

Next generation sequencing and the 2014 Ebola outbreak

Ellie Thomas and Salvador Carranza

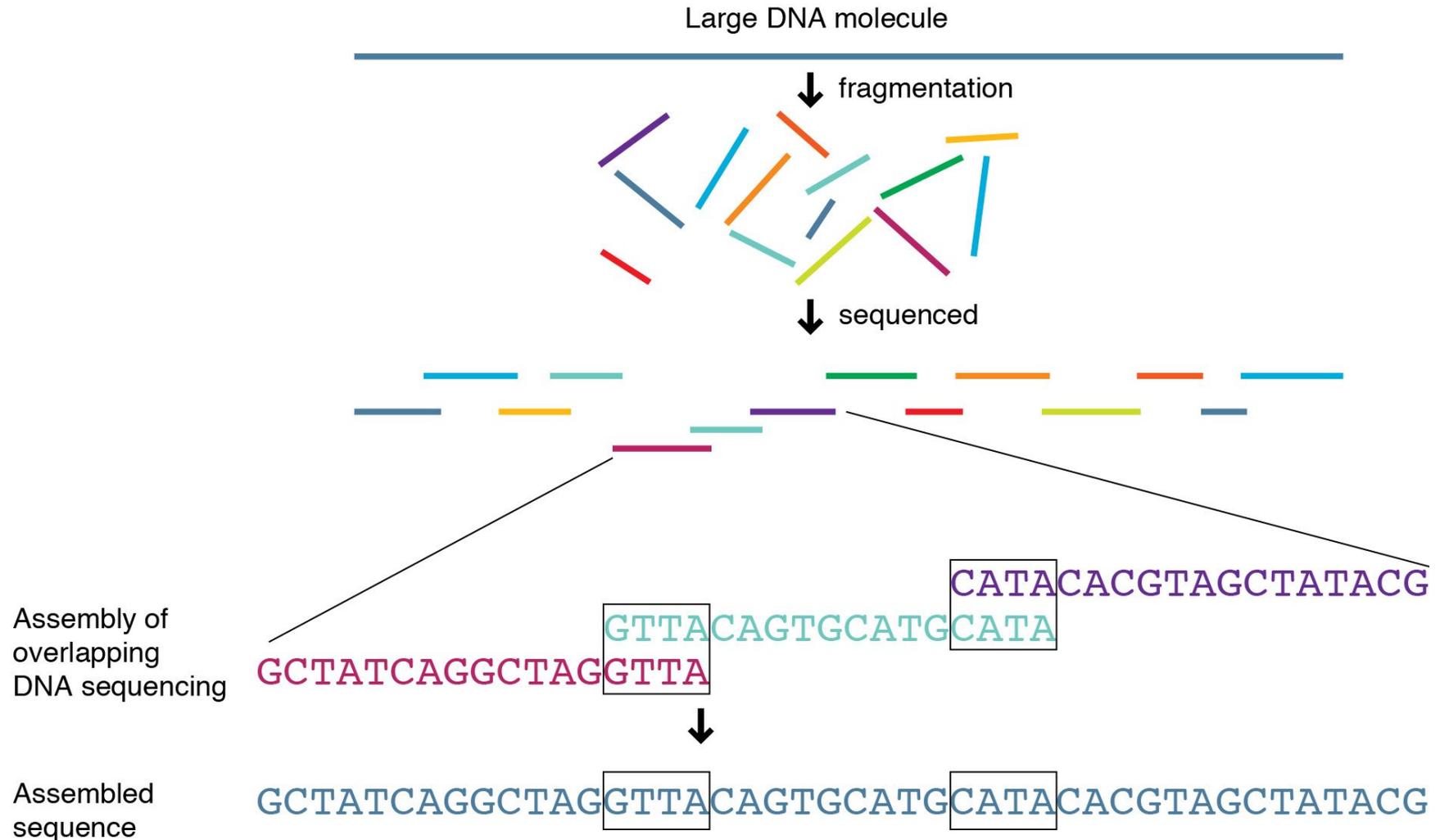
Ebola and Next Generation Sequencing

Next Generation Sequencing (NGS) methods

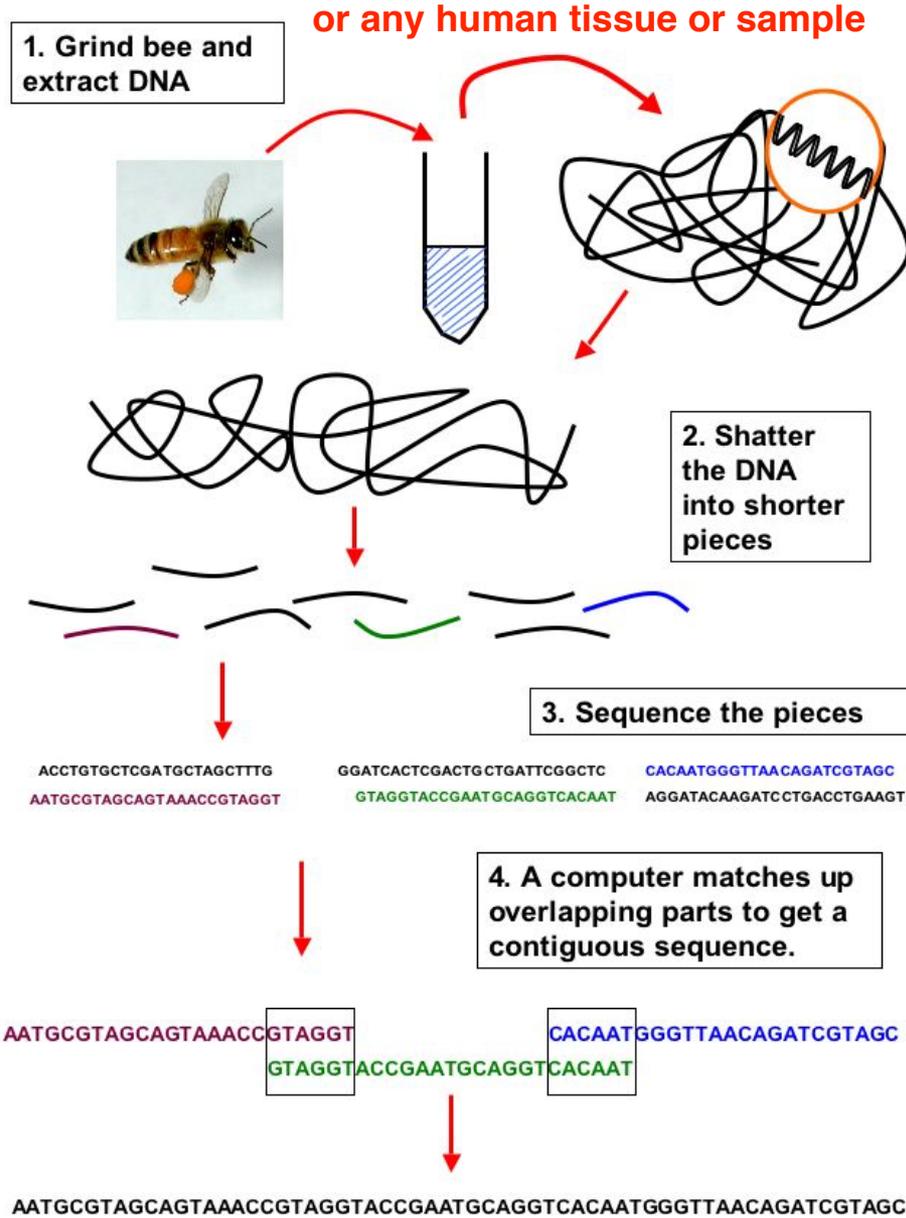
Ebola background

Paper: Genomic surveillance elucidates Ebola virus origin and transmission during the 2014 outbreak

