MOSQUITO DISEASE ECOLOGY

Becky Trout Fryxell, Ph.D
Medical & Veterinary Entomology
University of Tennessee
RFryxell@utk.edu
Twitter: @DrVectorBio

ENTOMOLOGY & PLANT PATHOLOGY

General Definitions

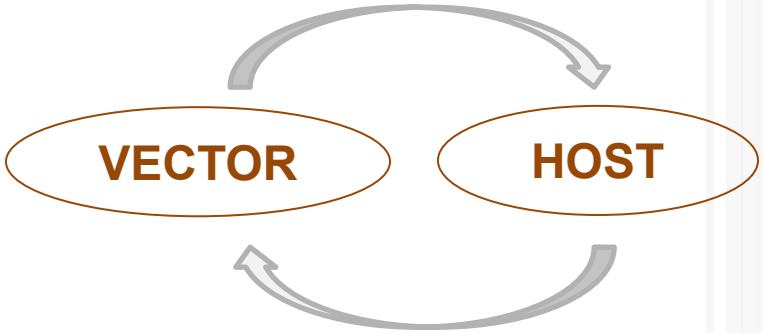
- What's an arthropod?
  - Exoskeleton
  - Segmented Body
  - Jointed Appendages
- What's an insect?
  - 3 Tagma (head-thorax-abdomen)
  - 6 Legs (3 pairs)
  - Compound Eyes
  - 2 Antennae (1 pair)

- What's a vector?
  - Arthropod that transmits a pathogen
  - Bridge spatial & ecological gap btwn animals & humans
  - Increase opportunities for emergence (disperse)
- What is a vector-borne disease?
  - Infectious agent transmitted by an arthropod (vector) that causes a negative immune response (disease)

The Etymology of the Entomologist's Epidemiology

- Vectors TRANSMIT agents…
  - Ex. viruses, bacteria, protozoa, nematodes
  - transmit is a verb
- Pathogens (agents) cause symptoms which leads to a disease diagnosis
- Vectors do not TRANSMIT diseases & they do not VECTOR agents
  - vector is a noun
- The host responds to infection of an agent and they produce a response…which is classified as a disease
- Correct: A vector transmits a pathogen which causes a symptom and the disease is diagnosed

Transmission Patterns



- **Mechanical** – 'flying needle'
  - No dvpt or replication- 'fomite'
    - *E. coli* or *Salmonella* on flies/roaches
- **Biological** – often necessary
  - Propagative
    - Multiplication of pathogen w/in vector (bacteria/virus)
  - Cyclodevelopmental
    - Development of pathogen w/in vector (*W. bancrofti*)
  - Cyclopropagative
    - Multiplication & Development of pathogen w/in vector (*Plasmodium falciparum*)

Biological Transmission Routes

- **Vertical:**
  - Trans-ovarial
    - From female (<male) to offspring
- **Horizontal:**
  - Trans-stadial
    - One life stage (instar) to the next
      - Larvae to pupae to adult –WNv in some mosquitoes
  - Trans-seasonal
    - One season to the next, especially in overwintering / diapausing vectors

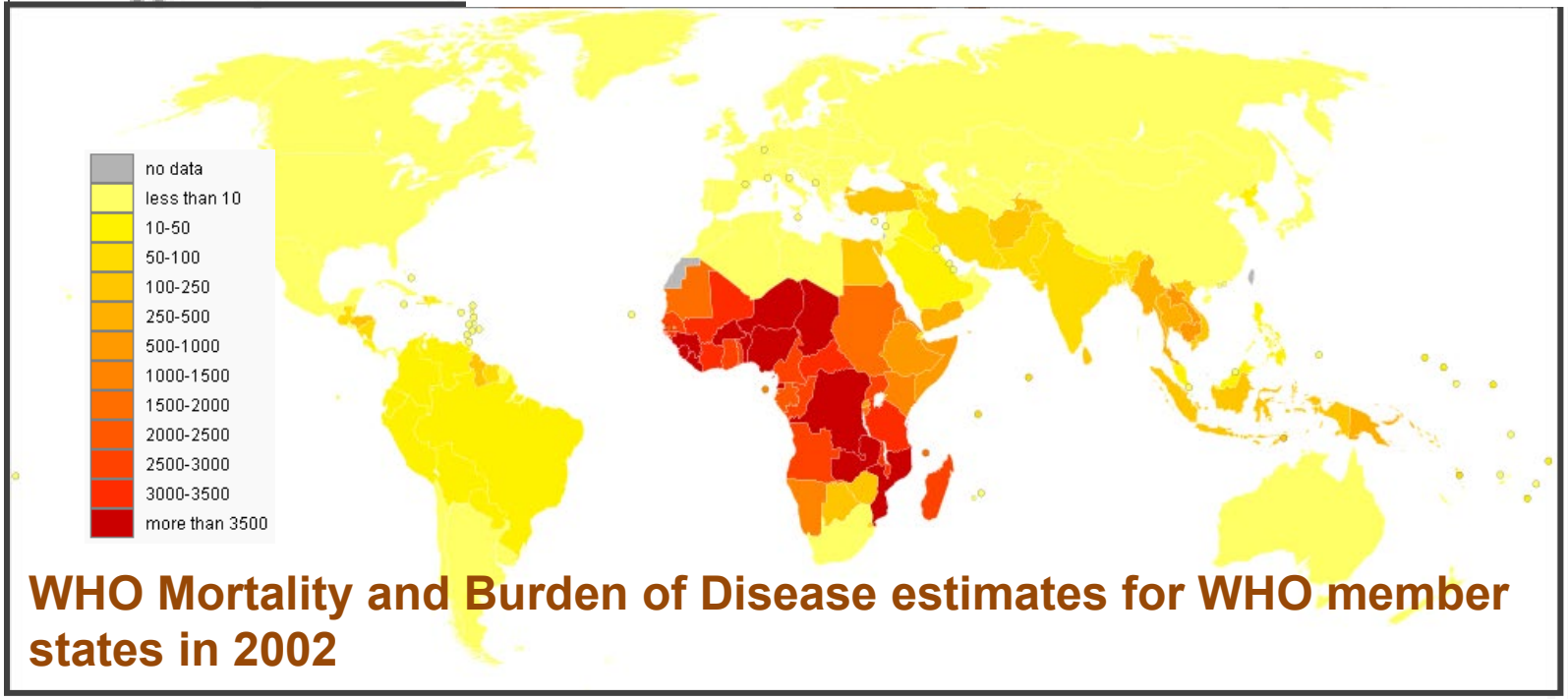
World Cases = Everyone in the Southeast + Midwest

Deaths = population of Knox county

Estimated malaria burden by WHO region in 2017

WHO Region	Malaria cases	Malaria deaths
African	200 million	403 000
Americas	976 000	630
Eastern Mediterranean	4.4 million	8300
South-East Asia	11.3 million	19 700
Western Pacific	1.9 million	3620
World	219 million	435 000

Source: *World malaria report 2018*



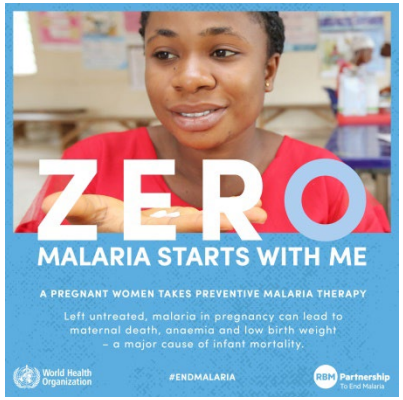
WHO Mortality and Burden of Disease estimates for WHO member states in 2002

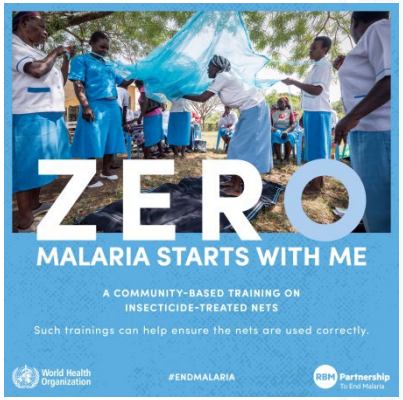
World Malaria Day (April 25)

- Every 2 minutes, a child dies from malaria
- Each year 200+million new cases are reported
- Management
  - ITNs, Case management, antimalaria pills, targeted insecticides

ZERO MALARIA STARTS WITH ME

- Grassroots campaign that aims to keep malaria
  - high on the political agenda,
  - mobilize additional resources, and
  - empower communities to take ownerships of malaria prevention and cure





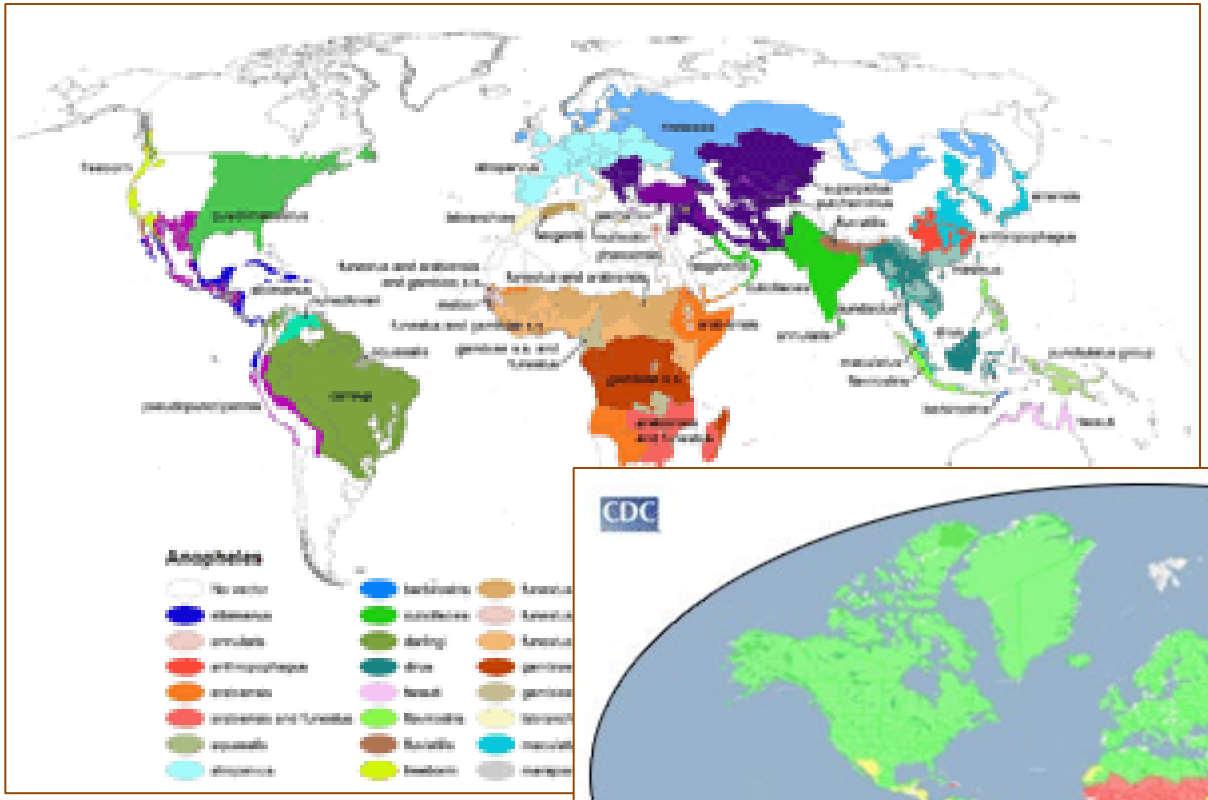
Political <u>will</u> to reduce malaria deaths
Strategic information to <u>drive</u> impact
Better guidance, policies, and strategies
A <u>coordinated</u> national malaria response

