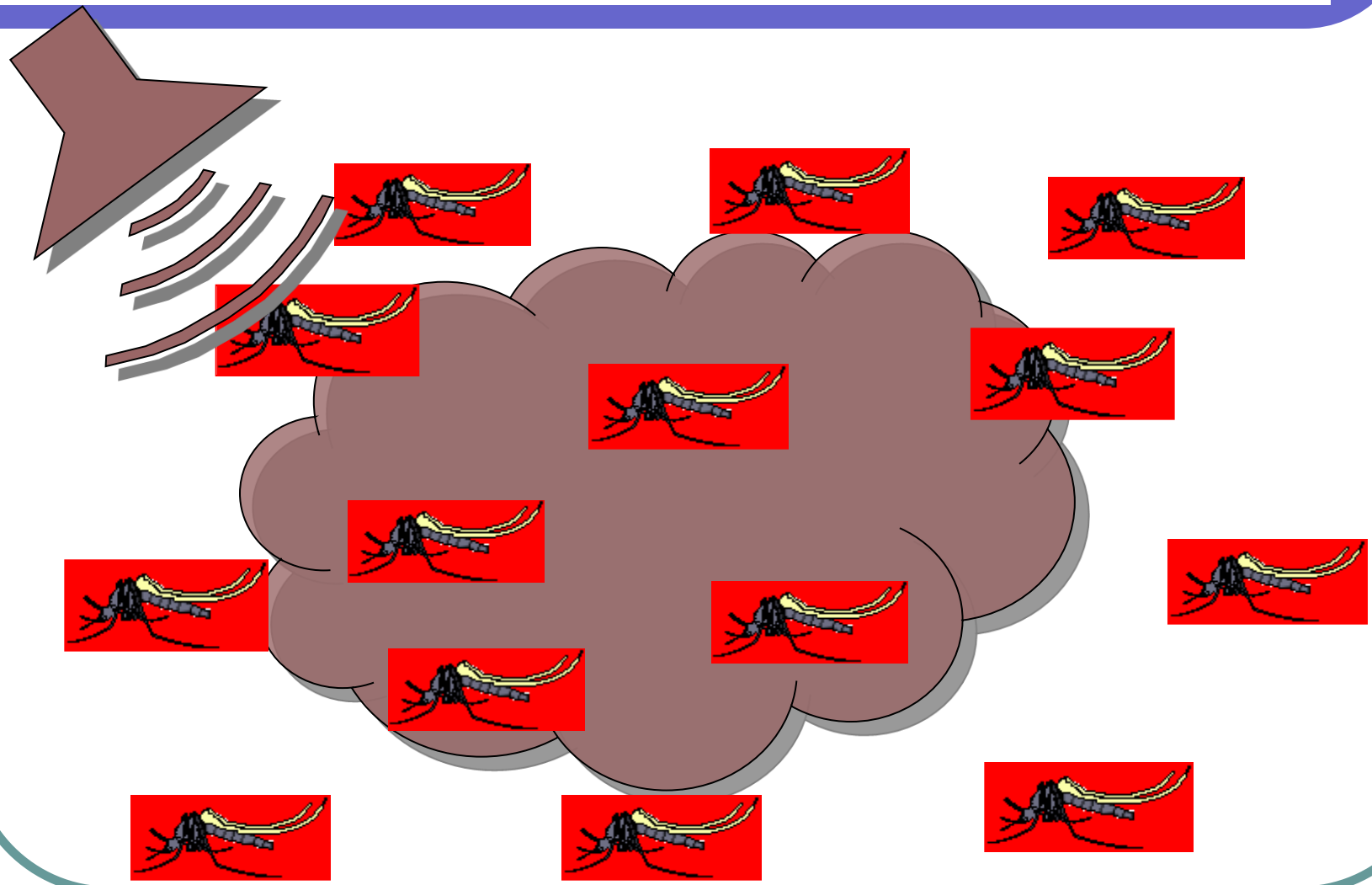
Spray again - more survivors



More resistance in population



More resistance in population



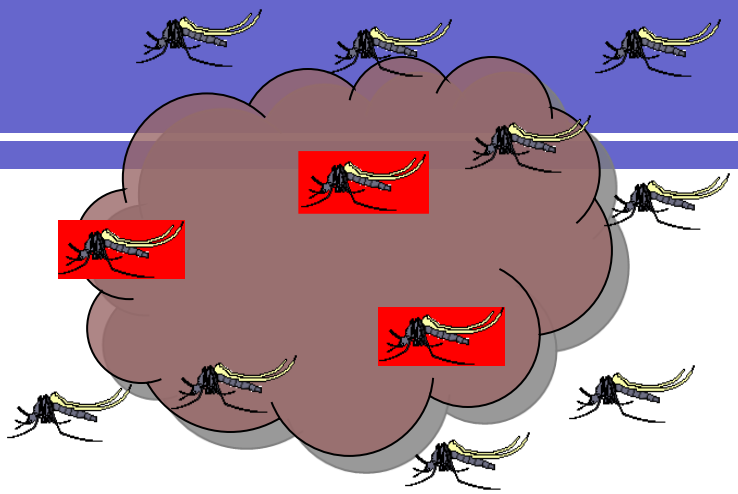
Traditional Definition

"**Insecticide resistance**" describes the ability of strains of insects to survive "**normally**" lethal doses of insecticide, the ability having resulted from selection of