The mission of Wildlife Services’ (WS) National Wildlife Research Center (NWRC) is to apply scientific expertise to resolve human-wildlife conflicts while maintaining the quality of the environment shared with wildlife. NWRC develops methods and information to address human-wildlife conflicts related to the following:

- agriculture (crops, livestock, aquaculture, and timber)
- human health and safety (wildlife disease, aviation)
- property damage
- invasive species
- threatened and endangered species
This past year has been a tumultuous one for health care professionals and disease specialists. The global impacts of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2, the virus that causes COVID-19) to public health have been enormous and have shown how interconnected we are to one another and the environment.

Approximately 75 percent of emerging infectious diseases in people come from animals. Because of this, Wildlife Services (WS) and other APHIS programs support a collaborative One Health approach to addressing animal diseases and pathogens, including SARS-CoV-2. Working across multiple disciplines and levels, the One Health approach seeks to achieve optimal health for people, animals, plants, and the environment by recognizing and considering the many interconnections among them.

Globally, hundreds of organizations are furthering the One Health approach through initiatives, policies, programs, and platforms. In the United States, Congress has appropriated millions of dollars to APHIS to lead efforts to combat the threat of zoonotic diseases and advance emergency preparedness.

I’m proud to share that WS employees, including National Wildlife Research Center (NWRC) researchers, biologists, and technicians, have risen to the challenge. They are providing scientific expertise in a variety of disciplines, such as wildlife disease, genetics, modeling, and animal behavior and ecology, to help guide wildlife disease surveillance and monitoring efforts, disease diagnostics, and agency responses to disease outbreaks. In this year’s report, you’ll learn how NWRC researchers are partnering with the Centers for Disease Control and Prevention, as well as State health departments and wildlife agencies, to monitor for SARS-CoV-2 in susceptible wildlife, such as mink and white-tailed deer. We are also helping to determine if wildlife species could serve as reservoirs for maintaining the virus outside the human population.

Unfortunately, SARS-CoV-2 is not the only infectious pathogen causing animal and human health concerns. NWRC’s expertise is also helping with efforts to mitigate the impacts of chronic wasting disease on deer and elk, African swine fever on domestic and feral swine, and rabies on terrestrial wildlife, such as raccoons, foxes, skunks, and mongooses. Working together with our Federal, State, and Tribal partners, WS and other APHIS programs are increasing and enhancing the Nation’s ability to prevent, detect, report, and respond to emerging and zoonotic diseases now and in the future.

It is with pleasure that I present to you the 2021 research accomplishments for NWRC.

Jason Suckow
Director
National Wildlife Research Center
Wildlife Services, APHIS-USDA
Fort Collins, CO
## Innovative Solutions to Human-Wildlife Conflicts

### National Wildlife Research Center Accomplishments, 2021

**Research Spotlights** ................................................................. 4  
SARS-CoV-2 Research in Wildlife ............................................... 5  
WS Wildlife Tissue and Serum Archives ....................................... 9  
Wildlife Forensics ........................................................................ 13  

**2021 Accomplishments in Brief** .............................................. 18  
Devices ...................................................................................... 19  
Pesticides ................................................................................... 21  
Other Chemical and Biological Methods ...................................... 23  
Disease Diagnostics, Surveillance, Risk Assessment, and Management ........................................ 27  
Wildlife Damage Assessments ..................................................... 32  
Wildlife Management Methods and Evaluations .......................... 38  
Wildlife Population Monitoring Methods and Evaluations .............. 44  
Registration Updates ................................................................... 48  
Technology Transfer ...................................................................... 50  
Awards ........................................................................................... 52  

**2021 Publications** ..................................................................... 54  

**Appendix I. List of 2021 NWRC Research Projects** .................... 68  

**Appendix 2. NWRC Research Contacts** ..................................... 69  

**Appendix 3. Acronyms and Abbreviations** ................................. 72
The health of animals, people, and the environment is connected. The One Health approach is a collaborative effort of the human health, veterinary health, and environmental health communities. Through this collaboration, APHIS achieves optimal health outcomes for both animals and people.

APHIS supports a One Health approach to addressing animal diseases and pathogens, including SARS-CoV-2.

The National Wildlife Research Center (NWRC) is part of Wildlife Services (WS), a program within the U.S. Department of Agriculture’s (USDA) Animal and Plant Health Inspection Service (APHIS). Our researchers are dedicated to finding biologically sound, practical, and effective solutions for resolving wildlife damage management issues. The following spotlights feature some of WS NWRC’s expertise and our holistic approach to addressing today’s wildlife-related challenges.

**SPOTLIGHT: SARS-CoV-2 Research in Wildlife**

On March 11, 2021, President Biden signed into law the American Rescue Plan (ARP) Act, also known as one of the COVID-19 stimulus bills. The ARP is a multifaceted, $1.9 trillion plan created to mount a national vaccination program, identify and address emerging strains of COVID-19, and safely reopen schools, among other activities. Under the umbrella of USDA activities funded in the legislation, the Secretary of Agriculture designated APHIS as the lead agency to develop a plan to conduct monitoring and surveillance of susceptible animal species for SARS-CoV-2 (the virus that causes COVID-19). The ARP allocates $300 million for this charge and the money is available until expended.

WS, along with other APHIS programs, is leading efforts to develop and carry out a robust early warning surveillance system to alert public health partners of potential disease concerns in animals and potentially prevent or limit the impacts of SARS-CoV-2 and future zoonotic disease outbreaks in people and animals. The system leverages and expands upon WS and APHIS’ Veterinary Services disease monitoring and surveillance capabilities and expertise.

The following sections highlight WS surveillance and research related to SARS-CoV-2 in wildlife.

**Initial SARS-CoV-2 Surveillance**

In fall 2020, before Congress passed the ARP, outbreaks of SARS-CoV-2 were confirmed on mink farms in Utah, Michigan, Wisconsin, and Oregon. WS National Wildlife Disease Program (NWDP) wildlife biologists and NWRC researchers conducted wildlife surveillance for the virus in small to medium-sized carnivores and other species around the infected farms.

Per One Health principles, which highlight the interconnections among people, animals, and the environment, we conducted this surveillance as part of investigations involving the Centers for Disease Control and Prevention (CDC); the U.S. Geological Survey; and State departments of agriculture, natural resources, and health. APHIS supports a One Health approach to addressing animal diseases and pathogens, including SARS-CoV-2.

More than 200 wild and invasive free-ranging animals found near infected mink farms—including raccoons, minks, skunks, opossums, rodents, and feral cats—were captured, sampled, and tested for SARS-CoV-2 at APHIS’ National Veterinary Services Laboratories (NVSL) in Ames, IA. Presumed escaped minks were closely associated with nearby barns and...
designated as domestic escapees based on their location, behavior, genetics, and appearance. Researchers identified wild minks by their brown coat color and size (smaller than farmed mink).

Surveillance results showed that 11 mink escapees in Utah and 1 in Oregon tested positive for antibodies to SARS-CoV-2. Further testing showed several of the mink not only had SARS-CoV-2 antibodies but also were positive for the SARS-CoV-2 virus. One wild mink from Utah also tested positive for the virus. No other sampled species had a detectable antibody response or tested positive.

While it is thought that infected farm workers introduced the virus to the farmed minks, it is unknown how the virus was transmitted to the wild mink. This was the first free-ranging native wild animal confirmed with SARS-CoV-2 in the United States.

APHIS is working closely with Federal, State and industry partners to develop a SARS-CoV-2 infection avoidance and monitoring program for mink farms. The voluntary program will offer incentives, guidance, and support to mink farmers dealing with the virus and may be adapted for other species and industries affected by it or other zoonotic pathogens.

**White-Tailed Deer Exposure to SARS-CoV-2**

Close to 300 million white-tailed deer live in the United States. They are found in every State except Alaska and enjoyed by many wildlife watchers, recreationalists, and hunters. Studies conducted in 2020 and early 2021 showed that white-tailed deer have protein (ACE-2) receptors capable of binding to SARS-CoV-2, allowing the virus to enter susceptible cells, and that captive deer experimentally exposed to the virus are susceptible to infection. Given these findings and the fact that white-tailed deer often come into close contact with people, APHIS further investigated SARS-CoV-2 in wild, free-ranging deer.

“Widespread human infections with SARS-CoV-2 combined with human-wildlife interactions create the potential for spillover between people and animals,” says NWRC’s assistant director, Dr. Tom DeLiberto. “Studying the susceptibility of certain mammals to SARS-CoV-2 helps to identify species that may serve as reservoirs or hosts for the virus, as well as understand the origin of the virus and predict its impacts on wildlife and the risks of cross-species transmission.”

We obtained serum samples from wild, free-ranging white-tailed deer opportunistically as part of wildlife damage management activities WS conducted in 32 counties in Illinois, Michigan, New York, and Pennsylvania from January 2020 to 2021. These samples were tested at NWRC and NVSL. Antibodies to SARS-CoV-2 were detected in 33 percent of the 481 samples collected. The results varied...
by State (in Illinois, 7 percent of 101 samples contained antibodies; in Michigan, 67 percent of 113 samples; in New York, 19 percent of 68 samples; and in Pennsylvania, 31 percent of 199 samples). None of the deer populations surveyed showed signs of clinical illness associated with SARS-CoV-2.

Additionally, in late 2021, NVSL confirmed SARS-CoV-2 infection in wild white-tailed deer in Ohio, based on samples collected between January and March 2021 by The Ohio State University College of Veterinary Medicine as part of ongoing deer damage management activities.

The finding that wild white-tailed deer were exposed to and infected with SARS-CoV-2 was not unexpected given that white-tailed deer are susceptible to the virus, are abundant in the United States, often have close contact with people—and that more than 114 million Americans were estimated to have been infected with the SARS-CoV-2 virus by the summer of 2021, according to the CDC.

Further research is needed to understand the significance of SARS-CoV-2 in free-ranging white-tailed deer, including how the deer are exposed to the virus and potential impacts, if any, to overall deer populations, other wildlife, and people. APHIS is working closely with Federal and State partners, including the U.S. Department of the Interior, the CDC, and the Association of Fish & Wildlife Agencies, to determine next steps.

Investigating SARS-CoV-2 in Other Wildlife

Although wild animals were likely the origin of SARS-CoV-2, it is largely unknown how the virus affects most wildlife species and if wildlife could serve as a reservoir for maintaining the virus outside the human population.

Through a series of experimental infection studies, Colorado State University, NWRC, and University of Queensland researchers evaluated the susceptibility of nine wildlife species to SARS-CoV-2. Results of captive animal studies showed that several species that frequently
In studies with experimentally infected captive animals, results showed that deer mice, bushy-tailed woodrats, and striped skunks (pictured) are susceptible to SARS-CoV-2 infection and can shed the virus in respiratory secretions.

Photo: Adobe Stock

come into close contact with humans and human dwellings, including deer mice, bushy-tailed woodrats, and striped skunks, are susceptible to infection and can shed the virus in respiratory secretions. In contrast, cottontail rabbits, fox squirrels, Wyoming ground squirrels, black-tailed prairie dogs, house mice, and racoons are not susceptible. These results expand the knowledge base of susceptible species and provide evidence that some human-wildlife interactions could result in SARS-CoV-2 transmission.

Wildlife Disease Dynamics: NWRC’s Newest Research Project

The SARS-CoV-2 virus, highly pathogenic avian influenza, and rabbit hemorrhagic disease are just a few of the emerging infectious pathogens and diseases making headlines these days. WS’ NWDP and NWRC have been at the forefront of the Nation’s efforts to monitor and understand the impacts of these pathogens on wildlife.

Since NWDP’s inception in 2003, NWRC has supported the program through cutting-edge disease research. In recognition of that strong partnership, NWRC formed a new research project in 2020 titled “Wildlife Disease Dynamics, Epidemiology, and Response,” led by research biologist and wildlife epidemiologist Dr. Susan Shriner.