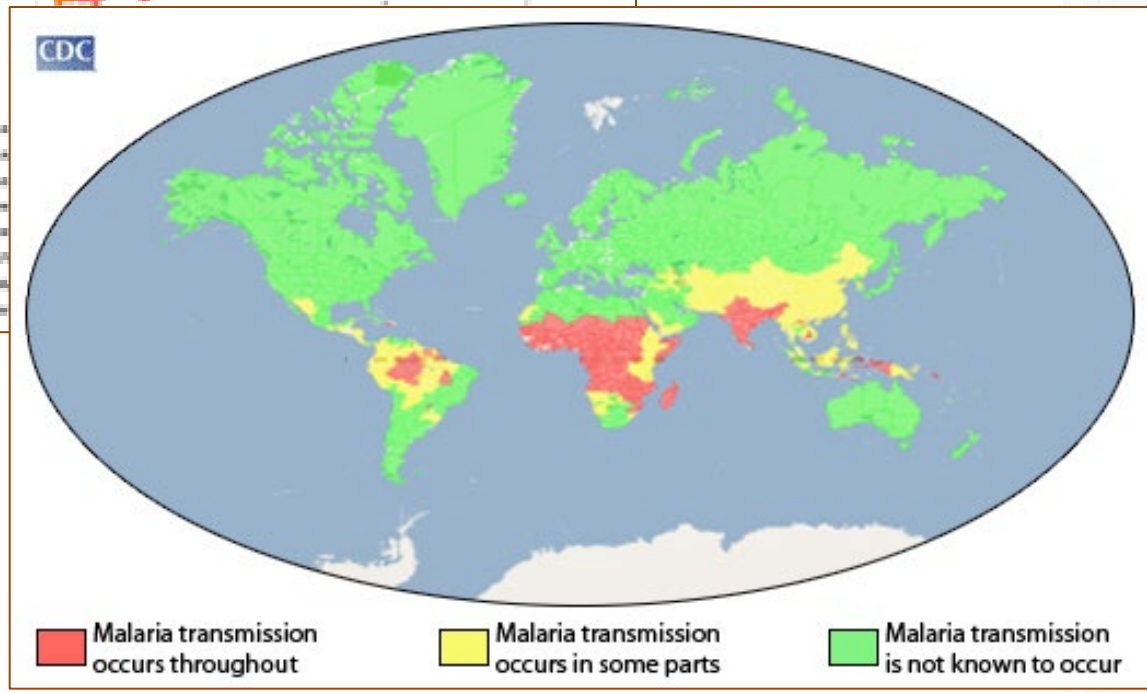
Human Malaria

- Vector: *Anopheles gambiae* Africa (30-40 spp.)
  - *Only horizontal transmission via mosquito*
- Pathogen: *Plasmodium* spp. (n = 5 species)
  - *P. falciparum* (most common)
- Host: Human
  - >219 million cases / year
  - 17% of childhood deaths
- Reservoir: None
  - 5 spp. w/ macaque reservoir
- Human risk factors
  - Young, Pregnant, Immunocompromised
  - Behavioral factors
- Symptoms in ~5-10 days
  - Cold stage, then hot stage, then sweating stage

MALARIA: ECOLOGY & DISTRIBUTION

Worldwide lots of vectors… why is malaria not everywhere?





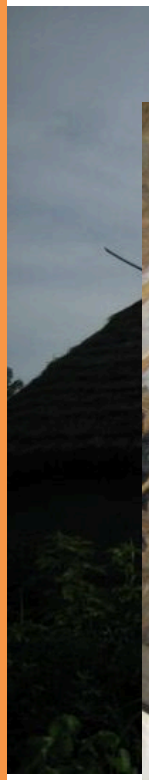
MALARIA: ENVIRONMENT IS A CHALLENGE

Water is everywhere during rainy season

Poor roads & infrastructure

Cultural Norm

MANAGEMENT: C

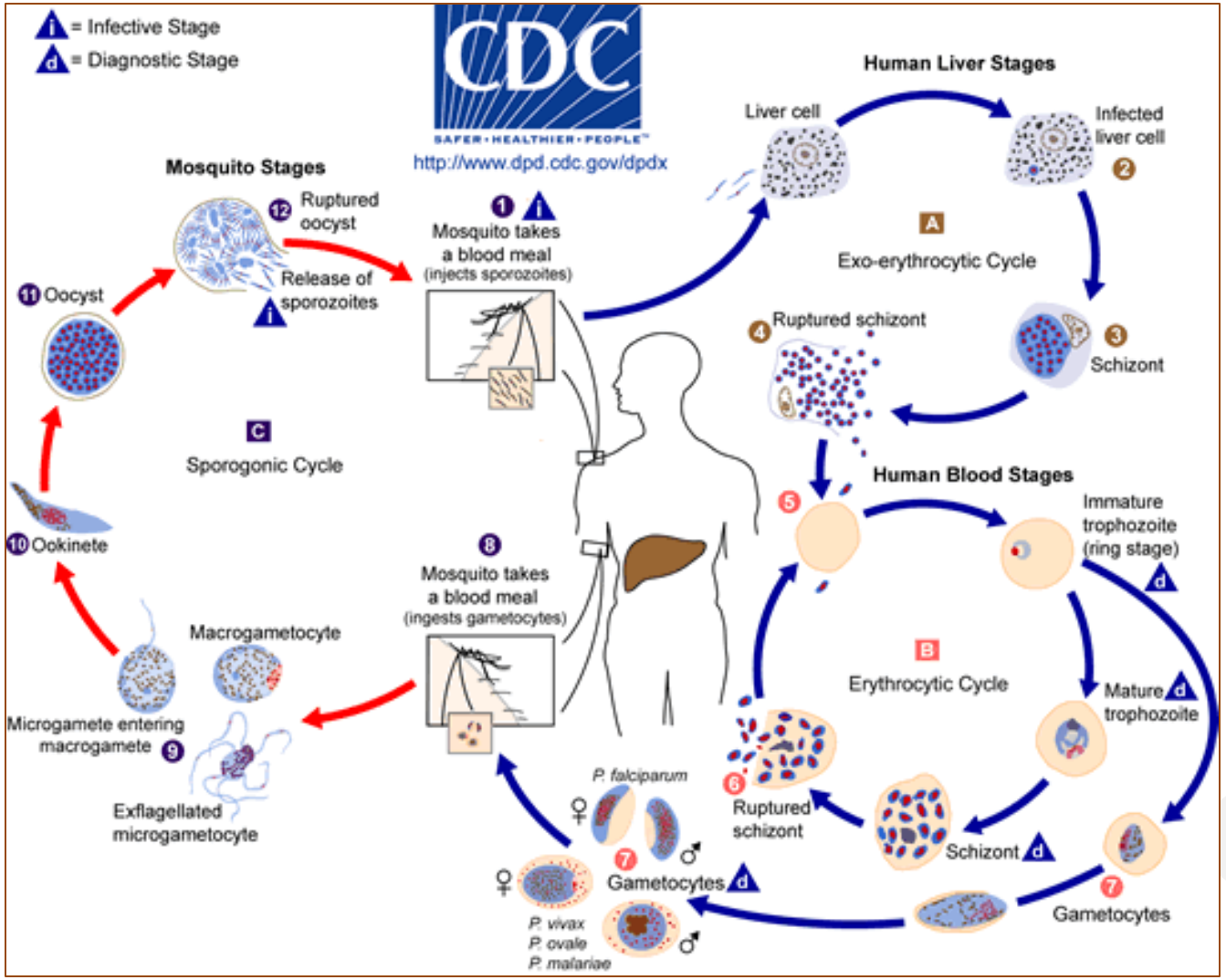


Mosquito Lesson: Disease Ecology

Mosquito Lesson: Disease Ecology

Plasmodium Life Cycle- liver / blood / mosquito

https://www.youtube.com/watch?v=2O3YrdUZQ5U&t=160s



Malaria Management

Integrated

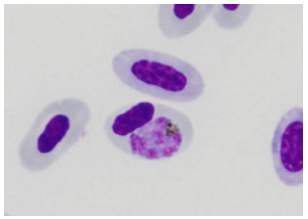
ITNs, Case Management,
Antimalarial Pills,
Targeted Insecticides

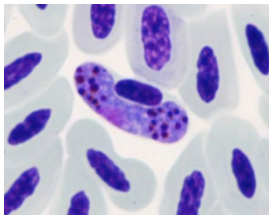
Pyrethroids Treatments

-Individual & Community Protection
-Strong Residuals
-Quick Knockdown
-Low Mammalian Toxicity

Avian Malaria

- Vector: mosquitoes (different genera for different locations)
- Protozoan Pathogen
  - *Plasmodium relictum* (spp.)
  - Haemoproteus spp.
- Host: Birds….local vs migrating birds
- Reservoir: migrating birds (?)
- Clinical Signs
  - Anemia, weakness

