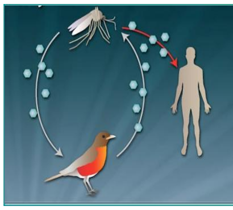


From Birds to People: The West Nile Virus Story

hhmi | BioInteractive

Click & Learn Transcript

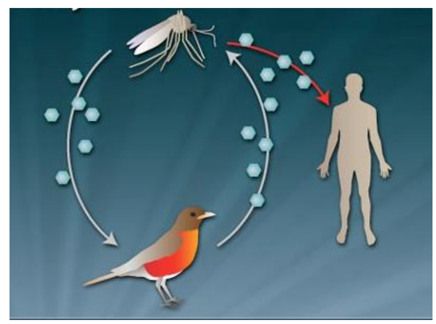
OVERVIEW

This document is a descriptive transcript of the Click & Learn From Birds to People: The West Nile Virus Story, which explains how the West Nile virus infects mosquitoes, birds, and people—with very different consequences. In addition to the written content on each slide, there are illustrations, maps, photographs and videos of Drs. Laura Kramer and Marm Kilpatrick, who discuss their research on the virus life cycle and how factors like temperature and climate affect the spread of the virus.

SLIDES

Slide 1: Learning Objectives

- Think about how infectious diseases spread, and how they can be contained.
- Learn about the many factors that affect the spread of a mosquito-borne infectious disease.
- Understand the difference between the infectious agent, vector and host, and that some hosts play a role in spreading a virus and others don't.
- Learn about what makes a vector effective in spreading an infectious disease.
- Understand that viral infection can have different effects in different hosts and in different individuals.
- Gain an appreciation for the range of studies needed to understand mosquito-borne diseases.



[Image: Illustration depicting the cyclical transmission process between mosquito, bird and back to mosquito, maintaining the infection. The illustration also shows a human to the right side of the cyclical image, depicting the one-way transmission of the virus from mosquito to human.]

Slide 2: Why study West Nile Virus?

West Nile virus (WNV) has harmed bird populations and ecosystems, and caused thousands of cases of serious illness in people. Studying how the virus replicates and spreads can help researchers find strategies to combat it. In addition, studying WNV can help answer important general questions about where infectious diseases come from, how they spread, and why they seem to come and go depending on the season or year.

Slide 3: WNV Has Different Effects on Different Species

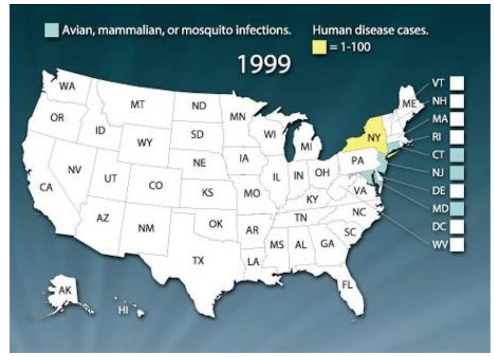
West Nile virus infects mosquitoes, birds, and people—with very different consequences: Mosquitoes are largely unaffected. Some birds get very sick and die. People can get sick, but symptoms are usually mild.



[Three images (left to right): mosquito sitting on human skin, a dead crow laying on the ground, and a doctor listening to a patient with a stethoscope]

Slide 4: WNV Has Harmed Bird Populations

West Nile virus first appeared in the US in New York City in the summer of 1999.
That year, nearly 5,500 crows died in 4 months!



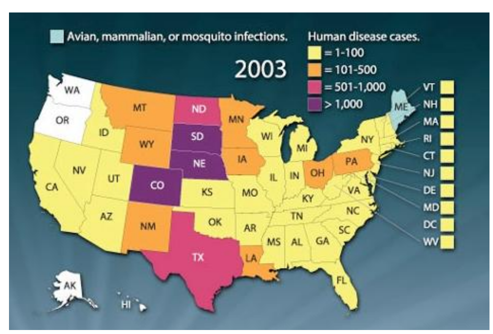
[Image: Map of the United States with the date 1999 above it. A legend at the top of the map explains color coding for the States: New York is colored in yellow to illustrate human disease cases, ranging from 1-100, caused by WNV infection. Connecticut, New Jersey, and Maryland are colored in blue to illustrate avian, mammalian, or mosquito infections, but no human cases. All other states have no color, indicating no infections or disease cases.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Laura Kramer explaining the devastation of the crow population. The video shows Dr. Kramer sitting in an office speaking and footage of various birds. Video length is 1 min. 24 sec. The following is a transcript of the video:]

> "One of the unique things about West Nile is that it was noted when West Nile first came that birds were dying. And people didn't relate the birds dying to people getting sick at first until some very astute people recognized that this was an unusual situation, and identified the virus as West Nile. West Nile is unique in the New World in that it does kill birds. It's been most noticed in the American Crows, which 95% of the infected birds die. Now, it might be different now, and nobody has looked that carefully, done studies, to see if they've become more resistant. But people would pick up dead crows because they are big, they're noticeable, and they would submit them to the labs and, most often, they would be positive. Other birds die, too. They're not as easily seen. So if you have like a little chickadee that dies, you're not going to see it in the same way that you would see, you know, a big black crow. And it's not just American Crows, but bluebirds and a number of other birds; their populations have been very seriously hurt by West Nile coming. The question is will this change over time? Are the birds becoming more resistant?"