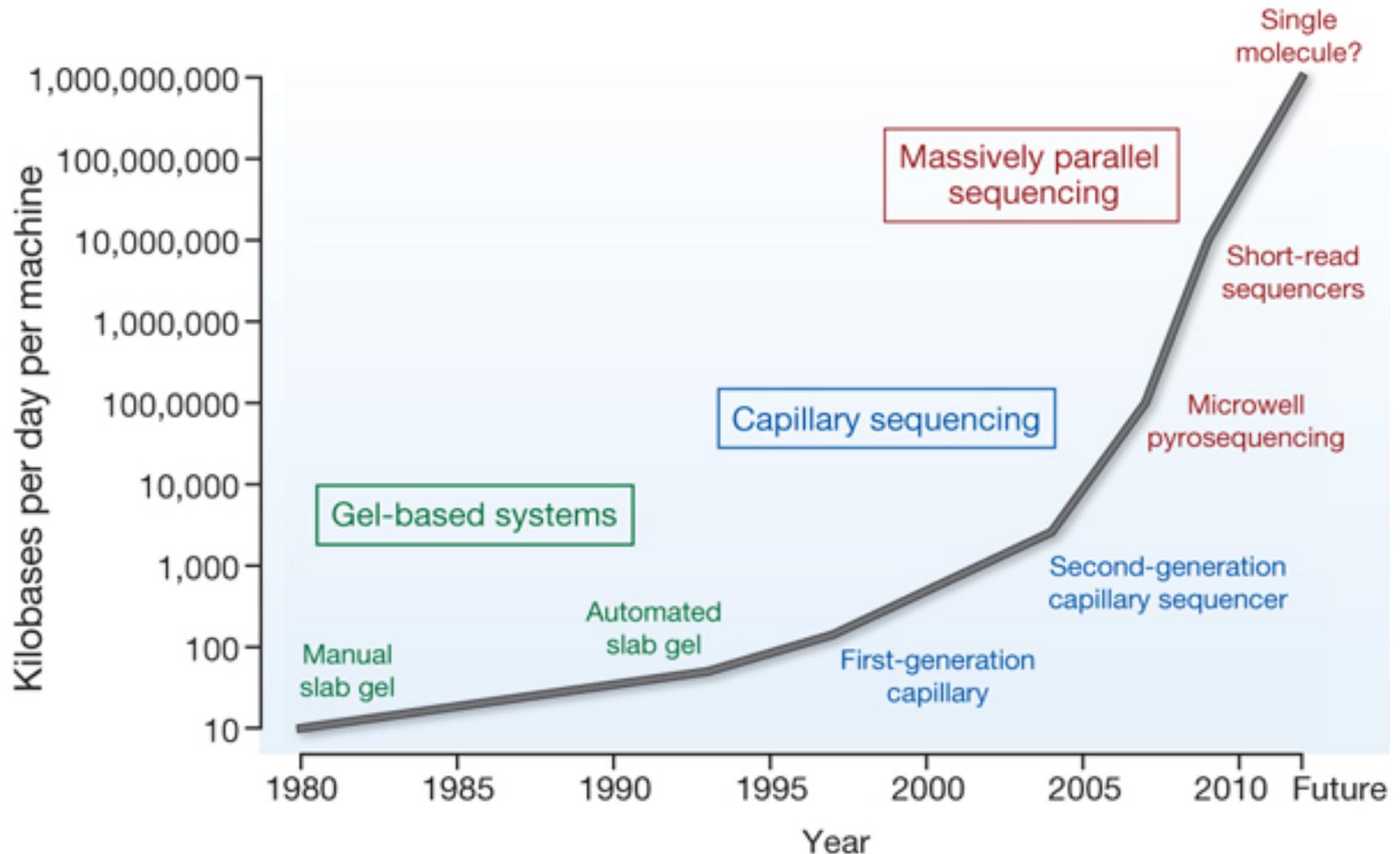
What is genomic sequencing?



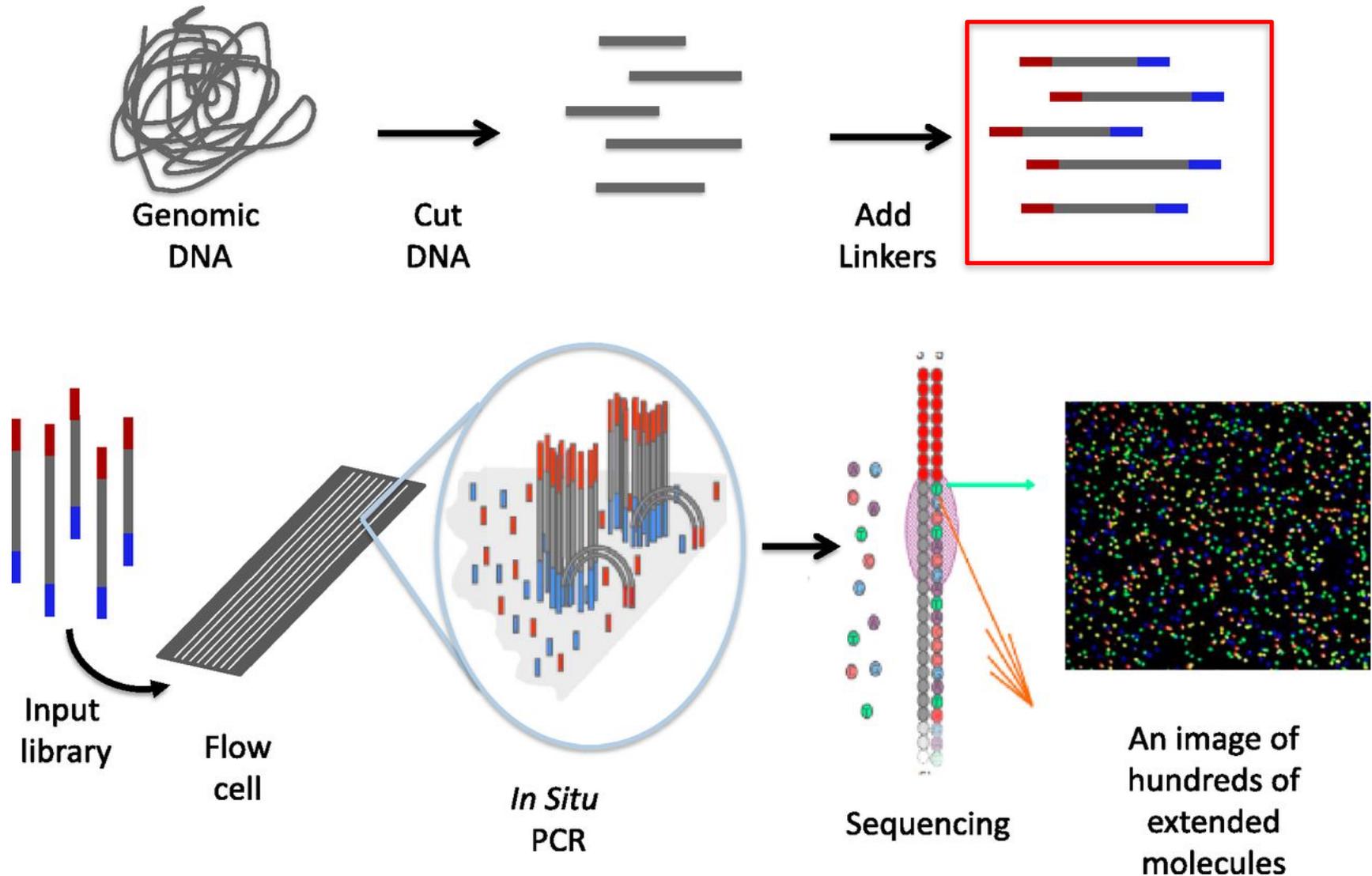
Genomic Sequencing example



Improvements in genome sequencing



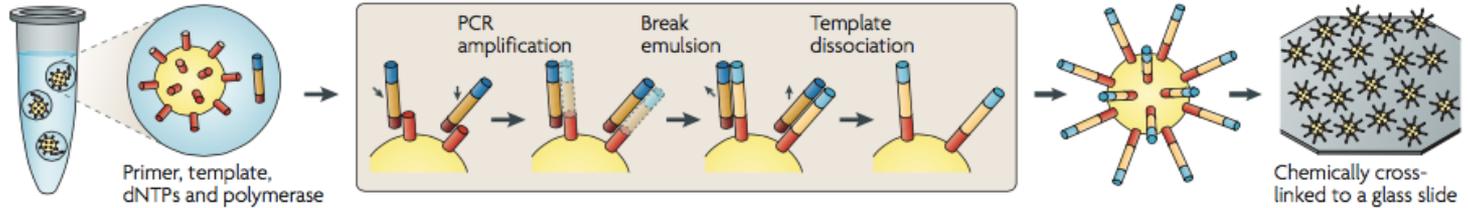
What is the workflow for Next Generation Sequencing (NGS)?



How are the adapters made?

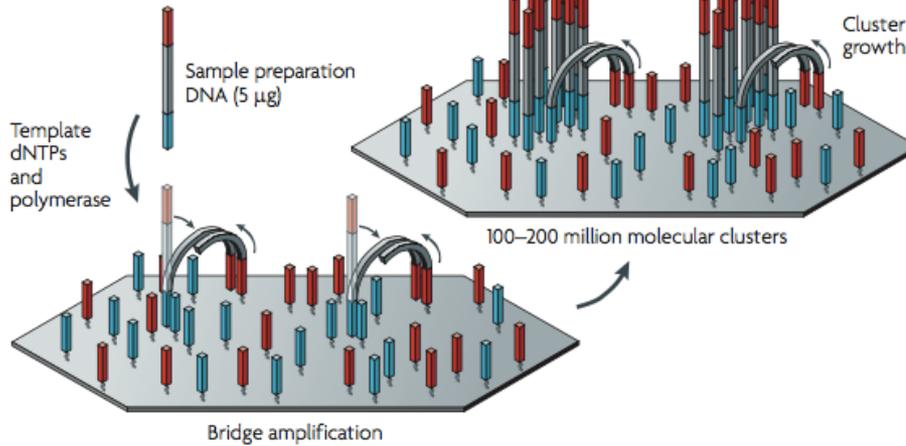
a Roche/454, Life/APG, Polonator Emulsion PCR

One DNA molecule per bead. Clonal amplification to thousands of copies occurs in microreactors in an emulsion



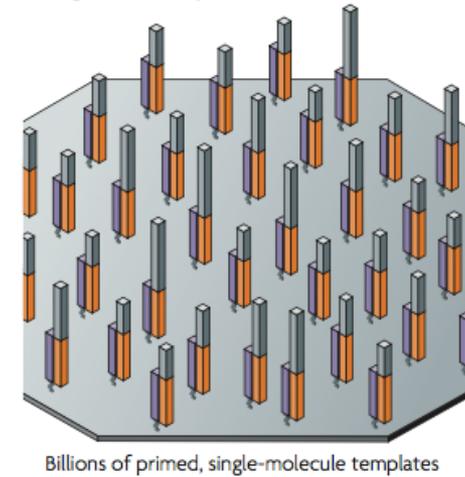
b Illumina/Solexa Solid-phase amplification