tolerant individuals in populations exposed to the toxicant for several generations.

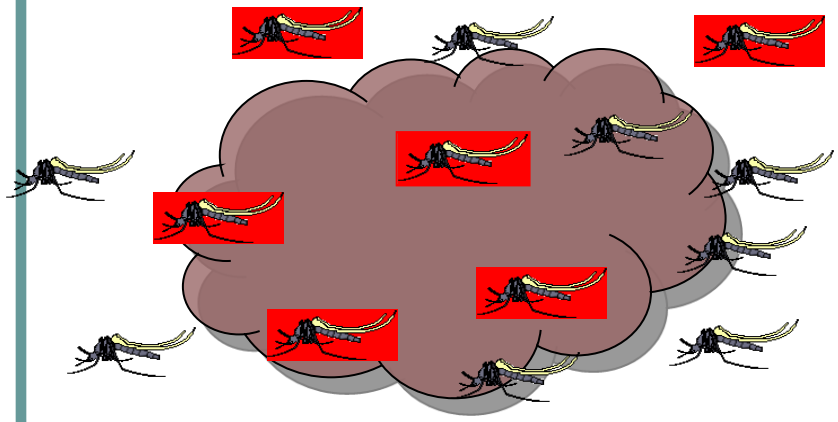
The time to act is before resistance reaches this level.

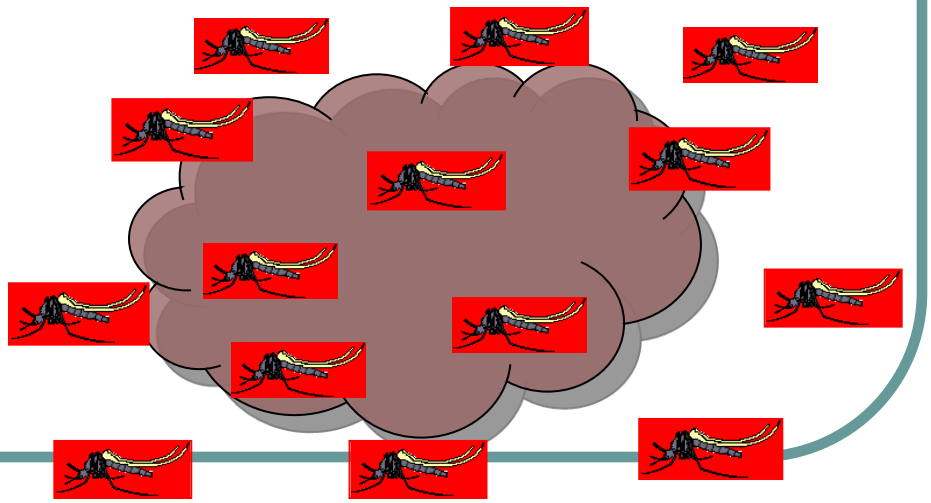
Resistance Definition

- **Insecticide Resistance** is a genetic change in response to selection by toxicants that **may impair control** in the field.