Slide 5: WNV Has Spread Rapidly

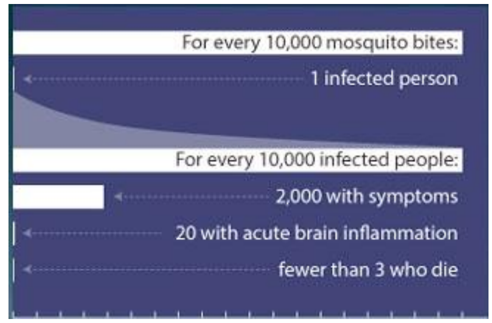
Since it was first detected in 1999, the virus spread rapidly throughout the US and peaked in 2002-2003.
In which parts of the US did most cases of human disease occur?



[Image: Map of the United States with the date 1999 above it. A legend at the top of the map explains the color coding for the States of the number of human disease cases: yellow equals a range of 1-100 cases, orange equals 101-500 cases, pink equals 501-1,000 cases and purple equals over 1,000 cases. Most States are colored yellow, with the exception of eight States that are colored orange (Montana, Wyoming, New Mexico, Minnesota, Iowa, Louisiana, Ohio and Pennsylvania), two States that are colored pink (North Dakota and Texas), and three States that are colored purpose (Colorado, Nebraska, and South Dakota). Maine is the only blue state, which indicates reported incidents of avian, mammalian, or mosquito infections, but no human disease cases. Washington, Alaska and Hawaii have no color, indicating no infections or disease cases.]

Slide 6: Human Symptoms of Infection Are Variable

Although the virus is widespread, symptoms in humans are typically mild. Some people, however, develop an inflammation of the brain. There is no vaccine for West Nile virus.



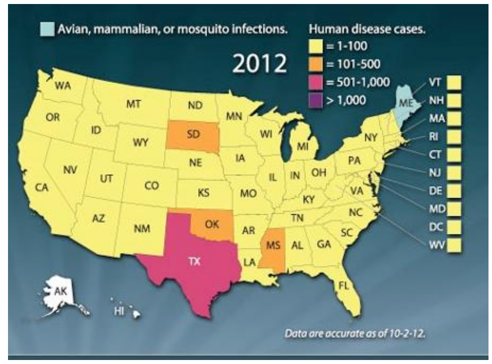
[Image: Chart showing that for every 10,000 mosquito bites, there is 1 infected person. For every 10,000 infected people, there are 2,000 with symptoms, 20 with acute brain inflammation, and fewer than 3 who die.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Marm Kilpatrick explaining the effects of the infectionin humans. The video shows Dr. Kilpatrick collecting data in the field, sitting outside on a bench speaking, pictures of the virus, maps of virus impact across the United States in different years, and footage of mosquitoes in traps. Video length is 1 min. 51 sec. The following is a transcript of the video:]

> “My name is Marm Kilpatrick, and I’m an Assistant Professor at the University of California Santa Cruz. The overall thrust of our research is really just to try to understand what are the factors that drive West Nile transmission.
>
> “West Nile virus is actually a virus that we believe came over from Africa in 1999; it was first detected in New York in 1999, and then subsequently spread across the entire U.S. in just about four or five years. And so, it made this amazingly rapid spread. It’s a virus that’s transmitted primarily between mosquitoes and birds. Occasionally, those mosquitoes will bite a bird and get infected, and then bite a human subsequently and infect the human, or a horse or other mammals like that. And so, that’s how people can get sick.
>
> “The number of cases varies enormously from year to year. We had some years where the number of reported cases was over 10,000, and then in other years there were numbers in the low thousands, with just one or two thousand cases. The virus doesn’t cause huge numbers of cases, but it can occasionally cause some fatalities, and it has, in fact, killed over 1,000 people in the U.S. I’ve calculated roughly the chance of getting West Nile virus from a single mosquito bite, and it’s probably on average between one in 10,000 and one in 100,000 mosquito bites. So, if you’re an outdoorsy kind of person, and you’re getting a large number of mosquito bites, then the chance of getting West Nile virus is much higher, whereas if you just get a few mosquito bites here and there, your chance of getting infected is relatively quite low.
>
> “About 80% of people that get infected with West Nile virus--nothing happens. You don’t even get sick. Your immune system is actually able to fight off the virus. In contrast, there is a small fraction of people that get infected that do, in fact, get sick, and then a much smaller fraction even of those people can go on to get serious symptoms. However, I should point out that a fraction of those people that do get serious illness frequently can have lifelong debilitating problems as a result of infection. And so, while the virus isn’t causing tens or hundreds of thousands of serious illnesses every year, it is causing hundreds to sometimes low numbers of thousands of permanently debilitating cases of encephalitis and things like that each year.”