One DNA molecule per cluster



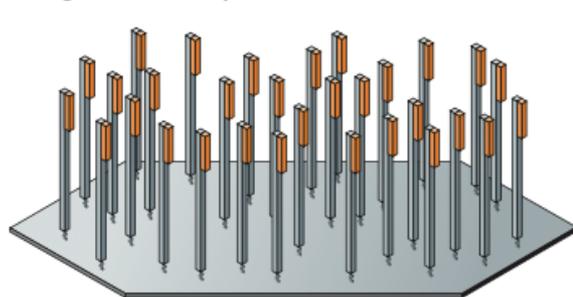
c Helicos BioSciences: one-pass sequencing

Single molecule: primer immobilized



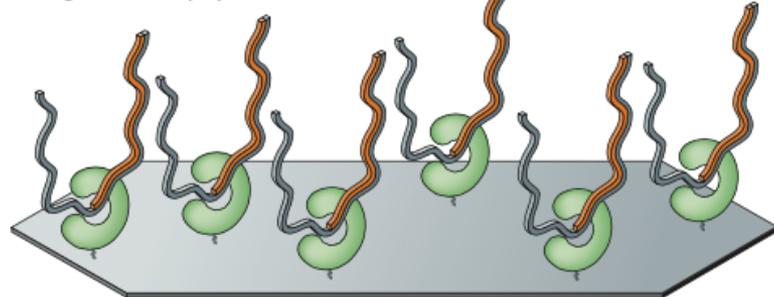
d Helicos BioSciences: two-pass sequencing

Single molecule: template immobilized



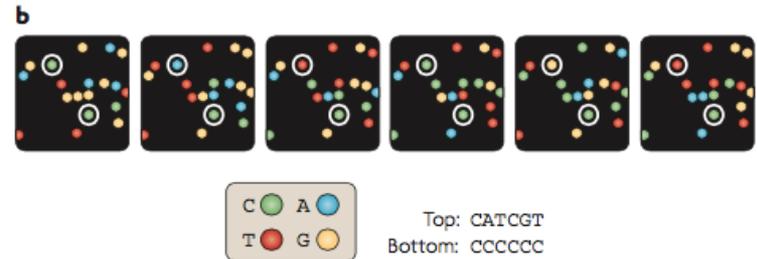
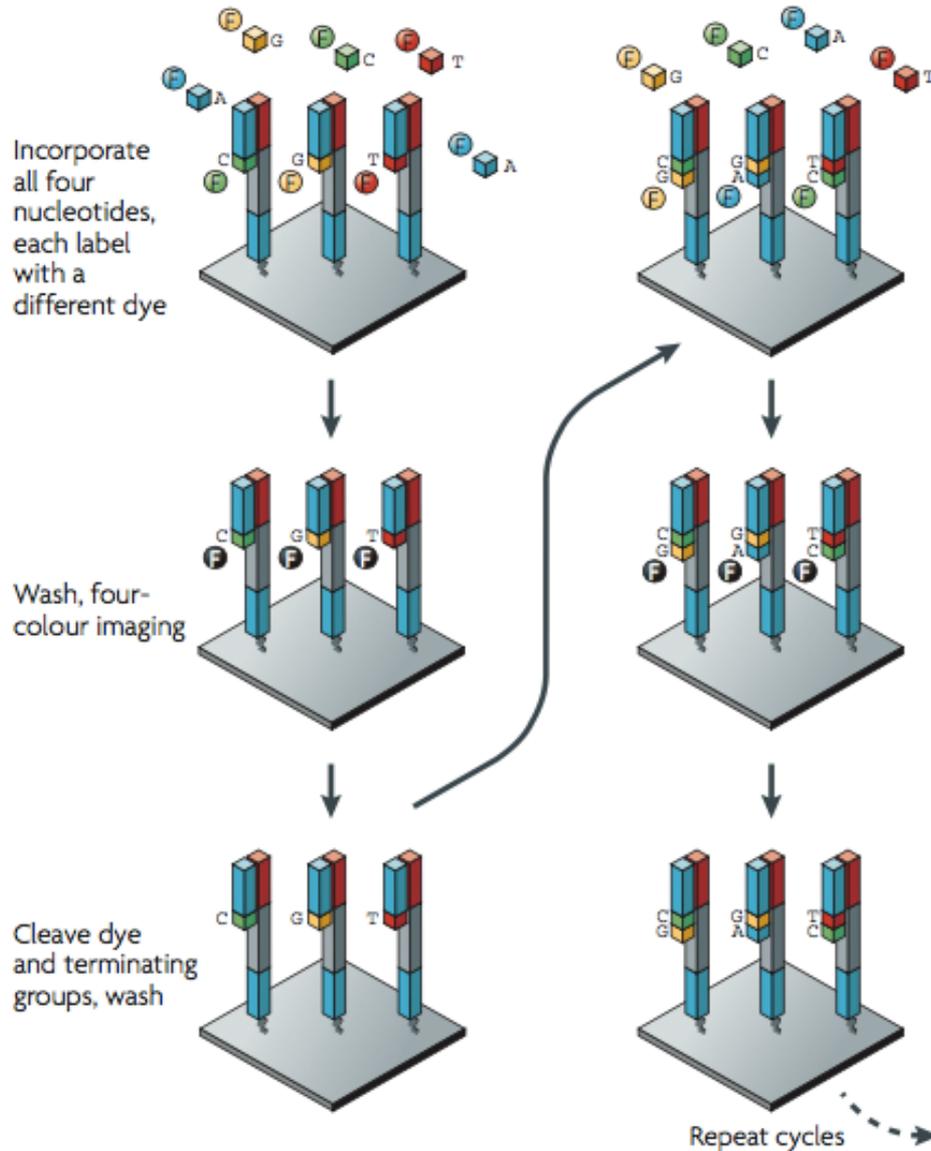
e Pacific Biosciences, Life/Visigen, LI-COR Biosciences

Single molecule: polymerase immobilized



How do you visualize base pairs?

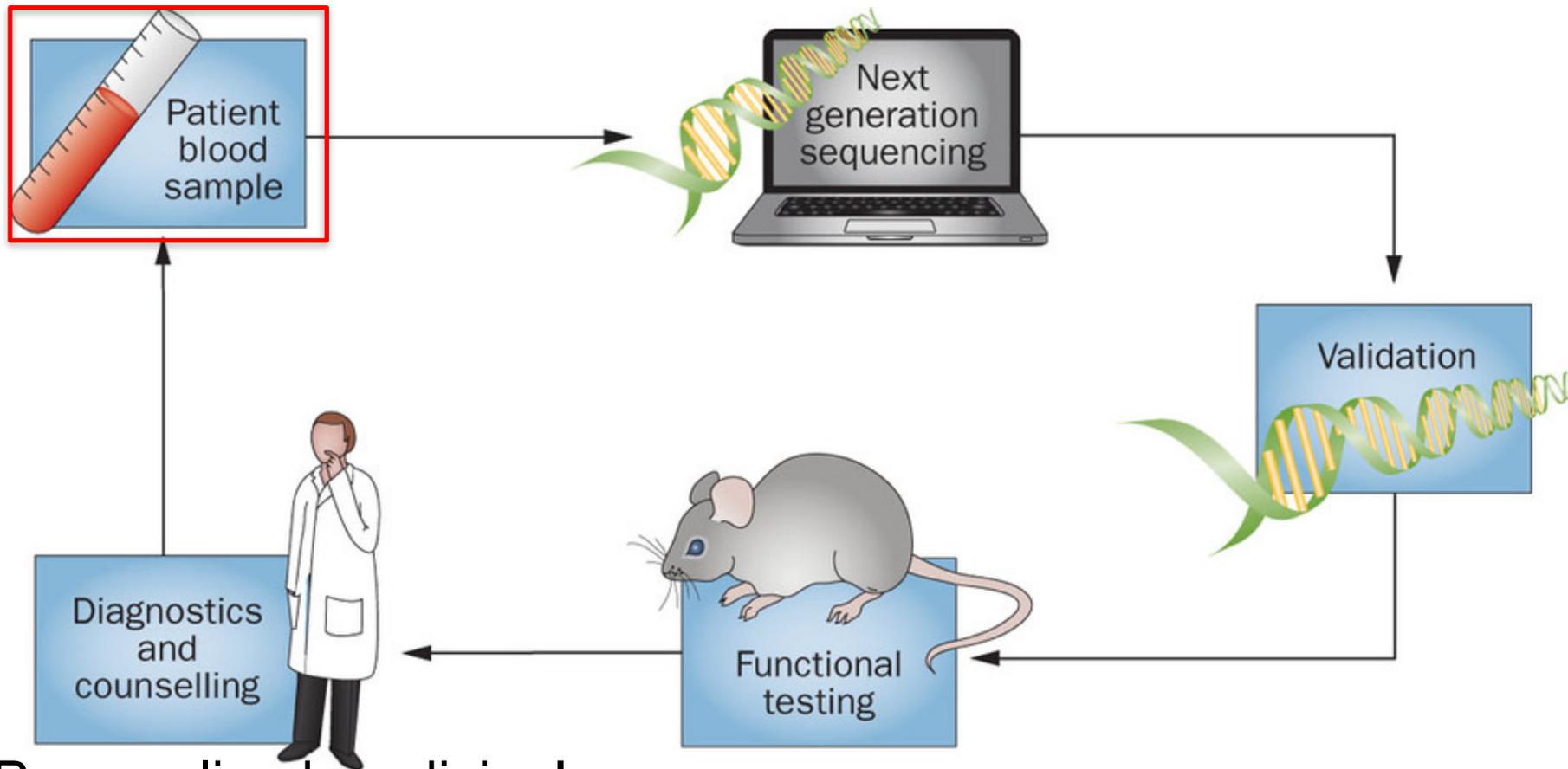
a Illumina/Solexa — Reversible terminators



<https://www.illumina.com>

What can you do with genomic sequencing?