At what point do you have a resistant population?





Sources of Selection for Resistance

- Mosquito control applications of insecticides
- Agriculture applications of insecticides, fertilizers and herbicides
- Urban applications of insecticides, fertilizers and herbicides
- Non-point source runoff into mosquito larval habitat

How to develop Insecticide Resistance in Vectors

- single class of insecticide
- long-residual action
- slow-release formulation
- apply to all life states, all generations
- treat all habitat where pest occurs

Factors Influencing the Selection of Resistance to Insecticides

A. Genetic

- 1. Frequency of R alleles
- 2. Number of R alleles
- 3. Dominance of R alleles
- 4. Penetrance; expressivity; interactions of R alleles
- 5. Past selection by other chemicals
- 6. Extent of integration of R genome with fitness factors

B. Biological

- 1. Biotic
- a. Generation turn-over
- b. Offspring per generation
- c. Monogamy/polygamy; parthenogenesis
- 2. Behavioral
- a. Isolation; mobility; migration
- b. Monophagy/polyphagy
- c. Fortuitous survival; refugia

C. Operational

- 1. The chemical
- a. Chemical nature of pesticide
- b. Relationship to earlier used chemicals
- c. Persistence of residues; formulations
- 2. The application
- a. Application threshold
- b. Selection threshold
- c. Life stage(s) selected
- d. Mode of application
- e. Space-limited selection
- f. Alternating selection

Testing Considerations

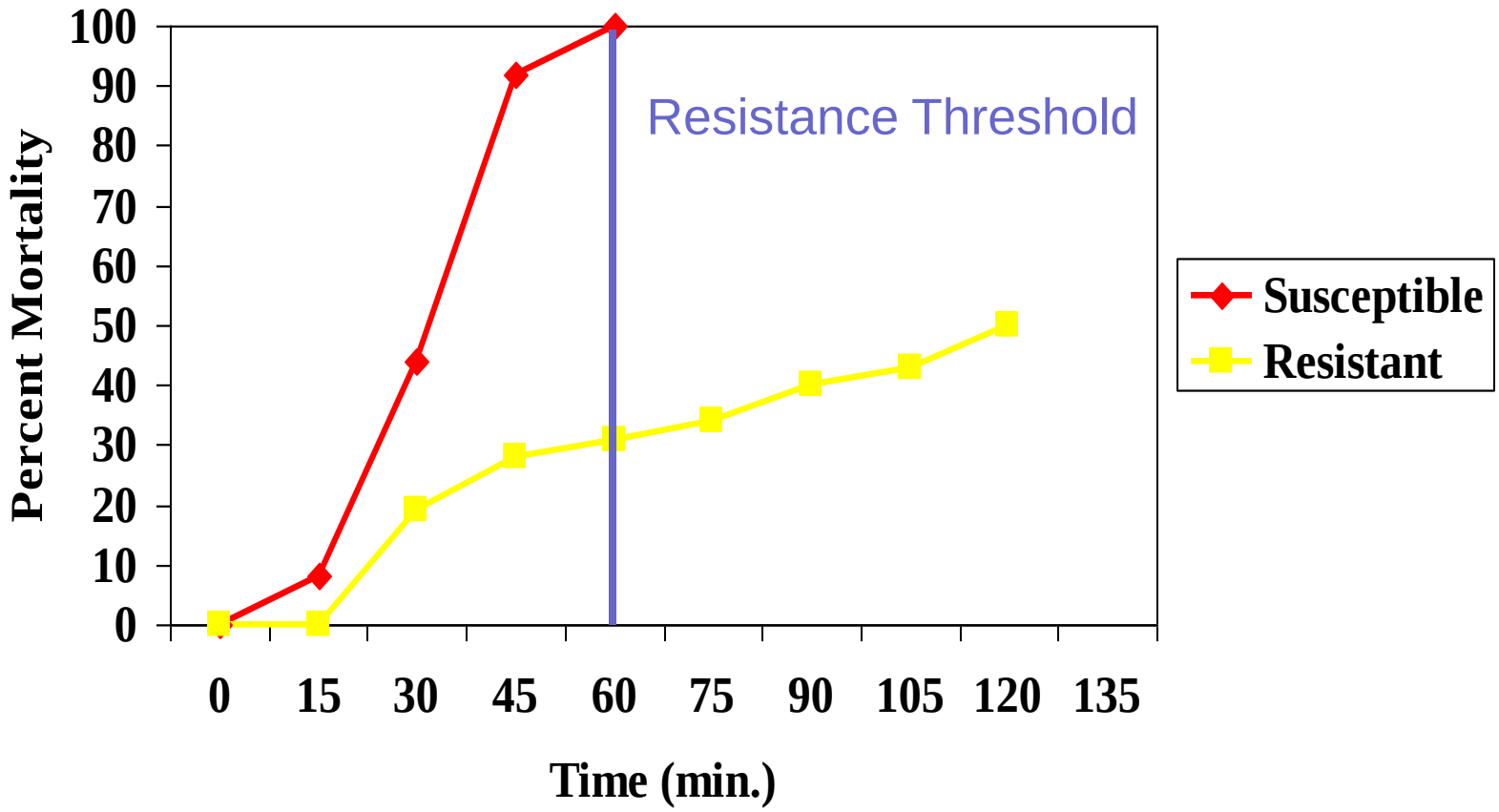
- There are multiple assays used to detect resistance.
- Assays do not correlate with operational control parameters. i.e. doses in assays ≠ label rates.
- Only caged field tests mimic operational control but are difficult to interpret unless done with susceptible mosquitoes to detect resistance.