Slide 7: 2012: A Severe Outbreak Year

As of October 2, 2012, there have been 3,969 cases of West Nile virus disease in people, including 163 deaths. 2012 is the worst year ever for West Nile virus infections since 2003.



[Image: Map of the United States with the date 1999 above it. A legend at the top of the map explains the color coding for the States of the number of human disease cases: yellow equals a range of 1-100 cases, orange equals 101-500 cases, pink equals 501-1,000 cases and purple equals over 1,000 cases. Most States are colored yellow, with the exception of three States that colored orange (South Dakota, Oklahoma, and Mississippi) and one that is colored pink (Texas). Maine is the only blue state, which indicates reported incidents of avian, mammalian, or mosquito infections, but no human disease cases. Alaska and Hawaii have no color, indicating no infections or disease cases. There are no purple states on this map.]

Slide 8: The WNV Genome Has Mutated

- By 2002, a new strain of West Nile virus had emerged. Compared to the strain that was introduced in 1999, this new strain had a shorter incubation time in the mosquito vector before it could be transmitted to other animals.
- Thus the new strain spread rapidly in the US, completely replacing the original strain.

[Image: Illustration depicting RNA as a gold helix with projections of blue, orange, pink, green and yellow bars representing the bases.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Laura Kramer explaining how the RNA genome of West Nile virus accumulates mutations, resulting in new genotypes. The video shows Dr. Kramer sitting in an office speaking, maps of virus impact across the United States in different years, a map depicting global temperatures, and footage of laboratory research. Video length is 1 min. 58 sec. The following is a transcript of the video:]

> "So the virus came from Africa into Israel. That's the same strain that was introduced to New York in 1999 and then spread across the U.S. in a matter of years. As the virus spread west from New York towards the West Coast, the virus had several mutations in the viral genome. There were three consistent changes that we saw in strains beginning in 2002, and it increased in number so that most of the strains we isolated were identified as this new genotype, so that by 2004, we didn't see the strain that was introduced. We don't see it anymore. It's been completely displaced by this new genotype, which is very minimally different. And so, our question was what allowed this displacement to occur so rapidly? What advantage did this virus, this new genotype have over the old genotype given that there were so few genetic changes?
>
> "We've done some laboratory studies with our colonized mosquitoes where we've looked at what a small difference in temperature will do. And we incorporated those temperature studies with the change in genotype of West Nile virus, trying to see whether those hot days that we had when West Nile was introduced into the U.S., and when West Nile hit the Rockies in 2002 and 2003--those very hot days--whether that allowed this displacement of the introduced genotype with the new genotype, whether it facilitated that. And we saw that yes, it did make a big difference. Small increases in temperature gave a greater advantage to the new genetic strains of West Nile compared to the introduced strains."

Slide 9: WNV Is a Model for Mosquito-Borne Infections

- West Nile virus is an important disease agent because of its effects on birds and the ecosystem, as well as humans.
- It can also serve as a model for understanding other infections spread by mosquitoes. Dengue and yellow fever are two infectious diseases also spread by mosquitoes.

[Image: Close-up photo of a mosquito sitting on human skin.]

Slide 10: The Mosquito Is a Vector For West Nile Virus

- West Nile virus does not generally spread directly from bird to bird, bird to human, or human to human.
- Having a vector—the mosquito—greatly increases the virus's ability to spread.
- Vectors are animals that spread infectious agents to other animals.

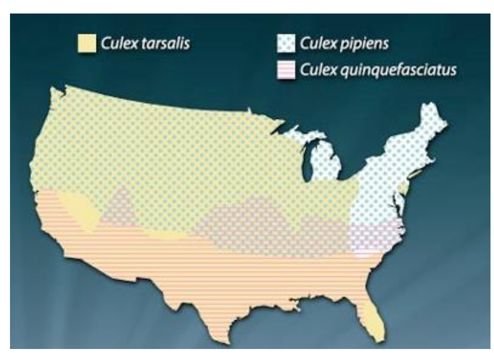
[Image: Outline drawing of a mosquito showing the position of its salivary glands and mid-gut.]

[On the right side of this slide is a clickable film graphic to launch a video showing how mosquitoes spread viruses, beginning with footage of a mosquito on human skin and then cutting to an anatomical drawing with animation demonstrating the the process. Video length is 29 sec. The following is a transcript of the video:]

> "And the mosquitoes will then land and will probe, and what you can see is now what happens to the virus as it enters the mosquito. So it's going to enter, and it's going to go into the midgut, and then it's going to start replicating. And it's actually going to go out through the midgut and then disseminate to the other body organs; but the most important is going to the salivary glands, because the salivary glands will then allow this mosquito to then deliver the virus back into the next person that it bites. Okay, so that's how it works."