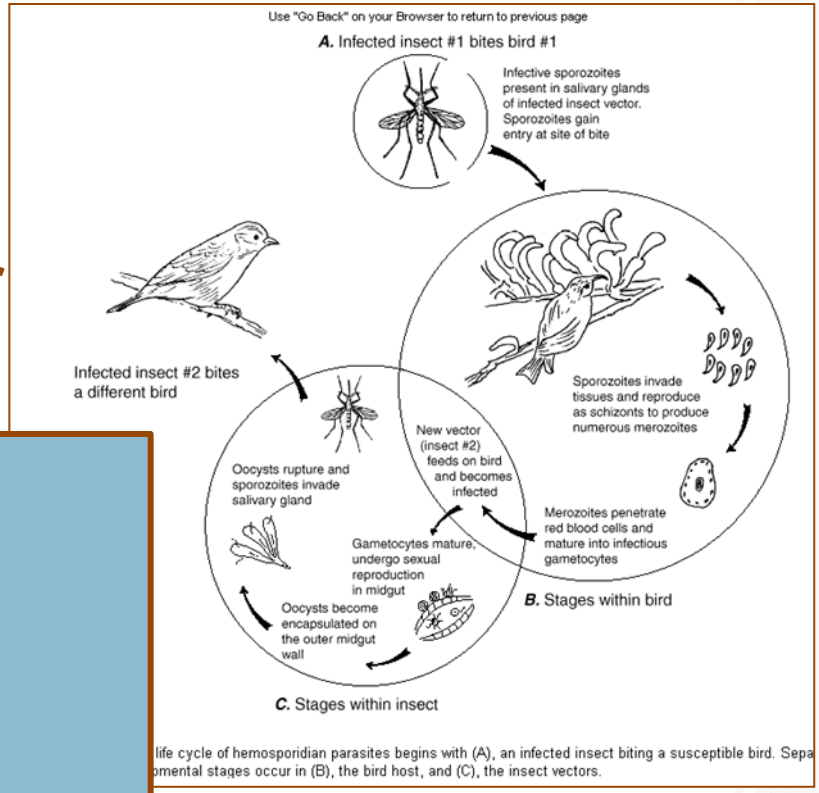


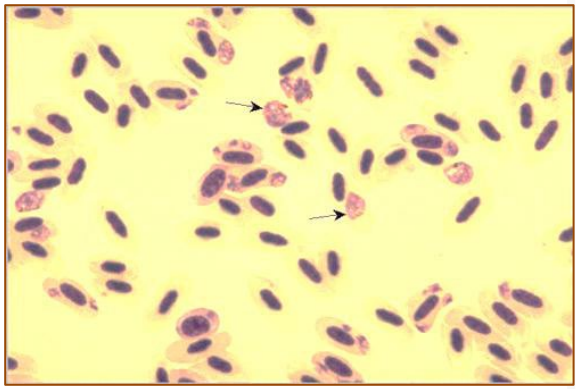


Mosquito Lesson: Disease Ecology

Hawaii, Galapagos Islands, & New Zealand

Extirpated the honeycreeper in Hawaii



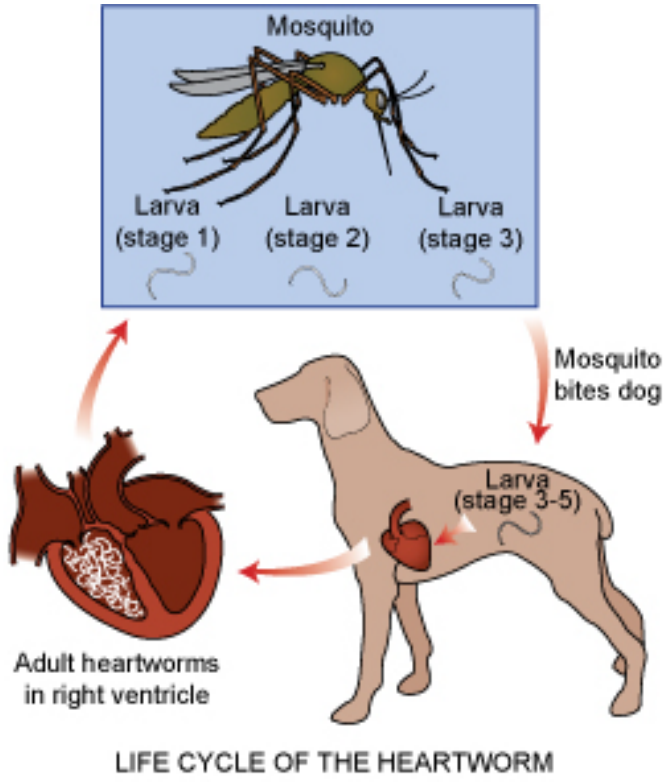


CANINE HEARTWORM – *DIROFILARIA* NEMATODES

- Biological vectors (cyclopropagative!)
  - They develop & replicate within mosquito host
- Vector- typically *Aedes vexans* mosquitoes
- Hosts: canines, coyotes, felines
  - some human cases
- Transmission & Life Cycle
  - L3 nematode transmitted to canine

http://www.youtube.com/watch?v=P6F9KApqkII

Clinical Signs Vary:
No sign – cough – cough/exercise intolerance – difficulty breathing hepatomegaly, ascites - death



LIFE CYCLE OF THE HEARTWORM

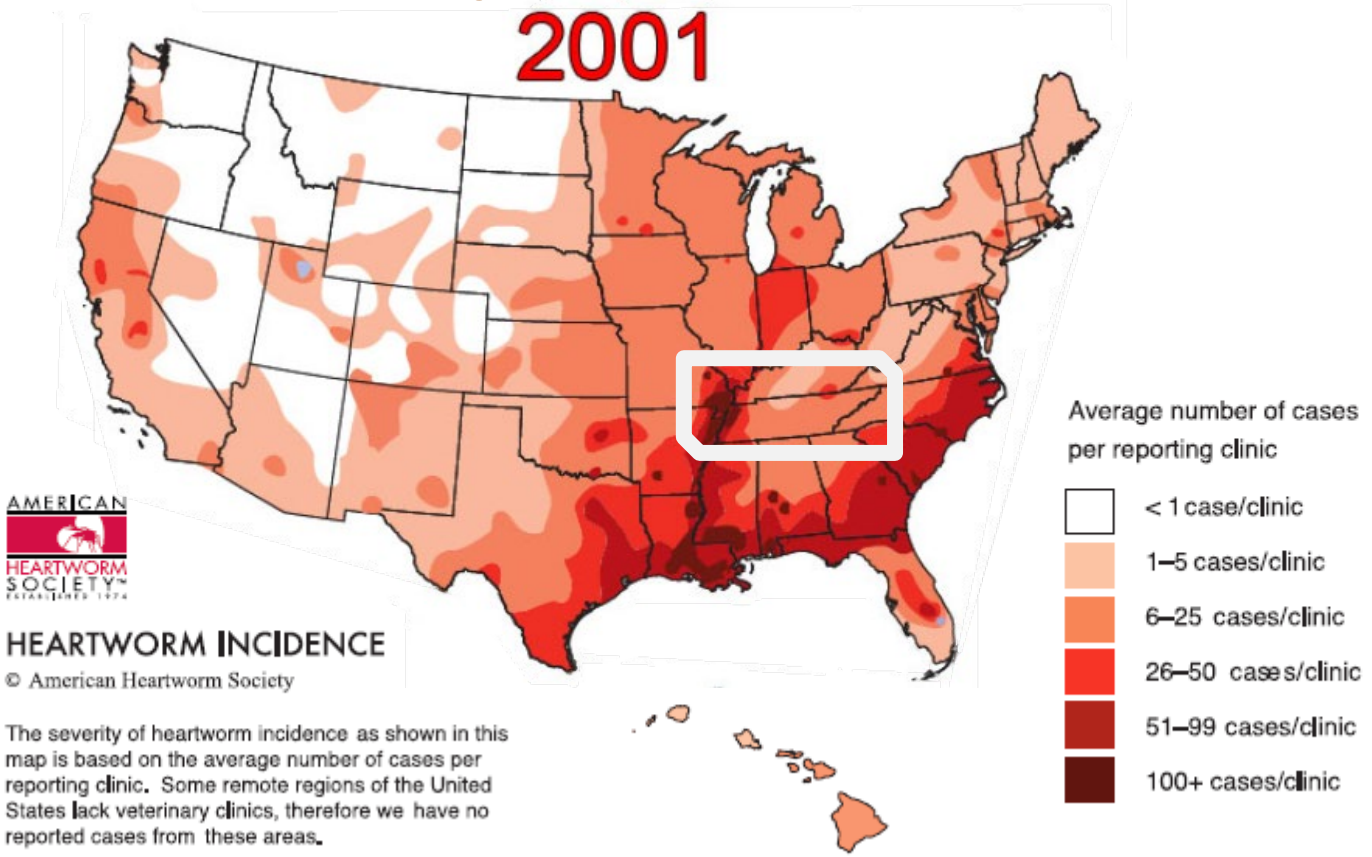
Heartworm Incidence

Treatments

- Organic arsenical compounds
  - Melarsomine dihydrochloride (intramuscular injection)
- Must keep exercise to a minimum

