

**United States Department of Agriculture** 

## Innovative Solutions to Human-Wildlife Conflicts

National Wildlife Research Center Accomplishments, 2018

**United States Department of Agriculture** Animal and Plant Health Inspection Service Wildlife Services National Wildlife Research Center 4101 LaPorte Ave. Fort Collins, CO 80521-2154 www.aphis.usda.gov/wildlifedamage/nwrc PHONE: (970) 266-6000 FAX: (970) 266-6032 Email: nwrc@usda.gov

The mission of the 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

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Cover Photos: Given its unique expertise and facilities for studying fish-eating birds, the NWRC Mississippi field station was asked to help evaluate the impacts of oil exposure to live birds as part of the Natural Resource Damage Assessment Trustee Committee's Deepwater Horizon oil spill injury assessment.

Photos by USDA Gail Keirn (facility and lab) and Katie Hanson-Dorr (infrared)

## **Message From the Director**

In his 1961 inaugural speech, John F. Kennedy, our Nation's 35th President, presented a vision for America's role in the world and laid the foundation for how individuals might commit to that vision. He famously said, "…ask not what your country can do for you—ask what you can do for your country."

The embodiment of this rhetorical challenge is the Government employee and his or her commitment to public service. Their commitment to serve the public good without expectation of personal gain is the thread that binds Government



Larry Clark, NWRC Director Photo by USDA, Gail Keirn

program's first responders to emergencies. These employees can be mobilized anywhere in the Nation within 24 hours. They represent the front line of the WS response and serve as liaisons to stakeholders and other State and Federal agencies on wildlife issues. Experienced in incident command operations, they are a valuable asset in any Federal emergency response. Activities are coordinated by NWDP staff through NWRC headquarters, which itself is integrated into the APHIS emergency response network. These first responders allow time to mobilize broader resources within WS if needed.

employees to their communities. At the National Wildlife Research Center (NWRC), we conduct research and develop methods to resolve human-wildlife conflicts for the protection of people, agriculture, and wildlife.

In this year's report, we highlight our efforts to learn more about the effects of oil spills on water birds, the development of safer rodenticides, and the role of wildlife pathogens on our Nation's food safety and security.

We also highlight our activities to support Wildlife Services' (WS) emergency response efforts. These activities, led by our National Wildlife Disease Program (NWDP), define what a collective effort in public service can accomplish. Thirty-two wildlife disease biologists, supervised by individual States within WS, serve as the This past year presented significant challenges to the NWDP program, but employees were up to the task. WS mobilized and responded to a virulent Newcastle disease outbreak in poultry in California; oil spills in Alabama, Illinois and Michigan; Hurricanes Harvey, Irma, and Maria; and a volcanic eruption in Hawaii. First responders were deployed a total of 887 days to these emergencies. This was in addition to their support of disease sampling for the feral swine program and other sampling initiatives for plague, tularemia, avian influenza, Lyme disease, avian botulism, West Nile virus, chronic wasting disease, and rabies.

When faced with a catastrophe, we can sometimes lose sight of how we, as individuals, can make a difference. Helen Keller once said, "I am only one, but I am one. I cannot do everything, but I can do something, and I will not fail to do the something that I can do." WS employees and their efforts to respond to emergencies epitomize what the collective action of individuals can accomplish. I and others in USDA are proud of our researchers, support staff, and first responders and the support they provide to the citizens of this country. It is with pleasure that I present to you this year's research accomplishments for the National Wildlife Research Center.

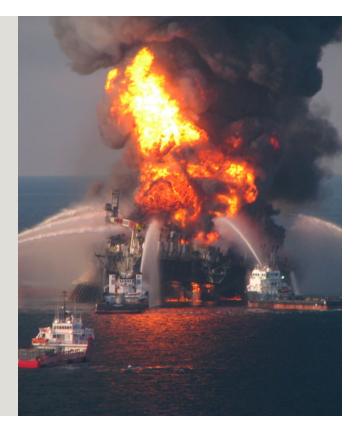
Larry Clark, Director National Wildlife Research Center Wildlife Services, APHIS-USDA Fort Collins, CO

# Contents

Research Spotlights
Effects of Oil Spills on Birds
2018 Accomplishments in Brief
Devices20Pesticides20Repellents22Other Chemical and Biological Methods23Disease Diagnostics, Surveillance, Risk Assessment, and Management27Wildlife Damage Assessments34Wildlife Management Methods and Evaluations40Wildlife Population Monitoring Methods and Evaluations50Registration Updates55Technology Transfer56Awards57
<b>2018 Publications</b>
Appendix 1. List of 2018 NWRC Research Projects
Appendix 2. NWRC Research Contacts
Appendix 3. Acronyms and Abbreviations

## **Research Spotlights**

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



The Deepwater Horizon oil spill in 2010 released more than 3 million barrels of crude oil into the Gulf of Mexico. As part of the response efforts, NWRC and other Federal, State, and university researchers investigated the impacts of oil exposure on live birds. *Photo by U.S Coast Guard* 

## **SPOTLIGHT: Effects of Oil Spills on Birds**

The Deepwater Horizon oil spill in the northern Gulf of Mexico began April 20, 2010, with the explosion and subsequent sinking of the Deepwater Horizon drilling rig and the deaths of 11 rig workers. Before it was capped, the uncontrolled well released more than 3.19 million barrels of crude oil, creating the largest offshore oil spill in the history of the United States. The oil covered approximately 43,000 square miles of ocean habitat, including more than 10 percent of the Gulf's coastline.

Thousands of birds died from the overwhelming effects of direct oiling, while many more were observed alive with varying amounts of oil on their skin and feathers. The Deepwater Horizon Natural Resource Damage Assessment Trustees conservatively estimated bird deaths at 51,600–84,500 individuals. But, for live birds, the nature and extent of injuries from oil exposure was not well understood. Studies show that oil can have a wide range of adverse effects on birds, including anemia, decreased nutrient absorption, altered stress response, and decreased immune function, when they ingest it at less than acutely lethal doses.

As part of the Natural Resource Damage Assessment Trustee Committee (which comprises the U.S. Department of the Interior [DOI], U.S. Environmental Protection Agency [EPA], USDA, U.S. Department of Commerce, and five affected Gulf States), a series of injury assessment activities began, which included Knowing the impacts of contaminants, such as crude oil, on wildlife aids in emergency response, restoration, and litigation efforts.

developing an avian toxicity testing program. The goal of the program was to provide scientific data on the impacts of oil exposure to live birds. These data would help inform the process for determining and quantifying bird injuries from the Deepwater Horizon oil spill.

NWRC researchers assisting with this program investigated the impacts of oil on birds through oral and external dosing. The goal of their studies was to evaluate the effects of low to moderate oil exposure and potentially repeated oil exposure that did not cause an immediate or quick death, but might cause physiological effects that ultimately affect bird survival, reproduction, and health.

The species chosen for these studies included the double-crested cormorant (*Phalacrocorax auritus*) and laughing gull (*Leucophaeus atricilla*). Double-crested cormorants are common, fish-eating seabirds that live in oceanic, coastal, and inland waterways and can be used as surrogates for other fish-eating species, such as pelicans, terns, and skimmers. The laughing gull is a small, black-headed bird that usually nests in large groups of up to 50,000 and was one of the most commonly oiled species found in the Gulf during the spill. Its diet consists of both terrestrial and aquatic invertebrates and seasonal berries. The laughing gull's abundance and flexible diet make it a useful model for studying the effects of oil on a broad range of species.

The findings from these and other bird toxicity studies were the focus of a December 2017 special issue of the journal *Ecotoxicology and Environmental Safety* and are highlighted below.

#### **Oral Dosing**

A series of oral dosing studies with captive laughing gulls and double-crested cormorants were conducted at NWRC's headquarters in Colorado and its Starkville, MS, field station.

Initial studies involved force-feeding (gavaging) oil to captive birds to determine toxicity. However, the appearance of oil in excreta within minutes of dosing in both species suggested the method did not allow enough time for the toxicants to be absorbed. The decision was made to modify dosing methods to increase the exposure time of birds to oil and to better mimic natural exposure.

A second round of studies exposed cormorants and laughing gulls to oil through contaminated food. Both species were given doses of 5 or 10 milliliters (ml) per kilogram of body weight per day through a daily allotment of oil-injected fish. Gulls were dosed for up to 28 days and cormorants were dosed for up to 21 days to see if birds developed hemolytic anemia or other anatomic, hematologic, and biochemical changes that warranted further investigation.<sup>1</sup>

<sup>1</sup> The NWRC is committed to the safe and humane treatment and handling of research animals. The Center abides by the Animal Welfare Act and works with the USDA's Animal Care program to ensure all animals are well cared for.



Laughing gulls were one of the most commonly oiled birds found during the Deepwater Horizon oil spill. Photo by Wikimedia Commons, Dick Daniels

Fifty percent of the laughing gulls fed oil-injected fish died. In addition, all gulls fed oil-injected fish showed a decrease in the number of red blood cells, along with evidence of Heinz bodies (denatured hemoglobin that indicates hemolytic anemia), signs of oxidative stress (inability of the body to detoxify), and increased liver and kidney weights.

"Double-crested cormorants fed oil-injected fish were also severely impacted. Fifty-six percent of treated cormorants died or were euthanized due to poor health," says NWRC supervisory research wildlife biologist and Mississippi field station leader Fred Cunningham. "Treated birds ate less and weighed less than nontreated birds. They also showed signs of reduced body temperature, lethargy, feather damage, and morbidity."

Similar to laughing gulls, cormorants also showed evidence of hemolytic anemia, increased Heinz bodies, increased liver and kidney weights, and lesions in the kidneys, liver, heart, and thyroid gland.

#### **External Dosing**

In addition to eating oil-contaminated fish, seabirds are often exposed to oil by swimming and diving in contaminated water. By coating cormorants' feathers with oil and allowing the birds to preen, NWRC researchers were able to study the impacts of continuous oil exposure and its resulting metabolic and thermoregulatory effects on the birds.

Researchers spread 13 grams of oil every 3 days for 15 days over 20 percent of captive cormorants' bodies, on the breast and back feathers only. This was to simulate the high end of "light oiling" (per the U.S. Fish and Wildlife Service category used for the Deepwater Horizon Natural Resource Damage Assessment).

Two of the 11 oiled birds died before the end of the study. Birds treated with oil ate more fish than unoiled birds, suggesting that oiled birds were trying to compensate for heat loss and thermal stress. Treated birds showed signs of oil toxicity, including deteriorated



Thousands of birds were coated with oil from the Deepwater Horizon oil spill. Studies were conducted to determine the effects of this exposure on bird survival, reproduction, and health.

Photo by Wikimedia Commons, LA Governor's Office

In the United States alone, rodents cause billions of dollars in damages each year to standing crops and stored foods, property, and native plants and animals.

feathers, abnormal feces, excessive preening, feather plucking, and lethargy. Furthermore, hemolytic anemia and clotting dysfunction, cardiac abnormalities, and liver and kidney damage in oiled cormorants were similar to those observed in the oral dosing study (*see previous section*). Since light oiling on dark brown birds such as the cormorant is hard to see from a distance, researchers note it is possible that birds suffering from oil intoxication in the wild may go unnoticed.

Data from this study were also used by NWRC researchers and their partners to test the accuracy of a bioenergetics model called Niche Mapper to estimate thermoregulatory impacts of oiling. Cormorants had significant increases in surface body temperatures after oil exposure. Niche Mapper accurately predicted surface temperatures and metabolic rates for unoiled and oiled cormorants. It also predicted a 13- to 18-percent increase in daily energetic demands due to increased thermoregulatory costs associated with oiling. Data showed oiled cormorants ate about 20 percent more food than nonoiled birds. Researchers note that Niche Mapper can give insight into the effects of sublethal oiling on birds by quantifying the thermoregulatory costs of diverting energy resources away from important life processes like maintenance, reproduction, and migration.

These studies highlight the harmful effects of oil toxicity on the overall health of birds and bird populations. Sublethal effects of oil intoxication include lethargy and decreased feather integrity. This may reduce a bird's ability to migrate and reproduce, resulting in long-term impacts for bird populations. The findings from this research offered valuable information for the Deepwater Horizon Natural Resource Damage Assessment and will aid future oil spill damage assessments as well.

#### **NEXT STEPS**

NWRC researchers will continue exploring the impacts of oil exposure on wildlife. Future studies will look at the impacts of oil on foraging behavior, thermoregulation, feather microstructure, and cardiac function in birds.

### SPOTLIGHT: Improving Rodenticides and Other Rodent Damage Control Methods

Rodents occur worldwide and provide many important ecosystem functions. Yet, some species can also cause serious damage to ripening crops, forest and nursery trees, rangelands, ornamental plants, and property, including cables and irrigation pipes. Rodents also eat and contaminate stored food, spread diseases, and contribute to the decline of native plants and animals on islands. Many tools are used to reduce rodent populations and their damage.

The summaries below highlight NWRC research to develop and evaluate such tools, including rodenticides, barriers, and other products and methods to eliminate or reduce damage from native and invasive rodents.

#### **Developing Safer, More Effective Rodenticides**

While there are many methods (e.g., traps, barriers, habitat modification, scare devices) to reduce rodent damage, rodenticides remain the most important tool



NWRC's rodent project focuses on the development and testing of new rodenticides and other tools to prevent rodent damage to agriculture, property, human health, and natural resources. *Photo by Wikimedia Commons* 

in the toolbox. Some rodenticides are lethal after one exposure (second-generation anticoagulants), while others require more than one dose (first-generation anticoagulants). The use and availability of rodenticides are becoming more restricted due to: increased manufacturing and registration costs, concerns about toxicity levels for nontarget animals, potential impacts to children, reduced effectiveness of some formulations, and concerns about humaneness to the targeted rodents. NWRC researchers are working to address many of these impacts by evaluating new rodenticide compounds and formulations.

"An ideal rodenticide is not only fast-acting and highly effective, but also species-specific, economical, and less hazardous to other animals and the environment," says Gary Witmer, a research wildlife biologist and leader of NWRC's rodent project. "We are investigating ways of modifying existing rodenticides to improve their efficacy while reducing hazards."

Recent studies show that using formulations with two active ingredients—an anticoagulant and an acute toxicant within one bait, but at lower concentrations than in rodenticides with only one active ingredient may lead to safer, more effective management tools. NWRC researchers and colleagues at the University of California-Davis combined cholecalciferol (vitamin D3) with diphacinone, a first-generation anticoagulant. Under normal conditions, vitamin D helps the body maintain appropriate levels of calcium. However, toxic doses of cholecalciferol cause high levels of calcium in the blood, which affects organ functions.

NWRC researchers found that adding a small dose of cholecalciferol to diphacinone-based rodenticides is effective against voles that are resistant to anticoagulants. This combination also reduces risks to other wildlife that may scavenge on dead or dying rodents; with lower concentrations of both compounds, there is less environmental residue and a quicker death. This new formulation, as well as one containing cholecalciferol and brodifacoum, were also effective with pocket gophers and ground squirrels.

Sometimes, simply reformulating an active ingredient can enhance a rodenticide's effectiveness. NWRC scientists did just that in another study: they compared the effectiveness of existing zinc phosphide rodenticides to formulations with reduced zinc phosphide concentrations or encapsulated zinc phosphide. In trials with wild-caught voles, zinc phosphide rodenticides with concentrations as low as 0.5 percent were still highly efficacious (80-percent mortality). The concentration of existing commercial products is 2 percent.