Chief Advantage of Bottle Bioassay

DIRECT MEASUREMENT OF THE CRITICAL TOXICOLOGICAL PARAMETER:

The length of time required for an insecticide to traverse intervening tissues to reach and interact with it's target in the presence or absence of any resistance mechanism(s).

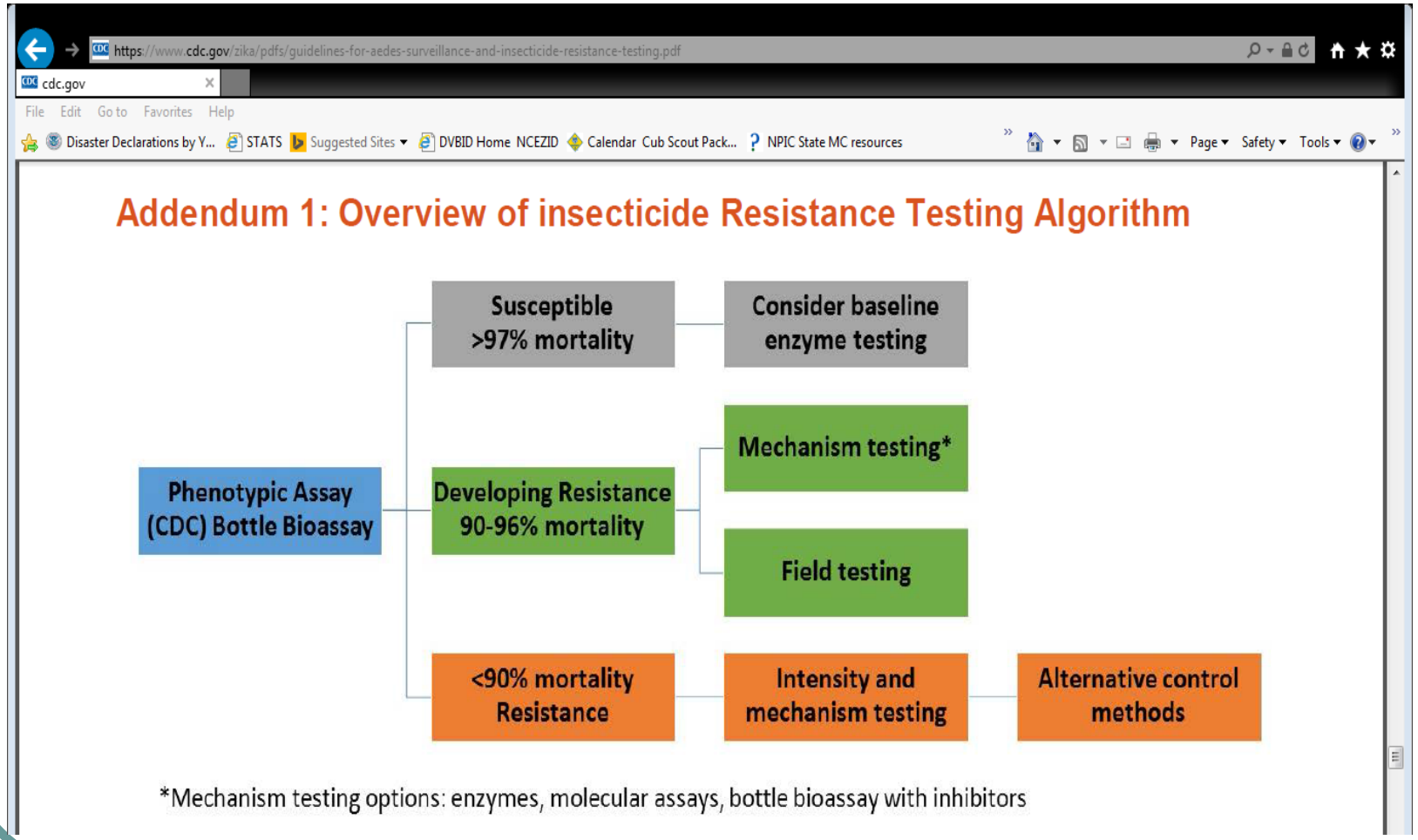
The upper range limit in minuets for survival of a representative susceptible population is selected as the resistance threshold.