“Our project supports NWDP, identifies and characterizes wildlife pathogens at the wildlife-agricultural interface, and develops tools for the identification and mitigation of disease risks to agricultural, public, and wildlife health,” says Shriner. “We work hand in hand with the program’s wildlife disease biologists to help address emerging wildlife disease issues.”

Over the next 5 years, Shriner anticipates the project will focus on understanding the dynamics of wildlife pathogens such as avian influenza virus and SARS-CoV-2, as well as evaluate current wildlife disease surveillance methods and conduct outbreak investigations. Her team will also assess North American wildlife and livestock’s ability to serve as hosts for emerging agricultural and zoonotic pathogens, including SARS-CoV-2.

Next Steps—With the passage of the 2021 ARP and efforts to strengthen our Nation’s ability to quickly detect and respond to emerging and zoonotic diseases in animals, NWRC will continue to provide leadership and expertise on wildlife diseases through several of its research projects. Next steps will likely include broader SARS-CoV-2 surveillance in wildlife such as deer, red fox, and other common species, as well as more in-depth studies to better understand the significance of the virus in free-ranging wildlife. These efforts will help advance knowledge about how animals are exposed to the virus and potential impacts, if any, to overall wildlife populations and people.
Spotlight: WS Wildlife Tissue and Serum Archives

WS’ National Wildlife Disease Program (NWDP) participates in wildlife disease monitoring and surveillance in all regions of the United States. The program’s wildlife disease biologists are trained in epidemiology, wildlife health, animal handling, surveillance, and sampling techniques. They collect thousands of tissue, serum, hair, and other samples annually from numerous wildlife species, including rodents, waterfowl, feral swine, deer, coyotes, raccoons, and other species. Such samples have great historical and scientific value to WS, as well as to conservationists, epidemiologists, and researchers around the world. They are an important source of specimens for retrospective studies.

NWDP established the WS Wildlife Tissue and Serum Archives in 2005. The initial contents included samples from avian influenza surveillance in wild birds, feral swine disease surveillance, and plague and tularemia monitoring. The archives have since expanded to include samples for diseases such as swine tuberculosis, pseudorabies, West Nile virus, leptospirosis, and others. The collection is unique in its quantity of samples, the diversity and broad geographic range of the species sampled, and the breadth of samples collected consistently over extended periods of time.

The following sections highlight how NWDP’s archived samples have been used in NWRC’s wildlife research.

Age and Environmental Factors Drive Avian Influenza

A challenge for many wildlife managers and disease ecologists is knowing the relative importance of wildlife population and environmental factors, and how they influence the spread of pathogens at local, regional, and continental levels.

NWRC, APHIS’ Veterinary Services, and university scientists combined data on low pathogenic avian influenza virus (AIV) from wild waterfowl samples in the WS Wildlife Tissue and Serum Archives with waterfowl banding and recovery data to identify the factors most likely to influence AIV’s spread in the United States.

Researchers studied: (1) demographics (age, sex); (2) environmental reservoirs (water temperature and local aggregation of birds); (3) hot spots (areas of high AIV prevalence); and (4) contact networks (flow and clusters of birds connected by similar migration patterns). These factors form the basis for five hypothetical systems used to explain the distribution of AIV over space and time.

Researchers tested the five hypotheses using statistical models and found that bird age and environment reservoirs were the two primary factors influencing continental-scale AIV infection in migratory waterfowl. Water temperatures, plus the seasonal movement and aggregation of young that had never been exposed to AIV, drove the spread of the virus. Researchers recommended that AIV
Five hypothetical systems driving the spread of avian influenza virus: (1) demography hypothesis, (2) environmental reservoir hypothesis, (3) hot-spots hypothesis, (4) contact network hypothesis, and (5) multiple mechanism hypothesis.

Graph: USDA, Wildlife Services
surveillance and monitoring efforts focus on assessing local densities of younger birds and water temperatures instead of tracking bird migration patterns or regional bird movements from areas of high AIV prevalence.

“Having access to avian influenza surveillance data from across the country and across multiple years allowed us to successfully model the movement of the virus on the landscape,” says NWRC project leader Dr. Alan Franklin. “The WS Wildlife Tissue and Serum Archives make this type of research and analysis possible.”

**SARS-CoV-2 in Deer**

Much is still being learned about SARS-CoV-2 (the virus that causes COVID-19 in people), but scientists know it can spread from people to animals in some situations, especially during close contact. Studying the susceptibility of certain mammals to this virus helps to identify species that may serve as reservoirs or hosts. It also helps us understand the origin of the virus and predict its impacts on wildlife and the risks of cross-species transmission.

The spotlight “SARS-CoV-2 Research in Wildlife” discusses recent surveillance for the virus in white-tailed deer. In addition to opportunistically collecting and analyzing samples from deer during WS wildlife damage management activities in 2020 and 2021, NWRC researchers evaluated 143 archived deer serum samples from the WS Wildlife Tissue and Serum Archives. The samples were collected in Illinois, Michigan, New York, and Pennsylvania from 2011 to 2020, before SARS-CoV-2 was detected in the United States.

The archived samples were screened using a commercially available SARS-CoV-2 antibody screening test known to be highly accurate when used for other species. However, it had not yet been validated for deer. To help allay concerns that the commercial test may have detected antibodies to another virus (a phenomenon known as cross-reacting),
a subset of samples was tested at NVSL using a different test specific to SARS-CoV-2. Both analyses resulted in identical findings. Researchers detected SARS-CoV-2 antibodies in only 1 of the 143 archived samples. The single sample was at the minimum threshold of detection and likely a false positive. This low-level detection is well within the expected false positive rate of the test used.

"When you graph the data, it’s clear that deer started to become exposed to the SARS-CoV-2 virus after the virus became prevalent in people in the spring of 2020," says NWRC’s assistant director, Dr. Tom DeLiberto. “However, it’s important to note that we still do not know if the deer were exposed through people, the environment, or other wildlife. Having the WS Tissue and Serum Archives was invaluable in this retrospective study and helped us determine when deer in the United States were first exposed.”

Mining Genetic Information From Feral Swine Archive

In 2015, NWRC’s Wildlife Genetics Project started to archive feral swine genetic samples. These samples have been opportunistically collected by WS field specialists and wildlife disease biologists in 39 States, Guam, and Puerto Rico while conducting feral swine damage and disease management activities. To date, WS personnel have collected nearly 27,000 feral swine genetic samples. Additional samples from feral swine in Canada and Mexico have been acquired through collaboration.

The archived samples provide NWRC geneticists with enough DNA to genotype or "genetically fingerprint" individual feral swine, allowing scientists to identify and distinguish among current feral swine populations as well as determine their origins.

Using archived samples, NWRC genetic analyses show that feral swine are overwhelmingly wild boar and domestic pig hybrids, which may have greater potential than domestic pigs to establish and become invasive. NWRC geneticists are leveraging the archival genomic resources to identify genes that likely contribute to a hybrid pig's heightened invasiveness.

NWRC geneticists have also used archived samples to develop a statistical test to differentiate the unique genetic attributes of feral swine from domestic breeds. Such a test helps States such as Missouri, Michigan, and Minnesota enforce local prohibitions on the possession or transport of feral swine. It also serves to deter the establishment of captive herds, which can lead to invasive populations should animals escape or be released.
Wildlife forensics, which involves the use of genetic technologies, aids in wildlife damage management.

“The genetic insights we’re gathering from these archived feral swine samples help us determine the effectiveness of current management efforts, as well as how feral swine may be spreading across the country,” says NWRC geneticist Dr. Tim Smyser. “Is a population the result of a failed eradication attempt or the illegal movement of feral swine by people? Did feral swine that were detected in Great Lakes States originate in Texas or Canada? The answers may help guide future management actions, policies, or regulations.”

Another line of NWRC research combines high-resolution genetic analysis with serological disease diagnostics. Specifically, researchers are evaluating whether genetic attributes influence feral swine susceptibility or resistance to infection from diseases, such as pseudorabies and brucellosis. If such genetic underpinnings exist, researchers plan to develop predictive models that use feral swine genetics data to determine potential disease risks across the country.

**Next Steps**—NWRC geneticists are working to incorporate feral swine-specific genetic markers into environmental DNA assays so that researchers and natural resource managers can identify the source of swine DNA found in water samples. Additionally, geneticists are examining the genomes of pigs in Africa that are susceptible and resistant to African swine fever. The information will be used to help determine the susceptibility of U.S. feral swine to the virus.

**Spotlight: Wildlife Forensics**

The field of wildlife forensics uses science-based processes and techniques to examine, identify, and compare evidence found at sites associated with wildlife incidents. While traditionally used to solve crimes against wildlife, such as poaching or illegal selling of animals or animal products, wildlife forensics also aids in wildlife damage management. DNA samples from saliva, hair, or blood on clothing and other items from people involved in wildlife attacks or from animal carcasses at predation sites can identify the animal species—and often individual animals—involved.

NWRC’s Wildlife Genetics Project uses wildlife forensics to address wildlife damage management issues. The summaries below highlight NWRC’s recent wildlife forensics efforts.

**Investigating Animal Attacks on People**

As urban wildlife and feral animal populations increase, so do associated conflicts. Often, agencies that end up handling these issues do not have access to useful wildlife damage management tools, such as forensic analysis. Attacks on people by coyotes and packs of feral dogs can challenge the resources and preparedness of these agencies.

On February 28, 2020, the Kentucky Department of Fish and Wildlife Resources requested NWRC’s assistance in a forensic evaluation of a suspected wild animal attack that resulted in the death of a 13-year-old child.
Researchers from NWRC’s Wildlife Genetics Project swabbed numerous bite marks and collected hairs from the child’s shoes, T-shirt, and jacket. Additional samples were taken by authorities from the child. The child lived with a dog, so the Kentucky State police collected oral swabs from the pet. Test results showed all DNA collected from these items came from several individual domestic dogs. One was the home dog, but because its DNA was not associated directly with the bite marks, it was not implicated in the attack.

“We identified three individual dogs’ DNA associated with the bite marks,” says Dr. Toni Piaggio, a geneticist and project leader of the Wildlife Genetics Project. “Of the samples collected from nine feral dogs in the area where the incident occurred, three matched samples obtained from the bite marks on the child, his T-shirt, jacket, and shoes.”

The feral dogs were captured and euthanized. This was the second human fatality in a rural area due to a feral dog pack attack (the first was in Florida in 2019). Feral dogs may be an emerging issue in wildlife damage management because they impact native wildlife and are considered one of the biggest threats to biodiversity worldwide.

In Chicago that same year (2020), a young boy was purportedly attacked by a coyote in a city park. Again, experts with the Wildlife Genetics Project were asked to determine the species involved and to compare DNA collected from the child’s clothing to that of a coyote that had been captured in the area. Laboratory results showed the DNA from the child’s clothing and wounds matched that of the coyote being held.

After the incident, NWRC and WS Illinois Operations helped Chicago Animal Care and Control develop a coyote attack preparedness plan and DNA sampling protocol for any future attacks. The plan has been requested by other entities in Illinois and elsewhere.

**Protecting Endangered Species From Predators**

Millions of dollars are spent each year in the United States to boost endangered and threatened species populations and their habitats. In some situations, local populations of these species are vulnerable to extinction due to predation by native and invasive wildlife.

Predation on ground-nesting birds and their eggs is a major concern for conservationists and wildlife managers. Accurately identifying the predatory species responsible is key to effective management. For instance, the greater sage-grouse (*Centrocercus urophasianus*) is a ground-nesting bird at risk of extinction in multiple U.S. States and Canada. Predation on sage-grouse nests is rarely seen,
and it is difficult to identify the responsible predator species from nest remains.

To help identify common mammalian predators, NWRC geneticists analyzed predator saliva DNA on sage-grouse eggshells and bird carcasses in Wyoming. Researchers monitored sage-grouse nests and hens using infrared trail cameras and radio telemetry. They also sampled for DNA on egg remains and/or hen carcasses when a nest failed or a hen was eaten.