Researchers also found that voles ate more encapsulated zinc phosphide-coated oats at the lowest concentration (0.5 percent). Encapsulating zinc phosphide has an added advantage: it improves taste and reduces bait shyness issues, which leads to more efficient toxicant exposure. "Bait shyness" is a learned aversion to toxic bait due to sublethal exposure to the toxicant. With this type of aversion, the rodent associates the bait's distinctive flavor with the toxic consequences of ingesting it. If a rodent does not succumb to the toxin at first exposure, it is highly unlikely that the animal will eat similar baits in the future.

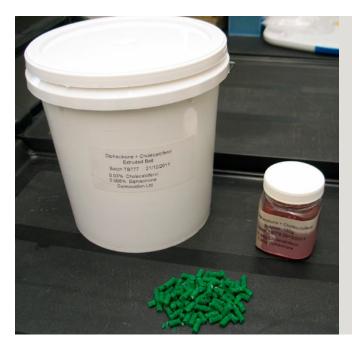
To explore ways to overcome bait shyness, NWRC researchers evaluated different components (lecithin, magnesium carbonate, sodium cyclamate, zinc sulfate) of current zinc phosphide bait formulations to determine which components may be contributing to bait shyness. They carried out a series of feeding preference trials with captive voles. They also tested encapsulation as a potential way to mask undesirable flavors in the zinc phosphide compound.

Results showed that numerous components of current bait formulations may contribute to bait shyness in voles. Voles avoided zinc sulfate and sodium cyclamate, so these flavor additives did not help to reduce bait shyness. But voles did prefer encapsulated bait, which suggests that encapsulation may alter the flavor profile of the bait and reduce bait shyness. Researchers recommend exploring the use of encapsulation in future bait development and varying the composition of zinc phosphide baits between applications to reduce bait shyness.

Such modifications to the formulations of existing registered active ingredients help improve the safety and efficacy of rodenticides.

#### **Exploring New Rodenticide Compounds**

Finding new compounds for use in rodenticides can be challenging due to the time and costs associated with registration. It may also be difficult to shift support away from well-established, existing products to lesser known or new products. Current rodenticide



NWRC research indicates that rodenticides may be more effective and safer when lower concentrations of an anticoagulant and an acute toxicant are combined into one bait. *Photo by USDA* 

research is exploring the use of new compounds or those currently registered or used for other purposes.

NWRC researchers tested the feasibility of using sodium nitrite (SN) and alphachloralose in rodenticides. SN is a preservative commonly used to cure meats, such as sausage and bacon. When eaten in high amounts over a short period of time, it is toxic to some animals. Alphachloralose is an organic compound that evokes a rapid onset of intoxication. It was developed as a house mouse rodenticide in Europe, where it is registered, manufactured, and sold for use in mouse control. Because SN and alphachloralose are used in other products, information about their toxicity and nontarget hazards already exists. This aids in future product registrations for these compounds.

Results from tests with captive rodents identified issues with both compounds. Previous NWRC studies with captive rodents determined that the median lethal dose of SN for various rodent species is about 246 milligrams/kilograms. However, all of the SN formulations tested showed limited effectiveness, with less than 20 percent of the house mice and Norway rats succumbing to the toxicant. For alphachloralose, efficacy was less than 35 percent in trials with wildcaught house mice that were fed a paste bait with 4.4-percent alphachloralose.

NWRC researchers note that issues associated with concentration and palatability of both SN and alphachloralose must be resolved before either may be an effective rodent control tool.

#### **Testing Barriers To Prevent Rodent Access**

"With their large and ever-growing incisors, rodents can chew through almost anything," says NWRC's Witmer. "Finding an effective barrier to keep them out of areas where they're not wanted takes persistence."

Burrowing rodents, such as Richardson's ground squirrels, can interfere with security systems at



Burrowing ground squirrels can interfere with security systems at nuclear missile silos. NWRC researchers tested various barrier systems to prevent ground squirrel access at military sites. *Photo by USDA* 

nuclear missile sites by passing through or digging under chain-link security fences, chewing on and triggering security sensors, and undermining facility infrastructure. Traditional methods of rodent control (i.e., rodenticides and traps) are not practical at these sites because of their remote locations and limited resources and personnel.

At the request of the U.S. Department of Defense (DoD), NWRC researchers assessed Richardson's ground squirrel populations, activities, and burrows at Malmstrom Air Force Base in Montana.

"When we visited missile sites in Montana, we noticed many active ground squirrels," says Witmer. "Many of the burrows we uncovered were fairly short, but some were elaborate and deep, measuring up to 12 feet long and 5 feet deep. We also observed the squirrels readily passing through and under the sites' 2-inch mesh chain-link fences."

Researchers conducted preliminary barrier trials in NWRC's outdoor rodent buildings in Fort Collins, CO, in hopes of identifying a more secure barrier system for the missile sites. Not only did the barrier need to keep out ground squirrels, but it also had to be see-through for security reasons. In trials with wildcaught ground squirrels, several barrier systems failed because the animals climbed over them or squeezed or clawed through them.

However, several above- and below-ground barrier systems were able to prevent ground squirrel intrusions. Effective above-ground barriers included (1) clear, polycarbonate plastic and (2) 2- by 4-inch woven wire fencing with two strands of electrified tape near the soil surface. Effective below-ground barriers included (1) a pea gravel-filled trench and (2) smallmesh expanded metal sheets. NWRC researchers shared this information with DoD, but note that the barrier systems still need to be field-tested at actual missile sites to confirm their effectiveness. NWRC research related to wildlife pathogens helps to ensure our Nation's food is safe, plentiful, and nutritious.

#### **Fertility Control in Rodents**

It's estimated that about 7 billion rats roam the planet on any given day. One of the more common species—the Norway rat—can produce up to 2,000 descendants per female each year. Given these high numbers, it's no wonder rodents can cause a lot of damage to agricultural crops and property, as well as spread disease.

In an effort to create new tools for reducing rodent damage, NWRC scientists teamed up with SenesTech, Inc., and the University of Arizona to test the effectiveness of a liquid bait to control fertility in Norway rats. Two chemicals (4-vinylcyclohexane diepoxide and triptolide) that target ovarian function in female rats were fed to captive Norway rats via a liquid bait. Triptolide is also known to affect spermatogenesis (sperm production) in males.

Treated females that were mated with treated males bore no offspring, while control pairs produced normal litter sizes of 9–10 offspring. There were fewer primordial follicles in ovaries of treated females than in control females, and the testes and epididymis of treated males weighed less than those of control males.

These results are promising, though NWRC researchers caution that it may not be possible to reproduce them in a wild setting where other food resources are available.

The EPA recently granted a registration for the liquid bait for use with Norway rats and black rats under the product name ContraPest.

#### **NEXT STEPS**

NWRC rodent project researchers plan to continue producing data sets to evaluate new active ingredients and formulations for rodenticides that could receive EPA registration. They will also continue studying new chemical repellents and fertility control compounds. Efforts are underway to evaluate a new self-resetting kill trap for small rodents, such as mice and voles. Future research will also focus on rodenticide hazards to nontarget species and ways to reduce those hazards.

## SPOTLIGHT: Wildlife Pathogens and Food Safety

Wildlife can carry and spread pathogens that infect poultry, livestock, and people, as well as threaten our Nation's food safety and security. For example, avian influenza viruses (AIV) can spill over from wild birds and other wildlife into poultry operations and become highly pathogenic. This causes poultry losses and trade embargos on U.S. poultry products. Pathogenic and anti-microbial resistant (AMR) bacteria can enter agricultural operations through wildlife feces, making livestock ill and contaminating food products as they are harvested and prepared for human consumption.

"In addition to pathogens that wildlife are suspected to spread, such as *Salmonella* and *E. coli*, new and novel wildlife pathogens continue to emerge in the United States and around the world, posing unknown threats to agricultural and human health," says NWRC research biologist Alan Franklin. "NWRC conducts research and surveillance for wildlife pathogens



Wildlife, such as these invasive feral swine, has been increasingly recognized as a threat to food safety because of its ability to transmit pathogens to agricultural crops and livestock. *Photo by USDA* 

and works to develop tools and methods for mitigating their spread."

The summaries below highlight recent NWRC research related to wildlife pathogens and food safety.

#### Identifying Wildlife-Related Disease Risks

Contaminated fruits and vegetables have become a main source of foodborne illnesses. A 2015 report by the Centers for Disease Control and Prevention (CDC) and the Interagency Food Safety Analytics Collaboration analyzed data from nearly 700 outbreaks of foodborne illness caused by either *Salmonella, E. coli* 0157:H7, *Campylobacter* or *Listeria* between 1998 and 2012. Results showed vegetables were responsible for the greatest number of illnesses, most due to *Salmonella* and *E. coli*. In some cases, wildlife were linked to outbreaks that have cost millions of dollars in healthcare costs and product recalls. In 2006, about 200 people in 26 States were diagnosed with an especially virulent strain of *E. coli* 0157:H7 found in spinach. The strain that caused the outbreak—which caused 3 human deaths and more than 100 serious illnesses—was also found in wild pig feces in the area. However, the extent to which wildlife and wildlife feces contaminate agricultural fields and products is poorly understood.

Five pathogens cause nearly 90 percent of foodborne illness attributed to leafy greens. Wildlife are either documented or hypothesized as the source of spread to agricultural crops and people for three of these pathogens: *Salmonella*, Shiga toxin-producing *E. coli* (STEC), and noroviruses. To better understand whether wildlife contaminate lettuce and spinach fields with these pathogens, NWRC scientists partnered with the Center for Produce Safety and Southern Colorado Farms to study wildlife activity at farms in the San Luis Valley of south-central Colorado. Although a variety of wildlife were found visiting crop fields, none of the three pathogens were found in either the collected wildlife feces or on the produce from the fields.

In other studies, NWRC scientists have sampled wildlife feces in urban areas, such as community parks, and found they contain pathogens that could be spread to people. Although the risk is low, pathogens could be touched or carried on the bottom of shoes and then accidentally spread and ingested from dirty hands. Thus, the risk of wildlife contaminating food or other resources may vary by region and depend on available sources of foodborne pathogens and the growing environment.

In addition to field studies, NWRC scientists worked with captive animals to learn more about how wildlife pathogens, such as AIV, are spread. For instance, studies have explored how long waterfowl shed AIV, how much of the virus they shed, and whether past infections affect viral shedding if birds are reinfected. NWRC has also examined the ecology and prevalence of AIV across the United States. This information helps identify "hot spots" where the virus has the highest chance of spreading from the environment into domestic poultry.

In other studies with captive mammals, NWRC scientists discovered that experimentally infected striped skunks and cottontail rabbits shed large amounts of avian influenza viral ribonucleic acid (RNA) and may influence the spread of viruses in certain areas. They have also found that captive raccoons are more likely to become infected with AIV from contaminated water versus eating contaminated animals or eggs. Insights about how wild animals carry and move AIV help regulators and producers develop better biosecurity methods for domestic poultry facilities.

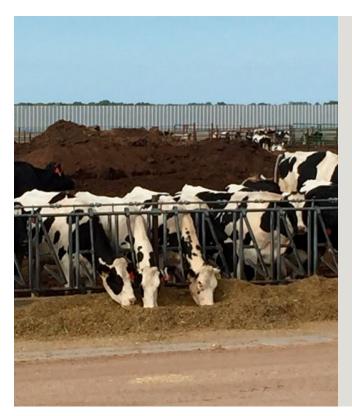
Another area of concern related to the spread of pathogens involves AMR bacteria. Since many existing drugs can no longer fight off AMR infections, these bacteria have a major impact on human and agricultural health. The healthcare burden from AMR bacteria exceeds \$4 billion annually. Reducing antibiotic use in agriculture has helped reduce AMR bacteria in the food chain in some situations, but there are examples where AMR bacteria still persist.

For instance, NWRC researchers tagged and monitored raccoons and several other mammal species at multiple livestock facilities in Colorado to learn more about wildlife's role in maintaining and moving AMR bacteria. Findings showed that raccoons had a higher prevalence of AMR bacteria than other wildlife species and most were infected with multiple strains of AMR bacteria. Raccoons infected with AMR bacteria often came from surrounding areas and had contact with cattle feed troughs, which may play a role in the presence of AMR bacteria at livestock facilities. Thus, maintaining good biosecurity and preventing wildlife on farms may help lessen the spread of pathogens.

Although it's often assumed that livestock facilities are the main source of AMR bacteria, other areas—such as wastewater treatment plants that allow antibiotic residues and antibiotic-resistance genes to pass through their systems—may also be a source. Samples from raccoons captured downstream from wastewater treatment plants had a higher prevalence of certain AMR types, such as erythromycinresistance. This suggests that wildlife may be infected with AMR bacteria from these sources. Because raccoons are often found near aquatic habitats and are also common in urban environments, this species may be an important way that AMR bacteria move to and from livestock facilities and urban areas.

#### **Improving Disease Detection and Diagnostics**

Being able to detect and diagnose diseases is key to monitor and prevent their spread. For this reason, NWRC researchers have developed many diagnostic tools and monitoring systems for pathogens affecting wildlife, people, and livestock.



When studying wildlife's role in maintaining and moving antimicrobial-resistant bacteria in livestock facilities, NWRC researchers found that raccoons frequently moved from surrounding areas into feed troughs used by cattle. *Photo by USDA, Gail Keirn* 

The 2014–2015 outbreak of highly pathogenic avian influenza in U.S. poultry resulted in approximately 48 million birds being euthanized and an estimated \$3.3 billion in economic losses. Biocontainment and emergency response measures highlighted the important role of aerosols in the virus' transmission within and between poultry facilities. In fact, aerosol sampling may offer another surveillance tool to monitor influenza viruses in agricultural production systems.

Researchers from the NWRC, Colorado State University's High Plains Intermountain Center for Agricultural Health & Safety, the University of Wyoming, and McGill University developed and tested a system to more effectively sample and detect aerosolized influenza viruses. The researchers paired liquid impingement (a widely used system in which aerosolized viruses are deposited into a liquid substrate) with anion exchange resin-based virus



Monitoring sylvatic plague exposure in rodent-eating carnivores, such as coyotes, is an important tool for assessing plague risk to people and wildlife. Photo by USDA, Natural Resources Conservation Service

concentration. To test this new system, various quantities of type A and type B influenza viruses were aerosolized within a custom-built container and sampled using liquid impingers (devices used to collect viral aerosols) with and without anion-exchange resin. Ultimately, adding anion-exchange resin to the sampling devices improved detection of type A and type B influenza viruses by more than six times and three times, respectively. The new technique is simple to perform, adaptable to existing methods, and cost-effective and will likely prove valuable in future influenza emergency response efforts.

"Although much of our research focuses on wildlife pathogens that impact agriculture, we also partner with the CDC and other Federal agencies to address human health concerns," says NWRC microbiologist Jeffrey Chandler.

Sylvatic plague is another disease that affects people and many other mammals. It is caused by the bacterium, *Yersinia pestis*. People usually get plague after being bitten by an infected flea or handling an animal that is carrying *Y. pestis*. Modern antibiotics are effective in treating plague, but without prompt treatment, the disease can cause serious illness or death.