Preventative

- Ivermectin, Milbemcyin, Selamectin, Moxidectin



YELLOW FEVER: INTRODUCTION TO USA

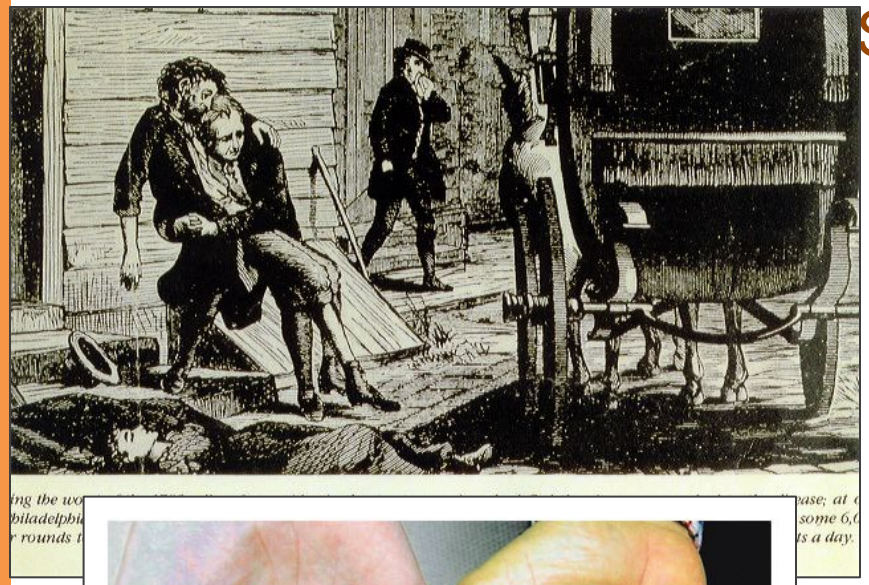
500,000 deaths

US capital moved from Philadelphia to Washington DC

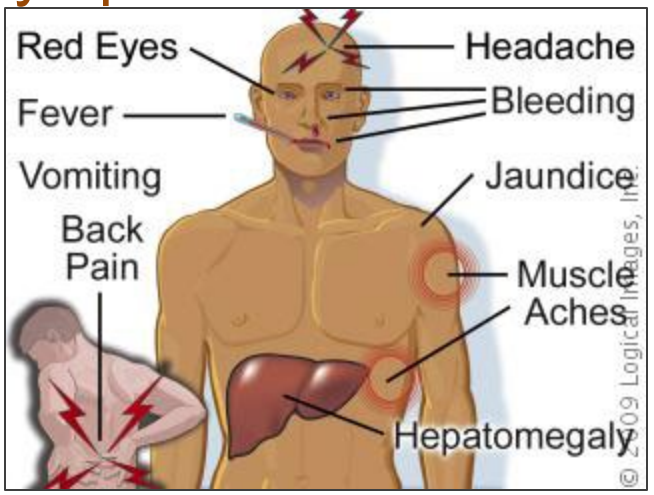
New York's Greenwich Village became 'the village' b/c it was safe haven outside of the city

Napolean abandoned his North American conquests b/c 23,000 troops died

Yellow Fever



Symptoms



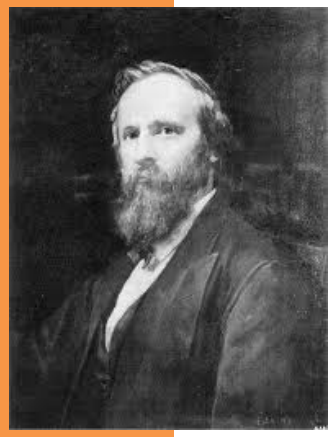
- Severe Symptoms
  - Jaundice / liver damage
  - Abdominal pain
  - Bleeding in mouths, eyes, GI... black vomit

Considered a “stranger’s disease”: Rite of passage to become acclimated to fevers and for the virus to hone in on new blood

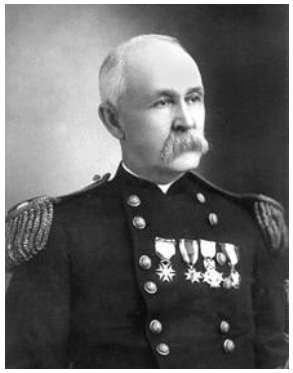
Yellow Fever: Memphis 1878

1878- Epidemic from Brazil to Ohio
Mississippi Valley 20k lives & $200 million
Worst urban disaster in American History
SFO earthquake, Johnston flood, Chicago Fire
Memphis- >5k lives (~1/3 population)
Fatality Rate:
70% Caucasian vs 6.7% African American

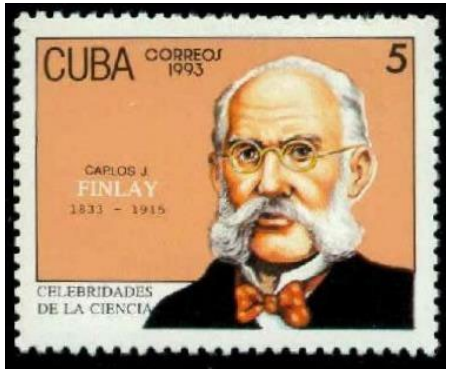
Memphis 1878



Pres. Hayes



George Sternberg



Carlos Finlay

Aedes aegypti

Germ Theory vs. Environment

George Waring
Redesigned the sewer system to eliminate germs

Finlay's Discoveries (Notes)

Hemorrhaging- infection in blood

Pathogen disappears once temperature drops

Epidemics were sporadic, *Aedes aegypti*
- multiple blood meals
- mosquito disappears below 60ºF

1881- presented his findings – *The mosquito hypothetically considered as the agent of transmission of yellow fever*

Seemed to solve the problem

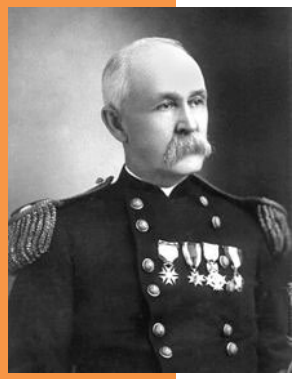
POST-1878

- 20 years - no epidemic
- 1898
  - USS Maine explodes & sinks in Havana
  - Spanish- American war (April to August)
  - US gains Philippine Islands, Puerto Rico, & Guam
  - Cuba became independent
  - August: YF quickly spread amongst the Americans Theodore Roosevelt drafted a request to Washington that it withdraw the Army from Cuba. By the time of his letter, 75% of the force in Cuba was unfit for service
- Money and political power to 'solve yellow fever'

"In no area did the US lag behind the rest of the world so much as in its study of the life sciences and medicine."

-John Barry author of *The Great Influenza*

Yellow Fever- Post War Cuba outbreak 1900

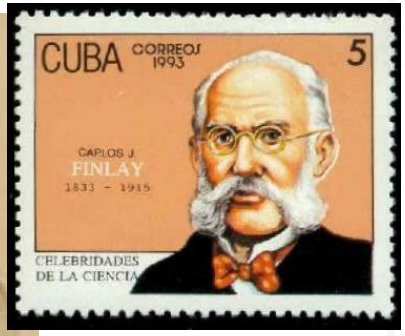


George Sternberg
(Surgeon General)

Reed

Sanitation
& Typhoid

Lazear



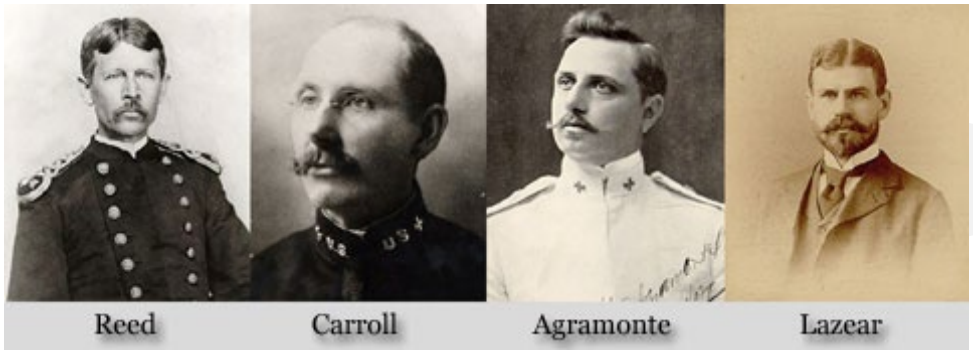
Contacted Finlay

Kept Records

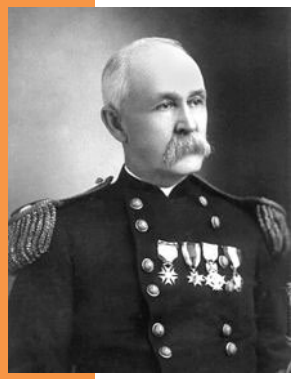
When & where people were sick
Autopsied all deaths
Detailed records of mosquitoes

May 23, 1900

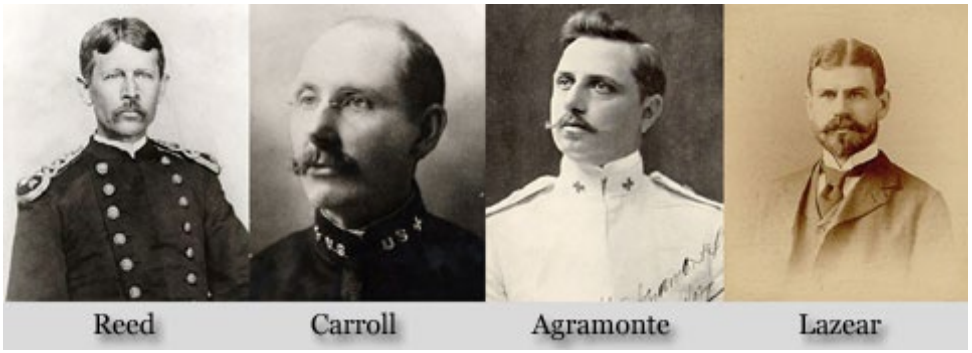
Sternberg wrote a letter to the army to send a medical board (~commission) to Havana to study all infectious disease
verbal focus on YF



Yellow Fever Commission



George Sternberg



Started work at the end of the season, so no active cases

Dr. Henry Rose Carter- noted 5-7 day period between cases

Reed & Carroll –test tubes & vomit of patient samples

*Reed got homesick and left for Washington DC

Lazear- mosquitoes from all over the island

Notes: symptoms, where fever broke, mosquito collections in the area

Epidemiology of Yellow Fever
Reed, Carroll, Agramonte, Lazear

Disprove Sarnelli bacteria

Cultures from patients with yellow fever

Cultures from autopsies

TABLE I.

Blood Cultures During Life.

Day of Disease.	Character of Attack.	No. of Cultures.	No. of Bouillon Tubes Inoculated.	B. Icteroides.
1st.	Severe.	3	4 (3 agar plates.)	Negative.
"	Well-marked.	1	4	"
"	Mild.	1	3	"
2nd.	Severe.	6	18	"
"	Well-marked.	1	2	"
"	Mild.	1	3 (6 agar plates.)	"
3rd.	Severe.	7	18 (6 agar plates.)	"
"	Mild.	2	4	"
4th.	Severe.	5	14	"
"	Well-marked.	2	6	"
"	Mild.	1	1	"
5th.	Severe.	5	12 (3 agar plates).	"
"	Well-marked.	1	3	"
"	Mild.	1	1	"
6th.	Severe.	4	6	"
"	Well-marked.	1	2	"
7th.	Severe.	1	2	"
"	Well-marked.	1	2	"
8th.	Severe.	2	6	"
"	Well-marked.	1	2	"
9th.	Severe.	1	2	"

Number of cultures..... 48
" " bouillon tubes inoculated..... 115
" " agar plates..... 15

TABLE II.