For 79 percent of the nests and 47 percent of the carcass samples, researchers identified the mammalian predator species using DNA. Eighty-six percent of the detected mammal predators were canids, including coyotes and dogs. Other predators included rodents, striped skunks, and cattle.

NWRC researchers acknowledge that identifying nest and hen predators is challenging given the lack of species-specific signs at nests and the difficulty in differentiating predators from scavengers using DNA evidence. The results suggest that the best approach to reducing nest and hen predation is to use multiple techniques, including field surveys, camera monitoring of depredation events, and DNA forensics-based methods.

In similar efforts, NWRC geneticists assisted U.S. Army Reserve biologists at Camp Ripley in Minnesota with identifying local populations of endangered and threatened species can be vulnerable to extinction due to predation by native and invasive wildlife. To help identify common predators of greater sage-grouse, NWRC geneticists analyzed predator saliva DNA on sage-grouse eggshells and bird carcasses.

Photo: USDA, Wildlife Services
NWRC geneticists developed new protocols for collecting DNA samples from depredated livestock carcasses. Knowing which predator species is responsible ensures that management actions to prevent depredations are appropriate.

Photo: USDA, Wildlife Services
predators responsible for attacking endangered Blanding’s turtle (*Emydoidea blandingii*) nests and hatchlings. DNA samples taken from five hatchling carcasses collected in 2021 identified several predators, including crows, chipmunks, and mice. The findings gave the biologists valuable information that helped them implement management strategies to reduce predation by these species.

**Collecting DNA From Livestock Carcasses**

WS aids landowners, State and Federal agencies, and others who request help with wildlife damage management issues, including livestock depredation by animals such as coyotes, wolves, mountain lions, and bears. Often, WS field specialists can determine the predator species responsible by looking at bite marks and other patterns on a carcass. But sometimes, the clues left at the scene are not enough, and more high-tech methods are needed. Knowing which species is responsible ensures that immediate and future management actions to prevent depredations are targeted and appropriate.

WS is investigating new uses for noninvasive DNA sampling (for instance, collection of hair, scat, and saliva) in predation damage management. Salivary DNA samples can identify the predator species and individual animal responsible for killing livestock. However, studies show that samples with low DNA quality and quantity can create challenges.

To improve salivary DNA sample collection techniques, NWRC geneticists collaborated with NWRC’s Utah Field Station and the nonprofit Wildlife Science Center in Minnesota to investigate differences in coyote, wolf, and mountain lion salivary DNA deposits and degradation on cattle and sheep carcasses.

“We found that wolf DNA was the most abundant and easily collected of the three species sampled,” says Piaggio. “For best results, the DNA should be collected within the first 12 hours of deposit; otherwise, it degrades, and our chances of identifying the animal’s genetic signature is reduced. This can be overcome, however, by taking more samples.”

Better results are also achieved when the parts of a carcass hide with saliva deposits are swabbed for DNA in the laboratory and not in the field. NWRC recommends that field specialists working on depredation incidents remove and ship sections of carcass hides to the NWRC genetics lab for DNA collection and analysis.

These findings resulted in new protocols for collecting DNA samples from depredated carcasses. To view the protocols for field collection of hides from depredated carcasses, see Appendix I in Piaggio et al. 2019, *DNA persistence in predator saliva from multiple species and methods for optimal recovery from depredated carcasses*, published in the Journal of Mammalogy.

**Next Steps**—NWRC’s Wildlife Genetics Project continues to provide technical expertise and services to new stakeholders, including urban wildlife managers, local municipalities, and police departments. Researchers are also assisting WS Operations with shorebird depredation investigations and management. Future work includes a collaboration with NWRC’s Utah Field Station and Utah State University to evaluate whether prey DNA can be detected by swabbing the oral cavities of coyotes that recently fed on depredated livestock.
2021 Accomplishments in Brief
WS NWRC employs about 150 scientists, technicians, and support staff who are currently devoted to 16 research projects (see Appendix I). Below are brief summaries of select findings and accomplishments from 2021 not already mentioned in this year’s report.

**Devices**

- **Best Management Practices for Trapping.**
  Traps and trapping are an important component of wildlife damage management, wildlife research, and conservation. The Association of Fish & Wildlife Agencies published more than 20 years’ worth of research on live-restraining traps used to capture mammals in a comprehensive monograph titled *Best Management Practices for Trapping Furbearers in the United States* (Wildlife Monographs, volume 201, issue 1). WS Operations and research personnel, as well as experts from many other State and Federal agencies, were an integral part of the research supporting the trapping guidelines. The monograph describes performance data collected for 84 different trap models, including cage, foothold, and foot-encapsulating traps, as well as a power-activated foot snare. The data was collected from trapped furbearers—such as muskrats, American beavers, raccoons, and coyotes—in 33 States from 1997 to 2018.
Key findings from the publication include the following:

- Selectivity was high for all trap types.
- Most traps had high capture efficiency.
- Cage traps had the lowest average injury score.
- Added-, offset- and laminated-jaw foothold traps performed better than standard jaw models.
- Trap-related mortality or significant injury to furbearers and nontarget animals was rare.

Though the data spans two decades, interest in evaluating trap designs and trapping techniques to improve animal welfare, capture efficiency, selectivity, practicality, and user safety remains high. To ensure these tools remain available, wildlife managers and others must address concerns and knowledge gaps through public outreach, trapper education, adaptive management, ecological research, and trap research and development.

Contact: Thomas DeLiberto

- **Improved Strategies for Handling Entire Feral Swine Sounders.** As feral swine populations expand throughout North America, researchers are increasingly tasked with trapping and marking entire sounders (family groups) to monitor them and gather information for management purposes. Capture and marking procedures are challenging, dangerous for both researchers and animals, and time consuming. NWRC researchers developed an integrated pig-handling system to efficiently sort, weigh, chemically immobilize, and mark multiple feral swine simultaneously in a controlled manner. To evaluate the system’s functionality, 121 pigs of varying ages and group sizes were captured and marked over 18 capture events in Texas. Using the pig-handling system, researchers chemically immobilized 51 large pigs weighing 90–223 pounds (lbs)/41–101 kilograms (kg) and manually restrained 170 smaller pigs (less than 99 lbs/45 kg), with injury rates below 4 percent. Average handling times for large pigs was approximately 72 minutes and less than 1 minute for smaller pigs. Sounders were released intact and routinely recorded together on motion-activated cameras after release. Incorporating a handling system into wild pig research and management is encouraged to facilitate safe handling procedures for both pigs and handlers.

Contact: Michael Lavelle
Pesticides

- Repellents for Perching Birds. NWRC researchers are investigating anthraquinone (AQ), a naturally occurring plant compound, for use in surface repellents to reduce fecal contamination from perching birds. European starlings, house sparrows, and pigeons often gather in groups, resulting in hazards to human health and safety, as well as monetary losses, due to the accumulation of their feces. The acidic nature of bird feces is corrosive to building materials and can cause unsafe walking surfaces. NWRC researchers evaluated three surface-application repellent formulations with captive European starlings: Airepel HC with castor oil, an AQ-based repellent; Airepel HC with castor oil that does not contain AQ; and MS2, a novel, inert formulation with a tacky, oily texture.

Results showed all three formulations reduced fecal accumulations beneath treated perches. Researchers recommend further testing of these repellent formulations under field conditions.

Contact: Scott Werner

- Crop Features Impact Repellent Applications and Effectiveness. Blackbirds cause significant damage to sunflower crops. Although their consumption of sunflower achenes (seeds) has been reduced by more than 80 percent in laboratory trials when the seeds are fully coated with an AQ-based repellent, researchers have been unable to replicate these results with intact sunflowers or in field trials. NWRC and North Dakota State University researchers evaluated the efficacy of an AQ-based repellent that is applied directly to mature sunflower plants, using a lab-based sprayer similar to one farmers would use to treat their crops. The repellents failed to reduce damage when the treated sunflower plants were exposed to blackbirds in a laboratory setting—even in the absence of disk flowers, which keep the repellent from reaching the seeds.

Researchers also tested the ability of ground rigs equipped with drop nozzles to deposit a repellent effectively in a field setting. Applications varied by tractor speed, tank pressure, spray action, nozzle type, tank mixture, and repellent application rates. Seeds and disk flowers were collected at application to determine repellent coverage.
and residues. Results showed that repellent coverage did not differ among treatments, that the amount of repellent residue on the seeds was considerably less than that on disk flowers, and that the repellent did not reduce bird damage. Researchers note that because field techniques cannot apply enough repellent on the sunflower’s face, the AQ concentrations on the seeds are considerably lower than those shown to reduce bird damage in laboratory trials (on loose seeds). More research is needed to overcome issues related to plant shape and structure, so that repellent can be deposited more effectively.

Contact: Page Klug

- Repellent Seed Treatment To Prevent Feral Swine Damage to Corn. Feral swine damage corn more than any other crop in the United States. Most of the damage occurs immediately after farmers plant seeds, when pigs root them up to eat. NWRC researchers evaluated the effectiveness of an AQ-based repellent to reduce feral swine consumption of unplanted seed corn in Alabama and Texas. Three AQ concentrations (0.5, 1.5, and 3 percent) were tested. Results showed that the 3-percent concentration had the greatest repellency rates (95 and 59 percent in Alabama and Texas, respectively). However, repellency decreased when larger numbers of feral swine appeared at the bait sites. Several nontarget species also visited the treated bait sites. Raccoons (Procyon lotor) ate the AQ-coated corn, but white-tailed deer (Odocoileus virginianus) and mule deer (O. hemionus) did not.

Overall, the results show promise for developing an AQ-based repellent for seeds to reduce feral swine damage. Future studies will test the repellency of the 3-percent AQ concentration on seed corn planted underground.

Contact: Scott Werner

- Impacts of Sodium Nitrite on European Starlings. Sodium nitrite (SN) is an inorganic salt commonly used to cure meat. SN is the active ingredient in a toxic bait called HOGGONE, which is being evaluated in Australia and the United States for use with invasive feral swine. To better understand the impacts of SN on nontarget bird species, NWRC researchers investigated the toxicity of this chemical in European starlings. Findings
showed that SN presented a moderate toxic hazard to European starlings. The exposure of 10 starlings to the current experimental formulation of HOGGONE (10 percent SN) resulted in the death of 1 starling during 4 days of exposure to the toxic bait.

Researchers note that SN has moderate potential to be developed as a toxicant for invasive European starlings, depending on its cost-effectiveness. Future studies with other North American songbird species are recommended to determine the nontarget risk of SN toxicity at bait sites and to limit the availability of SN bait to birds from the spillage that feeding feral swine cause. Researchers note nontarget risks can be minimized by following best practices for vertebrate pest baiting.

Contact: Scott Werner

**Other Chemical and Biological Methods**

- **Determining Mosquito Blood Meal Hosts Using Genetics.** Sequencing the DNA of fresh blood meals in invertebrates, such as mosquitoes, allows researchers to detect and identify host species and pathogens. Multiple vector-borne pathogens that occur in Puerto Rico—including dengue, Zika, chikungunya, and West Nile viruses—pose a potential threat to people and animals. Identifying species that mosquitoes feed on helps present a snapshot of how urban wildlife may contribute to pathogen transmission and provides important information to public health and disease managers.

NWRC, Colorado State University, and University of Southern Mississippi researchers collected 604 blood-engorged mosquitoes from 240 traps placed throughout neighborhoods in the San Juan Metropolitan Area during 2018 and 2019. Two mosquito species were collected: the southern house mosquito (*Culex quinquefasciatus*) and the yellow fever mosquito (*Aedes aegypti*). By analyzing the DNA in the mosquitoes’ blood meals, researchers determined that the southern house mosquito fed on 17 bird species, 7 mammal species, and 1 reptile species. Yellow fever mosquitoes fed on two bird species and three mammal species. The most dominant host species were those humans have introduced, such as domestic chickens, domestic dogs, rats, and iguanas. A variety of uncommon native bird species were also detected, demonstrating that this method can also be used as a biodiversity detection tool.