Monitoring *Y. pestis* exposure in rodent-eating carnivores, such as coyotes, is an important tool for assessing plague risk to people and wildlife. Since these types of carnivores eat rodents often and are thereby exposed to their fleas as well, testing one carnivore is considered the same as testing hundreds of rodents. To improve WS' ability to monitor *Y. pestis* exposure in wildlife, scientists from NWRC, CDC, and Colorado State University developed a new semi-automated, bead-based flow cytometric test<sup>2</sup> called the F1 Luminex plague assay. This test was shown to be 64 times more sensitive than traditional plague monitoring methods. It can detect *Y. pestis* exposure in coyotes, as well as felids and raccoons. Researchers are now evaluating the F1 Luminex plague assay as a

<sup>2</sup> Involves suspending particles that bind to anti-Y. pestis antibodies in a stream of fluid that is then passed through an electronic detection device

replacement for the passive hemagglutination test, the WS National Wildlife Disease Program's main tool for plague monitoring.

Another public health risk involves water quality and contamination by wildlife feces. Having sensitive, reliable, and user-friendly methods to test water quality and safety is important. NWRC, EPA, and university researchers developed and tested an anion-exchange resin-based system to aid in detecting male-specific F+ RNA (FRNA) coliphages (i.e., viruses that parasitize *E. coli* bacteria) from water. FRNA coliphages are microbial indicators of fecal contamination. Different genotypes of these coliphages are associated with specific animal hosts, making FRNA coliphages useful for identifying and tracking sources of fecal contamination.

This new system disperses anion-exchange resin in water samples. The resin absorbs the FRNA coliphages, which concentrates these organisms into a small sample. Reducing sample volume effectively is critical to improve downstream detection with modern molecular methods. In field studies, the anion-exchange resin method concentrated and detected FRNA coliphages equally or better than existing strategies and at a lower cost. Also of note, this system was effective in diverse water sources known to contain a variety of chemicals that can hinder FRNA coliphage detection. Therefore, such a tool may be useful for frequent or continuous water testing and identifying sources of wildlife-associated fecal contamination.

#### **Methods To Prevent Disease Spread**

Fresh produce can become contaminated with foodborne pathogens in many ways, including fecal contamination by wildlife.

"Working with our partners, we developed and tested a new method for reducing pathogens on tomatoes and spinach," says Chandler. "We use a process called prophage induction, which basically causes bacteria to be destroyed from within."

Prophages are viruses integrated into the genome of a host bacteria. When prophages are exposed to certain environmental conditions or chemicals (e.g., prophage inducers), they start to replicate, ultimately killing the host bacteria. In this study, NWRC researchers inoculated STEC and Salmonella enterica bacteria on tomatoes and spinach and exposed the bacteria to the prophage inducer mitomycin C. They then monitored the bacteria. Prophage induction reduced STEC contamination on tomatoes and spinach by 99.9 percent and 90 percent, respectively. Similarly, prophage induction reduced S. enterica contamination on tomatoes and spinach by 90-99 percent. These findings show that prophage induction is a potential strategy to control bacterial foodborne pathogens on fresh produce.

#### **NEXT STEPS**

NWRC scientists continue to examine the role of wildlife in maintaining pathogenic and AMR bacteria at the wildlife-agricultural interface. With increased cases of novel pathogens coming into the United States, NWRC scientists are also focusing on how wildlife may be involved in their spread and introduction into agricultural operations. In collaboration with other institutions, they are developing tools, such as biosensors, to detect and characterize pathogens in the field and, in support of national biodefense, predictive models to identify new pathogens.

## **SPOTLIGHT: Successful Technology Transfer**

From field specialists to researchers and managers, WS employees often experience "aha" moments when working to solve complex wildlife damage management problems. Once discovered, those ideas Wildlife Services experts work with private companies and universities to turn ideas into marketable products.

may go on to become practical, feasible, and costeffective wildlife damage management tools. They may also be patented.

"WS scientists and operational experts are some of the most creative people I know. My job is to help them transfer their creative ideas into patentable and licensable products for further development and manufacturing by the private sector," says John Eisemann, WS' Technology Transfer Program Manager. "Probably the biggest obstacle is getting our experts to realize the potential of their ideas."

Within the last few years, WS has received four new patents from the U.S. Patent and Trademark Office (PTO) and negotiated three licenses with private companies for past patents. License royalties from these patents bring money directly back to the inventors and NWRC.

"Recently, many of our patents have been related to repellent and vaccine development, but they don't have to be. We've also received patents on traps and other field tools," notes Eisemann.

The future success of wildlife damage management depends on new tools and technologies. Traditional methods and tools face increased scrutiny from regulatory agencies, and few private companies are financially willing or able to pursue new registrations or handle the risks involved in developing new products. Government plays a valuable role in absorbing these risks and encouraging innovation and discovery.

The Federal Technology Transfer Act of 1986 and later legislation changed how Federal laboratories,

such as NWRC, do business. These laws allow Federal laboratories, universities, and industry to form commercial partnerships that enhance the development of new technologies and move them to the marketplace, while at the same time offer financial benefits to Federal inventors. Under these laws, Federal laboratories must seek U.S. partners for technology transfer opportunities before working with foreign entities. This flexibility has allowed NWRC to work with a variety of small to midsize U.S. and foreign partners.

Below are examples of WS technology transfer success stories.

#### **Building a Better Snake Trap**

Invasive or non-native reptiles have been in Florida for over 100 years, and their populations have increased quickly in the last half century. Large exotic snakes and lizards, such as Burmese pythons, Argentine giant tegu lizards, and Nile monitor lizards, are all breeding in Florida. Developing new tools to control these species and the damage they cause is vital to help protect native wildlife and ecosystems.

In 2013, a wildlife biologist at the NWRC Florida field station received a patent for a live snake trap. The trap uses two trip pans to capture long animals such as large, heavy snakes (e.g., invasive Burmese python and monitor lizard). This is the first trap to require that two trip pans be depressed at the same time to close the trap door. The pans are spaced far enough apart that nontarget animals, such as small native snakes, raccoons, and opossums, are unlikely to trigger the trap. One of NWRC's main technology transfer goals is for end users to adopt our research and development. This often includes patenting and licensing inventions for commercial products. The patent for the live trap is licensed to a private company, which now has the product available for purchase on its website.

#### Animals Don't Always Like What They See

Birds and rodents damage agriculture by eating newly planted, maturing, ripening, and stored crops, as well as livestock feed. Their feces may also contaminate crops, feed, and equipment. Tools that prevent bird and rodent damage to these resources can save millions of dollars each year.

NWRC scientists have longstanding partnerships with private companies and industry groups to investigate bird and rodent repellent compounds, formulations, and application strategies for reducing wildlife damage. One such partnership with Arkion Life Sciences (Arkion) has resulted in five co-owned patented technologies and associated repellent products that are cost effective, practical, environmentally safe, and socially responsible and that are now marketed and sold nationally and internationally. Recent advances have also led to the development of a new repellent application strategy that takes advantage of both visual cues and post-ingestive consequences (e.g., an unpleasant taste or sickness in the animals that eat the repellents).

Anthraquinone (AQ)—a naturally occurring plant compound—was first patented as a bird repellent in 1944 to reduce bird damage to agricultural crops. At that time, it was assumed that the compound caused post-ingestive stress. Recent NWRC-Arkion research has shown that AQ can also cause avoidance behaviors in birds and mammals through visual cues related to the compound's absorption of the ultraviolet (UV) spectrum. As a result, NWRC and Arkion designed repellent products and application strategies that "trick" birds and mammals into overlooking food



WS employees develop new tools and technologies, such as this live trap for large snakes and lizards. Such products are often licensed to private companies for commercialization. *Photo by USDA, John Humphrey* 

items or deter them from sitting or perching on treated structures. NWRC research has also shown that if birds first come into contact with AQ, there are lower cost options for deterring them later: other less expensive compounds with similar UV spectral features are an effective substitute for AQ, or subsequent uses of AQ work at lower application rates.

Future NWRC-Arkion research efforts are focused on developing AQ-based products for foliar applications to crops; topical application to fruit and nut trees; structural applications in poultry, beef, and dairy facilities; and incorporated or topical applications to rodenticide baits. We expect applications of this repellent technology to increase throughout the United States and internationally, saving farmers money and helping to protect a wide variety of resources.



Anthraquinone (AQ) is used as a bird repellent in part due to visual cues related to the compound's absorption of the ultraviolet (UV) spectrum. These pictures show human-visible (left) and ultraviolet images (right) of the UV-absorbent feeding cue and the UV-reflective feeding cue used to test conditioned food avoidance among red-winged blackbirds. Note how the UV-reflective feeding cue is orange in color. USDA solely owns one patent, co-owns one patent, and co-owns three patent applications for AQ technologies with a private company. *Photos by USDA, Scott Werner* 

#### Automatic Aerial Bait Delivery System for Brown Treesnakes

The invasive brown treesnake causes severe ecological and economic damages on Guam. About 1–2 million brown treesnakes inhabit the 200-square-mile island. WS has worked actively with the Territory of Guam and with other Federal agencies to reduce the number of snakes on the island and prevent their spread. As part of this work, WS and its partners have used many control tools and strategies, including an oral toxicant made of acetaminophen.

In 2009, NWRC and Applied Design Corporation (ADC), a private engineering firm, entered into a series of cooperative agreements to design a bait cartridge, automated manufacturing system, and aerial bait delivery system to distribute acetaminophen to brown treesnakes in remote and inaccessible areas on Guam. The NWRC supplied information on the snake's ecology and behavior, guidance on Federal pesticide regulations, and early prototype concepts. Working together, NWRC scientists and experts at ADC designed a biodegradable bait cartridge and automatic delivery system that can disperse bait cartridges via helicopter or fixed wing aircraft. In 2018, ADC manufactured more than 41,000 baits for use on Guam, and we expect that number to grow.

ADC and NWRC filed jointly for a U.S. patent for the bait cartridge in 2014, and ADC plans to file two more patents related to its automated bait delivery system and bait manufacturing process. ADC plans to commercialize this technology for use in wildlife damage management, including large-scale control of brown treesnake populations on Guam and invasive species management worldwide.

"This project is a shining example of interagency and private partner collaboration," says Eisemann, WS' Technology Transfer Program Manager. "DoD and DOI have provided more than \$6.5 million for research and the development of the aerial baiting system."

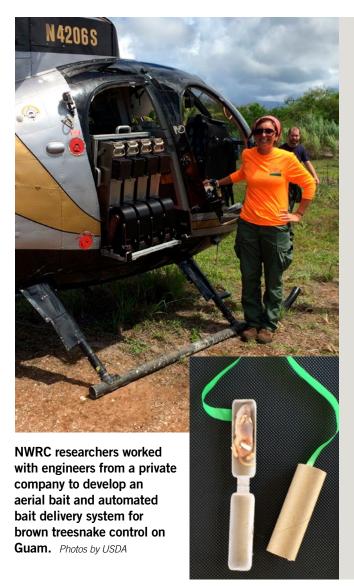
The DoD, DOI, USDA, the Government of Guam worked together to determine research needs and best management practices for implementing the aerial baiting program. They also collaborated with ADC to design and produce the aerial baiting system and Guam helicopter pilots to conduct early aerial baiting research. Because of the intense interest in snake management on Guam and other conservation projects in the region, WS has also stationed a helicopter on Guam to offer support services.

#### **Contraceptive for Wild Horses and Burros**

Overpopulation of wild horses and burros is a significant concern in the United States, as these animals can overgraze native plant species and compete with livestock and local wildlife for food and habitat. DOI's Bureau of Land Management (BLM) estimates that about 37,300 wild horses and burros (31,500 horses and 5,800 burros) are roaming on BLM-managed rangelands in 10 Western States. Per BLM, this exceeds by nearly 11,000 the number of wild horses and burros that can exist in balance with other public rangeland resources and uses. Management options right now are limited: in most cases, BLM can only remove horses and burros from the range and offer them for adoption or hold them in BLM's care indefinitely. Today, more than 49,000 wild horses and burros taken from BLM-managed lands are fed and cared for at short-term corrals and long-term pastures.

In 2013, EPA approved the GonaCon-Equine immunocontraceptive vaccine (GonaCon) for adult female wild or feral horses and burros. GonaCon was developed by NWRC scientists and is the first single-shot, multiyear wildlife contraceptive for use in mammals. This nonlethal tool gives wildlife managers another option in their work to reduce overabundant wild horse and burro populations in the United States.

WS has since licensed GonaCon-Equine to SpayFIRST!, a public-benefit company. This license allows the company to produce and sell the vaccine in the United States and internationally. SpayFIRST! expects to receive a product registration for horses from EPA in 2019 and is working with NWRC to develop a line of wildlife contraceptive products.



#### **NEXT STEPS**

The Technology Transfer program will continue working hard to promote the development and protection of intellectual property within WS. This includes creating new outreach materials to attract collaborators, better tracking and process management systems to streamline the process, and internal mechanisms to determine the value of patenting WS inventions. NWRC is also pursuing patents for several new wildlife damage management products. These include a unique feral swine feeder, a novel formulation for an orally delivered wildlife contraceptive, and a technology that makes livestock feed less attractive to birds.

## **2018 Accomplishments in Brief**

NWRC employs about 150 scientists, technicians, and support staff who are devoted to 16 research projects (see Appendix 1). Below are brief summaries of select findings and accomplishments from 2018 not already mentioned in this year's report.

#### **Devices**

 Bait Stations for Feral Swine. Invasive feral swine exist in the majority of U.S. States. Researchers are evaluating the use and delivery of toxicants, contraceptives, and vaccines for feral swine management, with a focus on developing strategies that minimize risks to nontarget species. NWRC and Australian partners developed a bait station for feral swine that accommodates the species' group feeding behaviors. The bait station also



NWRC researchers and Australian partners developed a bait station for feral swine that prevents white-tailed deer and raccoons from accessing the bait.

Photo by USDA, Wildlife Services

helps prevent white-tailed deer and raccoons from accessing the bait. In evaluating different designs, researchers found that bait stations made of two back-to-back troughs at 1.1 meters long were sufficient for delivering bait to large numbers of feral swine. In field studies, video footage showed that 80 percent of feral swine (33 of 41), 0 percent of white-tailed deer (0 of 7), and 17 percent of raccoons (1 of 6) accessed the bait stations. Future changes will add resistance to the bait station lids to completely exclude raccoons and identify baiting strategies that quickly acclimate feral swine to the stations.

Contact: Michael Lavelle

## **Pesticides**

 Secondary Hazards to Nontarget Species From Feral Swine Toxic Bait. Sodium nitrite (SN) is a meat preservative commonly used to cure meats, such as sausage and bacon. When eaten in high amounts over a short period of time, it is toxic to feral swine and other animals. To prevent secondary hazards to wildlife, NWRC scientists and partners in Australia and Texas investigated four types of micro-encapsulation coatings for an SN toxic bait. The micro-encapsulation coating helps protect SN from breaking down in the environment before feral swine eat it and helps mask its odor and salty flavor.