Personalized genomics

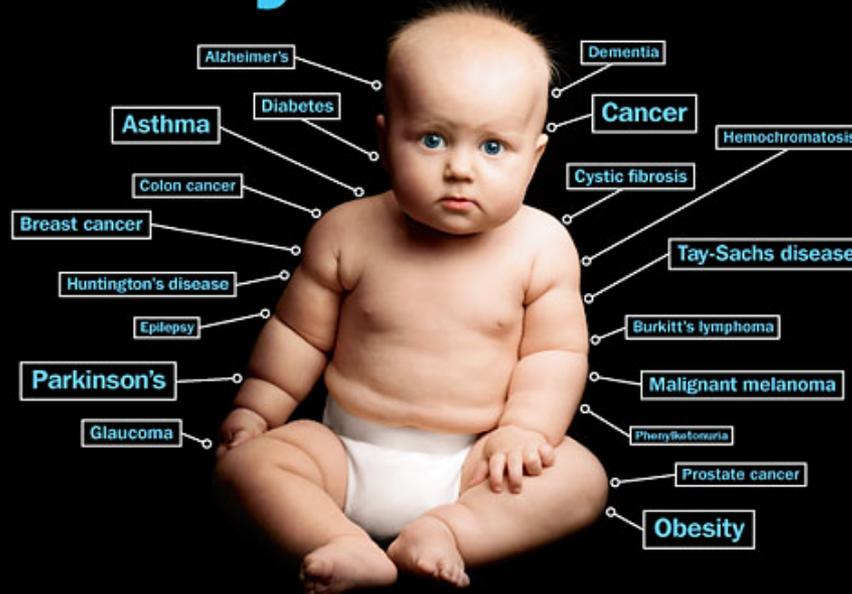


Personalized medicine!

Egypt Divided / Pot's Big Moment / Best of 2012 Movies, Music, Books & More

TIME

Want to Know My Future?



New genetic tests can point to risks—
but not always a cure

BY BONNIE ROCHMAN

Ebola and Next Generation Sequencing

Next Generation Sequencing (NGS) methods

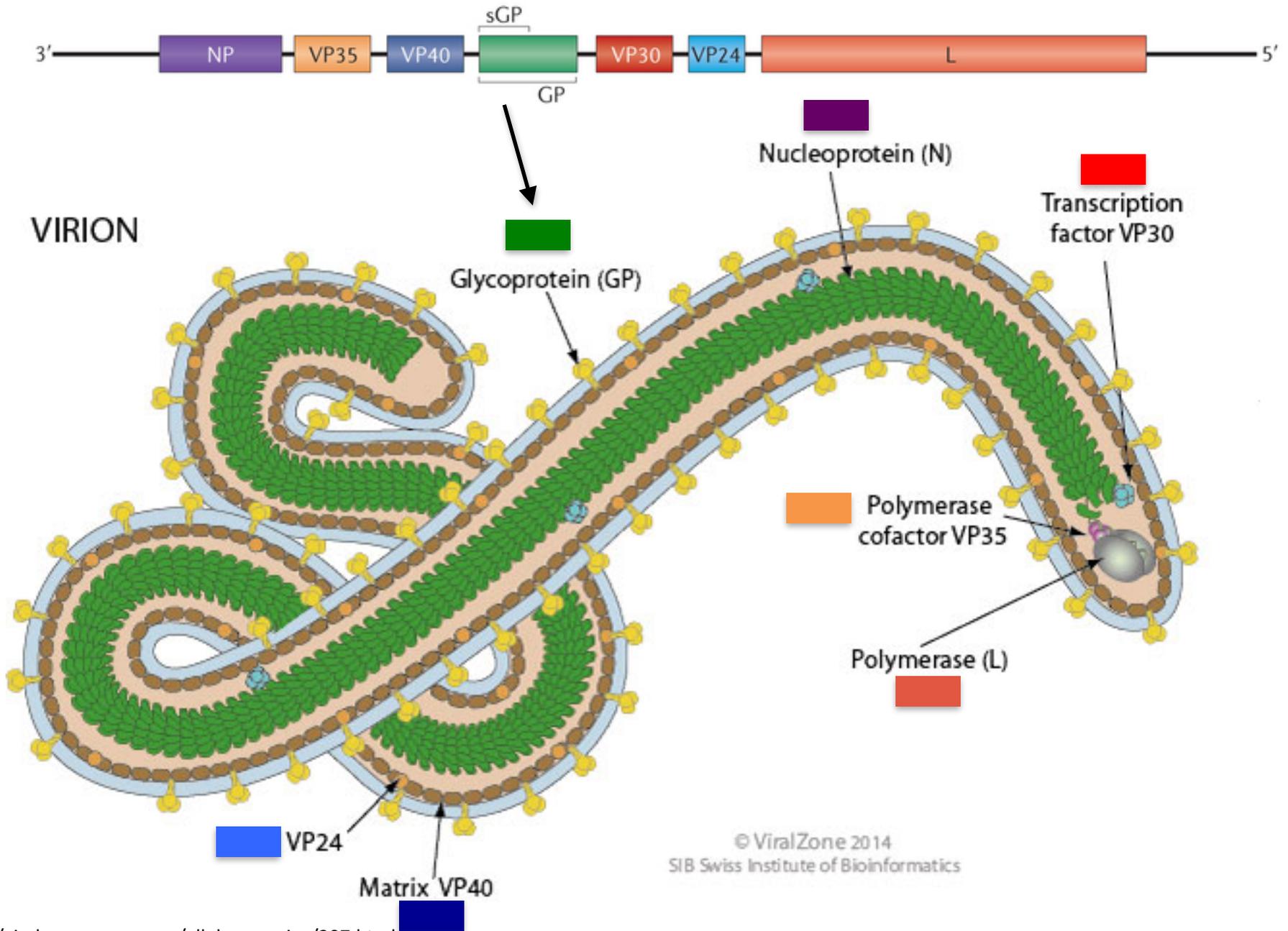
Ebola background

Paper: Genomic surveillance elucidates Ebola virus origin and transmission during the 2014 outbreak

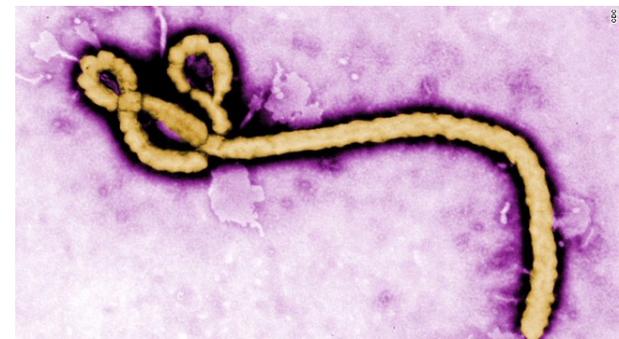
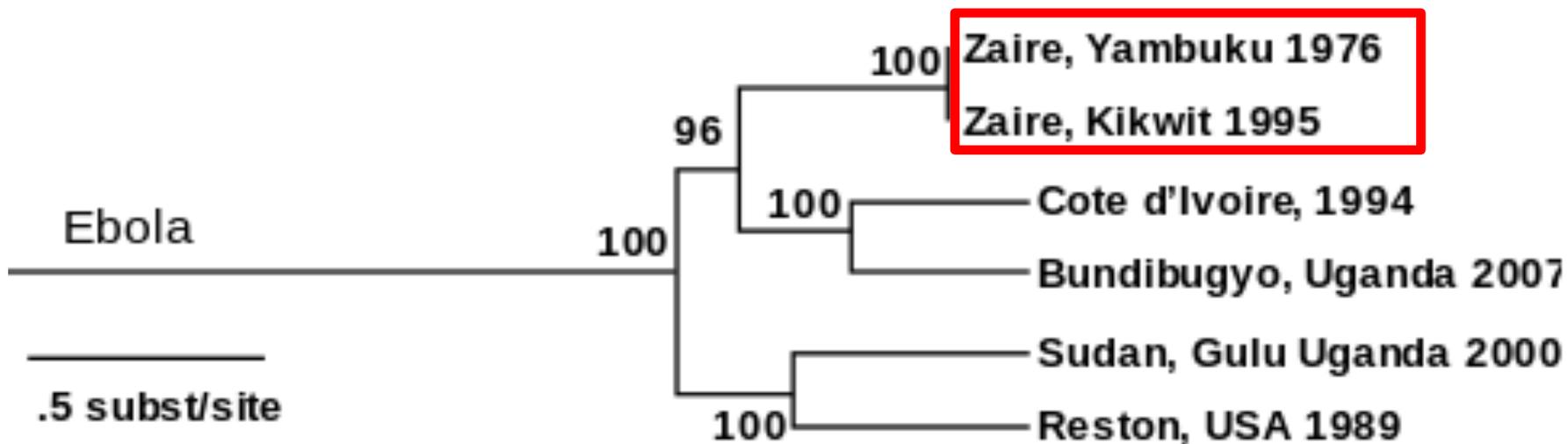
What is Ebola?