No. of Case.	Day of Disease.	Time of Autopsy.	Source of Culture.	B. Icteroides.
1	7th.	2 hours after death.	Blood, liver, spleen, kidney.	Neg'tive
2	6th.	13 hours after death.	Blood, liver, spleen, kidney.	"
3	4th.	8 hours after death.	Blood, liver, spleen, kidney.	"
4	8th.	4 hours after death.	Abdominal cavity, blood, liver, spleen, kidney, bile, duodenum.	"
5	4th.	4 hours after death.	Blood, liver, spleen, kidney, bile, duodenum.	"
6	6th.	6½ hours after death.	Abdominal cavity, blood, pericardial fluid, lung, spleen, kidney, liver, bile, duodenum.	"
7	6th.	50 minutes after death.	Blood, lung, liver, spleen, kidney, bile, jejunum.	"
8	6th.	½-hour after death.	Blood, lung, liver, spleen, kidney, urine, small intestine.	"
9	4th.	2 hours after death.	Liver, spleen, small intestine.	"
10	5th.	7 hours after death.	Liver, kidney, spleen, small intestine.	"
11	3rd.	½-hour after death.	Liver, kidney, spleen.	"

Etiology of Yellow Fever

Reed, Carroll, Agramonte, Lazear

Mosquito as a Host of the Parasite of Yellow Fever

Dr. Henry Rose Carter- noted 5-7 day period between cases

The circumstances under which Carter worked were favorable for recording with considerable accuracy the interval between the time of arrival of infecting cases in isolated farm-houses and the occurrence of secondary cases in these houses. According to Carter "the period from the first (infecting) case to the first group of cases infected, at these houses, is generally from two to three weeks."

Finlay's report

His present views on this subject may be stated in his own language: "First, reproduction of the disease, in a mild form, within five to twenty-five days after having applied contaminated mosquitoes to susceptible subjects. Second, partial or complete immunity against yellow fever obtained when even no pathogenous manifestation had followed those inoculations." (Medical Record, Vol. 55, No. 21, May 27, 1899.)

Without reviewing the cases regarded as mild forms by the author of this theory, we believe that he has not, as yet, succeeded in reproducing a well marked attack of yellow fever, within the usual period of incubation of the disease, attended by albumen and jaundice, and in which all other sources of infection could be excluded.

EPIDEMIOLOGY OF YELLOW FEVER

TABLE III.

INOCULATION OF NON-IMMUNE INDIVIDUALS THROUGH THE BITE OF MOSQUITOES (C. FASCIATED).

No. of Case.	Age.	Nativity.	Date of Inoculation.	Character of attack and No. of patients bitten.	Day of Disease.	Time of between infection of mosquito and inoculation.	No. of Mosquitoes.	Result.	Remarks.
1		U. S.	August 11th.	Mild.....1	7th.	5 days.	One.	Negative.	
2		U. S.	" 11th.	Very mild ..1	5th.	5 "	"	"	
3	24	U. S.	" 12th.	" ..1	5th.	6 "	"	"	
4	20	U. S.	" 12th.	" ..1	5th.	6 "	"	"	
5	24	U. S.	" 14th.	" ..1	5th.	8 "	"	"	
6	34	U. S.	" 16th.	" ..1	5th.	10 "	"	"	
7	22	U. S.	" 18th.	Severe.....1	2nd.	3 "	"	"	
8	20	U. S.	" 19th.	Very mild ..1	5th.	13 "	Two.	"	
				Severe.....1	1st.	3 "			
9	28	U. S.	" 25th.	Fatal. 1	2nd.	6 "	One.	"	
				Mild.....1	1st.	4 "			
				Severe.....1	2nd.	2 "			
10	46	England.	" 27th.	" 1	2nd.	12 "	One.	Positive.	Severe attack of yellow fever.
				Mild.....1	1st.	6 "			
				Severe.....1	2nd.	4 "			
				Mild.....1	2nd.	2 "			
11	24	U. S.	" 31st.	Fatal.....1	2nd.	12 "	One.	Positive.	
				Mild.....2	2nd.	4 and 10 days.			
				Severe.....2	2nd and 9th.	2 and 8 "	One.		Well marked attack of yellow fever.
				" 3	1st, 2nd and 2nd.	2, 8 and 16 "			
				Mild.....2	1st and 2nd.	6 and 10 "	One.		
				Fatal.....1	2nd.	12 "			
				Severe 1	1st.	2 "	One.		
				Mild.....3	1st, 2nd and 2nd.	4, 6 and 10 "			
				Severe.....3	All on 1st.	2, 4 and 8 "	One.		
				Mild.....1	2nd.	6 "			

Virus had to stay in mosquito for a period of time (extrinsic)

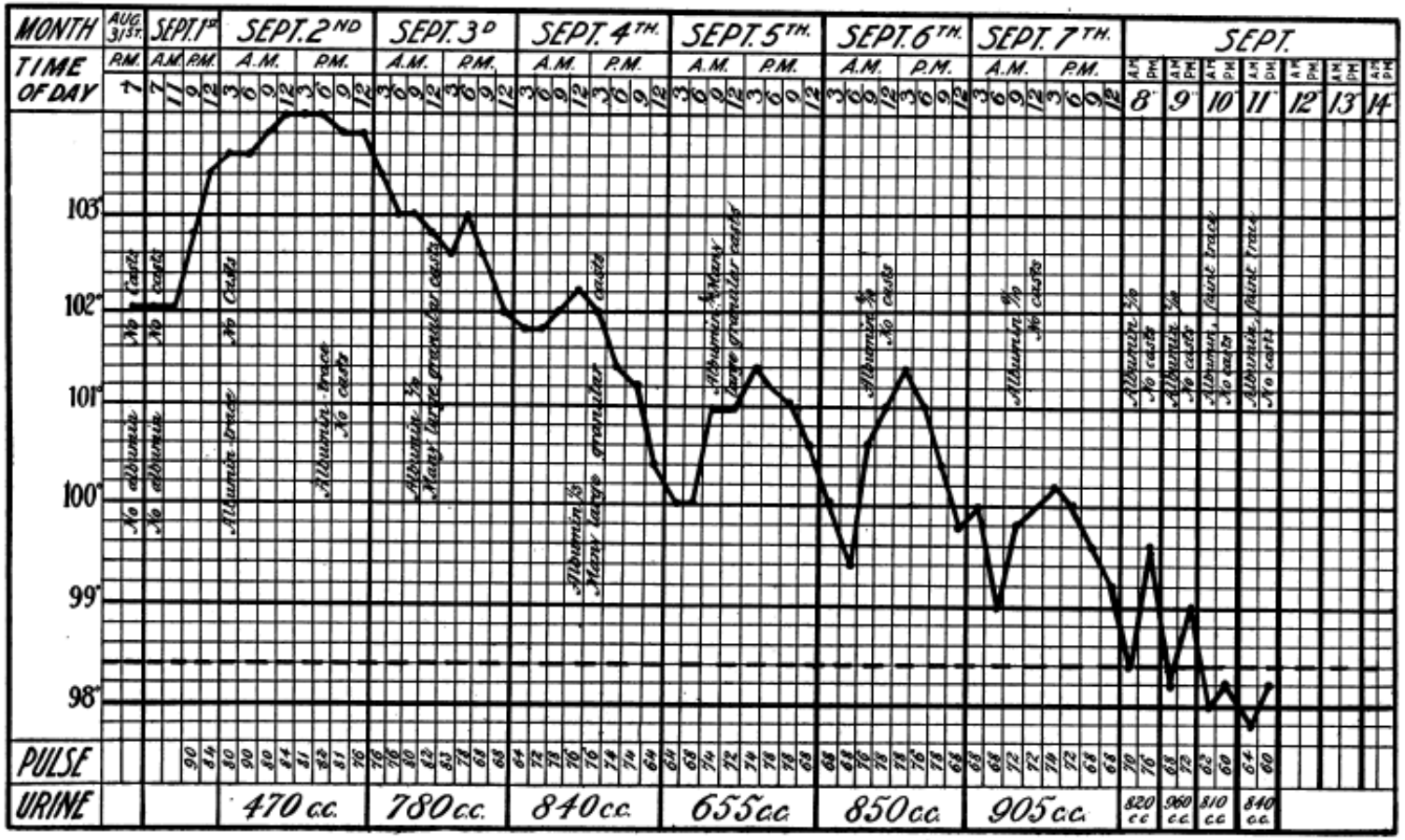
Mosquito Lesson: Disease Ecology

EPIDEMIOLOGY OF YELLOW FEVER

Case 10: Dr. Carroll- survived

Poor case b/c he contacted YF patients

Case I. Yellow fever following, within the usual period of incubation, the bite of an infected mosquito, (Culex fasciatus)



Epidemiology of Yellow Fever

Case 11: patient XY (William E. Dean)

4 bites = same mosquito as Carroll & 3 more

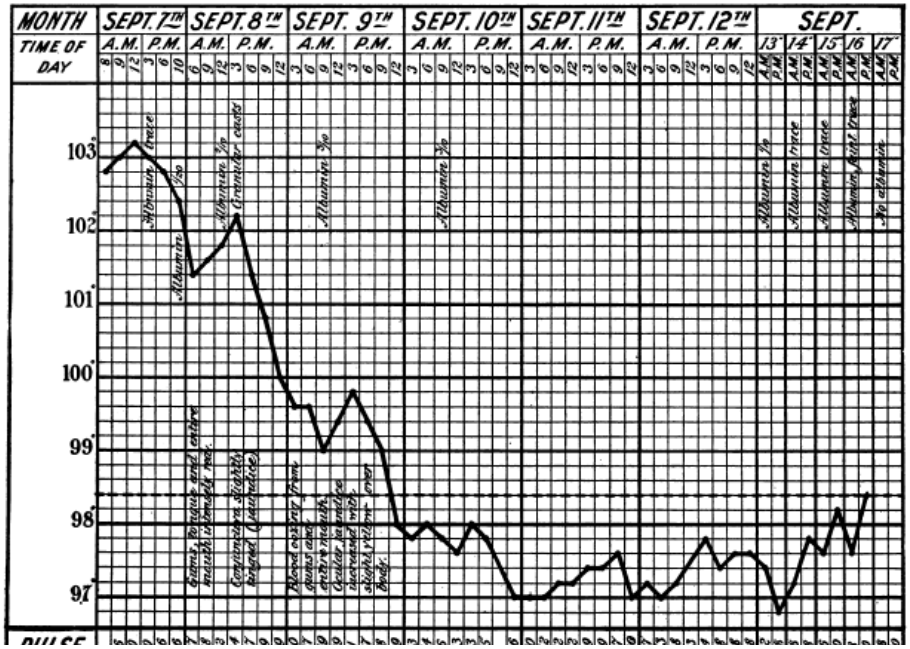
...serious infection

Excellent Case…

New to tropics

On base for >2 mo.