These findings provide a snapshot of the animal community in the San Juan Metropolitan Area, which potentially plays a role in the spread of mosquito-borne pathogens.

Contact: Toni Piaggio
**Virus Isolation Using Chicken Eggs Can Skew Results.** The growth and cultivation of viruses (also known as virus isolation) in model organism cells or tissues for research purposes is a common practice. Unlike bacteria which can be grown on agar (an artificial nutrient medium), viruses require a living host cell for replication. For instance, influenza A viruses are often grown in embryonated chicken eggs that are free of specific pathogens. Such growth in alternative host tissues and cells, however, basically constitutes a host jump that can lead to genetic changes. Colorado State University and NWRC researchers sequenced 70 avian influenza viruses from wild birds and compared their genomes before and after the viruses were isolated in embryonated chicken eggs. Results showed that the growth of the viruses in eggs led to genetic mutations, with some mutations becoming dominant. Researchers warn that such skewed populations could lead to flawed evolutionary inferences and biased subtype detection. For genomic studies of wild bird influenza viruses, researchers recommend directly sequencing the virus from host samples instead of growing it in embryonated chicken eggs.

*Contact: Toni Piaggio*

**Framework for Invasive Species Surveillance Using eDNA.** Every year, invasive species cause billions of dollars in economic losses and other damages in the United States. A significant portion of invasive species management is dedicated to assessing the presence of these species, whether it is initial detection of alien species of concern, tracking their spread, or monitoring for survivors of eradication efforts. Advances in molecular technologies allow for detecting a species through its environmental DNA (eDNA), even when its numbers are relatively low. In 2020, the National Invasive Species Council formed a task team of nearly 30 Federal scientists and invasive species experts to summarize the usefulness of eDNA sampling and analysis for invasive species surveillance programs. The overall framework helps managers decide if, when, and how to use eDNA for surveillance—and, if it is used, how to ensure managers and stakeholders will accept the results, while clearly understanding the method’s strengths and limitations.

*Contact: Toni Piaggio*

**Standardizing Assay Limits of Detection and Quantification for eDNA.** eDNA studies often use quantitative real-time polymerase chain reaction (qPCR) to detect low levels of target species’ eDNA in water, soil, or air samples. NWRC researchers and partners proposed a standardized process and reporting method for calculating and interpreting eDNA assay limit of detection (LOD) and limit of quantification (LOQ) for single-species qPCR studies.
Standardizing how LOD and LOQ are determined, interpreted, and reported for eDNA assays allows for more informed and meaningful comparisons of results between laboratories. It also provides a means for assessing assay quality and performance. Seven independent laboratories participated in an interlaboratory comparison of LOD and LOQ for 36 eDNA assays for a variety of species. This work established standards for minimum reporting for all eDNA studies.

Contact: Toni Piaggio

- **Impacts of Surgical Sterilization on Coyote Behavior.** Coyotes (Canis latrans) that feed on livestock often do so out of necessity, to provide adequate amounts of food for their pups. Surgical sterilization methods that preserve gonadal hormones, such as vasectomies, have successfully reduced livestock depredation by free-ranging coyotes without affecting behaviors such as territoriality and mate fidelity. To learn more about the impacts of sterilization on coyote behavior, NWRC and Tufts University researchers compared the behavior and reproductive hormones of untreated captive coyote pairs to pairs that had received different surgical sterilization treatments (for instance, vasectomy, spay, neuter, and ovary-sparing spay).

The behavioral findings showed that sterilization treatments did not create intolerance between coyote pairs or break down pair bonds. Testosterone concentrations of neutered and vasectomized males differed significantly from those of intact males, indicating that the sterilization treatments were successful and that the different techniques impacted hormones differently. There were no differences in estradiol or progesterone levels among female treatment groups. No sterilized pairs produced pups, but the intact pairs did. The results show that sterilization holds potential as a future management strategy to help reduce livestock depredation.

Contact: Julie Young
• **Combined GonaCon and Rabies Vaccination for Feral Cats.** Overpopulation of free-roaming and feral cats is a global problem, negatively impacting animal health and welfare, human health, and wildlife resources. Among many other things, these cats can spread diseases—such as rabies, toxoplasmosis, and leptospirosis—to people and other animals. NWRC and Israeli researchers examined the safety and efficacy of GonaCon immunocontraceptive vaccine (GonaCon) used in combination with a rabies vaccine in 16 mature feral female cats.

Results showed that in the short term, the combined vaccinations were safe and effective in the treated cats. Over the study period, no negative health concerns were detected. There were no differences in serum rabies antibody titers among groups, and the cats kept a protective titer throughout the study. Anti-gonadotropin-releasing hormone antibodies were detected in all but one of the GonaCon-vaccinated cats. Although fertility tests were not conducted, an evaluation of vaginal cells and ovarian tissues suggested that reproduction was suppressed in GonaCon-vaccinated cats. Such a combined vaccination approach may help to reduce human and animal health risks associated with feral cats.

**Contact:** Doug Eckery

• **Training Ferrets To Detect Avian Influenza.** The spread of highly pathogenic avian influenza in commercial poultry and backyard flocks in the United States in 2015 resulted in more than $800 million in damage and control costs, as well as the lethal removal of nearly 50 million domestic birds. Fecal sampling of wild waterfowl and their habitats is an integral part of surveillance for the early detection of emerging avian influenza viruses (AIV) that pose a threat to human and poultry health. To aid in developing new tools for early detection of AIV in the environment, researchers from NWRC, Colorado State University, and Monell Chemical Senses Center trained domestic ferrets to discriminate between the feces of avian influenza-infected and noninfected mallards. Efforts are underway to similarly train detector dogs to help with early detection of the virus in the environment.

*Photo: USDA, Gail Keirn*
ferrets to discriminate between the feces of AIV-infected and noninfected mallards. Whenever they detected a marked increase of acetoin in the odor of the feces, which indicates AIV infection, the ferrets displayed a conditioned behavior (active scratch alert).

Results show that ferrets can identify this signature odor and that it is specific for AIV infection. This odor identity is not compromised by days since infection, exposure dosage, individual duck identity, or husbandry methods. These findings suggest that using trained detector dogs could add a layer of surveillance screening to the current early detection system that would improve efficiency by decreasing the number of samples tested.

Contact: Susan Shriner

Disease Diagnostics, Surveillance, Risk Assessment, and Management

- **Influenza A Virus Reassortment in Birds and Mammals.** When influenza A viruses (IAV) infect the same host simultaneously, some of their genetic segments can mix—a process known as reassortment. This process is an important source of viral diversity and can lead to the spread of viruses to new host species. To investigate taxonomic differences in IAV reassortment, NWRC and Emory University researchers examined reassortment of two distinct avian IAVs within their natural host (mallards) and a mammal (guinea pigs). The animals were inoculated with both H3N8 and H4N6 viruses, two viral subtypes typically found in mallards that also circulate in other North American waterfowl. Subsequent samples were collected from the cloacas of the mallards and the nasal tracts of the guinea pigs, and viral genetic exchange was monitored.

Results revealed abundant reassortment in mallards, giving rise to highly diverse viral populations. In guinea pigs, reassortment rates were lower, with fewer unique genotypes and lower diversity. These findings indicate that mallards provide a more suitable host environment for avian IAV reassortment than mammals.

Contact: Susan Shriner

- **Dispersal of Antimicrobial-Resistant Bacteria by Gulls.** Disease experts suspect that gulls serve as reservoirs and disseminators of antimicrobial-resistant (AMR) bacteria, given the birds’ wide-ranging movements, use of human waste sites and agricultural production systems, and known propensity to carry pathogens. U.S. Geological Survey and NWRC researchers investigated the risk of long-distance dispersal of AMR bacteria (Escherichia coli) by landfill-foraging gulls in Alaska. Fecal material from glaucous-winged gulls (Larus glaucescens), herring gulls (Larus argentatus), glaucous gulls (Larus hyperboreus), and potential hybrids of these species was collected at seven community landfills and other gull congregation areas (such as beaches). Forty-two gulls were also captured and fitted with global positioning system (GPS) transmitters to track their movements.

Using a combination of phenotypic, genomic, and animal telemetry approaches, researchers determined that gulls likely acquire AMR bacteria from landfills. They may then disperse it across and between continents through their migratory movements. The frequency of AMR Escherichia coli...
coli detections in gulls was strongly correlated with the number of people in the local community. Satellite telemetry tracking of gulls inhabiting Alaska landfills showed that during the period they shed AMR bacteria, they migrated to Russia, Canada, and California. Researchers note this technology may be useful for optimizing surveillance of AMR in the environment and minimizing its spread.

Contact: Jeff Chandler

- Responding to an African Swine Fever Detection in Feral Swine. African swine fever (ASF) is a deadly pig disease that can significantly impact swine producers, their communities, and the economy. There is no ASF treatment or vaccine available. The only tools to stop the spread of the disease are depopulation and movement bans. Because feral swine can carry and spread ASF, any response to an outbreak in the United States would include surveying and culling them. In collaboration with the National Feral Swine Damage Management Program, WS Operations, and APHIS’ Veterinary Services, NWRC researchers developed a model to predict ASF transmission in feral swine. The model considers many factors, including feral swine density, movement, interaction and contact among swine; culling capacity (for example, how many swine would need to be removed daily, based on local conditions, to prevent disease transmission); and time (for instance, the period between the ASF introduction and initial detection).

The product of this modeling effort is an application that allows users to enter values for the various factors and receive an optimal culling radius for disease elimination. The application also shows the size of the culling area and the number of feral swine targeted for removal under different management conditions to aid in ASF preparedness and planning.

Contact: Kim Pepin

- Contact Among Feral Swine: Implications for Disease Risk. Feral swine are an invasive, social species that can transmit devastating diseases such as ASF to domestic swine. Contact among individual animals plays a fundamental role in the spread of infectious disease, affecting the length and severity of an outbreak within a population. NWRC researchers and partners placed proximity loggers and GPS devices on 48 feral swine in Florida and South Carolina to determine the effects of social structure, spatial distribution (home-range overlap
and distance), sex, and management (lethal removal and baiting) on contact rates and the risk of establishing ASF in U.S. feral swine populations.

Modeling results found that social group membership was the primary factor influencing contact rates among feral swine. In fact, contact rates among members of the same social groups (sounders) were 10 times higher than those among individuals in different sounders. Fewer contacts occurred among sounders whose home ranges were greater than 1.2 miles/2 kilometers (km) apart, while no contact occurred among sounders whose home ranges were greater than 2.5 miles/4 km apart. When contact and management data were combined with ASF information to simulate disease transmission and risk, results showed that indirect contact by feral swine resulting from the use of bait stations might increase the risk of disease establishment by up to 33 percent, relative to direct contact among feral swine. Low-intensity population reduction (removal of less than 6 percent of the feral swine population) had no impact on contact rates but did reduce the risk of ASF establishment relative to no population reduction. This suggests that even low levels of ongoing management can reduce the risk of an ASF introduction taking hold. This approach provides insight for optimizing disease control in spatially and socially structured feral swine populations.

In a related study, NWRC researchers and partners from the University of Florida, Colorado State University, Archbold Biological Station-Buck Island Ranch, and APHIS’ Veterinary Services placed GPS collars on 20 feral swine and 11 cattle on a cow-calf ranch in Florida. Important microbes feral swine carry that pose a risk to cattle include pseudorabies virus, Mycobacterium tuberculosis, Brucella abortus (agent of brucellosis), and antimicrobial-resistant strains of Escherichia coli or Salmonella sp. bacteria. Researchers used movement ecology theory and network modeling to estimate the effects of sex, distance/proximity, and cattle supplement availability on contact among individual animals plays a fundamental role in the spread of infectious disease, affecting the length and severity of an outbreak within a population. NWRC researchers and partners investigated direct and indirect contact between feral swine and livestock. Photo: USDA, Wildlife Services.
rates between feral swine and cattle; they also characterized feral swine use of cattle resources that humans provide (for example, mineral supplements, molasses, and water). Results showed that despite limited direct contact between feral swine and cattle, numerous indirect contacts occurred via natural resources across the landscape. Researchers note that indirect contact could be the primary mode of disease transmission between feral swine and cattle because the two species tend to use the same habitats at different times. They also recommend excluding feral swine from liquid molasses sites to help decrease indirect contact with cattle.