The ebola genome and its structure



Where did the Ebola (Zaire) strain originate?



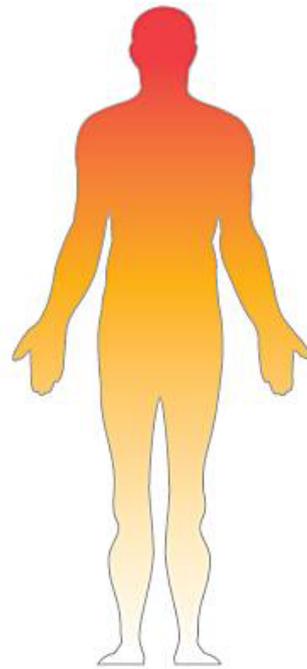
What does an Ebola infection look like?



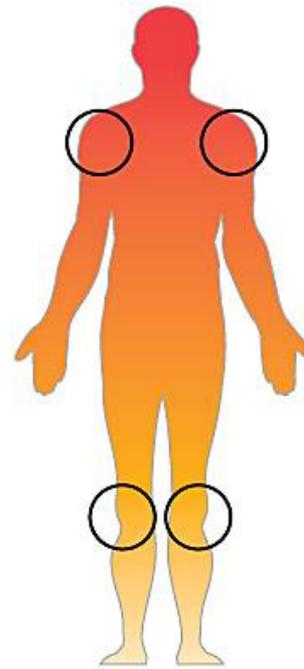
Virus's typical path through a human



Day 7-9
Headache,
fatigue, fever,
muscle
soreness



Day 10
Sudden high
fever, vomiting
blood, passive
behavior



Day 11
Bruising, brain
damage,
bleeding from
nose, mouth,
eyes, anus



Day 12
Loss of
consciousness,
seizures, massive
internal bleeding,
death

NOTE: Symptoms can start as early as two days after infection.

SOURCES: World Health Organization; BBC

Melina Yingling/McClatchy-Tribune

Stay Safe at School



If you are sick, call 117.



Tell your parent if you feel sick and need to stay home.
Staying home when you feel sick will protect you and your classmates.



Do not touch pee, poop, or vomit.
Tell your teacher if you see pee, poop or vomit on the floor.



Do not touch your friends or teachers.
Do not kiss, hug, or shake hands.



Stay at least 3 feet away.
Hold your arms out to the side and stay that far away from others.



Eat your own food and drink.
Do not share food, drinks, utensils, or cups. Use your own utensils and cups.



Play games, like singing songs and playing outside, without touching.
You can still play without touching your friends.



Remember to always wash your hands with soap and water.

**If you feel sick,
call 117
Together, we can get to zero!**

Ebola and Next Generation Sequencing

Next Generation Sequencing (NGS) methods

Ebola background

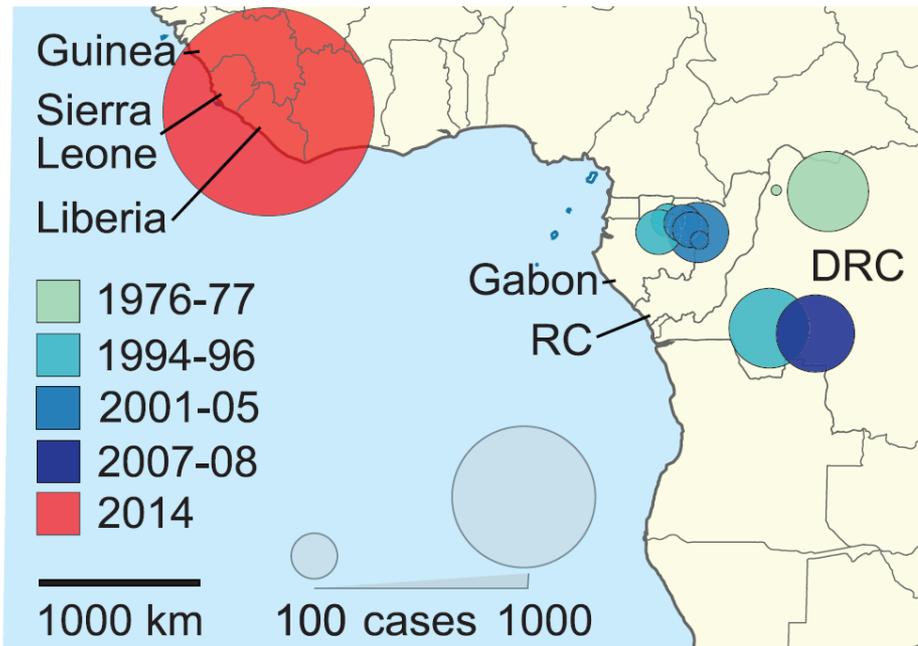
Paper: Genomic surveillance elucidates Ebola virus origin and transmission during the 2014 outbreak

Genomic surveillance elucidates Ebola virus origin and transmission during the 2014 outbreak

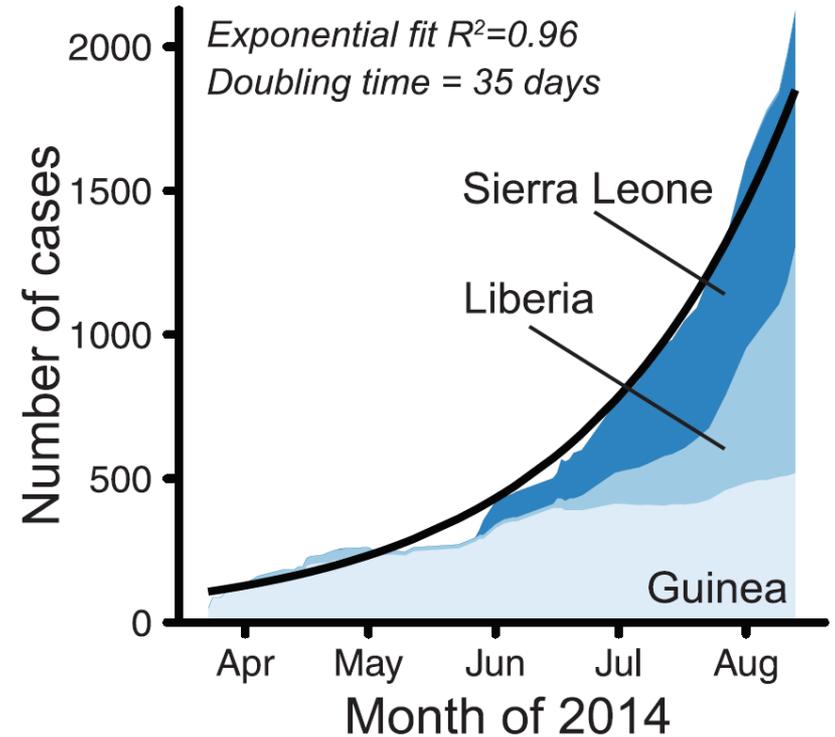
Stephen K. Gire,^{1,2*} Augustine Goba,^{3*†} Kristian G. Andersen,^{1,2*†} Rachel S. G. Sealfon,^{2,4*} Daniel J. Park,^{2*} Lansana Kanneh,³ Simbirie Jalloh,³ Mambu Momoh,^{3,5} Mohamed Fullah,^{3,5†} Gytis Dudas,⁶ Shirlee Wohl,^{1,2,7} Lina M. Moses,⁸ Nathan L. Yozwiak,^{1,2} Sarah Winnicki,^{1,2} Christian B. Matranga,² Christine M. Malboeuf,² James Qu,² Adrienne D. Gladden,² Stephen F. Schaffner,^{1,2} Xiao Yang,² Pan-Pan Jiang,^{1,2} Mahan Nekoui,^{1,2} Andres Colubri,¹ Moinya Ruth Coomber,³ Mbalu Fonnier,^{3†} Alex Moigboi,^{3†} Michael Gbokie,³ Fatima K. Kamara,³ Veronica Tucker,³ Edwin Konuwa,³ Sidiki Saffa,^{3†} Josephine Sellu,³ Abdul Azziz Jalloh,³ Alice Kovoma,^{3†} James Koninga,³ Ibrahim Mustapha,³ Kandeh Kargbo,³ Momoh Foday,³ Mohamed Yillah,³ Franklyn Kanneh,³ Willie Robert,³ James L. B. Massally,³ Sinéad B. Chapman,² James Bochicchio,² Cheryl Murphy,² Chad Nusbaum,² Sarah Young,² Bruce W. Birren,² Donald S. Grant,³ John S. Scheffelin,⁸ Eric S. Lander,^{2,7,9} Christian Happi,¹⁰ Sahr M. Gevao,¹¹ Andreas Gnirke,^{2§} Andrew Rambaut,^{6,12,13§} Robert F. Garry,^{8§} Humarr Khan,^{3†§} Pardis C. Sabeti^{1,2†§}