Slide 11: *Culex* Genus Mosquitoes Are the Main WNV Vector

There are hundreds of species of mosquitoes in the US, most of which are not infected with West Nile virus. Among those that are, the ones belonging to the genus *Culex* are considered to be the most important vectors.



[Image: A map of the United States illustrating the distribution of three species of mosquitoes belonging to the *Culex* genus, each depicted by distinct colors and patterns. *Culex pipiens,* depicted by blue dots, is found across the entire northern two-thirds of the US; *Culex tarsalis,* depicted by yellow, is found across most of the country, with the exception of Michigan and most of the northeastern and Mid-Atlantic States, except for parts of southern New Jersey and Long Island where it is present; *and Culex quinquefasciatus*, depicted by pink stripes, is found throughout much of the southern half of the country. All three species overlap across much of the middle of the country.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Laura Kramer explaining geographical distribution. The video shows Dr. Kramer sitting in an office speaking and photos of the different mosquito species. Video length is 1 min. 45 sec. The following is a transcript of the video:]

"West Nile has a different vector depending on the region of the United States, and depending on whether you're in the New World or the Old World or South America.

"And the *Culex* species we have in the U.S. that are the important vectors are *Culex pipiens*, which is the Northern house mosquito, which is what we have here in New York, and then, *Culex quinquefasciatus*, which is the Southern house mosquito, which is seen in Texas, Southern California, Florida. And then they hybridize, so there is a hybrid zone where you get *Culex pipiens* hybridizing with *Culex quinquefasciatus*, and that's sort of in the midsection of the U.S.

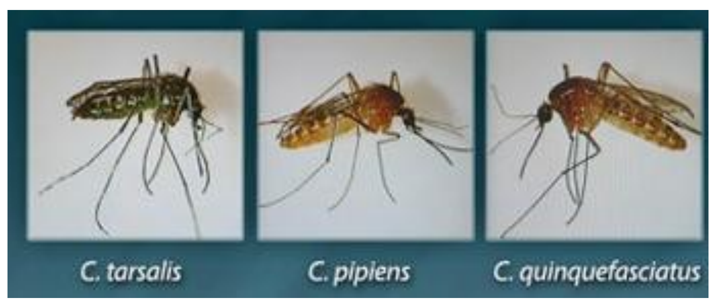
"The *Culex pipiens* were breeding very successfully when West Nile was introduced because there was a drought in New York, and people were not allowed to change water in their pools; there was water restriction. So the water became very organically rich, and *Culex pipiens* love organically rich water. So it allowed the *pipiens* to thrive and to be available for the virus to infect.

"Culex tarsalis are your more agricultural mosquito that you find in the central valley of California and in the Rockies. They transmit the virus better than *Culex pipiens*, so the virus took off in 2002 and 2003 because it hit the area of the country where *Culex tarsalis* was its vector. So, also the environmental conditions; it was very hot and dry those years. So the *Culex tarsalis* were thriving: the virus does better at hotter temperatures, and so the virus really took off at that time."

Slide 12: Why Are *Culex* Mosquitoes Effective Vectors?

An effective vector must:

- Be able to be infected by the virus;
- Feed on animals in which the virus can replicate effectively;
- Live long enough and feed frequently enough to spread the virus from animal to animal through its bites.



[Images: Photos of the three *Culex* mosquito species that are vectors for the West Nile virus (from left to right): *C. tarsalis, C. pipiens,* and *C. quinquefasciatus*.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Laura Kramer describing how the mosquitoes that spread West Nile virus meet all of these requirements. The video shows Dr. Kramer sitting in an office speaking and footage of trapped mosquitoes being studied in a lab. Video length is 53 sec. The following is a transcript of the video:]

"So when the mosquito first takes a blood meal, it goes into its stomach. Its stomach is called the mid-gut. The virus multiplies in those cells in the stomach or mid-gut, and then disseminates to the hemolymph, which is the mosquito's blood, and infects other tissues such as salivary gland tissues, fat bodies, nervous tissue. And then, from the salivary glands, the virus will be transmitted in the saliva. The mosquito has to take two blood meals to be an important vector. It has to take the blood meal that will infect it, and it has to take a blood meal to transmit. So it has to live long enough for the virus to complete its incubation period from being taken up by the mosquito, replicating in the mosquito, to be transmitted by the mosquito. And that's probably the most important factor. The mosquito also has to feed on a susceptible host. "

Slide 13: WNV Spread Requires Mosquitoes and Birds

Birds, humans, and other mammals are West Nile virus hosts—animals that provide an environment for viral reproduction—but birds are the most important hosts.