Contact: Kim Pepin

• Modeling Rabies in Mongooses. Small Indian mongooses (Urva auropunctata) were introduced from Asia to several Caribbean islands during the 19th century to control rodent populations on sugar plantations. These opportunistic carnivores quickly became invasive in Caribbean ecosystems, where they cause substantial damage to native species. In Puerto Rico, Cuba, Grenada, and the Dominican Republic, mongooses are the primary reservoir for rabies virus. To help inform applied research for mongoose rabies management, NWRC and Canadian researchers created a model and conducted uncertainty analysis to identify important factors and data gaps for understanding mongoose rabies dynamics in Puerto Rico.

The analysis revealed that transmission rates, infection mortality, and the location and size of initial outbreaks strongly influence rabies virus spread and persistence in mongoose populations. Other important mongoose population variables include habitat-specific densities and habitat influences on home range and dispersal. Researchers suggest using these results to design ecological studies and collect data to (1) improve models for spatial simulation of mongoose rabies dynamics and (2) control and guide the development of management strategies targeting mongoose rabies in the Caribbean.

Contact: Amy Gilbert

As part of WS’ efforts to develop and refine tools for mongoose rabies control, NWRC researchers evaluated the rabies antibody responses of mongooses after the delivery of Ontario Rabies Vaccine (ONRAB) via Ultralite baits.

Photo: USDA, Wildlife Services
• **Evaluating ONRAB for Mongooses in Puerto Rico.** Since their introduction in the 19th century, small Indian mongooses have become agricultural pests on Puerto Rico and a reservoir for rabies virus. As part of WS efforts to develop and refine tools for mongoose rabies control, NWRC researchers evaluated the rabies antibody responses of mongooses following the delivery of Ontario Rabies Vaccine (ONRAB) via Ultralite baits. These baits are used to control rabies in other wild carnivores, such as skunks, in North America.

Ultralite baits containing ONRAB were presented to 18 captive mongooses; sham baits were given to 6. Thirteen of the 18 mongooses and all 6 that were given shams punctured and/or ate the baits. Researchers collected blood samples from the mongooses and analyzed them for rabies antibodies at 0, 14, and 30 days after vaccination. By day 30, rabies antibodies were detected in 85 percent of the mongooses that had punctured and/or eaten the baits. However, further refinement of the bait may be warranted, as some mongooses did not interact with it, while others ate it but did not produce antibodies. Modifying the bait structure and shape—for example, making it soft and more cylindrical to suit the narrow shape of a mongoose’s mouth—could help reduce vaccine spillage and inefficient contact with the oral mucosa.

**Contact:** Are Berentsen

• **Evaluating ONRAB for Raccoons and Striped Skunks.** Since the 1990s, oral rabies vaccination (ORV) has been used to halt the westward spread of the raccoon rabies virus variant from the eastern continental United States. To help expand available ORV products, NWRC researchers conducted experimental field trials in West Virginia of ONRAB for use with raccoons (*Procyon lotor*) and striped skunks (*Mephitis mephitis*). As a follow-up to the first year and U.S. experimental trials of ONRAB in rural West Virginia, this trial continued for 5 years and evaluated two bait densities (75 and 300 baits/km^2). Changes in the level of rabies virus neutralizing antibodies (RVNA) were measured in raccoons and skunks before and after exposure to ORV during each year of the trial.

The increase in bait density from 75/km^2 to 300/km^2 corresponded to an increase in average post-ORV RVNA prevalence for both raccoon and skunk populations. Raccoon population RVNA levels increased from 53 to 82 percent, and skunk population RVNA levels increased from 11 to 39 percent. Raccoon rabies virus was locally eliminated in the study area during the trial and up to 3 years post-trial. Researchers concluded that multiple years of ORV application may be needed to achieve and maintain RVNA seroprevalence in target wildlife populations for the control and elimination of the raccoon rabies virus variant.

**Contact:** Shylo Johnson

• **Rat Lungworm in Hawaii.** *Angiostrongylus cantonensis*, or rat lungworm, is the most common cause of eosinophilic meningitis in people. This condition causes headaches, a stiff neck, tingling or pain in the skin, fever, nausea, and vomiting, and can be permanently debilitating or even lethal. People usually become infected with this parasite by ingesting its larvae in raw or insufficiently cooked snails, slugs, freshwater prawns, frogs, or fish. Infection may also occur by eating contaminated fresh produce, such as lettuce. In Hawaii, all three species...
of introduced rats and a variety of native and non-native snails are known to harbor *A. cantonensis* infections. NWRC and University of Hawaii researchers sampled for *A. cantonensis* infection in wild Polynesian and black rats (*Rattus exulans* and *Rattus rattus*) and snails (*Parmarion martensi*) in Hilo. Results showed the overall infection prevalence was 86 percent in snails and 64 percent in rats (77 percent in Polynesian rats and 48 percent in black rats). Infections varied with environmental and host-related factors. Body mass was a strong predictor of infection in all three species, with different patterns seen between sexes and species of rats. Infection prevalence and intensity for snails were high in May and February, but generally lower and more variable during the intervening months. Understanding how infections may vary across host populations aids in future disease surveillance and targeted management strategies.

**Contact:** Shane Siers

### Wildlife Damage Assessments

- **Using Weather Radar To Predict Bird Damage.** Weather radar isn’t just for forecasting the weather. Next Generation Weather Radar (NEXRAD) is also used by researchers to track large flocks of blackbirds, which could help reduce bird damage to sunflower crops. Using weather surveillance radar data from 2012 to 2019, NWRC, University of Oklahoma, and University of Notre Dame researchers monitored one large blackbird roost near Bismarck, ND.

  Results showed the blackbird numbers routinely peaked in mid- to late October—around the same time unharvested sunflower crops reach maturity. The estimated maximum number of blackbirds in the flock ranged from nearly 347,000 to more than a million per day. Researchers determined that if producers could harvest their sunflower 2 weeks earlier, they could save as much as $1,800 a year in damages from this single roost. Planting early, choosing early maturing sunflower varieties, and desiccating the crop could result in an earlier harvest to avoid peak blackbird numbers. Researchers emphasize that a harvest coordinated among neighbors would be vital to the success of an earlier approach. If one neighbor harvests early but others do not, the birds will still find sunflower fields, and those producers may suffer severe bird damage. Predicting blackbird arrival can help producers plan dates for planting, harvesting, and implementing management tools before large flocks arrive. These methods could be expanded to other crops that blackbirds damage, such as corn and rice.

  **Contact:** Page Klug

- **Feral Swine Impacts to Water Quality.** Riparian habitats play a vital role in filtering pollutants and sediment from water, which improves water quality and ensures adequate nutrient cycling. Riparian areas also provide valuable habitat for plants and animals, surface water storage, resources for agriculture and livestock production, and recreational opportunities for people. However, the livestock and wildlife living in and near riparian areas can contaminate streams through direct contact or indirectly through runoff polluted with feces and urine. Use of such contaminated water has been linked to disease outbreaks in people. Feral swine may significantly contaminate and alter the functionality of riparian ecosystems by digging and overturning soil, which can lead
to increased erosion and runoff. NWRC and Auburn University scientists compared the water quality of streams from watersheds on a privately owned property in Alabama with a dense pig population and a nearby national forest without an established pig population. Water samples were collected twice monthly for a year.

Results showed that watersheds with feral swine had elevated dissolved organic carbon and total nitrogen levels attributable to feral swine feces. Furthermore, watersheds with feral swine had *Escherichia coli* values that were 40 times higher than watersheds without feral swine. This study is the first to definitively link feral swine to the introduction of fecal material and waterborne pathogens in watersheds.

**Contact:** Kurt VerCauteren

- **Using Federal Crop Insurance Data To Estimate Wildlife Damage.** Wildlife damage to crops is a persistent and costly problem for many U.S. farmers that varies substantially across crops, regions, and years. Most existing estimates of crop damage have relied on field studies conducted by trained biologists or surveys distributed to farmers. However, NWRC researchers developed a new method of estimating wildlife damage that uses Federal crop insurance data. As a case study, researchers estimated damage for corn, soybean, wheat, and cotton—all economically important crops that are vulnerable to wildlife damage. The combined loss for the four crops was estimated at $593 million in 2017. The highest total estimated losses from wildlife were in soybeans ($324 million) and corn ($194 million); the highest estimated percentage losses were in soybeans (0.87 percent) and cotton (0.72 percent). The eastern and southern regions of the country were clearly the most susceptible to wildlife damage. Using crop insurance data is a reliable way to evaluate differences in crop
damages for the coming years. A better understanding of damage differences can also help guide research and development of new management techniques.

Contact: Stephanie Shwiff

- **Why Some Animals Thrive in Urban Areas.** Some species, such as striped skunks, raccoons, and coyotes, thrive in urban areas. These adaptable species are considered “ecological generalists” that can readily modify their diets and habitat use—traits that allow them to live in challenging environments, including urban settings, where they may encounter novel, complex, and changing situations. To better understand these species’ adaptive behaviors, NWRC, university, and artificial intelligence researchers conducted trials at NWRC facilities in Colorado and Utah with an automated device that measured the ability of these animals to alter their behavior in changing circumstances. Designed by the researchers, the device included two easily accessible buttons placed on either side of a food-dispensing chute. Captive raccoons, skunks, and coyotes were encouraged to press one of the two buttons to receive a food reward. Once the animals learned to press the correct button for food, researchers reversed the button settings.

Raccoons in the study generally engaged with the device and successfully navigated the change. Although the sample size for skunks was limited, researchers found that they were willing to approach and engage with the device rather quickly. Nevertheless, skunks did not complete a high number of reversals: only one of three demonstrated learning. Despite efforts to habituate six coyotes and motivate them to interact with the device, only one did so. This study demonstrates that raccoons and skunks have the intellectual flexibility to adapt their
behavior by forming and then reversing learned associations.

Contact: Shylo Johnson

- **Black Vulture Conflict and Management in the United States.** Black vulture (Coragyps atratus) populations are increasing and expanding their range in North America. This development, combined with the birds’ ability to adapt well to human landscapes, has contributed to increasing numbers of human-vulture conflicts. NWRC researchers collaborated with WS Operations biologists to summarize the status of and trends in black vulture conflicts, review available management strategies, identify knowledge gaps, and provide recommendations to enhance management and understanding of this species.

Results showed vulture conflicts with livestock are on the rise, as well as vulture damage to private and public property and collisions between vultures and aircraft. Researchers have learned more about rancher perceptions of vulture predation on livestock, including estimates of economic damage and use of mitigation strategies. However, a basic understanding of the underlying mechanism driving the conflict and the evaluation of existing tools and methods to mitigate damage are limited. For damaged property, little information is available about the economic losses, stakeholder perceptions, or tool efficacy. Regarding aviation safety, recent research on the flight behavior of black vultures has direct implications for reducing aircraft collision risks. Knowledge is still limited about which factors influence vulture roost site selection and the most effective ways to leverage the species’ sensory ecology and behaviors to reduce damage.

Contact: Bryan Kluever

- **Vulture Roosts.** Turkey vultures (Cathartes aura) and black vultures often roost in groups.
Two factors that may influence where they choose to roost are air currents and distance to food sources. A roost near roads, for instance, may be an attractive option because it provides the thermal currents that emanate from paved surfaces as well as carrion from vehicle collisions. To assess the role of human-based and natural landscape features on roosting habitat selection, NWRC researchers and partners analyzed data from 11 black and 7 turkey vultures fitted with GPS satellite transmitters. Results showed that habitat fragmentation and diversity influenced the roosting choices of both species in all seasons. Turkey vultures were most likely to roost at intermediate road densities in three of four seasons, but black vultures showed a positive relationship with roads only in fall. Both species were increasingly unlikely to roost as the landscape became more urbanized. These findings inform wildlife managers where current and future roosts may likely occur.