Chart II. Yellow fever following, within the usual period of incubation, the bite of an infected mosquito, (Culex fasciatus)



Taken in connection with Case 11, in which we have been unable to find any other source of infection than the bite of an infected mosquito, five days preceding the attack, the case of Dr. C. (Case 10, Table III) becomes strongly confirmatory of the same origin.

Epidemiology of Yellow Fever

In Lazear's notebook, Lazear wrote…

Sept. 13- guinea pig had been bitten by a mosquito that developed from an egg laid by a mosquito that fed on a number of YF cases

5 days later…

EPIDEMIOLOGY OF YELLOW FEVER

Case. Dr. Jesse W. Lazear, Acting Assistant Surgeon, U. S. Army, a member of this board, was bitten on August 16, 1900, (Case 6, Table III), by a mosquito (*Culex fasciatus*) which ten days previously had been contaminated by biting a very mild case of yellow fever (fifth day). No appreciable disturbance of health followed this inoculation.

September 13, 1900, (forenoon), Dr. Lazear, while on a visit to Las Animas Hospital and while collecting blood from yellow fever patients for study, was bitten by a Culex mosquito (species undetermined). As Dr. L. had been previously bitten by a contaminated insect without after-effects, he deliberately allowed this particular mosquito, which had settled on the back of his hand, to remain until it had satisfied its hunger.

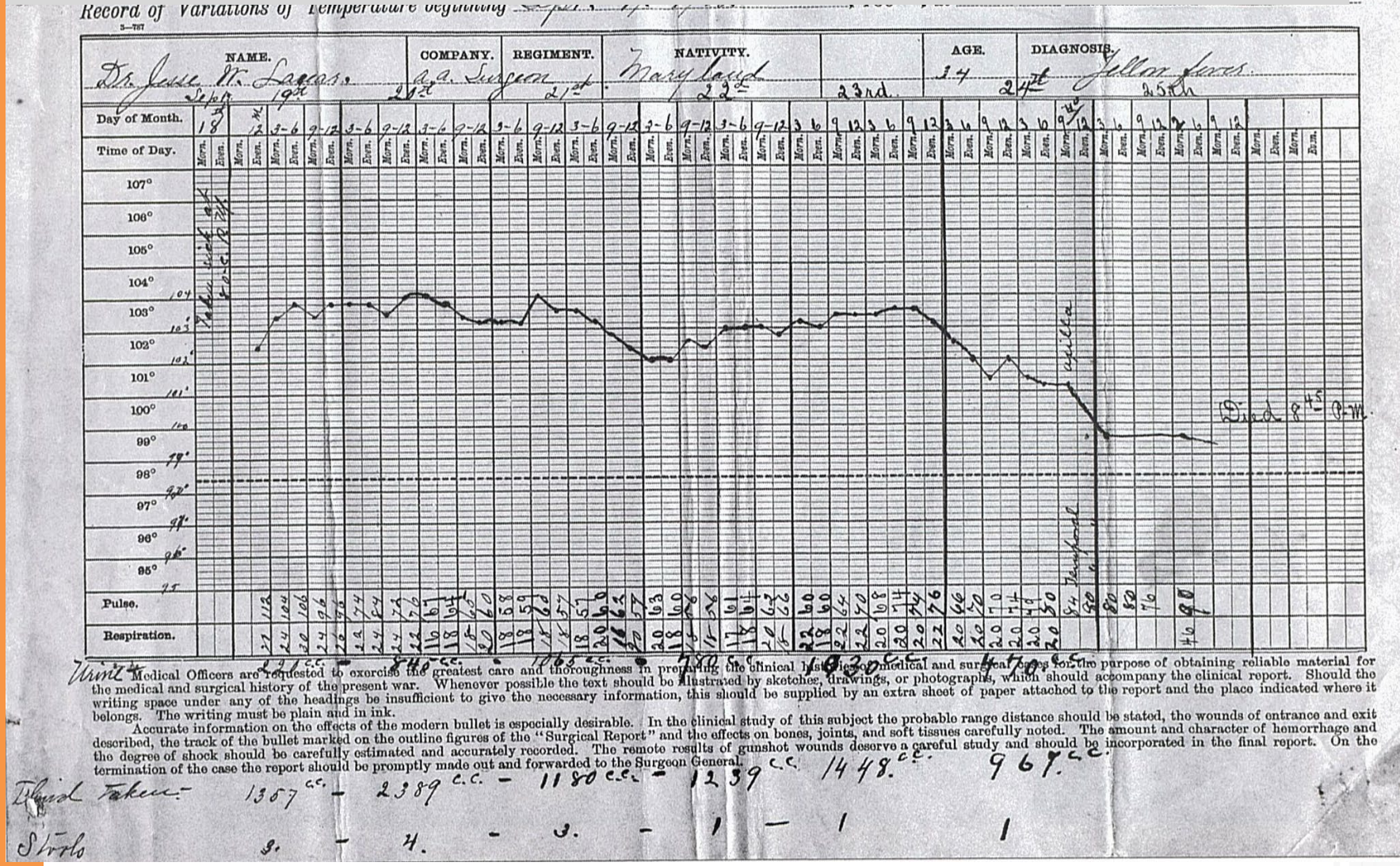
On the evening of September 18th, 5 days after the bite, Dr. L. complained of feeling "out of sorts," and had a chill at 8 p. m.

September 19th, 12 o'clock noon, T. 102.4°, pulse 112. Eyes injected, face suffused; 3 p. m., T. 103.4°, pulse 104; 6 p. m., T. 103.8°, pulse 106. *Albumen* appeared in the urine. *Jaundice* appeared on the third day. The subsequent history of this case was one of progressive and fatal yellow fever, the death of our much lamented colleague having occurred on the evening of September 25, 1900.

JESSE LAZEAR'S MEDICAL CHART

Mosquito Lesson: Disease Ecology

Lazear died from the virus passing from female to offspring (transovarial transmission)



EPIDEMIOLOGY OF YELLOW FEVER

From our study thus far of yellow fever, we draw the following conclusions:

1. *Bacillus icteroides (Sanarelli) stands in no causative relation to yellow fever, but, when present, should be considered as a secondary invader in this disease.*

2. *The mosquito serves as the intermediate host for the parasite of yellow fever.*

In response to this paper,

Italy: "silliest beyond compare"

US: "$10,000 was used to fund further mosquito experiments at a camp- to be named Camp Lazear"

Mosquito Lesson: Disease Ecology

Camp Lazear

Null Hypothesis: Mosquito transmitted

Alternate Hypothesis: Bacteria on clothing

Experiment 1:

Infected Clothing, No mosquitoes or sunlight

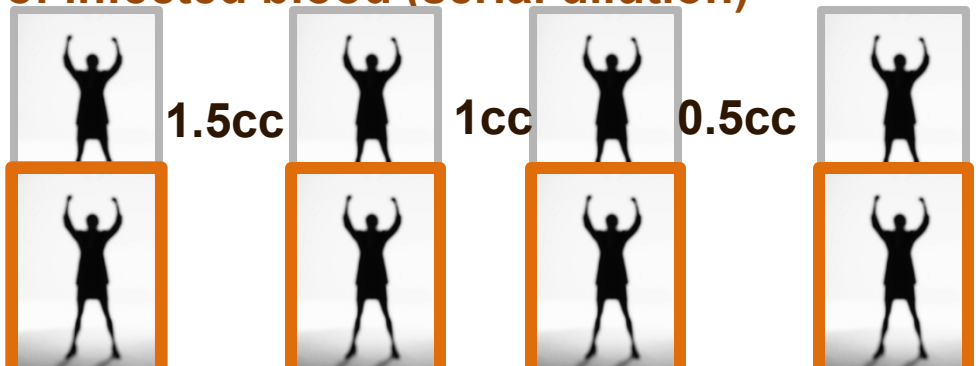
For $100 men locked themselves in the building and wore the infected clothes for 20 nights --- no yellow fever

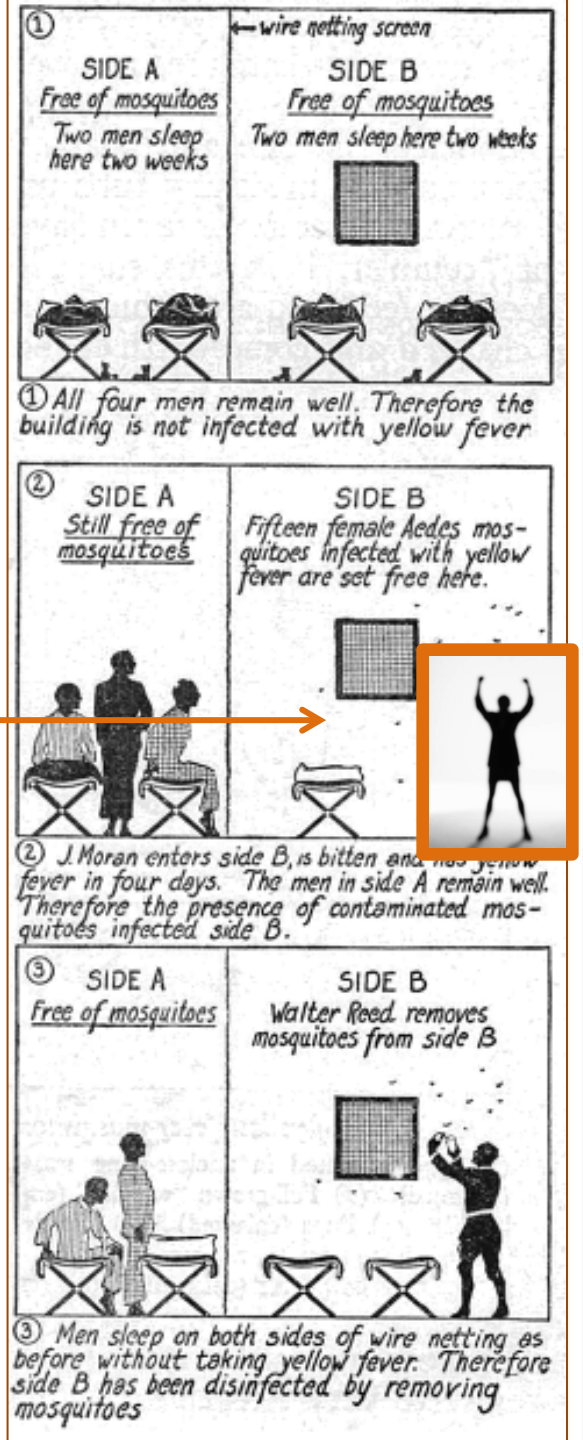
Experiment 2: Mosquito-borne

Experiment 3: Immunity

4 volunteers would receive a decreased dose of infected blood (serial dilution)