Results for all four types of micro-encapsulation coatings showed no risk of secondary poisoning for nontarget scavengers that ate the muscle, eyes, and livers of feral swine carcasses. Residual SN from the toxic bait was not detected in those tissues. The risk of secondary poisoning from eating feral swine vomit appeared low since about 90 percent of the SN was metabolized or broken down in the animals' stomachs before vomiting, and the residual SN continued to degrade after being exposed to the environment. Researchers note that secondary poisoning may be possible if scavenging animals eat 15 percent or more of their daily dietary requirements from digestive tract tissues or undigested bait from treated feral swine carcasses-but this is unlikely. APHIS will use results from this study to support the registration of an SN toxic bait for feral swine in the United States and Australia.

Contact: Nathan Snow

• Evaluation of Toxic Baits for Invasive Mongoose.

The small Indian mongoose is an invasive species on Hawaii, Puerto Rico, Croatia, Mauritius, and several Caribbean islands. Mongooses were introduced to these islands during the 19th century to control rats. Unfortunately, they also eat native birds, reptiles, and amphibians and serve as a reservoir for rabies in Puerto Rico. Numerous techniques, including shooting and trapping, have been used to protect native wildlife from mongoose predation with mixed results. Adding a toxicant for use in mongoose control may help these efforts.

NWRC researchers evaluated 10 commercial rodenticide baits, technical diphacinone powder, and 2 alternative acute toxicants



Mongooses were introduced to many Caribbean islands during the 19th century to control rats. Unfortunately, they also eat native birds, reptiles, and amphibians and serve as a reservoir for rabies in Puerto Rico. *Photo by Wikimedia Commons, Tony Hisgett* 

(para-aminopropiophenone [PAPP] and sodium nitrite [SN]) for use with Indian mongooses. PAPP, diphacinone, and bromethalin showed the most promise as toxicants for mongooses. Overall acceptance and mortality was low for hard pellets or block baits. In contrast, most baits formulated with fresh minced chicken were eaten by all mongooses in all trials. The exception was SN. Researchers noted the moisture in the chicken may have compromised the SN's encapsulation, rendering it unpalatable. SN may have potential if used at a lower concentration (less than 5 percent) and in another bait matrix. All of these active ingredients would need more laboratory and field evaluation before EPA approval and registration for use on mongoose. *Contact: Robert Sugihara* 

### **Repellents**

Repellent for Voles. Voles cause extensive girdling damage to orchards and vine crops in California. Although rodenticides are often used to control California vole (*Microtus californicus*) damage, they are rarely allowed in fields where fruit is on the trees for most of the year. Repellents have shown promise in lab studies, but often fail to prove highly effective in field trials. Recent lab studies have shown that low concentrations of anthraquinone (AQ, a naturally occurring compound) can cause up to 84-percent repellency in voles exposed to AQ-treated grains. Vegetation management, such as mowing, applying herbicide, or physically removing vegetation around the base of trees, may also help reduce vole damage.

NWRC and University of California researchers investigated the efficacy of combining an AQ application with vegetation management to minimize vole girdling damage to Clementine citrus trees. Results showed a 90–100 percent reduction in girdling damage to trees after a single application of AQ during trials in spring and summer. Removing vegetation around the base of trees further reduced damage during the summer, with no girdling observed on AQ-treated trees that were surrounded by bare soil. Furthermore, the efficacy of AQ did not decrease during the 5- to 6-week sampling period. Researchers recommend using AQ and vegetation management as part of an integrated pest management approach to mitigate vole damage in the summer. *Contact: Scott Werner* 

Repellent for Ground Squirrels and House

**Mice.** Chemical repellents are a useful wildlife management tool since they can be applied directly to a commodity or structure to prevent intrusion and damage. Rodents are major agricultural pests worldwide: they spread disease, damage property, and harm crops and food production. For example, in California alone, ground squirrels cause an estimated \$12–16 million in crop damage and \$8–12 million in physical damage to structures, levees, and earthen dams each year. House mice are the most widespread invasive mammal in the world. Commonly found in and around homes and farms, mice eat almost anything and are a main

NWRC researchers evaluated the effectiveness of an anthraquinone-based repellent in preventing damage by rodents. Richardson's ground squirrels and house mice only breached 25 percent and 21 percent of treated burlap barriers, respectively.

Photo by USDA, Hailey McLean



source of contamination at commercial animal facilities and in stored grains.

NWRC researchers evaluated the effectiveness of an anthraquinone (AQ)-based repellent in preventing structural damage by Richardson's ground squirrels and house mice. The researchers applied AQ-based repellent to pieces of burlap secured over the ends of polyvinyl chloride (PVC) pipes. Food was placed within the enclosed PVC pipes to encourage contact with the repellenttreated and untreated burlap barriers. Richardson's ground squirrels and house mice breached 55 percent and 100 percent of the untreated burlap barriers, respectively. Yet, they only breached 25 percent and 21 percent of the treated burlap barriers.

Results indicate that AQ-based repellents show promise for both ground squirrels and mice in preventing barrier breaches. The AQ treatments also decreased how much food the ground squirrels ate. Because AQ is a post-ingestive repellent, researchers conclude that the ground squirrels likely ate AQ when chewing through the treated barriers and then avoided untreated enrichment food associated with barriers. *Contact: Shelagh DeLiberto* 

## **Other Chemical and Biological Methods**

 Attractants for Feral Swine. Lethal control is the most common method used to reduce feral swine populations and damage, and many lethal strategies (e.g., baiting and trapping) rely on luring feral swine to a specific location. Identifying the most effective attractant for feral swine can vary based on the availability of natural foods, location, time of year, age, sex, and competition with other species. NWRC researchers studied numerous available feral swine attractants, such as food, scents, recorded swine sounds, and decoys or other visual attractants, to see which were most effective. Results showed that combining preferred foods with a scent or visual attractant helps lure feral swine from natural food sources. Placing attractants in the right locations will also increase the chance that feral swine will encounter them. For example, freshly turned soil is one attractant that is highly effective in enticing feral swine and has minimal appeal to nontarget species. Researchers recommend that future studies evaluate other potential feral swine attractants, such as fermented corn mash, flavor additives such as sweeteners and substances with umami and cheese-like features, fish oil, urine from female pigs in estrus, fruity scents, synthetic fermented eggs, blood and bone meal, and meat. *Contact: Mike Lavelle* 

 Body Odor Changes in Mammals. Body odors often contain information about the animal's sex, age, genetics, diet, reproductive status, and health. As a result, some animals can detect illness or infection among members of a group based on changes in body odors. Past studies by NWRC and Monell Chemical Senses Center have suggested that cytokines (i.e., substances secreted by cells of the immune system) are involved in producing disease-induced odors. But scientists don't know yet which cytokines contribute to disease-related odors or how they impact body odors overall.

NWRC and Monell researchers tested whether changes in body odor could be caused by proinflammatory cytokines tumor necrosis factor (TNF) and IL-1ß or indirectly by anti-inflammatory cytokine IL-10. The researchers treated captive mice with either lipopolysaccharide (LPS) or cytokines TNF and IL-1ß. Then, they evaluated changes in the mice's urine odors using trained mice (biosensors) and headspace gas chromatography. Trained mice noticed a similarity between LPS-associated odors and TNF-induced odors, but not IL-1ß- or IL-10-induced odors. Further, the chromatography analyses showed unique profiles of volatile compounds (chemicals that make up scents and odors) for each treatment. These findings support the value in continuing to develop tools that use body odor analysis to diagnose disease.

Contact: Bruce Kimball

Sex Pheromones as a Snake Attractant. Food-based attractants are commonly used in traps for invasive brown treesnake control on Guam. Though not well explored, using pheromones as attractants for trapping purposes may offer another option. Pheromones are chemical substances and odors animals produce to affect the behavior of other members of their species. There is substantial evidence that brown treesnakes use pheromones to find and choose mates. NWRC worked with James Madison University researchers to determine if methyl ketones (a closely related



Food-based attractants are commonly used in traps for invasive brown treesnake control on Guam. NWRC researchers are exploring the use of pheromones as attractants. *Photo by USDA, Wildlife Services* 

series of compounds that serve as pheromones in many snakes, including brown treesnakes) could be an attractant for brown treesnake removal. In this study, the researchers observed male brown treesnake responses to methyl ketone isolates and whole skin lipid extracts from female snakes. The male snakes showed only weak responses to the methyl ketones compared to the whole skin lipid extracts. While a pheromone control strategy is still possible, it may not be cost-effective or practical since the whole skin lipid extract is difficult to synthesize. *Contact: Bruce Kimball* 

- Permanent Sterility for Wild Horses. The current wild horse and burro population in the United States is nearly three times what the rangeland can support, which is detrimental for wild horses, wildlife, and rangeland. Range managers are exploring the use of contraceptives to decrease wild horse and burro populations, but no permanent contraceptive vaccine is available right now. One area of research is the control of ovarian follicular growth. By targeting follicular growth and oocyte (egg) development, there is potential to prevent ovulation and/or accelerate egg depletion in female mammals, thereby causing sterility. NWRC and Colorado State University studied the effects of vaccination against two oocyte-specific growth factors (Bone Morphogenetic Protein-15 [BMP-15] and Growth Differentiation Factor-9 [GDF-9]) on ovarian function in mares. Results showed that both treatments changed ovarian functions, with BMP-15 significantly decreasing ovulation rates and the size of ovulatory follicles. These findings support the development of a single-shot, permanent contraceptive for wild horses and burros. Contact: Doug Eckery
- Reimmunization Boosts GonaCon Effectiveness in Wild Horses. GonaCon-Equine is an injectable immunocontraceptive vaccine registered for use with female wild horses and burros. In a recent



A single-shot, permanent contraceptive for wild horses and burros continues to be a focus of NWRC's reproductive research.

Photo by Wikimedia Commons, Rick Cooper

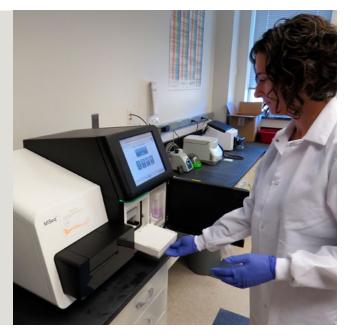
study, NWRC worked with Colorado State University and the National Park Service to examine the long-term effectiveness of this vaccine, both as a single immunization and as reimmunization, on reproduction and the side effects in free-ranging horses. Researchers randomly assigned 57 adult mares from Theodore Roosevelt National Park in North Dakota to either a GonaCon-Equine treatment group or a saline control group. After treatment, the mares were released and monitored annually to estimate foaling proportions, social behaviors, body condition, and injection-site reactions. After 4 years, the animals were recaptured, given a booster vaccination, released, and monitored for another 4 years.

With a single vaccination, the proportion of treated mares that gave birth was lower than that for control mares in the first 2 years after treatment. But with two vaccinations, the proportion of females giving birth was lower than that for control mares for 3 consecutive years. About 62 percent of immunized mares had some visible intramuscular swelling at the vaccine injection site. However, none of these mares had any signs of lameness, altered gait, or abnormal range of movement throughout the 8 years they were observed in this study. Results showed that GonaCon significantly reduced foaling in treated mares and that, with practical use, feral horses will need a reimmunization for the vaccine to offer sustained reductions in population growth rates over time. *Contact: Doug Eckery* 

**Contraceptive Vaccine for Farm-Raised Salmon.** Aquaculture provides 47 percent of the fish available for human consumption. Concerns exist that some farmed fish (i.e., salmon) may inadvertently escape into bodies of water with wild fish populations. Immunocontraception could offer a way to prevent escaped salmon from cross-breeding with wild salmon. NWRC partnered with Colorado State University and the Nofima Marin Sunndalsøra Research Station in Norway to explore this idea by treating 503 Atlantic salmon with an experimental contraceptive vaccine. The vaccine contained a peptide derived from the beta subunit of luteinizing hormone (LH) joined to two different carrier proteins (bovine serum albumin [BSA] and keyhole limpet hemocyanin [KLH]) and then formulated with one of four immunostimulants. Researchers evaluated specific antibody responses to the peptide and each carrier protein.

Vaccines with both *Aeromonas salmonicida* and *Vibrio anguillarum* stimulated much greater LH peptide antibody production than any of the other three immunostimulants evaluated. Results show it is possible to stimulate the immune system of Atlantic salmon to produce antibodies to a contraceptive vaccine. Further research is needed to improve the contraceptive vaccine's formulation and see if the amount of resulting antibodies is enough to reduce fertility in Atlantic salmon. *Contact: Doug Eckery* 

• Combining Fertility Control With Lethal Removal. Fertility control may be a complementary tool to use during large-scale culling programs for feral swine. To better understand how fertility control could be useful in feral swine management, NWRC researchers used models to compare the effects of different amounts of fertility control across a range of culling intensities, population growth rates, and conditions affecting the number of animals moving into a population from surrounding areas



NWRC and university partners developed a geneticsbased tool to identify the plants and animals found in feral swine feces. Results confirm feral swine sometimes feed on game species (i.e., deer, quail) and species of conservation concern. *Photo by USDA, Gail Keirn* 

(i.e., immigration). Adding fertility control reduced feral swine abundance much more than culling alone, and its effects were highest in populations with immigration. However, these substantial benefits from fertility control only occur over a narrow range of population conditions, especially when culling intensities were not high enough to cause population declines (i.e., high population growth rate or low culling intensity) and when immigration was present. Fertility control could be a helpful enhancement to culling in areas where an influx of new animals from surrounding areas is not preventable.

Contact: Kim Pepin

Genetic Tools for Analyzing Feral Swine Diets.

Foral swine are generalist feeders, able to evol

Feral swine are generalist feeders, able to exploit a variety of foods. Yet, their diets remain poorly understood since it's often impossible to identify partially digested food material by traditional stomach content analyses. To overcome this limit, NWRC and university geneticists developed a deoxyribonucleic acid (DNA) sequencing-based approach to identify the plants and animals found in feral swine feces. They also developed and evaluated blocking primers to reduce the amplification and sequencing of feral swine DNA in the samples, thus providing greater returns of sequences from diet-related items. Results for feral swine fecal samples in California and Texas showed acorns, northern bobwhite quail, deer, and elk; this suggests feral swine foraging may impact these important native species. Also of note, the food items of greatest interest (i.e., from game species and species of conservation concern) were found more often when researchers applied the blocking primers. Contact: Toni Piaggio

• Genetic Clustering of Feral Swine Populations. Until the early 1980s, feral swine populations generally remained only in Hawaii, California, and the southeastern States despite a long and varied history of feral swine introductions. The range of feral swine has since expanded, with populations now established in more than 40 States. Their expansion has come with increased economic and ecological costs. To learn more about the genetics of regional feral swine populations, NWRC researchers and multiple Federal and university partners sampled and analyzed DNA from about 1,000 feral swine from 35 States and 47 wild boar from Spain and Iran.