Contact: Bryan Kluever

**Cost of Cormorant Damage to Catfish Farms.** To reduce losses, catfish farmers in the southern United States often attempt to scare away fish-eating birds, such as double-crested cormorants (*Phalacrocorax auratus*), from their farms. Despite these efforts, cormorants continue to feed on farmed catfish. NWRC and university scientists conducted an economic analysis to determine the costs of bird damage management, based on survey responses from 88 percent of all farms in the Delta region of Mississippi and Arkansas. These farms account for more than 60 percent of all catfish production nationwide. Estimates of the revenue lost from catfish eaten by cormorants were developed from a concurrent study on cormorant distribution, abundance, and diet in the region.

Catfish farmers spent on average $704/ha (plus or minus $394/ha) to scare birds, making it one of the top five costs of raising catfish. The most expensive aspects of scaring birds were manpower (39 percent of all bird-scaring costs) and trucks used to scare birds (34 percent). The profitability of the catfish farms improved by 4–23 percent over 27 different production categories when the effects of bird predation were removed. In fact, all but one of the previously unprofitable farm production categories became profitable. Industry-wide, catfish losses averaged $47 million. Total direct economic effects (including both the increased costs to scare birds and the revenue lost from fish eaten by cormorants) averaged $65 million. This information is
useful to fish farmers, policymakers, and others striving to reduce the economic damages associated with fish-eating birds.

Contact: Brian Dorr

- **Cost of Scaup Damage to Baitfish and Sportfish Farms.** Although the lesser scaup (*Aythya affinis*) is not widely known to be a fish-eating bird, it will eat farmed fish. NWRC researchers estimated the cost of scaup damage to various species of baitfish and sportfish using data on the abundance, distribution, and dietary habits of scaup that visited Arkansas baitfish and sportfish farms during the winters of 2016–2017 and 2017–2018. Total annual costs to scare birds from baitfish and sportfish farms were $622 (plus or minus $742/ha). The greatest costs of scaring birds were manpower (56 percent), truck usage (32 percent), levee upkeep for vehicle access to scare birds (9 percent), firearms and ammunition (2 percent), and pyrotechnic devices (1 percent). The combined annual economic losses, calculated as reduced revenue from fish losses to scaup plus expenditures to scare birds, averaged $683/ha for golden shiners (*Notemigonus crysoleucas*), $695/ha for fathead minnows (*Pimephales promelas*), $663/ha for sportfish, and $673/ha for goldfish across the 2 study years. The fish losses to scaup alone averaged over $1 million per year for the Arkansas baitfish industry. Total estimated costs to the Arkansas baitfish industry on average were $5.5 million per year. This study provides important estimates of the costs related to ongoing and possibly increasing conflicts between lesser scaup and baitfish and sportfish farms.

Researchers also used the data to analyze and model the distribution and abundance of scaup using baitfish and sportfish farm characteristics. Results showed that scaup appeared more frequently at larger golden shiner and fathead minnow ponds stocked at greater densities. Farm-level models suggested that farms further from major rivers and with an average pond size of approximately 20 acres/8 ha were most likely to attract scaup. Producers can apply these findings to implement bird harassment efforts at times and locations where scaup predation is more likely.

Contact: Brian Dorr

- **Food Habits of Wintering Cormorants in the Mississippi Delta.** Double-crested cormorants impact U.S. commercial aquaculture and are considered the primary bird predator in catfish aquaculture facilities in the Mississippi Delta. The Delta covers 35,000 square miles and includes parts of Mississippi, Arkansas, and Louisiana. Recent changes in aquaculture practices, regulatory policies, and decreased overall acres in production prompted NWRC and university researchers to assess cormorant consumption of catfish in relation to their night roosts. Sixty-nine cormorants were collected from night roosts from October through April, coinciding with peak cormorant migration and seasonal residency within the region.

Stomach content analysis showed, on average, that catfish made up 33 percent of a cormorant’s overall diet, which is less than reported in previous studies. There was no difference between the amount of channel catfish (*Ictalurus punctatus*) versus hybrid catfish (*I. punctatus x I. furcatus*) eaten. Most catfish were eaten during the months of February and March. Analysis showed the best model for predicting catfish consumption was based on the amount of catfish aquaculture within 19 miles/30.6 km
of a night roost. This research also shows that catfish consumption drops rapidly for cormorants from roosts that are more than 6 miles away from farms. These findings will help inform cormorant management decisions. Researchers note that even though cormorants have shifted their diet to more naturally occurring fish species, aquaculture remains an important regional food source.

Contact: Brian Dorr

- **Estimating Wildlife Strike Costs at U.S. Airports.** Costs associated with aircraft-wildlife collisions, or wildlife strikes, in the United States are widely acknowledged by the aviation community. Estimating the cost of wildlife strikes to civil aviation relies on strike- and cost-related information reported by aircraft operators and airport staff to the Federal Aviation Administration’s National Wildlife Strike Database (NWSD). The accuracy of these estimates, however, is undermined by the skewed nature of reported cost data. Often, these estimates also fail to account for differences in observed strike characteristics (for example, type of aircraft, size of aircraft, type of damage, and size of the animal struck). NWRC and Colorado State University economists used modern machine-learning techniques to provide a more accurate measure of strike-related costs accrued by the U.S. civil aviation industry. Using NWSD data from 1990 to 2018, machine-learning models determined that wildlife strikes cost a minimum average of $54 million annually. This estimate is lower than previous estimates ($187 million) calculated using more traditional statistical methods.

Contact: Stephanie Shwiff

Wildlife Management Methods and Evaluations

- **Use of Sonic Net To Disperse Blackbirds.** Blackbird damage to agricultural crops is common across the United States. NWRC and College of William and Mary researchers evaluated the use of a novel tool called a Sonic Net to deter mixed-species blackbird flocks from predating on maturing
sunflower crops in North Dakota. The Sonic Net masks communication among birds by delivering “pink noise”—basically white noise (with reduced higher frequencies) that overlaps with the frequencies the birds use to communicate with one another. If birds cannot hear predators or the warning calls of other birds, their perceived predation risk increases, and they relocate to an area perceived to be less risky. Sonic Nets were placed in three sunflower fields actively used by mixed blackbird flocks. Each field contained two 0.2-ha plots with individually marked sunflowers. After treating 1 plot per field for 20 days with the Sonic Net, researchers measured the total area damaged on the marked sunflowers in all 6 plots.

The Sonic Net treatments reduced damage to sunflowers by 27 percent, 64 percent, and 23 percent, respectively. Researchers predict that the effect of the Sonic Net treatment may be greater in other crop phases and types, such as in the establishment phase or ground cover crops. Both lack high vegetation, which can obstruct the Sonic Net’s sound.

Contact: Page Klug

Avoiding Nontargets While Baiting Feral Swine. Toxic baiting of invasive feral swine (*Sus scrofa*) is a potential new tool for population control and damage reduction in the United States. However, there are concerns that nontarget species, especially songbirds, may be exposed to these baits.
NWRC researchers conducted an analysis of a 2018 baiting study to identify daily and landscape factors that may have influenced the use of bait sites by nontarget songbirds, ground birds, raccoons, and white-tailed deer. Results showed that no single strategy can prevent all these species from visiting bait sites. However, offering the bait at night, in feral swine-specific feeders, and in more uniform habitats can minimize songbird visits. Researchers also recommend placing bait stations away from sites where residual particles of grain from pre-baiting are available on the ground, removing spilled bait as quickly as possible, and employing a bird frightening device the morning after toxic baiting. Managers can increase feral swine visits to sites by allowing time for pigs to attract other pigs to the bait sites and baiting along linear habitat areas, such as edges of crop fields or riparian corridors.

**Contact:** Nathan Snow

### Hunter Survey Respondents

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<th>Percentage</th>
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<tr>
<td>91%</td>
<td>were white</td>
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<td>96%</td>
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<tr>
<td>52</td>
<td>average age (in years)</td>
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<tr>
<td>58%</td>
<td>bachelors degree or higher</td>
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<tr>
<td>73%</td>
<td>hunted feral swine</td>
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<tr>
<td>48%</td>
<td>owned or managed land (of those, 32% experienced feral swine damage)</td>
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<tr>
<td>11%</td>
<td>trapped and sold live feral swine</td>
</tr>
<tr>
<td>1%</td>
<td>provided feral swine guiding or outfitting services to paying hunters</td>
</tr>
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</table>

More than 37,000 Texas resident and nonresident licensed hunters were surveyed about their tolerance for feral swine.

Graphic: USDA

**Hunter Tolerance for Feral Swine.** In the United States, recreational hunters play a unique role in both controlling and spreading invasive feral swine. Many States encourage hunting to help manage feral swine populations; others may want to maintain or establish feral swine populations for future hunting opportunities. NWRC and Texas A&M AgriLife Extension Service researchers surveyed 37,317 Texas resident and nonresident licensed hunters about their “tolerance” for feral swine—that is, their ability and willingness to accept the costs or negative aspects of living with wildlife, and their desire for positive interactions with wildlife. A common indicator of tolerance is often the maximum acceptable number of target animals in an area. Of those surveyed, 83 percent were intolerant of feral swine: about 63 percent preferred to see the swine population reduced and 20 percent preferred to see it completely removed. Sixteen percent of hunters were tolerant of feral swine: 14 percent preferred to see the population remain the same and 2 percent preferred to see it increase. Researchers found that hunter tolerance could largely be explained by:

- motivations and preferences for hunting feral swine
- level of concern for feral swine damage
- overall attitudes toward feral swine

The most important motivations for hunting feral swine were to obtain a trophy animal, followed by obtaining meat. Respondents’ overall concern about feral swine damage was high, with the greatest concern about damage to pastures. Their overall attitudes toward feral swine were largely negative. Researchers note, however, that this should not be interpreted to mean that most hunters
support efforts to rid Texas of feral swine. This finding has significant implications for feral swine management in Texas, as hunters may resist efforts focused on eradication rather than strategic population reduction. Results of this research are also useful in expanding current knowledge about human tolerance for wildlife, including species that are non-native and invasive, and in identifying important factors affecting how hunters perceive and interact with feral swine.

Contact: Stephanie Shwiff

- **Evaluating the Effectiveness of Mountain Beaver Management.** Mountain beavers (*Aplodontia rufa*) are burrowing rodents found in the humid, wet forests and steep mountain areas of the Pacific Northwest. Their clipping and browsing of new conifer seedlings reduce seedling growth or kills trees, which reduces stocking density and delays stand development. NWRC and Oregon State University researchers evaluated the effectiveness of trapping and toxicant baiting to reduce mountain beaver damage across two mountain ranges in western Oregon. Damage estimates were collected on untreated and treated forest plots containing Douglas fir (*Pseudotsuga menziesii*) seedlings at 1-, 6-, and 12-month intervals after planting and the start of management activities.

Overall, mountain beavers damaged 76 percent and 46 percent of seedlings in untreated and treated plots, respectively. Seedling heights also differed after 1 year: seedlings in untreated plots were 4 inches/10.6 centimeters (cm) shorter on average than those in areas with trapping and baiting. The mean cost associated with preventing mountain beaver damage through trapping and baiting in the study was $62/acre ($154/ha)—less than the estimated mean cost of $74/acre ($182/ha) to replant seedlings in gaps where damage occurred. The study indicates that an integrated, proactive approach that includes trapping and baiting may be less expensive overall than the reactive approach of replanting seedlings in gaps created by mountain beavers.