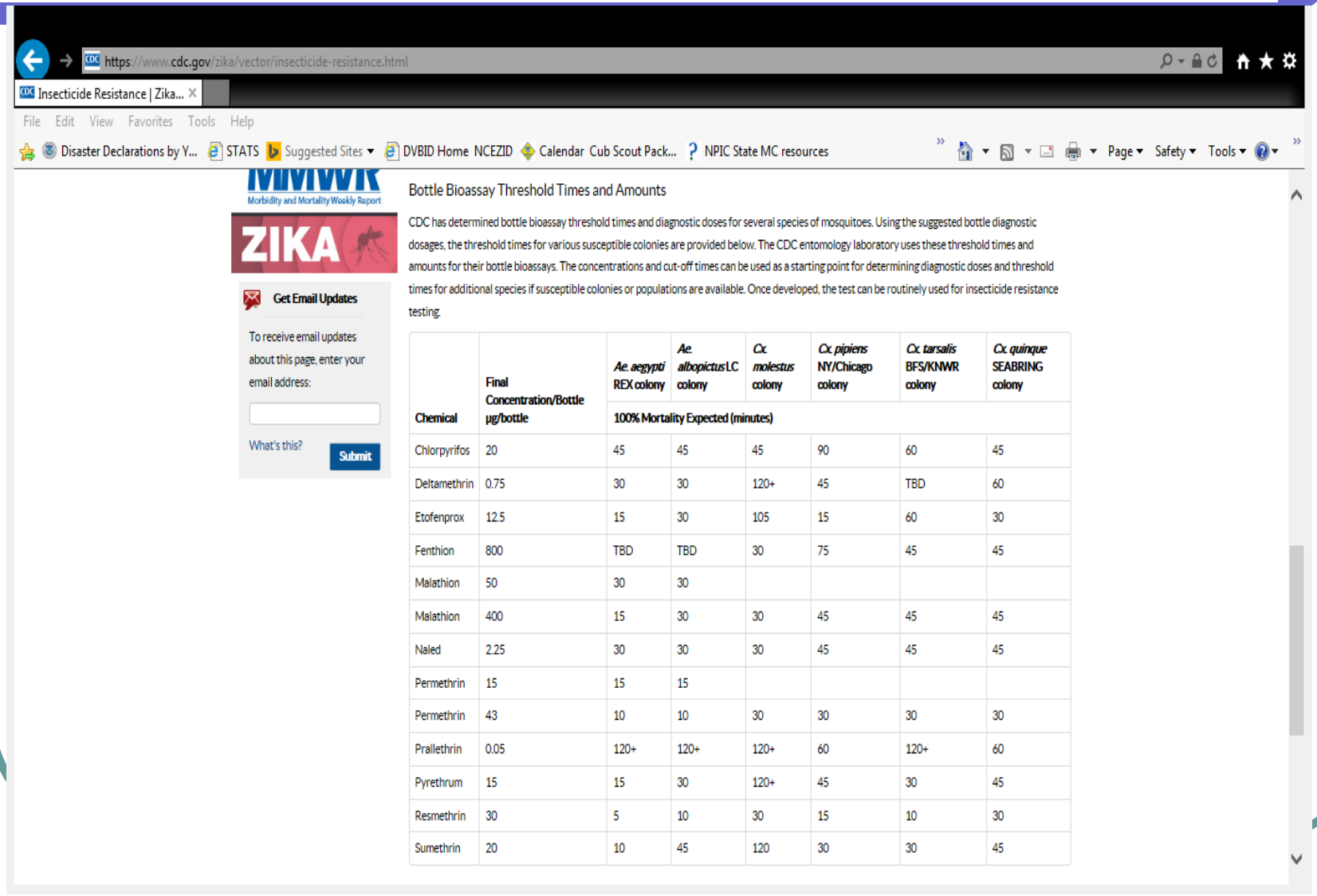
What do we do with this information?

- Rotate chemicals?
- More emphasis on other control methods?
- Change decision points to reduce use?

www.cdc.gov/zika



www.cdc.gov/zika



Bottle Bioassay Threshold Times and Amounts

CDC has determined bottle bioassay threshold times and diagnostic doses for several species of mosquitoes. Using the suggested bottle diagnostic dosages, the threshold times for various susceptible colonies are provided below. The CDC entomology laboratory uses these threshold times and amounts for their bottle bioassays. The concentrations and cut-off times can be used as a starting point for determining diagnostic doses and threshold times for additional species if susceptible colonies or populations are available. Once developed, the test can be routinely used for insecticide resistance testing.

Chemical	Final Concentration/Bottle µg/bottle	*Ae. aegypti* REX colony	*Ae. albopictus* LC colony	*Cx. molestus* colony	*Cx. pipiens* NY/Chicago colony	*Cx. tarsalis* BFS/KNWR colony	*Cx. quinque* SEABRING colony
		100% Mortality Expected (minutes)					
Chlorpyrifos	20	45	45	45	90	60	45
Deltamethrin	0.75	30	30	120+	45	TBD	60
Etofenprox	12.5	15	30	105	15	60	30
Fenthion	800	TBD	TBD	30	75	45	45
Malathion	50	30	30				
Malathion	400	15	30	30	45	45	45
Naled	2.25	30	30	30	45	45	45
Permethrin	15	15	15				
Permethrin	43	10	10	30	30	30	30
Prallethrin	0.05	120+	120+	120+	60	120+	60
Pyrethrum	15	15	30	120+	45	30	45
Resmethrin	30	5	10	30	15	10	30
Sumethrin	20	10	45	120	30	30	45

Greatest challenges

- Documenting presence of resistance
- Documenting resistance mechanisms
- Understanding what the outcomes of resistance management strategies produce

What am I?

Hint: Not a resistant mosquito larva.

CDC