Researchers identified 10 and 12 distinct genetic clusters using two approaches. Genetic clusters represent a cohesive breeding population of animals with limited movement of individuals among clusters. Identifying these clusters and their association with specific geographic areas shows that it's possible to target discrete breeding units for removal. Isolating populations is key to successful eradication. Information on genetic clusters may also help in evaluating management and eradication strategies—officials can see if animals in an area are from a failed eradication or from people recently moving them there illegally. *Contact: Tim Smyser* 

#### Disease Diagnostics, Surveillance, Risk Assessment, and Management

• Effects of Live Bird Market Turnover Rates and Size on Avian Influenza Persistence. The spread of influenza A viruses (IAVs) in live bird markets in Southeast Asia continues to threaten human and animal health. The constant flow of new birds into these markets to replace those sold ensures an endless number of potential hosts. This in turn supports the ongoing transmission and evolution of IAVs. But scientists don't yet know how specific factors—market size, turnover rate, and critical community size<sup>3</sup>—impact the persistence of IAVs in live bird markets. Working with the Imperial



Live bird markets in Southeast Asia facilitate the transmission and evolution of influenza A viruses in birds. Models developed by NWRC researchers and international partners show that market turnover rates and the average daily market population size play a key role in virus persistence. *Photo by Wikimedia Commons* 

College London, Shantou University Medical College, and the University of Hong Kong, NWRC scientists developed a stochastic (random) model to better understand these unpredictable factors and how they may impact the spread of IAVs.

Results showed that market turnover rates and the average daily size of the market played a key role in determining the likelihood of persistence. A single IAV strain needs a market size of about 11,800 birds with a typical turnover rate (50 percent) to persist. And two co-infecting strains (two strains in a single host) need only a 4.2-percent increase in this estimate to persist. These results show the importance of controlling market size to prevent self-sustaining IAV transmission in live bird markets. *Contact: Kim Pepin* 

 Findings From Avian Influenza Surveillance in Wild Birds. After detections of highly pathogenic (HP) influenza A viruses (IAVs) in wild birds in East Asia, wild bird sampling for IAVs increased throughout much of North America. Research and surveillance efforts focused on detecting Eurasian-origin HP IAVs and understanding whether wild birds could

<sup>3</sup> The minimum population size needed at the market for a single IAV strain to persist 1 year after infection starts

maintain and disperse such viruses. In a review of these efforts, experts from the WS National Wildlife Disease Program, U.S. Geological Survey, Canadian Food Inspection Agency, USDA Agricultural Research Service, University of Manitoba, and the University of Georgia identified five key findings: (1) wild birds may disperse IAVs between North America and adjacent regions via migration, (2) HP IAVs can be introduced to wild birds in North America, (3) HP IAVs may cross the wild bird-poultry interface in North America, (4) the likelihood of encountering and detecting a specific virus may be low, and (5) population immunity of wild birds may influence HP IAV outbreaks in North America.

Experts suggest that future research focus on understanding the role of wetlands in maintaining IAVs in wild bird populations, the ways in which IAVs circumvent biosecurity measures at poultry facilities, how IAVs adapt genetically to multiple bird hosts, and the immune response of wild birds to emergent HP IAVs in poultry. *Contact: Tom DeLiberto* 



After the detection of highly pathogenic influenza A viruses (IAVs) in wild birds in East Asia, wild bird sampling for IAVs increased throughout much of North America. A review of those efforts identified five key findings, including that wild birds may disperse IAVs through migration. *Photo by USDA, Sara Harmon*  • Avian Influenza Infection in Cottontail Rabbits.

Influenza A viruses (IAVs) have been reported in some wild mammals in habitats shared with waterfowl. While recent NWRC studies show that cottontail rabbits can shed significant amounts of these viruses, scientists have not evaluated the minimum infectious dose and the efficiency of various infection routes. To explore these topics, NWRC conducted a dose-response study that evaluated oral and nasal routes of infection in 36 cottontail rabbits. The nasal route of infection proved to be the most efficient, with all infected cottontail rabbits shedding viral RNA even at low inoculation doses. The oral route of infection was less efficient, but still produced infection rates of 50 percent or more at relatively low doses. These results suggest that cottontail rabbits are highly susceptible to IAVs at low doses-the same levels of exposure that are routine in waterfowl-contaminated environments. Furthermore, this study supports earlier observations that cottontail rabbits could pose a biosecurity risk to poultry operations since a low level, viruscontaminated water source could be enough to replicate some IAVs in this species. Contact: Jeff Root

Virulent Newcastle Disease Persistence and Spread in Wild Birds. Newcastle disease is a highly contagious and deadly respiratory and neurological virus in poultry. Virulent forms of the virus can have devastating economic effects on domestic poultry production worldwide. Low-virulent strains of Newcastle disease virus (NDV) occur throughout the world in both domestic and wild birds. To evaluate the role wild birds may have in introducing and transmitting virulent NDV in the United States, NWRC worked with the U.S. Department of Homeland Security and the U.S. Department of Energy's Oak Ridge Institute for Science and Education to conduct a qualitative risk analysis.

Findings showed that the legal and illegal movement of live birds, animal products, byproducts, and

animal feed, as well as spillover from wild birds and spontaneous virus mutation, all present some level of risk to the United States. While strict U.S. import regulations for live birds and their products have helped limit NDV risks, there are still concerns about the illegal movement of birds and bird products; spillover from wild birds (i.e., doublecrested cormorants, pigeons, and doves); and NDV vaccines used in domestic poultry. Researchers recommend more monitoring of wild birds to better understand their impact on NDV persistence, along with surveillance and reporting systems to detect and control disease outbreaks in at-risk populations such as backyard poultry. *Contact: Sarah Bevins* 

• Oral Rabies Vaccine Bait Flavors for Skunks.

Wildlife are the leading cause of rabies infections in animals and people in the United States. While the U.S. number of human deaths due to rabies is low, one study has estimated that over 20,000 people are exposed to this disease and receive postexposure vaccination for it each year. To eliminate the rabies virus and keep it from circulating in wild carnivores, the WS' National Rabies Management Program distributes millions of oral rabies vaccine baits each year. Most of these baits target raccoons, but skunks are another important spillover source of raccoon rabies—so wildlife managers need effective oral rabies vaccine products for them, too.

In a recent study, NWRC researchers tested skunks' preference for six different flavors of placebo Ontario Rabies Vaccine Bait (ONRAB), a product permitted in Canada for use with skunks. The researchers also tested the dose of vaccine needed to protect the skunks from rabies infection; this information helps evaluate if it's possible to reduce vaccine volume and dose without compromising efficacy. Results showed that skunks preferred chicken, cheese, and egg flavors over the plain flavor, but they did not show strong flavor preferences. Also, a relatively high dose



NWRC researchers tested the efficacy of a new oral rabies vaccine bait called ONRAB. The bait has shown promise for controlling rabies in raccoons and skunks in Canada. *Photo by USDA, Wildlife Services* 

of vaccine was needed to protect skunks against rabies. These findings aid in further refining ONRAB baits for delivery to skunks in the United States. *Contact: Amy Gilbert* 

• Efficacy of Ontario Rabies Vaccine Bait for Raccoons. The RABORAL V-RG product is the only aral rabies vaccine permitted for use with free

oral rabies vaccine permitted for use with freeranging raccoons and coyotes in the United States. However, another product—the Ontario Rabies Vaccine Bait (ONRAB)—has shown promise for controlling rabies in raccoons and striped skunks in Canada. NWRC researchers evaluated the efficacy of ONRAB for use on raccoons in the United States. Across two experiments, fifty captive raccoons were given either sham or live vaccine baits and then challenged with a lethal dose of rabies virus and monitored for 90 days. Seventythree percent of raccoons in the first experiment and 91 percent of raccoons in the second experiment were protected from rabies infection. All sham-vaccinated raccoons succumbed to rabies. The efficacy results of the second experiment were within recommended standards for animal rabies vaccines in the United States.

These results complement recent field data showing the potential of ONRAB to control and prevent rabies in free-ranging raccoon populations. From 2012 to 2014, NWRC researchers and the WS National Rabies Management Program distributed ONRAB baits in areas of New York, Vermont, and New Hampshire. Prevalence of the rabies virus-neutralizing antibodies in bloodsampled raccoons was 27 percent before the trial and 68 percent after baiting (averaged over 3 years). NWRC shared the captive trial and field data with the USDA Center for Veterinary Biologics (CVB). This information will aid decisions as CVB is considering a request from Artemis Technologies (ONRAB's manufacturer) to permit the product for broader use in the United States. Contact: Amy Gilbert

 Oral Rabies Vaccination Baiting Strategies for **Raccoons.** The seroconversion rate for raccoons after an oral rabies vaccination (ORV) application using RABORAL V-RG is about 30 percent. This is far below the recommended threshold of 60 to 90 percent needed to interrupt rabies virus transmission. Scientists don't vet know whether the low seroconversion rate is a result of vaccine performance, lack of access by raccoons to the bait, competition with other species, overall unattractiveness of the bait, or incomplete bait ingestion. To gain a better understanding, NWRC researchers partnered with WS field specialists to compare seroprevalence in raccoons before and after ORV applications using cluster baiting from a helicopter and hand distribution of single baits.

Overall, the helicopter cluster ORV application delivered more baits than hand distribution, but was less effective in reaching the core areas raccoons used. Seroprevalence did not change as a function of baiting strategy (helicopter

NWRC researchers and WS biologists investigate the prevalence of rabies virusneutralizing antibodies in the blood of raccoons before and after the distribution of oral rabies vaccine baits.

Photo by USDA, Anson Eaglin



versus hand baiting). The average increase in seroprevalence after ORV application was only 8.9 percent. Since both cluster and hand-baiting methods did not achieve the herd immunity needed to disrupt rabies transmission in raccoons, it will be important to evaluate more baiting strategies and other factors. *Contact: Are Berentsen* 

- Porcine Reproductive and Respiratory Syndrome Virus in Feral Swine. Porcine reproductive and respiratory syndrome (PRRS) virus is of economic importance to the U.S. commercial swine industry, costing an estimated \$664 million—or \$1.8 million per day-in productivity losses each year. Although the virus may have come into the United States in 1987 from Europe by imported wild boar, the ongoing role invasive feral swine and wild boar have in transmitting and maintaining PRRS in our country is uncertain. To gain insight on this issue, NWRC researchers analyzed sera samples collected by WS disease biologists from 5,506 feral swine in 26 States for PRRS antibodies. Antibodies to the virus were found in only 1.2 percent of the samples tested. This suggests that feral swine are not a likely source of PRRS infection for domestic swine. Contact: Kerri Pedersen
- Porcine Epidemic Diarrhea Virus in Feral Swine.
   Porcine epidemic diarrhea virus (PEDV) is an emergent pathogen in the United States. It can cause 90- to 95-percent mortality in young, naive pigs and substantial weight loss and dehydration in adult domestic swine. The virus was first documented in the United States in April 2013 and spread rapidly throughout domestic herds, costing the industry more than \$400 million. NWRC researchers have shown for the first time that PEDV also spilled over into feral swine populations. After analyzing blood samples from 7,997 feral swine, the researchers detected PEDV antibodies in 0.1 percent of the samples. Another 3.2 percent of the samples showed feral swine also had been exposed



The role of feral swine in spreading and transmitting pathogens, such as porcine reproductive and respiratory syndrome virus and influenza D, is the focus of several NWRC studies. *Photo by USDA, Wildlife Services* 

to transmissible gastroenteritis virus (TGEV). Like PEDV, TGEV is found only in swine and can cause high mortality rates in piglets. TGEV has been present in U.S. domestic swine for more than 50 years. Since the TGEV-positive feral swine were found throughout the United States and over the entire sampling period, it is likely that TGEV is endemic among feral swine, although continual spillover from domestic swine cannot be ruled out. Whether PEDV will display a similar pattern over time is unknown. *Contact: Sarah Bevins* 

 Influenza D Virus in Feral Swine. Influenza D virus (IDV) was first isolated from domestic pigs in 2011 and has since been identified in domestic cattle, camelid, and small ruminant populations across North America, Europe, Asia, South America, and Africa. It is genetically similar to influenza C virus, which causes respiratory disease in people. In the United States, feral swine serve as an important vector between domestic and wild animals for multiple transboundary diseases, such as influenza. NWRC and Mississippi State University scientists investigated the seroprevalence and transmissibility of IDV in feral swine. During 2012– 2013, they tested 256 swine from four States. Fifty-seven (19 percent) were seropositive for IDV. The researchers also tested 96 archived influenza A virus–seropositive feral swine samples from 16 States. Forty-one of these samples (42.7 percent) were IDV seropositive. Studies with captive feral swine showed that IDV-inoculated individuals shed virus 3–5 days post-inoculation and seroconverted at 21 days post-inoculation; 50 percent of naive feral swine allowed to interact with inoculated swine shed virus, seroconverted, or both.

These findings suggest that IDV has been circulating in feral swine across multiple U.S. States and can be spread among feral swine. Although the economic impact of IDV on commercial livestock remains unknown, feral swine may play a role in the ecology of IDV. Further studies are needed to understand whether other wild animals are infected by IDV and to what extent interspecies transmission contributes to IDV maintenance in domestic and wild populations. *Contact: Fred Cunningham* 

 Risks Related to African Swine Fever Introduction Into the United States. African swine fever (ASF) is a highly contagious viral hemorrhagic disease of pigs, warthogs, and bushpigs. The disease is endemic to Africa and parts of Eastern Europe. Currently, ASF is not found in the United States. Its introduction could cause severe economic impacts due to the loss of production from infected animals and trade restrictions. As part of a risk analysis, researchers with NWRC, the U.S. Department of Homeland Security, and the U.S. Department of Energy's Oak Ridge Institute for Science and Education identified vulnerabilities that could lead to ASF introduction or persistence in the United States or other ASF-free regions.

The analysis showed that, if an ASF introduction occurs, disease spillover events from domestic

swine into feral swine populations could complicate eradication efforts. The virus could also infect tick species found in the United States. Also of note, the current regulatory systems in place for importing live animals, animal products, byproducts, and feed are comprehensive, with considerable Federal oversight. Despite the robust regulatory framework, the illegal import of animals and their products is difficult to control, manage, or regulate. Recommendations for future research include: (1) ASF vaccine development and approval, (2) research on distribution and host preferences of common tick vectors in areas with high populations of domestic and feral swine, (3) integrating ASF into existing swine surveillance, and (4) sampling illegally imported and confiscated wild pig products to test for ASF.

Contact: Sarah Bevins

 Arboviruses in White-Tailed Deer. Arboviruses are a group of viruses transmitted by mosquitoes, ticks, or other arthropods. Common arboviruses include West Nile virus, St. Louis encephalitis virus, and bluetongue virus. White-tailed deer are abundant in the United States, making them good sentinels for monitoring arboviral activity across the country. To better assess the exposure of white-tailed deer to seven arboviruses, specialists with WS' National Wildlife Disease Program and the University of Texas tested 1,508 deer samples collected from 2010 to 2016 for antibodies to eastern equine encephalitis (2.5 percent), Powassan (4.2 percent), St. Louis encephalitis, (3.7 percent), West Nile (6 percent), Maguari (19.4 percent), La Crosse (30.3 percent), and bluetongue (7.8 percent) viruses. The tests detected at least one arbovirus in 51.3 percent of the white-tailed deer sampled and exposure to more than one arbovirus in 17.6 percent of the samples. The specialists note that white-tailed deer can offer a rapid, cost-effective way to monitor arboviral activity. Contact: Kerri Pedersen



Arboviruses are transmitted by ticks and mosquitoes to wildlife and domestic animals. In a recent national surveillance effort by WS and university scientists, 51 percent of the 1,500 white-tailed deer samples tested were positive for at least 1 arbovirus.