Contact: Jimmy Taylor

- **Evolutionary Impacts of Human–Wildlife Conflict.** Human–wildlife interactions, including conflicts, are increasingly common as growing urban and suburban areas create more opportunities for these encounters. Wildlife damage management techniques often aim to deter, relocate, or remove the animals causing the conflict, which may lead to selective pressures that shape animal population size, structure, and distribution. Moreover, the intensity of conflict management can vary considerably by species, public perception, policy, religious and cultural beliefs, and geographic region. NWRC and university researchers developed a conceptual model that combines human–wildlife conflict, wildlife management, and urban evolution to address how certain processes drive wildlife adaptation in cities.
Investigating human-wildlife conflict as an evolutionary phenomenon may provide insights into how conflict arises and how management plays a critical role in shaping urban wildlife characteristics.

Contact: Stewart Breck

- **Using Beavers as a Management Tool.**

  Using beavers as natural engineers to increase damming is becoming a popular management strategy known as beaver-related restoration. Methods used include beaver translocations, mimicking beaver dams with human-made equivalents, and increasing woody food/construction material for beavers. Expected outcomes from increased beaver dams are highly variable but may include improving watershed conditions (the result of raising the water table in valley floors), improving fish and wildlife habitat, and reducing flood levels. However, such expectations are rarely stated, and most are not evaluated after beaver-related restoration is implemented. Without proper planning and understanding of all the natural
processes at work, the actual success or failure of beaver-related restoration remains unknown. For such efforts to succeed, beavers must survive, remain in place, and build dams that ultimately result in desired conditions. A multidisciplinary team of researchers recently developed an assessment framework to support beaver-related restoration. Applying this adaptive management framework will help managers determine if beaver-related restoration is appropriate, and if so, help them achieve the desired outcomes. The researchers note that because nature is intrinsically messy, successfully implementing beaver-related restoration requires practitioners to state their goals and acknowledge that the responses of animals and landscapes are often unpredictable.

Contact: Jimmy Taylor

- **Factors Leading to Successful Island Rodent Eradications.** Eradications of invasive rodents on islands eliminate their negative impacts on native plants and animals. However, these efforts have been less successful in the tropics than in temperate regions, triggering research and reviews. A team of international scientists, including a researcher with the NWRC Hawaii Field Station, evaluated 35 eradication attempts on 17 tropical islands. Researchers compared the project management of each attempt, as well as operational and environmental factors. The efforts that failed were characterized by operational faults, such as poor planning, low quality bait, and gaps during bait application. In some cases, the operational faults were unequivocally the cause of the failure; in others, it was difficult to discriminate operational faults from confounding environmental factors (for example, land crabs eating bait or year-round rodent breeding). A leading cause of failure appeared to be that some rodents did not receive a lethal dose of toxin. Though this problem can arise on both temperate and tropical islands, there may be less room for errors (such as gaps in bait coverage) on tropical islands, where land crabs can eat bait.

The findings on factors leading to eradication successes (for instance, expert-reviewed plans, realistic funding and permits, and high standard baiting operations) reflect current best practice recommendations. Researchers note that strict adherence to best practices can increase overall rates of eradication success.

Contact: Shane Siers

- **Hard Versus Soft Bait for Rodent Eradications.** Rodenticide-based eradication attempts can fail if rats do not eat enough bait. A recent review of a failed attempt to eradicate invasive Polynesian rats from Wake Atoll suggests that some rats may not have eaten a lethal dose of rodenticide due to dietary and/or sensory preferences developed from regular access to human food. Human food may be higher in fats and oils and be softer or chewier than the hard pellet formulation of the rodenticide Brodifacoum 25W Conservation (B-25W) used in the eradication attempt.

To test this theory, NWRC researchers captured rats from two areas on Wake Atoll: one where rats may have regular access to human food, and another uninhabited part of the island where rats presumably have less access and, therefore, are less likely to be preconditioned. The rats were fed both a "soft" sachet formulation of a brodifacoum-based bait (FINAL Soft Bait with Lumitrack) and the harder pellet formulation of B-25W.
The rats overwhelmingly preferred the pellet formulation. No rats in the trial ate any of the FINAL bait, and all the rats that ate B-25W died. While these results suggest that any future baiting operations on Wake Atoll should continue to use hard pellets, researchers caution that dietary preferences of local rodent populations may differ on other islands. In these cases, alternative baiting choices may be more appropriate and effective.

Contact: Shane Siers

Wildlife Population Monitoring Methods and Evaluations

- Asian Longhorned Tick Surveillance. *Haemaphysalis longicornis*, the Asian longhorned tick (ALHT), is native to eastern Asia but has become invasive in several countries, including Australia, New Zealand, and the Eastern United States. The medical and veterinary communities are concerned about the establishment of ALHT in the United States because of its potential as a livestock pest and vector for disease. WS research and operations personnel, in cooperation with other collaborators, conducted ALHT surveys on potential wildlife hosts in Virginia and New Jersey. This surveillance found 51 ALHT-infested animals, including raccoons (*Procyon lotor*), Virginia opossums (*Didelphis virginiana*), red foxes (*Vulpes vulpes*), woodchucks (*Marmota monax*), eastern cottontail rabbits (*Sylvilagus floridanus*), striped skunks (*Mephitis mephitis*), and white-tailed deer (*Odocoileus virginianus*). Data also confirmed that ALHTs in all three of their active life stages (larva, nymph, and adult) were present.
on numerous hosts and in their habitats. Because many of these wildlife hosts are peridomestic (frequently living near people and human habitats), ALHT could become an issue for domestic animals and people.

Contact: Sarah Bevins

- **Factors Influencing Pronghorn Fawn Survival.** Pronghorns (*Antilocapra americana*) are an iconic symbol of U.S. deserts and plains and a valued game animal for hunters in many Western States. In Idaho, pronghorn populations have not rebounded from intensive hunting in the late 1980s. Because fawn survival impacts population growth, NWRC, Utah State University, and Idaho Department of Fish and Game researchers investigated factors affecting the survival rate of 217 radio-collared pronghorn fawns in Idaho. Data showed the leading cause of fawn mortality was coyote (*Canis latrans*) predation (58 percent), followed by unknown causes of mortality (18 percent), predation by unknown animals (12 percent), predation by bobcats (*Lynx rufus*, 6 percent), predation by golden eagles (*Aquila chrysaetos*, 3 percent), and other causes (3 percent). Models showed that the body mass index of newborn fawns and levels of 2,6-diaminopimelic acid in the fecal material of pronghorn mothers—both of which are linked to diet quality—were positively related to fawn survival. Models also showed that the presence of more rabbits and hares resulted in greater fawn survival, likely because they served as an alternative prey for coyotes and other predators. Researchers conclude that management actions to enhance forage quality or restore habitat for higher quality forage may aid in reestablishing pronghorn herds.

Contact: Eric Gese

- **European Starling Use of Urban and Rural Landscapes.** Since their intentional introduction into the United States in the 1800s, European starlings (*Sturnus vulgaris*) have become the fourth most common bird species in both urban and rural areas. Wildlife resource managers need better information about starling movement and habitat-use patterns to effectively manage populations of these birds and the damage they cause. NWRC researchers compiled data from six radio-telemetry studies conducted between 2005 and 2010 to compare the movements and habitat use of radio-tagged starlings in urban and rural habitats.

Key findings indicated that urban roosts contained smaller numbers of birds (fewer than 30,000) than more rural roosts (more than 100,000). Birds from city-center roosts occasionally switched to the outlying major roosts. Human-related food sources (for instance, feedlots, shipping yards, and landfills) were their primary foraging sites. Birds traveling to roosts from primary foraging sites in rural landscapes would often pass over closer minor roosts to reach major roosts in stands of emergent vegetation in large wetlands. The minimum estimated home range for tagged birds was
approximately 60 miles². Wildlife resource managers can use this information to predict potential roosting and foraging sites—and areas to monitor when carrying out programs in different landscapes.

**Contact:** Page Klug

- **Genetic Variation in European Starlings.** European starlings were introduced to New York in 1890 and subsequently became one of the most widespread and numerous bird species in North America. Genetic comparisons of starling individuals and populations can identify factors that helped facilitate this rapid and successful expansion. NWRC and Cornell University researchers investigated patterns of genomic diversity and differentiation using genome sequencing of 166 starlings from dairies and feedlots in 17 States. Consistent with this species’ high dispersal rate and history of rapid expansion, researchers found few genetic differences among birds from different locations. However, researchers did find some gene sequences correlated with temperature and/or precipitation, suggesting that local adaptation may have evolved rapidly. This survey of genomic signatures of expansion in North American starlings is the most comprehensive to date and complements ongoing studies of worldwide local adaptation in these highly dispersive and invasive birds.

  **Contact:** Scott Werner

- **Feral Swine Movement Behavior and Resource Selection.** Understanding how invasive species such as feral swine move through and use resources in the landscape gives insights into how their populations survive and expand. It also allows managers to predict the movements of animals in different landscapes and optimize
damage management activities. NWRC and Savannah River Ecology Laboratory researchers used GPS data from 49 feral swine in the Southeastern United States and hidden Markov models to define movement paths and assign behaviors (such as resting, foraging, and traveling) for individual swine. They also compared the connection between these behaviors and food availability (for instance, in seasons offering high or low access to forage).

Female feral swine were most active during twilight hours in the high-forage season and showed more variable movements in the low-forage season, while male feral swine exhibited nocturnal activity patterns in both seasons. The feral swine used bottomland hardwoods and dense canopy cover while resting, foraging, and traveling during both seasons. Males used shrub and grassy habitats, as well as bottomland hardwoods, while foraging in the low-forage season compared to the high-forage season and used roads, paths cut for power lines, and streams more often than females while traveling. Feral swine establish populations and home ranges in a variety of landscapes, but these results show that male and female pigs exhibit clear differences in movement behavior. As a result, managers can increase the effectiveness of techniques, such as trapping and toxicant baiting, by targeting feral swine in habitats where they prefer to forage or travel.

Contact: Kim Pepin

- **Black Bear Impacts on Mountain Lion Feeding Behavior.** Black bears (*Ursus americanus*) and mountain lions (*Puma concolor*) are both considered apex predators (species that have no natural predators). For more than 80 years, mountain lions have been the sole apex predators in the Great Basin of Nevada. However, black bears have recently recolonized the area and are known to scavenge on mountain lion kills. To evaluate the impacts of these bears on mountain lion foraging behavior in the Great Basin, NWRC researchers and partners in Utah and Nevada investigated kill sites of 31 mountain lions between 2009 and 2017, in areas with different bear densities. The researchers analyzed both the number of nights mountain lions spent feeding on a
particular prey item and the type and proportion of prey in their diet.

Results showed that the duration of mountain lion feeding bouts was driven primarily by the size of the prey being eaten, local bear density, and the presence of dependent kittens. The proportion of mule deer in mountain lion diets across all study areas declined over time, was lower for male mountain lions, increased with the presence of dependent kittens, and increased with higher bear densities. In sites with feral horses (*Equus ferus*), a novel large prey, mountain lion consumption of these animals increased over time. These findings suggest that higher bear densities over time may reduce mountain lion feeding bout durations and influence prey selection when alternative, but more dangerous, large prey are available.

*Contact: Julie Young*

**Registration Updates**

- **Broadening Acetaminophen Application Scenarios for Brown Treesnake Control.**

In 2003, acetaminophen was registered as a pesticide by the U.S. Environmental Protection Agency (EPA) for use in brown treesnake control on Guam. The original approved usage was limited to inserting single 80 milligram (mg) tablets into dead newborn mice and hand-placing them in PVC pipe bait stations in and around forested areas and along fence lines. Over the years, the label has been amended to

Some invasive brown treesnakes in Guam weigh up to 2,000 grams. NWRC research indicates that these larger snakes require higher doses of registered acetaminophen to ensure mortality. In 2020, the EPA label was amended to allow multiple acetaminophen tablets to be applied per bait when targeting unusually large brown treesnakes.