Fig1: Ebola outbreaks, historical and current

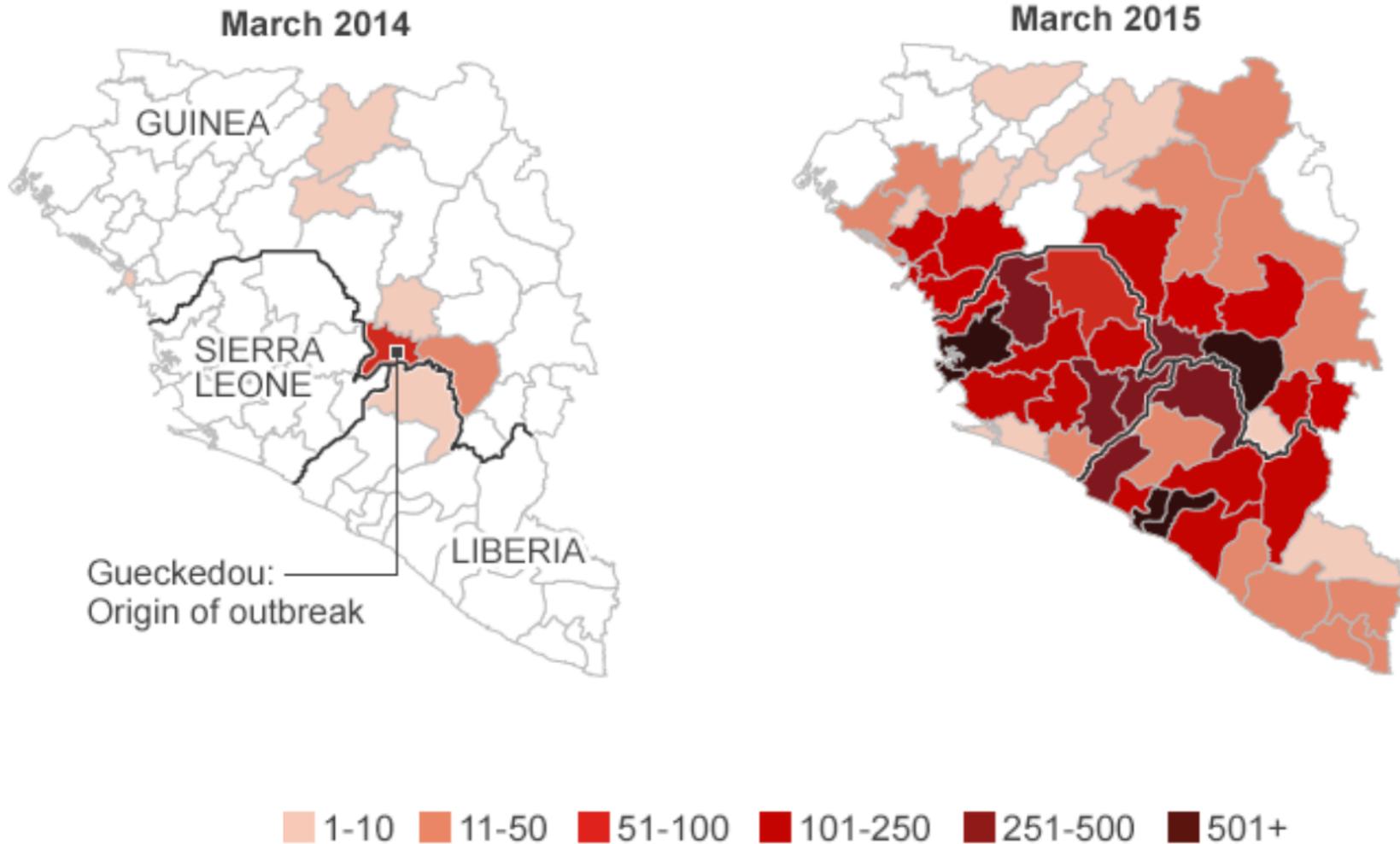
A



B



What was the severity?



TRANSMISSION

In Africa a particular species of **fruit bat** is considered to be the possible host for Ebola virus

Primary host Fruit bat



Secondary hosts

Gorillas



Chimpanzees



Antelopes



Pigs



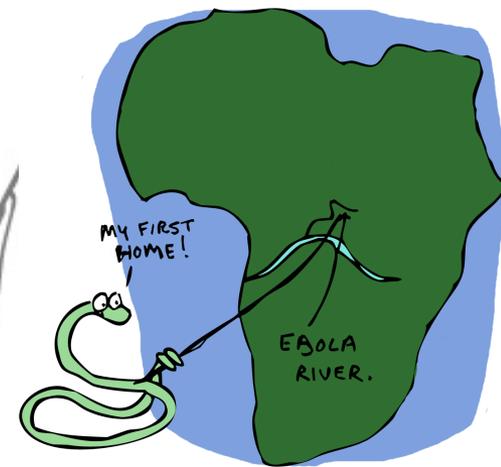
1

Humans are infected by close contact with the **blood, secretions, organs or other bodily fluids** of infected animals

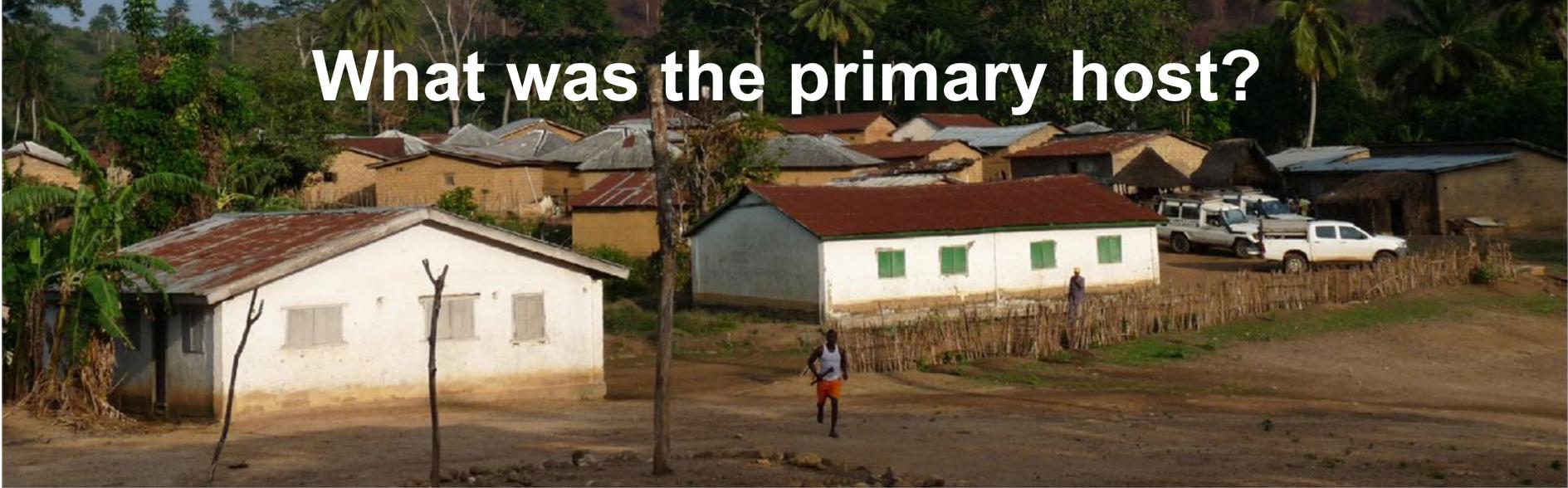


2

Ebola then spreads through human-to-human transmission, with infection resulting from direct contact **through broken skin or mucus or other bodily fluids** of infected people



What was the primary host?



Who was the first victim?