Photo by USDA, Agricultural Research Service

 Impact of Field-Edge Habitat on Wildlife Abundance and Disease in Adjacent Crops. Although many areas next to crop fields offer valuable wildlife habitat, farmers may worry this results in increased wildlife activity and damage to adjacent crops.
 Wildlife use of crop fields may also pose a food safety risk by spreading foodborne pathogens, such as *Salmonella* and *E. coli*. To better understand these issues, researchers with the NWRC and the University of California, Davis, studied rodent and rabbit use of field-edge habitats near orchard and row crops and subsequent pathogen prevalence in Yolo and Solano Counties in Sacramento Valley.

Researchers documented a greater number of mammal species in hedgerows versus conventionally managed field edges where vegetation is controlled, but this diversity did not lead to more wildlife use of adjacent crop fields. In walnut orchards, Salmonella and non-0157 Shiga toxin-producing E. coli were detected from two (1 percent) and four (2 percent) rodents, respectively. No pathogens were detected in the row crops. A subset of fecal samples from rodents captured in walnut orchards were positive for Giardia (25 percent) and Cryptosporidium (24 percent) but prevalence was not linked to field-edge habitat type. Overall, the risk of damage or contamination by foodborne pathogens was not greater in the hedgerow-bordered crop fields, though damage could vary by the stage and type of crop and wildlife species present. Contact: Richard Engeman

Hedgerows next to agricultural crops offer valuable wildlife habitat, yet some farmers worry about increased risks of wildlife damage and foodborne pathogens. NWRC and university researchers found that the risk of wildlife damage or contamination to crops near hedgerows was low in two California counties.

Photo by USDA, Natural Resources Conservation Service



 NWDP Surveillance Accomplishments. Each year, WS' National Wildlife Disease Program (NWDP) conducts and coordinates wildlife disease monitoring and surveillance throughout the United States. Below is a summary of its 2018 efforts.

ISSUE	SURVEILLANCE EFFORTS
Avian Influenza	More than 20,800 samples were collected from wild birds and tested for highly pathogenic avian influenza. Suspect samples were further evaluated at the National Veterinary Services Laboratories.
Avian Health	Samples were collected from over 500 wild birds. In most cases, paired serum and swab samples came from 14 States. These samples will be tested for a variety of avian diseases, depending on the species and where they were collected.
Feral Swine Diseases	More than 2,800 feral swine from 36 States and Guam were sampled. Serum samples were tested for antibodies to classical swine fever, swine brucellosis, and pseudorabies virus. Guam samples were also screened for exposure to Japanese encephalitis virus. Selected tissues were collected for genetic research, and archived serum samples were used in research that established feral swine as carriers of influenza D.
Plague and Tularemia	Over 1,100 dried blood samples on filter paper were collected, mainly from coyotes. An NWRC scientist has developed a new assay method to detect plague antibodies, which will be used in the future. Similarly, a new assay method for tularemia is under development. These new assays will allow NWDP to screen blood samples in-house rather than sending samples to outside laboratories.
Leptospirosis	In 2018, NWDP completed a multiyear project to assess the prevalence, distribution, and host range of <i>Leptospira</i> exposure in wildlife. Serum samples from 4,534 animals were tested. The results indicate that <i>Leptospira</i> infection is common and widespread in a number of species.

Table 1. Wildlife disease surveillance activities for 2018



WS' National Wildlife Disease Program conducts wildlife disease monitoring and surveillance throughout the United States on species such as feral swine and coyote. *Photo by USDA, Wildlife Services* 

### Wildlife Damage Assessments

Economic Benefits of Eliminating Feral Swine
 Damage to Crops. Feral swine are known to cause damage to crops and other types of property. A recent survey of feral swine damage to six crops in 11 States showed a loss of nearly \$190 million. Production losses are only part of the overall impact from feral swine. Preventing commodities from reaching the market restricts supply, resulting in higher prices for consumers. Without feral swine damage, market supply would increase, thus pushing prices down. This is clearly a benefit for consumers, but the outcome for producers is less

obvious. Producers experiencing the reduction in feral swine damage would be better off only if the increase in crop quantities made up for the lower prices. Producers who don't see an increase in production would be worse off. These changes in the well-being of consumers and producers are known as welfare changes.

To better understand welfare changes, NWRC and university economists used a partial equilibrium model to calculate how prices and quantities of corn, soybeans, wheat, rice, and peanuts in nine States might change if feral swine damage were eliminated. Results showed that producers and consumers would benefit by a net surplus gain of \$142 million in the first few years, followed by \$89 million per year over the long run. *Contact: Aaron Anderson* 

Feral Swine Damage to Crops at Planting. Most information on feral swine damage to agricultural crops comes from farmer or producer surveys. Actual in-field damage measurements are rather uncommon since they require more resources, labor, and coordination with farmers. NWRC researchers and WS field specialists assessed feral swine damage to freshly planted corn, cotton, and peanut crops in 46 fields in Alabama. Fourteen of the fields were under agreements with WS for feral swine control due to their past history of feral swine damage. Results showed that professional feral swine control greatly reduced the prevalence of damage among fields-the 14 fields that were within the areas where professional swine control operations took place were not damaged. For the 32 fields not receiving such protection, 7 (21.9 percent) received some level of damage. Of those, 40 percent (4 of 10) peanut fields, 15.4 percent (2 of 13) cotton fields, and 11.1 percent (1 of 9) corn fields were damaged. Damage levels were highly variable, both between and within crops. Losses were typically low (less than 1.3 percent),



NWRC researchers and WS field specialists assessed feral swine damage to freshly planted corn, cotton, and peanut crops in 46 fields in Alabama. One field experienced a loss of 54.2 percent—32,401 kg of crop lost, valued at \$15,779. *Photo by USDA Clint Turnage* 

but there were very notable exceptions where more substantial losses occurred. For example, one peanut field experienced a crop loss of 54.2 percent (32,401 kilograms of crop lost), valued at \$15,779. *Contact: Richard Engeman* 

**Economic Benefits of Falcons To Prevent Bird** Damage to Fruit Crops. "Conservation Biological Control," or CBC, is a term used to describe changes made in an environment to protect or enhance native predator populations and reduce the impact of pest species. One simple CBC practice is to install artificial nest boxes and roosting sites for falcons, such as the American kestrel. Kestrels are widespread, highly mobile, generalist predators that hunt in open habitats, including human-dominated landscapes. Kestrels using orchard nest boxes in the fruit-growing region of northwestern Michigan eat insects, mammals, and fruit-eating birds. NWRC recently worked with Michigan State University to find out whether kestrel activity around nest boxes and artificial



A cost-benefit analysis showed high economic returns for fruit producers who install artificial nest boxes for American kestrels in their orchards. The falcons help to reduce the abundance of fruit-eating birds.

Photo by Wikimedia Commons, Bill Bouton

perches increases predation, as well as perceived predation, risks to fruit-eating birds.

In this study, the researchers surveyed fruit-eating bird abundances in cherry orchards with and without kestrel boxes. They also conducted a cost-benefit analysis of nest box installation and used regional economic modeling to estimate the impacts of increased cherry production in Michigan. Results showed that fruit-eating bird counts were much lower at orchards with active kestrel boxes. Furthermore, benefit-cost ratios for kestrel nest boxes indicated that for every dollar spent on nest boxes, \$84-\$357 of sweet cherries would be saved from fruit-eating birds. Regional economic modeling predicted that increased sweet cherry production from reduced bird damage would result in 46–50 jobs created and \$2.2–\$2.4 million in increased income for the State of Michigan over a 5-year period. Contact: Stephanie Shwiff

- **Calculating Strike Risks for Different Bird Species.** Bird collisions with aircraft (also known as bird strikes) cost the aviation industry more than \$1 billion each year. Identifying which bird species pose the most risk to aviation may help airport managers develop targeted management methods and strategies. NWRC and the WS Aviation Hazard Program developed a model to estimate strike risks for different bird species. The model combines the relative hazard score (RHS) and bird strike frequency for common bird species found at airports. RHS is the percentage of total strikes for each species that results in damage, substantial damage, or a negative effect on the aircraft's flight (e.g., delay, emergency landing). It provides an index of severity, but not frequency. Of the 11,364 strike records and 79 bird species studied, red-tailed hawks, Canada geese, turkey vultures, pigeons, and mourning doves pose the greatest risk (i.e., frequent and damaging collisions) to aircraft in the United States. Researchers encourage airport wildlife biologists to adapt the model to their airport-specific strike data and use standardized bird surveys, corrected for detection bias, to further prioritize management efforts at their airports. Contact: Travis DeVault
- Species Flight Maneuverability and Frequency of Bird Strikes. Bird strikes involve financial loss to commercial, civil, and military aviation worldwide and are a source of mortality for birds. Purdue University and NWRC researchers investigated whether certain biological traits of birds, such as their body mass (takeoff time and distance), eye size (visual acuity), brain size (cognitive ability), and wing loading and wing aspect ratio (maneuverability), increased or decreased their risk of collisions with aircraft. Wing loading is the ratio of body mass to wing area and reflects the wing's ability to turn relative to the bird's body. Models comparing the traits of 93 bird species with bird strike frequency showed that birds with

Of the 11,364 strike records and 79 bird species studied by NWRC researchers, redtailed hawks (pictured), Canada geese, turkey vultures, pigeons, and mourning doves pose the greatest risk to aircraft in the United States. Photo by USDA, Christopher Loftis



greater maneuverability (i.e., lower wing loading) had a higher frequency of bird strikes. Examples of bird species with greater maneuverability include swallows and songbirds. Bird species with lower maneuverability include waterfowl and raptors. Researchers speculate that species with greater maneuverability may fly slower and take off at shorter distances to avoid aircraft. They may also be hazed less than other more obvious species at airports given their smaller body size and people's belief that they cause less damaging bird strikes. *Contact: Bradley Blackwell* 

# Reducing Airplane Collisions With Large Mammals. Airplane collisions with large mammals during takeoff and landing pose a significant risk to U.S. aircraft safety and cost airlines millions of dollars in repairs each year. Many large mammals are attracted to airports because of their surrounding habitats. Giving airport managers options for alternative land cover on and near airports may help reduce mammal-airplane collisions. To explore these options, NWRC, Mississippi State University, and University of Georgia researchers compared white-tailed deer and coyote use of two experimental fields: one with mixed native warmseason grasses and one with switchgrass (*Panicum*)

*virgatum*). Observing the fields via remote cameras, researchers found that coyotes and deer used the switchgrass field much less than the mixed native warm-season grass field—27 percent and 51 percent less, respectively. Considering that deer and coyotes are among the most hazardous mammal species to aircraft, fields of switchgrass may be a better alternative land cover. Researchers plan further studies to compare deer and coyote use of switchgrass to more traditional airport land covers, such as turf grasses and row crops.

Contact: Travis DeVault

# • Competitor or Prey? How Coyotes View Pets. Over

the past several years, the number of urban coyote attacks on people and pets has increased. To better understand whether coyotes view pets as prey or instead as competitors or a threat, Utah State University, Colorado State University, and NWRC researchers analyzed the diet of coyotes in the Denver metropolitan area. Results showed only small percentages of trash and domestic pets in the coyotes' diets. The presence of pets did not coincide with an increase in pet conflicts, supporting the hypothesis that coyote conflict with pets is driven mainly by competition or a threat



Many large mammals are attracted to habitats near or on airports. NWRC and university researchers studied white-tailed deer use of switchgrass—an alternative land cover for use around airports. Deer were observed 51 percent less in the switchgrass fields than in mixed native grasses. *Photo by Mississippi State University, Ray Iglay* 

response. Coyotes ate mostly native plants and animals, with rodents and rabbits being the most prevalent diet items. Coyotes ate rodents and nonnative plants more often in high-density housing areas and deer, corn, and native plants more often in low-density housing areas. Coyotes also ate more fruit and insects during the summer and autumn and more mammals and birds during the winter and spring. As human–coyote conflicts increase in urban areas, this information may offer insights that can promote better coexistence. *Contact: Stewart Breck* 

Predicting Livestock Predation. Endangered Mexican wolf (*Canis lupus baileyi*) predation on livestock is a primary concern affecting wolf recovery because it causes economic losses and negative attitudes toward wolves. To help reduce the potential for Mexican wolf predation on cattle in the wolf's recovery range, NWRC, New Mexico State University, the U.S. Geological Survey, and Wildlife Services experts developed a risk model based on confirmed depredation incidents, landscape and human variables, and cattle and natural prey populations. The model identified specific vegetation features and higher relative abundance of elk as two key factors linked to increased predation risks. Other factors that influenced depredation risk were gentle and open terrain and greater distances from roads and developed areas. The research team used these factors to create a risk map that identifies areas in Arizona and New Mexico with relatively high potential for cattle depredations by endangered Mexican wolves. Researchers note these areas should be the focus of future mitigation efforts including nonlethal damage management methods, especially during calving season. Contact: Stewart Breck



Endangered Mexican wolf predation on livestock is a main concern affecting wolf recovery. As part of a multiagency effort, NWRC researchers identified vegetation characteristics and other factors that increase livestock predation risks in the wolf's recovery range. Photo by Wikimedia Commons

Economic Impacts of Black Bear Damage to Douglas Fir Trees. Black bear damage to timber resources is a concern among timber harvesters, yet methods to assess bear damage are known to be inaccurate. Having improved methods is important for reliable data. In recent studies, NWRC and university partners worked on this issue by using ground surveys to quantify black bear damage to Douglas-fir stands in western Washington and Oregon. This area was a prime location to base the surveys, as black bears forage on western conifers in early spring when other food sources, such as berries, are unavailable. The researchers used growth and yield models, harvest simulation models, and net present value models to evaluate economic impacts at the forest stand scale using two damage scenarios. The first damage scenario (Salvage) accounted for mortality and volume losses of both fully and partially girdled trees while valuing removal of partially damaged trees. The second damage scenario (Total Loss) assumed complete loss of all black bear-peeled trees regardless of the amount peeled.