Photo: USDA, Shane Siers
allow for (1) manual or automated aerial dispersal of baits from aircraft over large areas or places not practically accessible by ground-based methods, (2) bait stations in urban and residential areas, and (3) the use of alternative baits, including rodents and birds of all sizes, lizards, and artificial bait. Recent NWRC research confirmed that a single 80 mg dose, if ingested, is lethal to brown treesnakes under 250 grams, which make up most of the snakes on Guam. However, some individual snakes grow much larger (up to 2,000 grams), and research indicates that these snakes require higher doses to ensure mortality. In December 2020, the label was amended again with EPA to allow multiple tablets to be applied per bait when targeting unusually large brown treesnakes and to permit more flexibility in bait station spacing. These label modifications allow for a broader range of application scenarios and more effective targeting of these invasive snakes.

Contact: Emily Ruell

Feral Swine Toxicant Development Update. WS continues to make steady progress toward registering a toxic bait for feral swine called HOGGONE, which contains sodium nitrite (SN) as the active ingredient. The program has partnered with Australia’s Centre for Invasive Species Solutions and Animal Control Technologies Australia to have HOGGONE registered for operational use in that country. Meanwhile, NWRC is working through the EPA requirements for a U.S. registration. Although effective at reducing feral swine numbers, the initial HOGGONE formulation resulted in unacceptable levels of nontarget hazards to passerine birds. This led NWRC researchers to modify the bait station, bait formulation, and baiting strategy, all of which have been evaluated in small-scale field trials in Alabama, Texas, and Queensland, Australia, during the last 2 years. The bait station has been modified to accept small, compacted trays that limit the ability of feral swine to spill the bait on the ground while feeding. HOGGONE (now renamed HOGGONE 2) has also been reformulated to reduce the risks to nontarget species by (1) increasing the microencapsulation coating around the SN, (2) decreasing the SN concentration by 50 percent to minimize the amount of SN deployed, and (3) using more finely milled grains to reduce the bait’s attractiveness to birds. The baiting strategy has been modified to reduce the attractiveness of the bait sites to nontarget animals by (1) decreasing the amount of pre-baiting time, (2) increasing the distance between pre-baiting sites and bait stations to avoid any remnant particles of whole-kernel corn that might attract birds, and (3) incorporating a deterrent device to scare nontargets away until a biologist can arrive to remove any spilled bait. EPA approved an amendment to the experimental use permit (EUP) for HOGGONE 2 in April 2021; NWRC researchers tested the above modifications with HOGGONE 2 in the field in northern Texas and southern Alabama in the summer of 2021. The EUP study’s final report will be submitted to EPA as part of the future Federal (Section 3) registration application for HOGGONE 2.

Concurrently, NWRC continues working to complete the remaining registration data for the Section 3 registration application, including an additional winter-spring field trial in 2023, product chemistry, ecological effects, toxicology, and food residue data required after EPA designated HOGGONE a “food-use” pesticide in 2018. The reformulation of HOGGONE into HOGGONE 2 also
necessitated repeating some of the product chemistry registration data developed for the original formulation.

WS anticipates submitting the full year-round Section 3 registration application to EPA in the fall of 2023. EPA has 25 months to evaluate the submitted registration data and food tolerance petitions. WS anticipates a final decision from the agency by late 2025 or early 2026.

Contact: Emily Ruell

**GonaCon-Deer Pesticide Label Updated.**

In April 2021, the EPA approved a label amendment for the GonaCon-Deer registration. The label changes will help improve field use of GonaCon-Deer to manage white-tailed deer. Changes include:

- allowing for “booster” doses to be administered by hand injection or remote darting. (The first vaccination of a female deer must still be administered by hand injection.)
- clarifying language that requires the marking of vaccinated animals.
- adding instructions for remote darting, including requiring that applicators retrieve darts whenever possible.

We expect use of remote darting to improve the feasibility and cost effectiveness of controlling deer populations with GonaCon-Deer. Hand injection of GonaCon-Deer is costly and time consuming; it also limits the percentage of a population that can receive booster vaccinations, thereby reducing the effectiveness of the product for population control. Also, hand injection requires capturing and immobilizing the animal, which can be a safety risk to animals and applicators. Allowing booster doses to be administered to marked animals via remote darting removes the need to repeatedly capture and immobilize the same animal.

Contact: Emily Ruell

**Technology Transfer**

- **Patents, Licenses, and New Inventions.**

In fiscal year (FY) 2021, NWRC scientists were awarded one U.S. patent and eight foreign patents. In addition, NWRC scientists submitted four utility patent applications. See the following table for details on issued patents and patent applications. NWRC scientists also submitted three U.S. provisional patent applications and one invention disclosure to the NWRC Technology Transfer Office.

Contact: John Eisemann
### Patent Table

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<th>INVENTION TITLE</th>
<th>NWRC INVENTORS AND COOPERATOR CO-INVENTORS</th>
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Technology Transfer Agreements.
WS partners with universities, private companies, and others to promote research and development for new products that help manage wildlife damage. WS formalizes these partnerships through a variety of intellectual property agreements. In FY 2021, NWRC entered into two Confidentiality Agreements, four Data Sharing Agreements, six Material Transfer Agreements, eight Material Transfer Research Agreements, and two Cooperative Research and Development Agreements.

Contact: John Eisemann

Awards

2021 NWRC Publication Award. Each year, the NWRC Publication Awards Committee, composed of NWRC scientists, reviews over 125 publications generated by NWRC colleagues. The resulting peer-recognized award honors outstanding contributions to science and wildlife damage management. In 2021, the committee presented the award to Dr. Kurt VerCauteren for his work on the book *Invasive Wild Pigs in North America: Ecology, Impacts, and Management* (CRC Press, 479 pp).

This publication arose from a multidisciplinary collaboration between NWRC, the University of Georgia, Auburn University, Michigan State University, and Mississippi State University. VerCauteren and his co-editors assimilate and organize information on wild pigs (also known as feral swine)—the most destructive vertebrate species ever introduced into the United States. The book addresses all aspects of wild pig biology, ecology, damage, and management in a single, comprehensive volume that managers, researchers, policy makers, and other stakeholders can build on. The impact of this book on WS operations, stakeholders, the public, and future research endeavors will be substantial.

NWRC Employee of the Year Awards.
The winners of this award are nominated by their peers as employees who have clearly exceeded expectations in their contributions to the NWRC mission. The winners this year are:

- Dr. Susan Shriner
  research grade scientist
  Wildlife Disease Dynamics, Epidemiology, and Response Project
  Fort Collins, CO

- Dr. Tim Smyser
  support scientist
  Wildlife Genetics Project
  Fort Collins, CO

- Kathlyn Stauffer
  biological science technician
  Managing Ungulate Damage and Disease Project
  Fort Collins, CO

- Corey Perrillioux
  facility manager
  Administration Unit
  Fort Collins, CO

Presidential Migratory Bird Federal Stewardship Award. APHIS received the Presidential Migratory Stewardship Award in recognition of WS’ efforts to conserve large native migratory birds and protect the people of Hawaii. The Council for the Conservation of Migratory Birds, led by the U.S. Fish and Wildlife Service and composed of many Federal agencies with migratory bird responsibilities, chose the winner in May 2021.
WS conducted a joint research-operational project to reduce human-wildlife conflicts associated with two culturally significant bird species: the Hawaiian goose, or nēnē, and the Laysan albatross, or mōlī. Since both species are large, they pose a potential aircraft strike risk near airfields in some areas of Hawaii. WS research and operations personnel collaborated with numerous partners not only to reduce the aircraft strike hazards these species present, but also to identify, preserve, enhance, and support important breeding and foraging habitats for the birds elsewhere on the island. The project employed innovative nonlethal management tools, such as canine teams for dispersing birds, mitigation translocation, and egg swaps. Targeted research gauged the impact and effectiveness of these tools, which will guide future management strategies.
The transfer of scientific information is an important part of the research process. NWRC scientists and other WS experts publish in a variety of peer-reviewed journals that cover a wide range of disciplines, including wildlife management, genetics, analytical chemistry, ornithology, and ecology. (Note: 2020 publications that were not included in the 2020 NWRC accomplishments report are listed here.)


List of 2021 NWRC Research Projects

Defining Economic Impacts and Developing Strategies for Reducing Avian Predation in Aquaculture  
*Project Leader: Fred Cunningham*

Developing Control Methods, Evaluating Impacts, and Applying Ecology To Manage Carnivores  
*Project Leader: Julie Young*

Developing Methods To Manage Damage and Disease of Feral Swine and Other Ungulates  
*Project Leader: Kurt VerCauteren*

Development of Injectable and Mucosal Reproductive Technologies and Their Assessment for Wildlife Population and Disease Management  
*Project Leader: Jason Bruemmer*

*Project Leader: Stephanie Shwiff*

Evaluation and Development of Wildlife Repellents and Repellent Application Strategies  
*Project Leader: Scott Werner*

Genetic Methods To Manage Livestock-Wildlife Interactions  
*Project Leader: Antoinette Piaggio*

Improving Methods To Manage Healthy Forests, Wetlands, and Rangelands  
*Project Leader: Jimmy Taylor*

Methods and Strategies for Controlling Rabies  
*Project Leader: Amy Gilbert*

Methods and Strategies To Manage Invasive Species Impacts to Agriculture, Natural Resources, and Human Health and Safety  
*Project Leader: Steven Hess*

Methods and Strategies To Manage Rodent Impacts to Agriculture, Natural Resources, and Human Health and Safety  
*Project Leader: Aaron Shiels*

Methods Development and Damage Management of Depredating Birds and Invasive Wildlife  
*Project Leader: Bryan Kluever*

Methods Development To Reduce Bird Damage to Agriculture: Evaluating Methods at Multiple Biological Levels and Landscape Scales  
*Project Leader: Page Klug*

Understanding and Exploiting Wildlife Behavior To Mitigate Wildlife Collisions With Aircraft, Other Vehicles, and Structures  
*Project Leader: Brad Blackwell*

Wildlife-Borne Pathogens Affecting Food Safety and Security: Developing Methods To Mitigate Effects  
*Project Leader: Alan Franklin*

Wildlife Disease Dynamics, Epidemiology, and Response  
*Project Leader: Susan Shriner*

More information about these projects is available on the NWRC web page at:  
[www.aphis.usda.gov/wildlifedamage/nwrc]
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### Acronyms and Abbreviations

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<td>AIV</td>
<td>avian influenza virus</td>
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<td>ALHT</td>
<td>Asian longhorned tick</td>
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<td>AMR</td>
<td>antimicrobial resistant</td>
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<td>APHIS</td>
<td>Animal and Plant Health Inspection Service</td>
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<td>AQ</td>
<td>anthraquinone</td>
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<td>ARP</td>
<td>American Rescue Plan</td>
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<td>ASF</td>
<td>African swine fever</td>
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<td>BMI</td>
<td>body mass index</td>
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<td>CDC</td>
<td>Centers for Disease Control and Prevention</td>
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<td>DAPA</td>
<td>2,6-diaminopimelic acid</td>
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<td>DNA</td>
<td>deoxyribonucleic acid</td>
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<td>eDNA</td>
<td>environmental DNA</td>
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<td>EPA</td>
<td>U.S. Environmental Protection Agency</td>
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<td>GPS</td>
<td>global positioning system</td>
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<td>IAV</td>
<td>Influenza A virus</td>
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<td>LOD</td>
<td>limit of detection</td>
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<td>LOQ</td>
<td>limit of quantification</td>
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<td>NEXRAD</td>
<td>Next Generation Weather Radar</td>
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<td>NVSL</td>
<td>National Veterinary Services Laboratories</td>
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<td>NWDP</td>
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<td>NWSD</td>
<td>National Wildlife Strike Database</td>
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<td>NWRC</td>
<td>National Wildlife Research Center</td>
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<td>ONRAB</td>
<td>Ontario rabies vaccine</td>
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<td>ORV</td>
<td>oral rabies vaccine</td>
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<td>qPCR</td>
<td>quantitative real-time polymerase chain reaction</td>
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<td>RVNA</td>
<td>rabies virus neutralizing antibody</td>
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<td>SN</td>
<td>sodium nitrite</td>
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<td>USDA</td>
<td>U.S. Department of Agriculture</td>
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<td>WS</td>
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