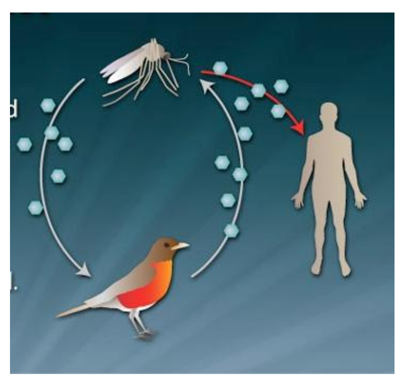
[Image: Illustration of the cyclical transmission of the virus from mosquito to bird and back to mosquito.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Kilpatrick explaining the role of birds in West Nile virus transmission. The video shows Dr. Kilpatrick sitting outside on a bench speaking, footage of mosquitoes in traps and birds, and photos of the different mosquito species. Video length is 1 min. 2 sec. The following is a transcript of the video:]

> "There is actually over 3000 mosquito species in the world, and there is about 150 or 170 in North America. And of those, over 60 have now been found infected with the virus. In most areas, there is really only a couple species that are involved in transmission, and those important species tend to be the ones that feed on birds and, in particular, feed on certain species of birds that either are highly preferred by mosquitoes or are highly infectious once they become infected, or both.
>
> "In this area in Washington, D.C., the most important vector of West Nile virus is a species called *Culex pipiens*, or the Northern house mosquito. And that mosquito is a really good vector because it feeds primarily on birds about 95 to about 85% of the time, depending on which month you're talking about. And the birds that it feeds on are moderately to highly infectious. And as a result, that mosquito can keep a, a cycle going quite intensely. And in addition, it actually does feed on humans some of the time. And so, the way that most human cases appear to occur both here in Washington, D.C. area as well as Chicago, New York, and places like that, appears to be when a *Culex pipiens* mosquito bites a bird, becomes infected, and then that same mosquito goes on later to bite a human."

Slide 14: Humans Are a Dead-End Host

When there are plenty of susceptible birds, the virus will infect the birds, replicate to high amounts, and spread to other birds and animals. When key species of birds are not available, mosquitoes might shift to biting more humans. Although the virus can infect humans and cause disease, it will not spread.



[Image: Illustration depicting the cyclical transmission process between mosquito, bird and back to mosquito, maintaining the infection. The illustration also shows a human to the right side of the cyclical image, depicting the one-way transmission of the virus from mosquito to human.]

[At the bottom of this slide is a clickable film graphic to launch a video of Drs. Kilpatrick and Kramer explaining the mosquitoes' host preferences. The video shows Dr. Kilpatrick sitting outside on a bench speaking, animation of a mosquito landing on human skin and feeding, and Dr. Kramer sitting in an office speaking. Video length is 2 min. 13 sec. The following is a transcript of the video:]

> [Dr. Kilpatrick:] "The main vectors of things like yellow fever and dengue fever are actually mosquitoes that feed primarily on humans. If humans are infectious for a disease like they are for dengue virus, you

get a quite intense human disease cycle where the mosquito feeds on a human, infects them, they become infectious, they feed another mosquito, and it kind of keeps cycling around. In contrast, for something like West Nile virus, humans are basically a dead end host. If a mosquito infects a human with West Nile virus, even if they get subsequent mosquito bites, they won't pass it on. And as a result, humans are kind of just accidental collateral damage, if you will, for something like West Nile virus."

[Dr. Kramer:] "One of the outcomes of the research that we have done with Marm Kilpatrick is to show that *Culex pipiens* not only is the most important, in this part of the country, is the most important enzootic vector, but it's also a very important bridge vector, and that it will feed on humans. In the communities that Marm was studying, mosquitoes preferentially were feeding on robins. And the robin feeding was greater early in the year, and then as the robins leave the area, we would see the proportion feeding on robins go down naturally because the robins weren't available. But at the same time that the robin population was going down, the house sparrow population was still very high. But the mosquitoes didn't seem to then go to the house sparrows for their blood meal, but instead then fed on humans. And this is important because you have the virus intensifying in transmission as the virus is being transmitted from mosquito to robin, or mosquito to bird, and then the robins leave, and you have built up all this virus in the mosquito population, and then the mosquitoes begin to feed on humans later in the summer when the robins leave the area. Had the mosquitoes fed on humans equally early in the summer, then you wouldn't have this buildup in virus because humans are dead end hosts, meaning that they can't infect the mosquito. The only way to transmit between humans is through blood transfusion or organ transplantation."

Slide 15: Some Birds Are Better Hosts Than Others

The factors that affect whether a bird is a good host for West Nile virus include its susceptibility to infection, the level of virus in its blood, and how long this high blood level of virus is maintained. That has to do with how well the bird's immune system fights off the virus.

[Image: Drawing of four different birds: robin, crow, blue jay, and scrub jay.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Kilpatrick explaining that crows, blue jays, and scrub jays are actually more infectious than robins. The video shows Dr. Kilpatrick sitting outside on a bench speaking and footage of various birds and horses. Video length is 57 sec. The following is a transcript of the video:]

"One of the things that is important in understanding the transmission of this virus is that some hosts are more infectious than others. And what it means to be infectious is that when a mosquito that's infected bites you and infects you, how well does the virus do in your blood, and then how well does your immune system actually knock down that virus? And so, what it means to be infectious is that the virus replicates quite well, and your immune system can't knock the virus down fast enough.