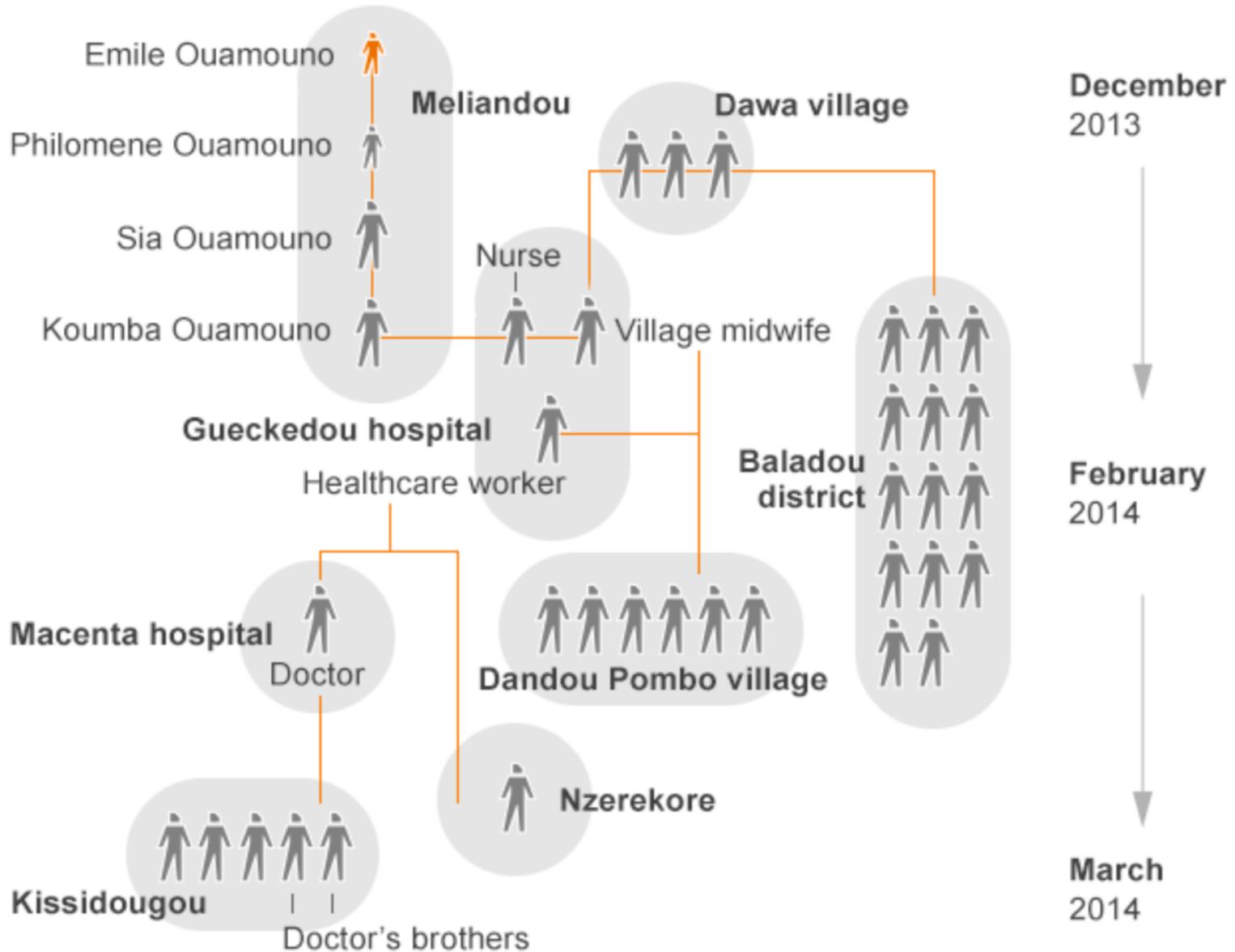


Fig1d: How did they sequence Ebola genomes?

	# Total	# Patients	% Coverage	x Coverage
Batch 1	15	12	99.77%	555 [16-23,042]
Batch 2	84	66	99.91%	2,186 [6-18,623]
Combined	99	78	99.88%	2,048 [6-23,042]
Replication	140	78	99.39%	2,109 [8-17,537]

Fig S5: How did Ebola avoid detection?

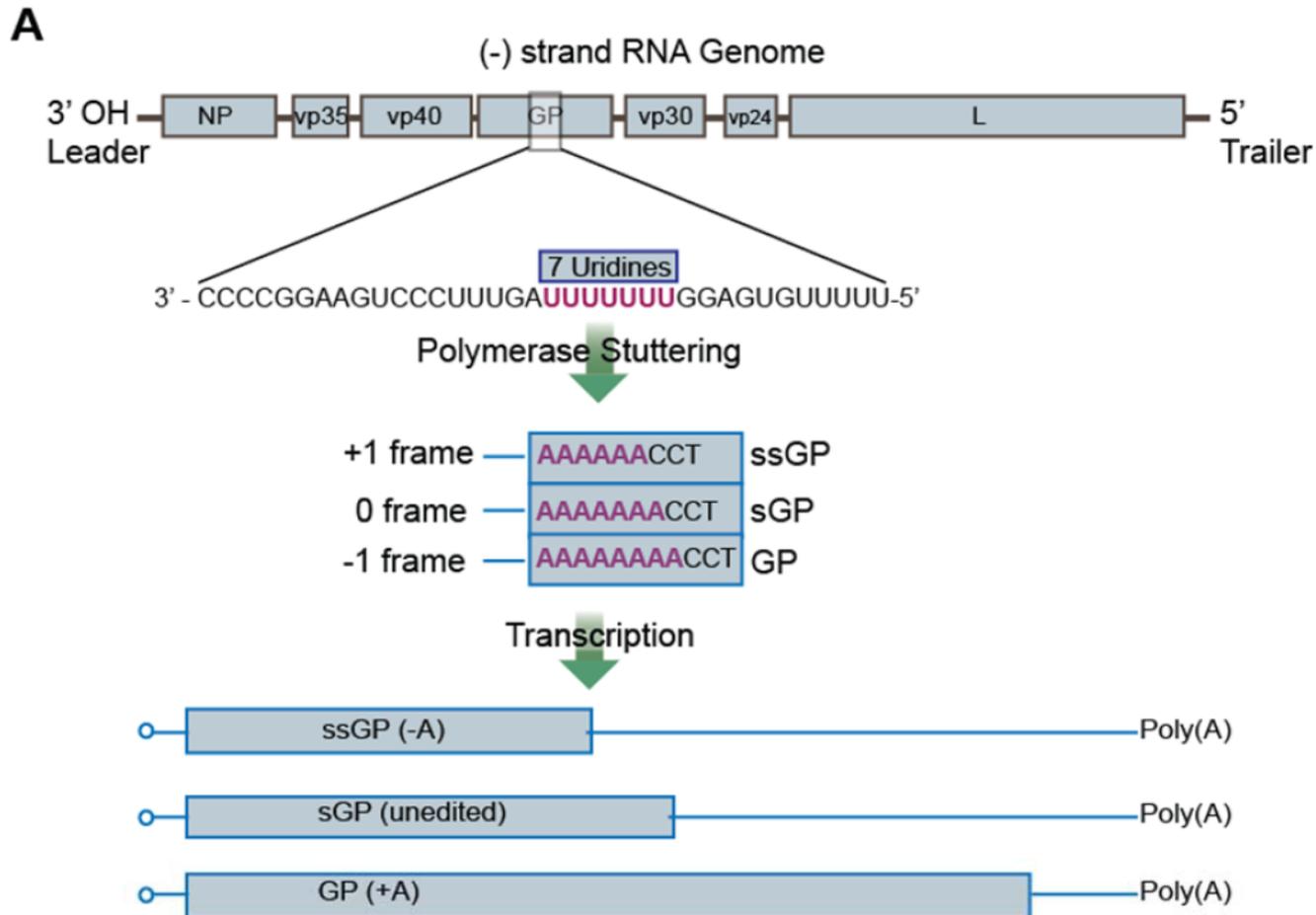


Fig2a/b: What are the relationships between outbreaks?

A

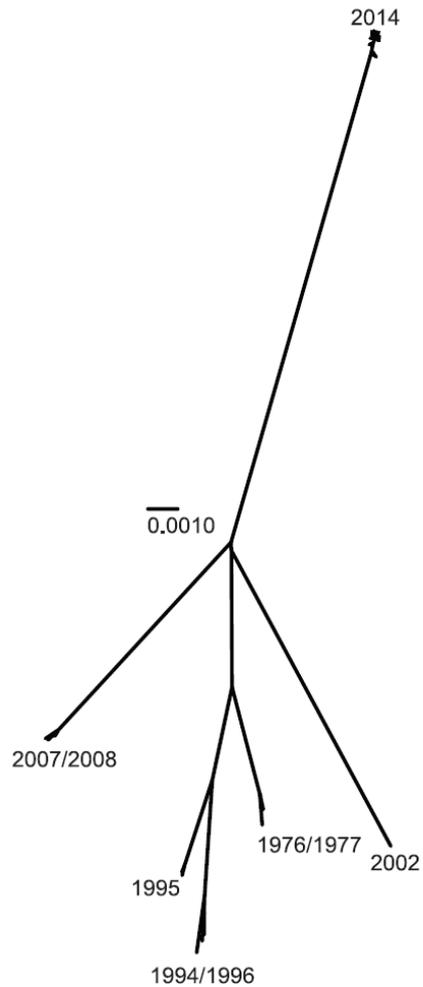


Fig4a: Which notable gene is affected?