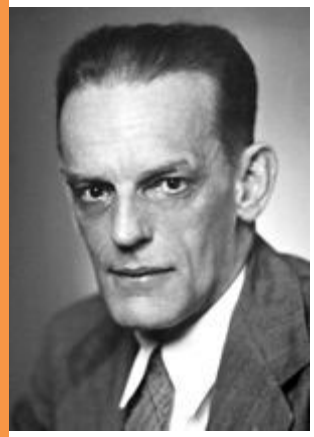
2cc 1.5cc 1cc 0.5cc





Yellow Fever: Eradication from US

- Rockefeller Foundation's International Health Board- vector control
- Mosquito Control - same mosquito transmits Dengue fever- DDT



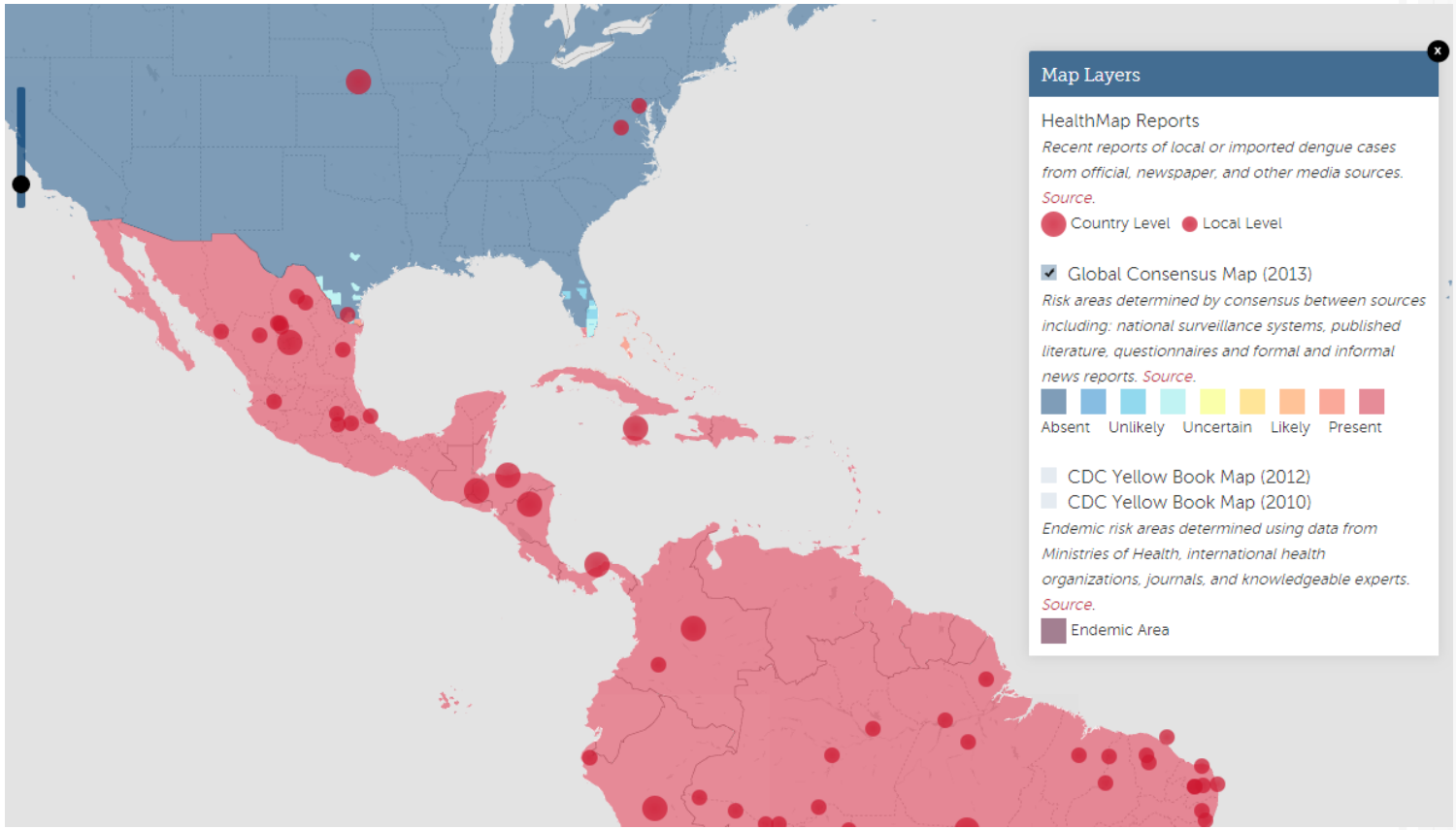
- Max Theiler- developed vaccine in 1930s and won Nobel Prize in 1951
  - Same vaccine

- Limit outdoors during peak hours
- Proper clothing
- Insect repellent

(https://www.epa.gov/insect-repellents/find-repellent-right-you)

DENGUE: CONSTANT US THREAT

- 4 related viruses make vaccine development difficult
- Symptoms show 4-7d after bite & last for 3-10d
  - HIGH fever & headache, eye pain, joint pain, rash, low white cell count https://www.cdc.gov/dengue/index.html

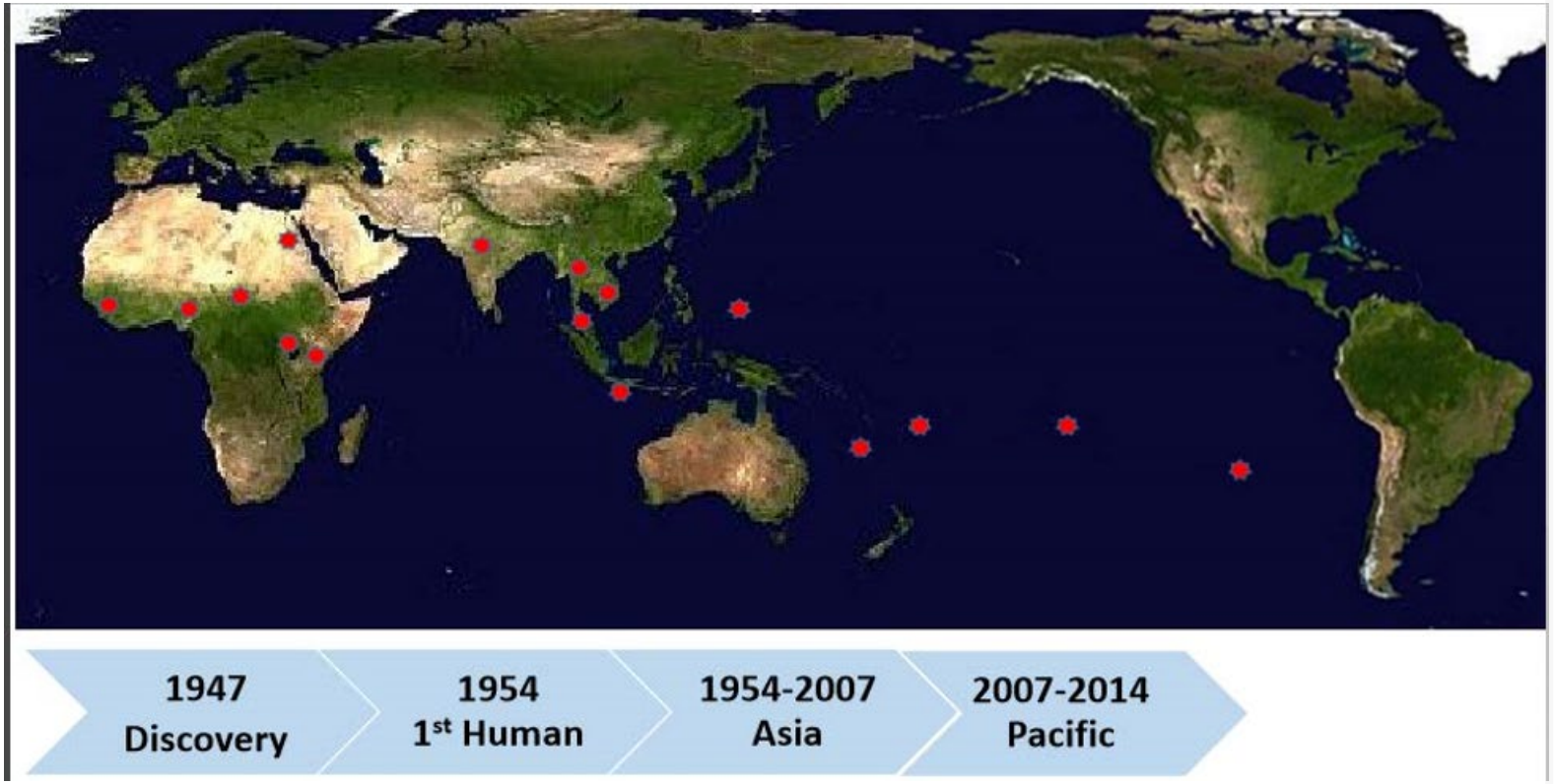


Zika Virus

- Rockefeller Foundation
  - Arbovirus Discovery
    - 1947 sentinel macaque
    - 1948 mosquito
      - *Aedes africanus*
    - 1954 human

- **Other hosts**
  Elephants, goats, hippos, lions, rodents, zebras…etc.