"Certain species are quite infectious and other ones not so much. American Crows and blue jays--that whole family of crows and jays--appear to be quite infectious for West Nile virus, whereas most mammals turned out to be not infectious at all. And then, some other species kind of fall somewhere in the middle. So American Robins, of the 53 species that have been studied for an infectiousness so far, are about ninth. And so, they're kind of moderately infectious but not nearly quite as infectious as, say, a scrub jay or a blue jay or something like that. And we still actually don't fully understand what it is that makes a bird infectious or not. It's some sort of relationship between the virus infecting the host and the host's immune system."

Slide 16: Robins Are the Main Host in North America

Which bird is the main host for West Nile virus has to do with many factors:

- The interaction between the virus and the bird's cells, which determines how well the virus replicates;
- How accessible the bird is to the mosquito vector, which depends on the bird's abundance, where it lives, and whether it has defensive behaviors to protect it from getting bitten; and
- The mosquito feeding preferences.

[Image: Illustration of a robin with text encircling it that reads: "Moderately infectious - Fairly abundant - Frequently fed upon"]

[At the bottom of this slide is a clickable film graphic to launch a video of Drs. Kramer and Kilpatrick explaining host preference. The video shows Dr. Kramer sitting in an office speaking, footage of birds and scientists performing labwork, Dr. Kilpatrick sitting outside on a bench speaking, and a graphic depicting a graph of the abundance of different bird types and the source of the mosquito blood meals. Video length is 3 min. 5 sec. The following is a transcript of the video:]

[Dr. Kramer:] "The American Robin is the preferred host for *Culex pipiens* mosquitoes. We used several different approaches to determine which was the most important avian host. And one of those approaches was to do point counts to look at the population of birds in the area. How was it made up? Is it made up largely of house sparrows? What's the proportion of one bird to another? And we saw that robins, just from the point counts, make up a very low proportion of the birds in each area. Maybe two to five percent, two to seven percent of the birds are robins. Most of the birds are house sparrows because they breed continually.

"We also looked at the blood meals of these mosquitoes. And we can do that by taking the abdomens where the blood is, in the mid-gut, and then doing a molecular assay, a PCR assay, to a certain gene in the vertebrate blood meal, and then sequence it and determine what host that mosquito fed on. And we saw that even though robins make up a very low proportion of the population in a community, mosquitoes preferentially were feeding on robins. Is it because it only likes robins? You know, robins are the chocolate cake of the bird world? But that's obviously not the case. It may be that robins don't have as much defensive behavior so that the mosquitoes are more successful feeding on the robins. It's not that they prefer robins, but the mosquito blood meals you are getting are ones where they were successful feeding, and the house sparrows won't let them feed and the robins will. I mean, that's one possibility."

[Dr. Kilpatrick:] "So one of the interesting things that our research, as well as some other folks have found is that mosquitoes don't simply fly around and bump into individual hosts and feed on them. They appear to have either substantial feeding preferences, or at least when they bump into different hosts, some hosts might defend themselves better than others. For example, American Robins are one of the most highly fed-upon hosts in this area here, and in fact, American Robins actually make up over half of all the blood meals even though they are only two to five or ten percent of the birds. And so, while they're not a rare bird, they are not nearly as abundant as you would expect given how many blood meals come from those birds. And so, what it appears is that mosquitoes are actually able to feed on American Robins quite frequently. It turns out that American Crows are actually preferentially fed upon by these mosquitoes. But it turns out that crows, although they're loud and obnoxious and kind of noisy, aren't that abundant. And so, they're not, in fact, infecting very many mosquitoes with the virus. In contrast, birds like American Robins, which are relatively much more abundant than crows, they're actually infecting many, many more mosquitoes with the virus than American Crows are. And so, our research suggests that American Robins appear to be the most important host actually infecting mosquitoes, and much more important, let's say, than an American Crow or than a house sparrow, or things like that, primarily because they're fed on so frequently by mosquitoes. And that enables them, A, to become infected, and then B, once they're infected, they're actually moderately infectious. But given that they are moderately infectious, then given that they're fed on by a huge number of mosquitoes, they can pass it on to a bunch more of them."

Slide 17: Location, Location, Location

Some birds build their nests high up in trees and others closer to the ground—on gutters, eaves, outdoor light fixtures, and other structures. Where a bird lives affects how many mosquito bites it gets.

[Image: Photo of a robin and her hungry young in a nest built on a tree limb.]