Black bear damage caused economic losses ranging from 4 to 16 percent (Salvage) and 17 to 46 percent (Total Loss) of net present value at the stand scale. Damage costs in the Total Loss scenario were on average four times greater than those in the Salvage scenario. A second study using the same methodology at a landscape scale found that the economic impact under both damage scenarios was less than 0.35 percent of net present value, suggesting that black bear damage is not evenly distributed. The study also showed that aerial surveys overestimated bear damage by about fivefold due to misclassification with root disease and failed to detect partially peeled trees that contributed to economic loss. This approach may help improve forest management plans that assess damage by black bears and other wildlife species. Contact: Jimmy Taylor

 Seed Predation and Dispersal by Invasive Rose-Ringed Parakeets. Rose-ringed parakeets (Psittacula krameri) are an invasive species in Hawaii with more than 6,000 individuals residing on Kauai. These birds destroy crops, but impacts to other native and non-native species are largely unknown. NWRC researchers teamed up with WS field specialists to analyze the diets of 64 rose-ringed parakeets from 5 sites on Kauai. All parakeets harvested had vegetation in their crops and gizzards, and 80 percent of the contents (by mass) was seed. The most common food items were corn (found in 67 percent of all birds and averaged 31 percent of their diet) and yellow guava (found in 97 percent of all birds and averaged 30 percent of their diet). Parakeets could be dispersing invasive yellow guava seeds, as 66 percent of the birds sampled had an average of three intact guava seeds in their crops and gizzards. These findings of a diverse plant diet, frequent seed predation, and potential to disperse invasive plant seeds implies that land managers in agricultural, urban, and natural areas should be concerned with the current expansion of these invasive birds on Kauai and elsewhere. Contact: Aaron Shiels



An analysis of 64 invasive rose-ringed parakeets' diets on Kauai showed the birds could be spreading invasive plant seeds across the island. *Photo by Wikimedia Commons* 

# Wildlife Management Methods and Evaluations

 Hazing Programs for Urban Coyotes. As coyotes expand and adapt to living in urban areas, they become more tolerant of people, which often leads to human-coyote interactions and conflicts. Hazing (scaring) involves using deterrents to move an animal out of an area or away from a person or to discourage an undesirable behavior for a short period of time. In an effort to see if hazing urban coyotes can reduce human-coyote conflicts, an NWRC researcher partnered with Jefferson County Open Space officials to train Denver Metro Area residents for participation in coyote hazing trials. As part of the program, 207 trained volunteers recorded 739 observations of coyotes, 96 (13 percent) of which involved a person hazing a coyote. In more than 70 percent of the hazing attempts, the coyote moved 10 feet away from the person doing the hazing, which demonstrates the importance of this tool as a short-term strategy for reducing negative interactions. Only in rare cases did hazing have no impact on deterring coyotes; this was mainly when people had dogs on leashes. Researchers speculate that the competitive



A study of urban coyotes in the Denver Metro Area showed that community-level hazing of urban coyotes may be an effective, short-term tool, but it does not alter coyote behavior over the long term.

Photo by U.S. Department of the Interior, National Park Service

interactions between coyotes and dogs makes hazing less effective.

In a separate study, casual park visitors were asked to self-report on their experiences with coyotes as well as their knowledge and use of hazing techniques learned from county park signs, staffed education stations, emails, and social media posts. Based on 495 self-reported results, most park visitors indicated they would haze a coyote in the future and that the educational effort influenced their decision to haze. Results show that community-level hazing of urban coyotes can be an effective, short-term tool for establishing a safety buffer during a negative coyote encounter, but did not support the notion that hazing would alter coyote behavior over a longer timeframe. *Contact: Stewart Breck* 

Influence of Habitat Variation on Predation Damage **Management.** Lethal predator control programs are used to protect endangered and threatened species and to benefit economically valuable prey species, such as deer, pronghorn, elk, and livestock. Managers often lack mechanisms to evaluate program effectiveness and may only be able to record the number of predators removed and prey survival or population growth. Yet in doing so, other factors may hide or exaggerate the impact of the program and its results. Hierarchical models can help explore complex interactions across multiple data sets or spatial scales, especially when there is a need to account for variation and uncertainty. NWRC, Utah State University, Brigham Young University, and Utah Division of Wildlife Resources scientists used such a model to evaluate the effectiveness of aerial covote removal in relationship to space use by mule deer during fawning season.

Results showed that coyote removal was more likely in open shrubland with low to moderately rugged terrain. A coyote's risk of being removed by aerial operations declined as ruggedness or tree cover increased. Such habitats require pilots to fly more attentively (due to human safety concerns) and likely make it harder for the flight crew to spot coyotes on the ground. Models also showed that deer select fawning sites with fairly rugged terrain near unpaved roads, shrublands, aspen, conifers, or hardwoods. These findings highlight a mismatch between where coyotes are most easily removed through aerial operations (i.e., more open habitat) and preferred fawning grounds. The effectiveness of aerial coyote removal can vary based on landscape features, and managers may benefit from targeting the more favorable areas (i.e., those with a high likelihood of coyote removal and high deer populations).

Contact: Julie Young

Effects of Coyote Removal on Sage Grouse Survival. Greater sage-grouse (Centrocercus urophasianus) populations have declined across western North America, and the species now occupies only 56 percent of its historical range. Predation from coyotes, badgers, golden eagles, ravens, striped skunk, and other species influences sage-grouse nest success. NWRC and Utah State University researchers investigated whether removing coyotes improved female sage-grouse survival or nest success in the Bighorn Basin of Wyoming. The researchers fitted 69 female grouse with very high frequency (VHF) necklace-style transmitters and monitored them. Results showed no differences in female grouse survival or nest success between sites with targeted birds, non-targeted birds, and no coyote removal. Removing coyotes, the main predator of nests and adult females identified within this system, did not improve female survival or nest success during the 2-year study. However, the researchers recommend long-term monitoring, which may offer a more robust understanding of this complex relationship. Contact: Julie Young



Radio transmitters were placed on greater sage-grouse hens to study the effects of coyote removal on their survival and nest success. *Photo by USDA, Wildlife Services* 

 Evaluating Livestock Protection Dogs To Prevent Wolf Predation. Nonlethal tools for reducing livestock predation, such as livestock protection dogs (LPD), are widely used by sheep producers in the United States. However, common LPD breeds, collectively called "whitedogs," appear less effective against wolves than coyotes. NWRC and Utah State University researchers looked into this issue in a recent study about behavioral differences among LPD breeds. The study compared the behavior and response to predation threats of whitedogs in the United States with three European breeds (Kangals, Karakachans, and Transmontanos). The breeds were selected for their boldness toward carnivores, history of use in areas with wolves, lack of aggression toward people, and size. In 2015 and 2016, researchers tested the dogs' responses to simulated encounters



NWRC and Utah State University researchers compared the behavior and response of three large, European breeds of livestock protection dogs (Kangal pictured here) to larger predators, such as bears and wolves.

Photo by USDA, Julie Young

with a wolf (decoy) in Idaho, Montana, Oregon, Washington, and Wyoming.

Results showed few significant behavioral differences among the tested breeds. Kangals tended to be more investigative when engaging a decoy, Karakachans more vigilant, and Transmontanos more able to decipher a threat. Transmontanos also spent less time scanning for threats than whitedogs, and Karakachans moved more than whitedogs. While these subtle behavioral differences may help producers choose an appropriate LPD breed for their needs and circumstance, results suggest that behavioral differences among breeds are less common than previously suggested. *Contact: Julie Young* 

• Learned Behaviors in Wolves. Coyotes, wolves, bears, and other carnivores often learn to overcome their fear of novel items in their environment, allowing them to access new food and other resources. For instance, some female black bears in Yosemite National Park have taught their cubs to overcome their fear of people in order to raid campgrounds for food. Nonlethal methods to prevent carnivore conflicts are often put in place after learning has already occurred. This can diminish their ability to prevent future conflicts. To learn how past experience (i.e., conditioning) influences a wolf's motivation and persistence, researchers with NWRC; Colorado State University; Montana Fish, Wildlife and Parks; and the Wildlife Science Center monitored investigative (sniffing, licking, scratching, nudging) and work (pawing, chewing, tugging, pinning) behaviors of both experienced and non-experienced captive wolves seeking a food reward.

Wolves that were already conditioned to the placement of a novel food reward in their pens began investigative (11 times) and work (4 times) behaviors faster than non-conditioned wolves. These results support the idea that increased learning and experience reduces a wolf's fear of new situations and objects and expedites exploratory behaviors. From a management perspective, this highlights the importance of using nonlethal methods proactively to curtail wolf learning and subsequent rewards (i.e., successful



Coyotes, wolves, bears, and other carnivores often learn to overcome their fear of novel items in their environment, allowing them to access new food and other resources. Using nonlethal predation damage management methods proactively may help curtail such learning.

Photo by Wikimedia Commons, Steve Jurvetson

predation on livestock). Further, where learning has already occurred, wildlife managers and livestock producers should use prevention measures continuously and actively to deter future conflicts. *Contact: Stewart Breck* 

Simple Fix To Reduce Bear Conflicts. Black bears are quick to take advantage of food left out by people. Black bears forage on garbage, bird seed, dog food, and other food items commonly found around homes and businesses. This has led to an increase in conflicts between bears and people in cities and towns across America. Unfortunately, these conflicts often end badly for the bears, with many being killed or moved to protect public safety and prevent property damage. To help address these issues, NWRC researchers partnered with Colorado Parks and Wildlife to evaluate whether the use of bear-resistant trash containers in Durango, CO, could reduce bear-human conflicts in this mountain town.

The town was divided into two treatment and two control areas for comparison. Residents in the treatment areas received bear-resistant containers free of charge, while residents in the control areas continued to use their own trash containers. Trash-related conflicts were 60 percent lower, and compliance with local wildlife ordinances increased 39 percent in the areas with bearresistant containers. Researchers recommend that municipalities within or adjacent to bear habitat consider bear-proofing measures, such as giving residents bear-resistant trash containers, putting in place bear-proofing ordinances or regulations, and increasing the enforcement of existing laws. *Contact: Stewart Breck* 

Modifying Cattle Feed To Prevent Bird Consumption. Invasive European starlings cause damage to commercial dairies by eating feed meant for dairy cows. Large foraging flocks of starlings alter the physical composition of cattle feed, often eating the most nutritious parts and possibly affecting milk production. To better determine if production losses occur as a consequence of foraging starlings, NWRC and Colorado State University researchers conducted controlled feeding experiments with captive starlings. Results showed starlings ate the high-energy portion of the cattle feed, reducing the availability of starch and crude fat. Using the dairy National Research Council production model equations, these nutritional losses could reduce the productivity of dairies.

Large foraging flocks of invasive European starlings alter the physical composition of cattle feed, often eating the most nutritious parts and possibly affecting milk production in dairy cows. Photo by Nick Dunlop



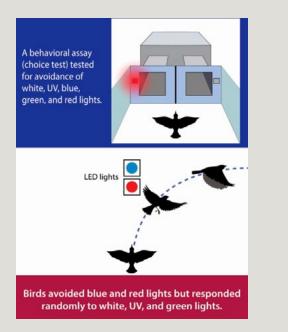
Researchers also assessed whether changes in the diameter of pelleted feed could deter foraging starlings. Six pelleted feed treatments of varying diameter were offered to captive starlings. With pellets of 0.95-cm diameter or larger, starlings consumed the feed at least 79 percent less. Preventing starlings from eating cattle feed by modifying the size of the rations may force the birds to leave a dairy in favor of other feeding sites. Researchers note the added time and cost of feeding milled or pelleted supplements to cattle may make economic sense when compared against the cost of lost feed and production. *Contact: Scott Werner or Randy Stahl* 

Bird Response to Approaching Vehicles. Animal responses to approaching predators or risky objects (e.g., vehicles) vary across species due to differences in experience, anti-predator strategies, and sensory capabilities. Within species, responses to approaching threats can also vary due to individual age, sex, condition, risk tolerance, and habituation. Recent research aimed at enhancing a bird's ability to detect an approaching vehicle indicates that speed and awareness of the approaching vehicle influence a bird's alert and escape behaviors. Yet, little is known about how repeated, non-injurious interactions with vehicles may impact a bird's behavior, if at all. To explore this issue, NWRC and university researchers exposed brown-headed cowbirds to a simulated vehicle approach using a video viewing chamber. The researchers collected data on alert and flight behaviors within and among individual birds in response to a virtual, oncoming vehicle.

Results showed low repeatability in alert and flight behaviors by brown-headed cowbirds, suggesting that an individual bird's avoidance behavior to an approaching vehicle often varies. The findings have implications for efforts to reduce bird-vehicle collisions. Past NWRC studies have shown that vehicle approach speed is a key factor in birds and mammals having ineffective avoidance responses. This study shows that approach speed may be even more important when avoidance responses vary. Reducing vehicle speeds (e.g., via lowering posted speed limits on roads) may be the best way to reduce animal-vehicle collisions, especially in areas where collisions are frequent and such regulations are practical. *Contact: Travis DeVault* 

Assessing Bird Avoidance of High-Contrast Lights. Birds frequently collide with man-made objects and vehicles (e.g., buildings, cars, airplanes, power lines). Lights have been suggested as a way to alert birds and minimize the chances of collisions. But little is known about what kinds of lights work best to deter birds-bird vision is different from human vision, and bird species also differ in how they perceive objects. In a recent study, Purdue University and NWRC researchers explored this issue. They used perceptual models to find out which LED (light emitting diode) lights are more visible to brown-headed cowbirds, based on the lights' specific wavelengths and color differences (high chromatic contrast). The researchers then evaluated the birds' response to the lightsavoidance, attraction, or neutral-with a behavioral test. Individual birds were released into an arena where they moved in a single direction and had to choose a left or right exit. One of the exit routes included a lit LED light, the other an unlit LED light.

Findings suggest that brown-headed cowbirds significantly avoid exit routes with lit LED lights that have peaks at 470 nm (blue) and 630 nm (red), but do not avoid or prefer LED lights with peaks at 380 nm (ultraviolet) and 525 nm (green) or broad-spectrum (white) LED lights. Researchers note the findings are limited only to steady lights under diurnal ambient light conditions and a single bird species. However, the approach could be applied to a wide set of conditions and species. Identifying wavelength-specific lights for use as



Lights have been proposed as deterrents to reduce bird collisions with man-made objects and vehicles. Combining perceptual models and behavioral choice tests, Purdue University and NWRC researchers determined that brown-headed cowbirds avoid blue and red lights. *Graphic by Purdue University* 

visual deterrents may help reduce bird collisions with stationary and moving objects. *Contact: Brad Blackwell* 

 Landscape Structure and Bird Strikes. Landscapes in and around airports may attract birds and other wildlife, which can result in wildlife-aircraft collisions. Pairing known wildlife attractants with dispersal, repellents, and population management may reduce bird strikes within the airport boundaries. Yet, the effectiveness of these techniques is limited to areas close to the ground, and they are not suitable once the aircraft is beyond the airport boundary and airborne. In recent years, the number of damaging strikes outside airport boundaries (more than 152 meters above ground level and over 1.5 miles from runways) has increased. NWRC researchers studied the effects of landscape features on the rate of damaging bird strikes at various distances

(3-, 8- and 13-kilometer [km] radii extents) from 98 civil airports.

Results showed that the rate of damaging strikes was influenced by large open areas and close proximity of wetlands, water, and cultivated crops at the 8- and 13-km extents. Within 3 km of an airport, increasing landscape diversity and crop area increased the strike rate. Researchers conclude that landscape structure and composition are predictors of the damaging bird strike rate at multiple spatial scales. These results can help promote collaborative management among wildlife professionals, airport planners, and landowners near airports to create an environment with a lower probability of damaging bird strikes. Efforts should focus on minimizing the amount of crops, especially corn, and increasing the distances between patches of open water. Contact: Travis DeVault

• Evaluating Avian Radar Systems for Tracking Birds at Airports. Avian radar technologies have the potential to track bird movements and activity in areas where human–wildlife conflicts might occur (e.g., airports, wind-energy facilities). However,



Landscapes in and around airports can attract birds and other wildlife. NWRC researchers evaluated the landscape features around 98 airports and their potential influence on bird strikes. *Photo by Wikimedia Commons* 

the capabilities and limits of these technologies are relatively unknown, and ground-truthing studies are needed to help wildlife managers understand the biological meaning of radar information. WS research and field experts partnered with University of Illinois researchers to evaluate the efficacy of three X-band marine radar sensors for tracking birds and flocks of birds observed at Chicago O'Hare International Airport. The radars were equipped with parabolic dish antennae set at 2, 4, and 8 degrees above the horizon, respectively. The researchers used field observations of birds or flocks to determine how often the three radar sensors gave corresponding information of bird targets.