ZIKA SLOWLY SPREADS EAST- INDIRECT



- **African vectors:**
  - ***Aedes aegypti, Aedes albopictus, Aedes luteocephalus, Aedes vittatus***
- **Asian vectors**
  - ***Aedes aegypti & Aedes albopictus***
- **Only 14 human cases documented**

ZIKA: YAP ISLANDS & FRENCH POLYNESIA

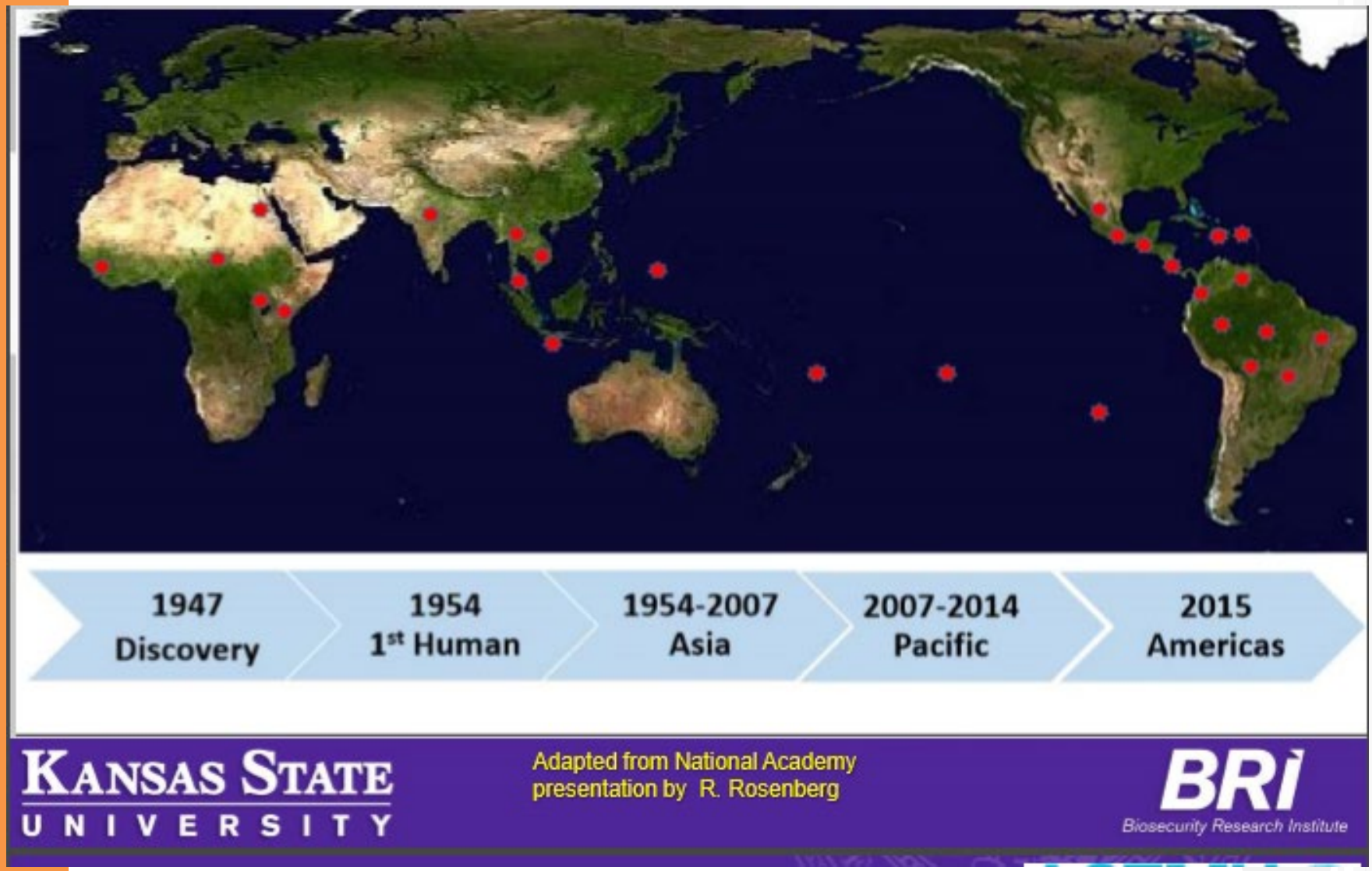
YAP 2007-2014 outbreak

- 49 confirmed cases (*59?*)
- 73% of Yap residents >3yrs infected
  - 18% symptomatic & 82% asymptomatic
- Co-infections
  - Zika, Chikungunya, & Dengue
- *Aedes hensilli* suspected vector

2013 FP

- 66% of population
  - 28,000 symptomatic & ~140,000 asymptomatic
- 1st suspected case of Guillain-Barré syndrome
- Retrospective Case Review (2015)
  - Increase in microcephaly
  - Evidence of perinatal transmission
- Asian Genotype in *Ae. aegypti & Ae. albopictus*

Mosquito Lesson: Disease Ecology



ZIKA: SEXUAL TRANSMISSION

- Senegal, 2008 - Entomologists
- Vaccinated against yellow fever
- Sick with rash

Probable Non–Vector-borne Transmission of Zika Virus, Colorado, USA

Brian D. Foy, Kevin C. Kobylinski, Joy L. Chilson Foy, Bradley J. Blitvich, Amelia Travassos da Rosa, Andrew D. Haddow, Robert S. Lanciotti, and Robert B. Tesh

Clinical & serologic evidence indicated 2 American scientists contracted Zika virus infections while working in Senegal in 2008. One of the scientists transmitted this arbovirus to his wife after returning home. Direct contact is implicated as the transmission route, most likely as a sexually transmitted infection.

- Sexual transmission in US
- Semen collected from a man with Zika virus had virus particles detectable by RT-PCR 62 days after fever
  - RT-PCR of blood at that time was negative(!)
  - Summer 2016: female to male transmission

Update: Interim Guidance for Prevention of Sexual Transmission of Zika Virus — United States, 2016

Weekly / April 1, 2016 / 65(12):323–325

Mosquito Lesson: Disease Ecology

ZIKA: CONFIRMED LINK TO MICROCEPHALY

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ORIGINAL ARTICLE

BRIEF REPORT

Zika Virus Associated with Microcephaly

Jernej Mlakar, M.D., Misa Korva, Ph.D., Nataša Tul, M.D., Ph.D., Mara Popović, M.D., Ph.D., Mateja Poljšak-Prijatelj, Ph.D., Jerica Mraz, M.Sc., Marko Kolenc, M.Sc., Katarina Resman Rus, M.Sc., Tina Vesnaver Vipotnik, M.D., Vesna Fabjan Vodušek, M.D., Alenka Vizjak, Ph.D., Jože Pižem, M.D., Ph.D., Miroslav Petrovec, M.D., Ph.D., and Tatjana Avšič Županc, Ph.D.

N Engl J Med 2016; 374:951-958 | March 10, 2016 | DOI: 10.1056/NEJMoa1600651

1st Trimester = miscarriage

2nd Trimester = microcephaly

3rd Trimester = stillbirth

Virus has an affinity to developing cells 7) urine 8) saliva 9) tears

Next Generation Mosquito Management

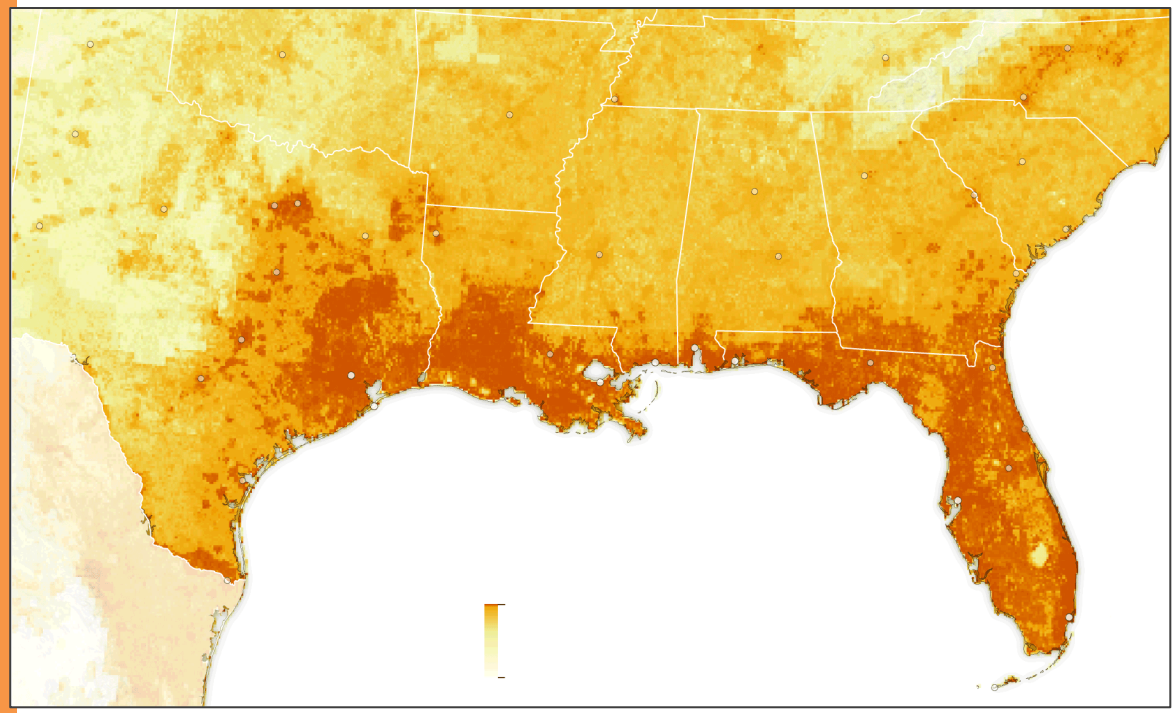
- Community engagement
- Abuzz app detects mosquitoes via wingbeat frequency

- *Wolbachia*-infected mosquitoes
  - To displace; affects mating (Mosquito Mate) https://mosquitomate.com/?v=3.0
  - To replace; affects pathogen (O'Neill Lab) https://research.monash.edu/en/persons/scott-oneill
- "GMO" mosquito
  - Oxitec self-limiting mosquitoes

  http://www.oxitec.com/our-solution/technology/ *

  https://www.youtube.com/watch?v=BTYelY7aqNY
  - CRISPR/CAS9 - gene drive - https://www.nature.com/articles/s41598-017-02744-7
  - RNAi – gene slicing https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5371932/

MOSQUITO-BORNE PATHOGENS *WILL LIKELY* CONTINUE TO BE A DISEASE OF POVERTY



Remove containers holding water

Wear repellents

Fix window screens