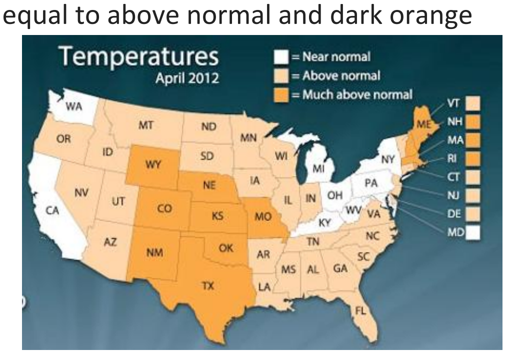
[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Kilpatrick explaining how distance from the ground affects infection rate. The video shows footage of a scientist setting mosquito traps and Dr. Kilpatrick sitting outside on a bench speaking. Video length is 35 sec. The following is a transcript of the video:]

"Because we're studying a virus that infects primarily birds, we wanted to know, is the density of mosquitoes that we see on the ground, is that the same as in the canopy? So we actually placed half of our traps in the canopy and half at kind of closer to the ground level to try to get at that question. And what we found, quite interestingly, is that especially in urban areas, the density of mosquitoes in the canopy can often be almost tenfold higher, or even higher than that, in the canopy traps than they are at ground level. And what that suggests to us is that birds that are roosting up high in these canopies can actually be subject to many, many, many more mosquito bites than they would be to someone lower on the ground."

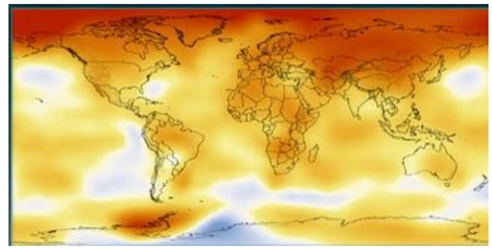
Slide 18: Climate Affects WNV Transmission

Some years are worse than others for West Nile virus transmission. Changes in temperature affect both the mosquito life cycle and virus replication in different hosts. The mild winter, early spring, and hot summer of 2012 are thought to have contributed to the increase in West Nile virus cases.

[Image: Map of the United States titled "Temperatures April 2012". A legend at the top of the map explains the color coding for the States as white equal to near normal, light orange equal to above normal and dark orange equal to much above normal. Most of the States are orange, with light orange primarily distributed in the west, north, mid-west and southeast and dark orange distributed among eight States in the middle and southwest and three States in the northeast. White is distributed among seven states in the northeast and Midwest, and in Washington and California in the west. The caption at the bottom of the image reads: January – April 2012 was the warmest such period on record for the contiguous US.]



Slide 19: Research Needed to Understand Climate's Role
Understanding the precise effects of temperature on transmission will help scientists predict the spread of the virus as temperatures rise due to global warming.



[Image: A world map illustrating with color variation above average annual temperatures. The map ranges in color from red at the top, in the mid-and upper-northern latitudes, to yellows and oranges with some white areas in the middle and lower (southern) sections of the map, with the white areas becoming more dominant in the southern area of the map.]

[At the bottom of this slide is a clickable film graphic to launch a video of Dr. Kramer explaining studies she is conducting in collaboration with Dr. Kilpatrick. The video shows footage of scientists in the lab working with mosquitoes, mosquitoes in traps, and Dr. Kramer sitting in an office speaking. Video length is 1 min. The following is a transcript of the video:]

> "We're also doing a lot of temperature studies with Marm where we're bringing field mosquitoes into the lab and trying to model what will climate warming do: What will an increase of one to two degrees do to the virus cycle? Will it make a difference?
>
> "So we will do studies where we infect the mosquitoes and look at how well they transmit the virus, how early they can transmit the virus, how many days does it take from the moment they take the blood meal until they can transmit. And this is temperature dependent because all these viruses replicate better at higher temperatures. So besides affecting the virus, temperatures will also affect the mosquito itself. The mosquito at a hotter temperature won't live as long, but it will transmit earlier. So we're looking at the tradeoff: does the higher temperature facilitate the virus transmission, or does the mosquito not live long enough to transmit, even though the virus is replicating, multiplying faster in the mosquito?"

Slide 20: Studying WNV from Multiple Angles
To understand how West Nile virus spreads and find ways to stop that spread, researchers must conduct careful studies of virus transmission, taking into account all of the factors that affect it.

[Images: Four photos of scientists (from upper left, clockwise): scientist in the field wearing equipment to trap mosquitoes, scientist holding a petri dish of three mosquitoes, scientist in the field holding a mosquito trap, and scientist in the lab.]