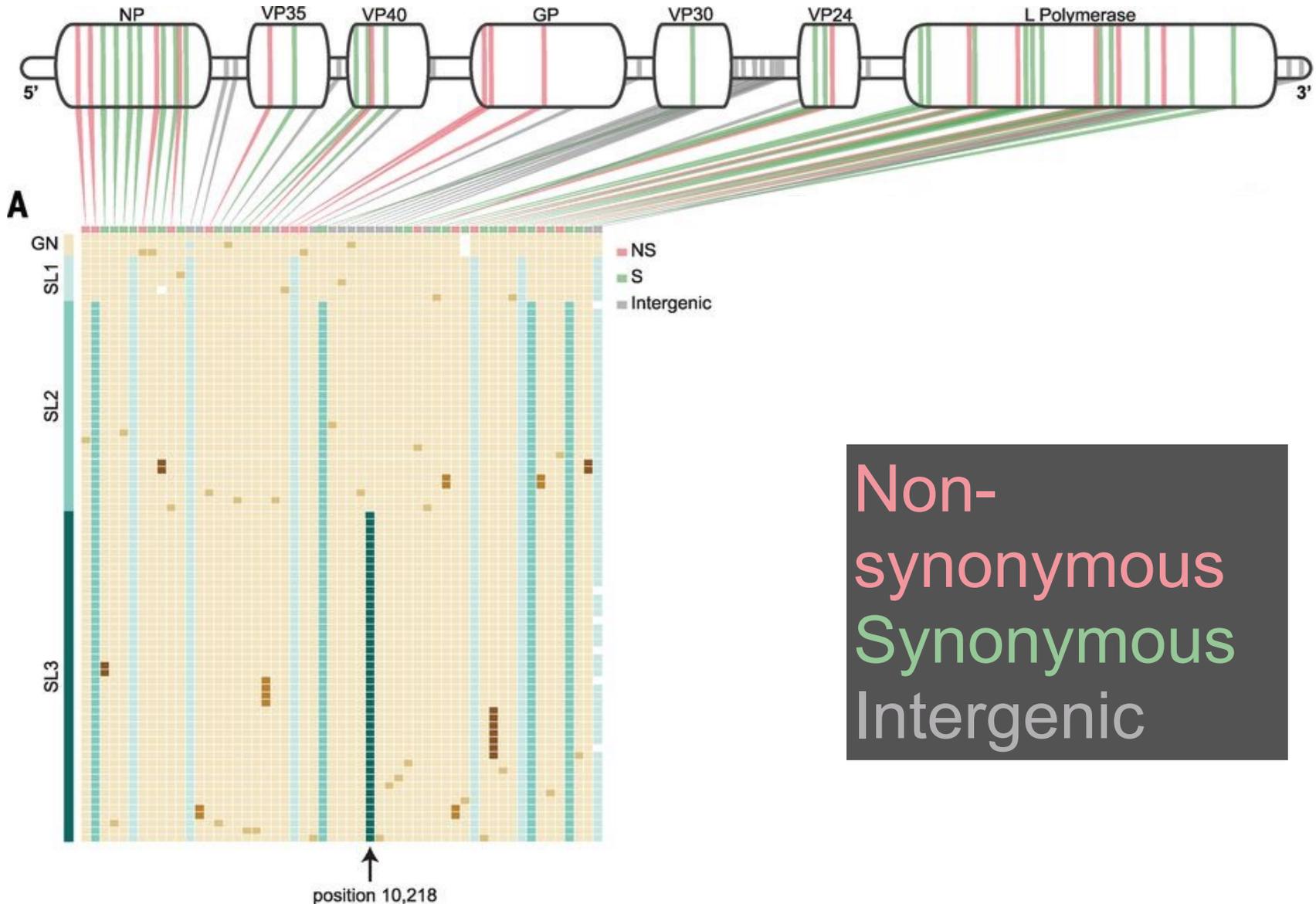


Fig 4b: Which clusters spread throughout Sierra Leone?

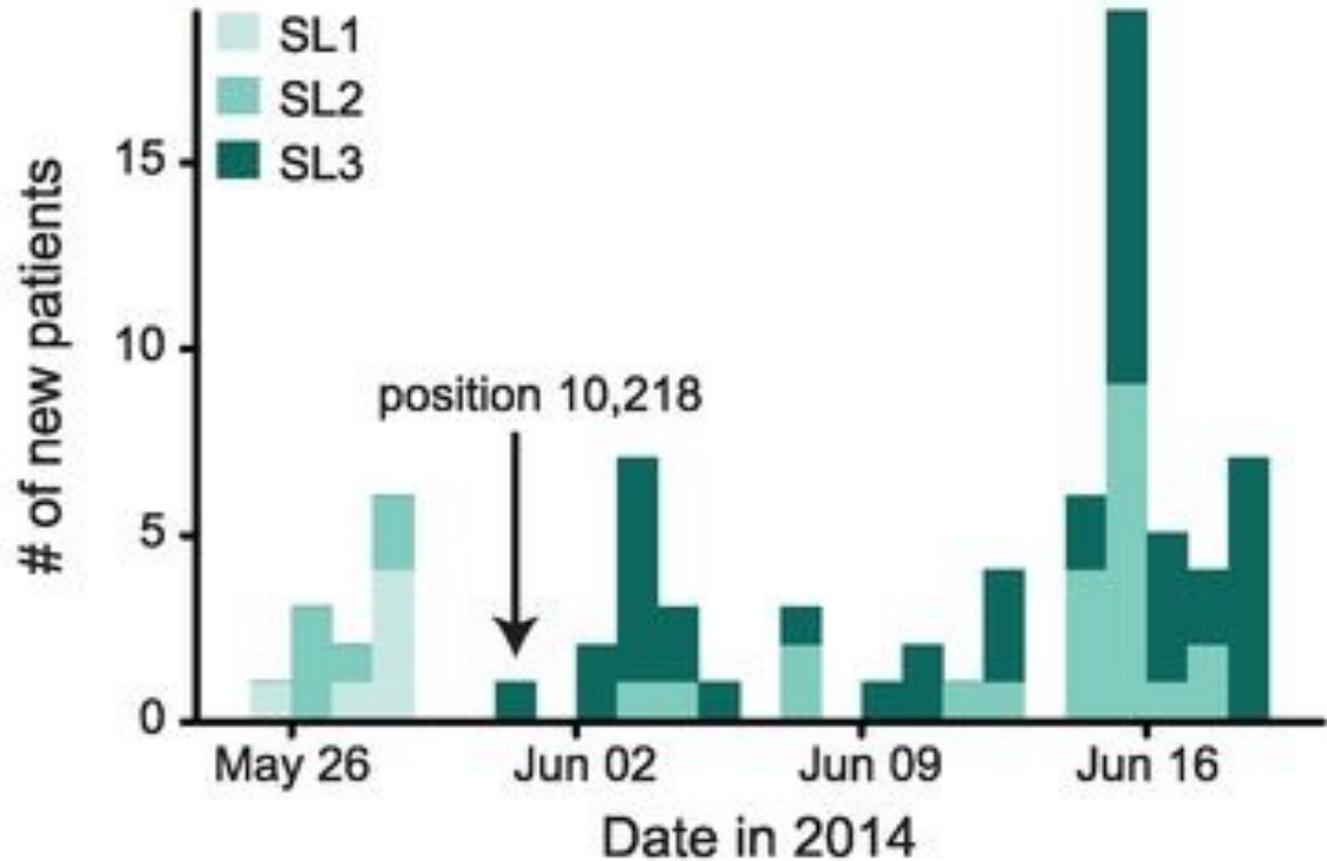
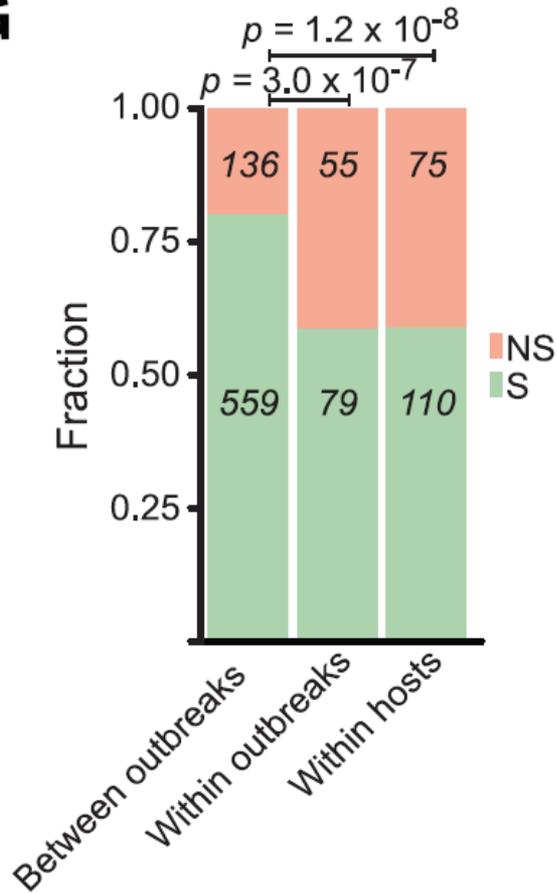
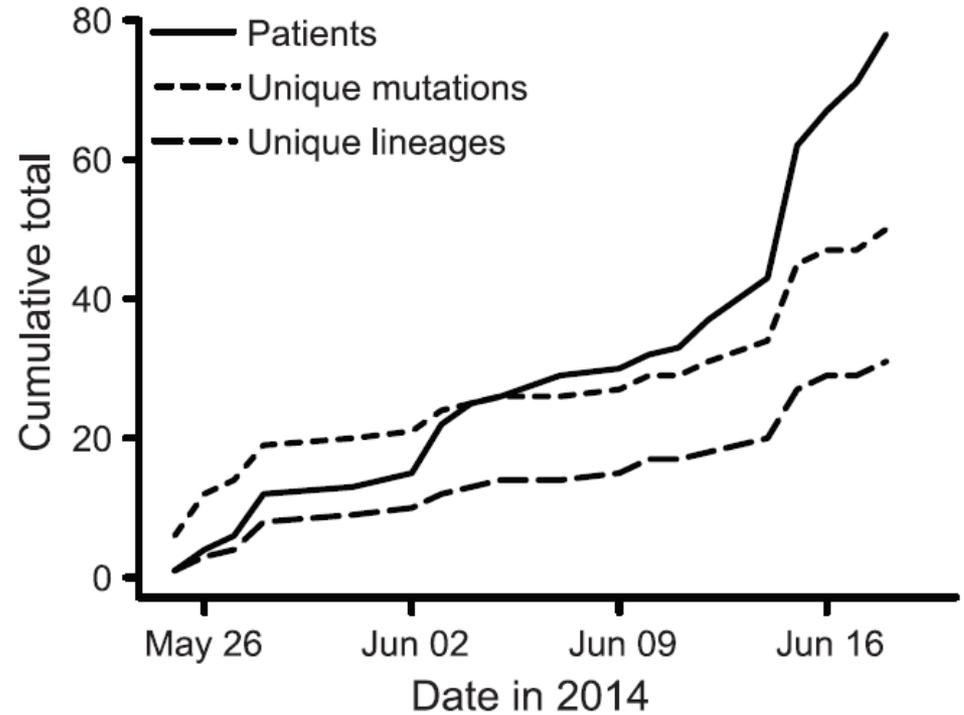


Fig4g/h: Why were mutation rates higher?

G



H





Summary

28,638 cases leading to 11,316 deaths

Achieved 99.88% sequence coverage

Genomic makeup between recent and historical outbreaks was determined

Sierra Leone outbreak stemmed from 2 genetically distinct lineages

Traced outbreak back to baby likely bit by natural reservoir



time for **questions**