[At the bottom of this slide is a clickable film graphic to launch a video describing how Drs. Kramer and Kilpatrick collaborate to study West Nile. The video shows Dr. Kilpatrick in the field, sitting outside on a bench speaking, scientists performing labwork, scientists setting up mosquito and bird traps in the field, and Dr. Kramer sitting in an office speaking. Video length is 1 min. 23 sec. The following is a transcript of the video:]

> (Footage of Dr. Kilpatrick in the field, freeing a bird from a net, says, "He thinks I'm a cat and I'm about to eat him.")
>
> [Dr. Kramer:] "We have a close collaboration with Marm Kilpatrick and the work that we do with Marm is field-based, and then we do lab testing of his field collected samples, and then we design studies that

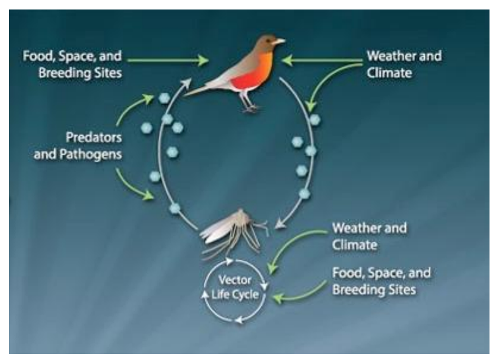
we want to do in the lab to clarify questions that come up during the field based part of the project."

[Dr. Kilpatrick:] "One of the interesting things about West Nile virus is that because you have a number of different mosquito species that can transmit the disease, as well as the number of different bird species that are fed on by these mosquitoes, in order to really get a handle on what are the possible factors driving transmission, you really have to measure everything at once. And so, we have a team of folks that measure many different aspects of the mosquito side of things. And we have a set of field sites that span this gradient I was talking about, and trap mosquitoes with a number of different kinds of traps, and those traps measure mosquito abundance as well as the fraction that are infected. In addition, we try to measure mosquito feeding patterns. So we actually want to know what are the mosquitoes feeding on at these sites, and we do that by trying to collect some mosquitoes that have blood in their stomach. And then, we sequence the DNA in that blood and find out who they fed on. Well, now, we actually need to combine that data with what's available to those mosquitoes in the first place. So we need to also go out and measure the available host community. So we actually go out and measure the density of birds at these sites, as well as the density of other hosts including things like squirrels and mammals, and humans as well."

Slide 21: Summary
Factors that affect West Nile virus spread:

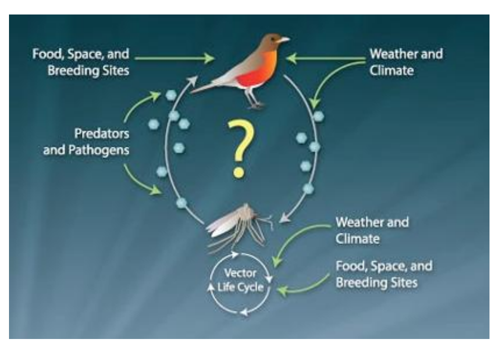
- The virus genetic makeup;
- The type of mosquito it infects;
- The mosquito's feeding preferences;
- The host's susceptibility to infection;
- The host's prevalence in areas where mosquitoes live;
- Climate and temperature.

[Image: This illustration depicts the cyclical transmission process between mosquito, bird and back to mosquito, maintaining the infection. The illustration depicts inputs to the bird of "Food, Space, and Breeding Sites" on one side and "Weather and Climate" on the other. Underneath the mosquito is a circle of arrows labeled "Vector Life Cycle" with inputs of "Weather and Climate" and "Food, Space and Breeding Sites". In the middle of the cycle between the bird and mosquito are inputs labeled "Predators and Pathogens."]



Slide 22: Testing Your Knowledge
Can you think of some ways to stop West Nile virus from spreading? Take a couple minutes to think about the answer before moving to the next slide.

[Image from the previous slide with a large question mark in the center, between the bird and mosquito.]



Slide 23: Some Strategies for Combating WNV

Human vaccines: This strategy would not decrease the spread of the virus, since humans are a dead-end host, but would reduce the incidence of serious illness and death among people. Although several vaccines are in development, none are yet available for use.

Bird vaccines: This strategy targets the main amplification host. An effective bird vaccine would prevent birds from getting sick and dying, but also protect humans from infection. One vaccine, licensed for the use in horses, has been tested in birds with some success. Drs. Kilpatrick and Kramer showed that the vaccine can reduce the amount of virus in robins.

Mosquito control: This strategy targets the vector; it would protect both birds and humans from getting bitten and infected. Mosquito control methods include pesticides, biological-control agents, and trapping.

Slide 24: Credits

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Slide 25: References and Sources

CDC West Nile website http://www.cdc.gov/ncidod/dvbid/westnile/index.htm

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Photos and animations:

Mosquito, frozen crow, and robin nest, Thinkstock/iStockphoto; doctor with patient, Thinkstock/AbleStock.com; elderly man with baby, iStockphoto; temperature map, NASA/Goddard Space Flight Center Scientific Visualization Studio; mosquito life cycle animation, Drew Berry, Walter and Eliza Hall Institute; WNV EM, CDC/P.E. Rollin; stock footage of robin, blue jay, bluebird, sick bird, crows, and horse, iStockphoto; *Culex* mosquitoes photos, courtesy of Laura Kramer.