In total, there were 972 sightings of individual birds or flocks on the airfield that had the potential to be observed by the radars. Of these, 143 (15 percent) were tracked by at least 1 radar sensor. All confirmed tracks of individual birds or flocks were 4.8 km or less from the radars. Larger bodied birds, birds/flocks flying at higher altitudes, and birds/flocks flying closer to the radars increased the radars' ability to detect and track them. Overall, the radar tracking rates for birds or flocks observed at the airport were lower than expected based on findings from other studies. The results show that an individual bird or flock's distance from the radar has a strong influence on whether or not the radar sensor tracks it. When using avian radar to detect and track birds, wildlife managers could best apply this tool by placing the radar system within 4 km of the landscape, habitat, or bird's suspected flight path. Contact: Brian Washburn

• Factors Affecting Translocation Success With Red-Tailed Hawks. One common way to reduce human-wildlife conflicts is capturing and moving (translocating) problematic wildlife. In particular, raptors near airports are often translocated since they pose an aviation safety hazard. Although this method has strong public support, little is known about its efficacy or what factors might influence the return of translocated birds to an airport. From 2010 to 2013, WS airport biologists captured, banded, and translocated 577 red-tailed hawks at Chicago O'Hare International Airport and monitored for their return. An NWRC researcher then analyzed the results to evaluate the efficacy of translocation.

About 82 percent of the translocated hawks were not seen again at Chicago O'Hare International Airport after their release, and their fate is unknown. The other 18 percent (102 hawks) returned to the airport. Results showed that hawks older than 1 year of age were 2.4 times more likely to return to the airport after translocation than younger birds. Odds of returning to the airport went up 4 times when translocation occurred during the breeding season, and 12 times for each subsequent translocation event involving the same hawk. The cost of one translocation event to the release sites that were 81, 121, 181, and 204 km (50 to 127 miles) from the airport was \$213, \$284, \$362, and \$426, respectively. Wildlife management programs at airports can increase their effectiveness and reduce costs by using release sites at least 80 km from the airport, translocating only younger birds during the non-breeding season, and translocating each individual hawk only once. Contact: Brian Washburn

 Translocating Short-Eared Owls From Airports. Wildlife-aircraft collisions (wildlife strikes) pose a serious safety risk and cost civil aviation at least \$957 million per year in the United States. Aircraft collisions with birds accounted for 97 percent of all reported strikes. A review of available wildlife strike information suggests short-eared owls (*Asio flammeus*) are frequently struck by aircraft during the winter months at numerous airports within the Lower Great Lakes Region of the United States. This species favors grassland habitats for nesting, roosting, and foraging; thus, the large, grassy areas at airports can be attractive to these birds. To better understand the efficacy of translocation to prevent short-eared owl collisions with aircraft, WS airport biologists and an NWRC researcher live-captured, banded, and released 32 short-eared owls about 64 to 80 km (40 to 50 miles) away from airports during 2012 to 2015. Only one short-eared owl was sighted again after its release; it was found on a different airport from where it was captured. While these results are promising, researchers need more information on the survival rate of translocated short-eared owls before they will know the full impacts of this nonlethal technique. *Contact: Brian Washburn* 

#### Culling Strategies To Improve Efficiency.

Determining efficient and effective culling strategies to reduce invasive and pest species is challenging. Managers must often determine when, where, and how resources should be distributed to have the biggest impact. For some species with seasonal births, it may be most effective to cull just prior to



To better understand the efficacy of translocation in preventing short-eared owl collisions with aircraft, WS airport biologists and an NWRC researcher live-captured 32 short-eared owls at airports. The birds were banded and then released different distances away from airports to see if they would return. *Photo by USDA, Megan Baker* 

birth pulses, before a substantial number of new individuals are added to the population. NWRC researchers used a spatial simulation model of feral swine population dynamics, which accounted for birth seasonality and the timing, spatial pattern, and intensity of culling, to determine the efficiency of different feral swine management strategies in space and time. Three key findings arose: first, since feral swine births peak in the spring and fall, increasing culling for this species in the summer and winter rather than other times of year could improve efficiency overall. However, because feral swine do not have distinct, large birth pulses, the timing of culling was not as impactful as spatial targeting. Second, culling in a wave-like pattern across the landscape (i.e., zoning) was the most efficient strategy regardless of culling intensity. And third, lower culling intensities can be as effective as higher intensities if used in conjunction with zoning. Contact: Kim Pepin

 Raccoons Use Ingenuity To Access Resources. The WS National Rabies Management Program works to improve methods for vaccinating raccoons and other wildlife against rabies. Knowing how raccoons evaluate and solve problems may help in developing such methods. In a recent study, University of Wyoming and NWRC researchers used the Aesop's fable test to evaluate the ability of raccoons to understand cause-and-effect. In Aesop's Fable "The Crow and the Pitcher," a thirsty crow discovers how to raise the level of water in a narrow vessel by displacing it with pebbles. In a similar exercise with captive raccoons, two of eight animals learned to displace water in a tube to retrieve floating marshmallows after watching researchers demonstrate the task. Researchers note more of the raccoons might have learned the task if they had more time to familiarize themselves with the stones and the water tube. These findings are an important first step in expanding our knowledge of causal understanding in raccoons. Contact: Shylo Johnson



The WS' National Rabies Management Program works to improve methods for vaccinating raccoons and other wildlife against rabies. Knowing how raccoons evaluate and solve problems may help in developing such methods. *Photo by University of Wyoming, Lauren Stanton* 

 Integrated Approach to Brown Treesnake Control. Guam's native ecosystem, agriculture, and infrastructure have been severely impacted by invasive brown treesnakes (BTS). Managing the snakes focuses on preventing their dispersal from Guam to other locations, detecting and removing them from other non-native locations, reclaiming areas on Guam as snake-free for the reintroduction of native wildlife, and protecting small sensitive sites (e.g., power stations, bird nesting sites) on Guam from snake intrusions. WS experts recently summarized Guam's successful collaborative BTS control efforts. Integrating available tools, such as traps, oral toxicants, spotlight searches, detector dogs, barriers, cargo fumigation, and public outreach, has prevented the spread of BTS from Guam. From 2007 to 2017, WS field specialists removed 232,686 BTS on Guam with 0 reports of snakes escaping to other locations. Furthermore,

no live BTS have been discovered in Hawaii since 1994 nor found in cargo since the implementation of Guam's detector dog program in 1993. New tools that reduce labor and costs would allow the control program on Guam to be expanded. Guam's BTS control program is the world's first large-scale invasive snake management effort. As such, the methods and concepts for addressing BTS provide a foundation and model for developing invasive snake management efforts elsewhere, such as for the invasive Burmese python in Florida. *Contact: Richard Engeman or Aaron Shiels* 

 Invasive Snakes Are Inaccessible to Control Tools While Digesting Large Meals. Many snakes are adapted to eating large prey items at infrequent intervals. Digesting large prey is physically demanding, and large prey bulges can impair snake movements. To cope with the demands of digestion, some snakes stop feeding and hide. The invasive brown treesnake (BTS) is a nocturnal, arboreal snake that was accidentally introduced to the island of Guam. Traps and bait stations used in BTS damage management rely on snakes actively seeking food. If snakes stop foraging after feeding on large meals, this could make management of this invasive snake more difficult. NWRC and U.S. Geological Survey scientists measured differences



Researchers studying invasive brown treesnakes on Guam discovered the species drastically reduces its level of activity after eating large prey items, which may impact brown treesnake management on the island. Photo by U.S. Geological Survey, Michael Hogan



Elevated and floating bait stations for potential use in future rodent eradication efforts were field tested in intertidal zones on Wake Atoll in the Pacific Ocean. *Photo by USDA, Dean Foster* 

in BTS activity, movement, habitat use, and detectability (i.e., the probability of finding a snake when searching or trapping for it) between snakes that had eaten a large meal and those that had not.

Compared to unfed snakes, snakes in the feeding treatment group showed drastic decreases in hourly and nightly activity rates, differences in refuge height and type, and a strong decrease in detectability by trapping and visual surveys for about 5–7 days. Researchers note that BTS management strategies need to account for these periods of unavailability and prevent the spread of BTS into new environments where large prey items are abundant. *Contact: Shane Siers* 

• Rodenticide Baiting in Intertidal Habitats on Tropical Islands. Successful rat eradications on islands are paying tremendous conservation dividends, but failed eradications are economically and environmentally costly. In order for a rodenticidebased eradication effort to be successful, every rat in every habitat must be exposed to enough toxic bait to receive a lethal dose. A review of the failed rat eradication effort on Wake Atoll in the central Pacific Ocean suggests that limited bait in the intertidal habitats of the atoll's lagoon may have caused the failed eradication. The habitat was not treated by aerial broadcast due to concerns about the loss of bait to tidal action and possible contamination of the marine environment. To prepare for a second eradication attempt on Wake Atoll, NWRC and Colorado State University researchers developed two alternative bait application strategies.

Both strategies distribute enough bait for a long enough period of time to successfully target rats while minimizing bait entering the ocean. Camera traps and experimental bait provisioning methods used during a demonstration showed rats foraging in the targeted tidal zone habitat and consuming placebo bait. Placebo bait was made available to rats through hand broadcast and elevated and floating bait stations to prevent bait spillage into the marine environment and minimize bait interference by crabs. These strategies will likely be considered during a future eradication attempt on Wake Atoll and are useful for rodent suppression efforts in other wetland areas. *Contact: Shane Siers* 

# Wildlife Population Monitoring Methods and Evaluations

 Using Camera Traps To Determine Species Reinvasion Potential. Wildlife managers are often tasked with protecting specific habitats and areas from invasive species damage. This may involve removing invasive species despite a substantial risk of reinvasion by neighboring populations. To help managers determine the status of invasive feral swine in an area of California and the risk of reinvasion after removal, NWRC researchers developed a dynamic occupancy model. The model monitors changes in the distribution of feral swine based on camera-trap data. Occupancy analysis (i.e., presence or absence) using passive detectors, such as camera traps, is a common way to track low-density populations. Dynamic occupancy models evaluate patterns in occupancy status over time and determine factors related to local extinctions, colonizations, and detections.



Using camera-trap data, NWRC researchers developed a model to monitor changes in the distribution of invasive feral swine. The information helps wildlife managers optimize their resource and removal efforts.

Photo by Wikimedia Commons, Western Arctic National Parklands

The NWRC's model showed that with an average of one camera per 4 km<sup>2</sup>, researchers could predict the absence of feral swine with 95-percent certainty 4 months after the last animal was detected. Wildlife managers can use this to help determine when they should switch from area-wide surveillance to surveillance focused on reinvasions. By knowing where animals are most likely to be found and whether they are truly absent from a site, managers can optimize their resources and removal efforts.

Contact: Amy Davis

Human Role in Feral Swine Movement. Feral swine are the most widely distributed invasive wild ungulate in the United States, yet little is known about the factors that influence their dispersal and colonization. NWRC and University of Florida researchers used a population genetics approach to describe patterns of dispersal and colonization among feral swine populations in Florida. By genotyping 482 swine from 39 locations, researchers revealed genetically distinct subpopulations. Some of the subpopulations showed patterns of significant interbreeding or genetic isolation that are not easily explained by natural dispersal and suggest the animals were moved by people. Data also showed that transition holding facilities for feral swine are not secure, which likely results in escapes or intentional releases into surrounding areas. These findings highlight the role of transportation and escapes from holding facilities in maintaining and expanding invasive feral swine in Florida. Contact: Tim Smyser

#### Improving Population Density Estimates.

Knowing wildlife population densities (number of animals per unit area) is essential for wildlife management and conservation. However, many methods for estimating density must overcome inherent challenges, such as changes in animal movements and the ability to detect an animal. University of Georgia, USDA Forest Service, and NWRC scientists used field data from feral swine populations in South Carolina and simulations to evaluate the effectiveness of five density estimation methods (i.e., biomarker Lincoln-Peterson estimator [LPE], camera LPE, camera spatially explicit capture-recapture [SECR], trap SECR, and removal) to account for changes in animal movement, underlying population density, and the probability of detection.

Results showed animal movement had the greatest impact on the accuracy of the estimators. Furthermore, the estimators did not perform well when detection probability was low. Researchers recommend modifying sampling designs to maximize detections of animals. For instance, placing detectors (i.e., cameras, traps) closer together to detect species that move less and for shorter distances and farther apart for species that move often and for greater distances. Researchers also found that LPE-based estimators provided lower density estimates than other field techniques. Camera SECR and trap SECR methods gave the highest density estimates. While the removal estimators were somewhat biased when density and scales of movement were low, they exhibited high accuracy when population density and capture rates were high. Researchers caution that animal movement and its impact on estimator accuracy requires accurate post-hoc sampling area measurements or the use of methods that implicitly account for spatial variation. Table 2 summarizes the five density estimation methods and when to use them.

Contact: Amy Davis

• Coyote Genetics in the Eastern United States. Understanding the genetic structure and

composition of coyotes in the Eastern United States is not only relevant for documenting their colonization history, but also gives insight into the biology of the species and its interaction with people. Experts with the U.S. Fish and Wildlife

ESTIMATOR	FIELD DATA USED	USE WHEN
Biomarker LPE	Biomaker Camera Corral Trap	<ul> <li>computationally simple method is necessary</li> <li>an assumption of circular home ranges is acceptable</li> <li>scale of movement and detection rates are fairly high</li> <li>some degree of inaccuracy and/or imprecision is acceptable</li> </ul>
Camera LPE	Camera Corral Trap	<ul> <li>computationally simple method is necessary</li> <li>an assumption of circular home ranges is acceptable</li> <li>scale of movement and detection rates are fairly high</li> <li>some degree of inaccuracy and/or imprecision is acceptable</li> </ul>
Camera SECR	Camera	<ul> <li>recaptures at multiple spatial locations are likely</li> <li>fairly accurate and precise density estimates are required</li> <li>mismatches between grid size and movement patterns of animals are unlikely or can be minimized</li> </ul>
Trap SECR	Camera Corral Trap	<ul> <li>recaptures at multiple spatial locations are likely</li> <li>fairly accurate and precise density estimates are required</li> <li>mismatches between grid size and movement patterns of animals are unlikely or can be minimized</li> </ul>
Removal	Corral Trap	<ul> <li>population densities are fairly high and a reasonable capture rate can be attained</li> <li>a simple method of data collection is preferred</li> <li>the target population is already being managed by culling</li> <li>data on movements of animals in the study area can be gathered or inferred</li> </ul>

 Table 2. Comparison of density estimation methods using data and simulation from feral swine populations in

 South Carolina

Service, NWRC, WS-Virginia Operations, University of Idaho, and West Virginia University investigated the genetic structure and patterns of hybridization in 121 coyotes from the States of West Virginia and Virginia. The results were surprising. Six to 16 percent of the samples showed evidence of hybridization between coyotes and wolves or coyotes and domestic dogs, with several individuals showing high domestic dog ancestry.

Such interactions could have tremendous ecological implications, especially if genes from domestic dogs facilitate adaptations to humandominated environments. Furthermore, the analysis suggested that the sampled coyotes are genetically diverse and do not display any evidence of genetic structure, meaning the populations do not experience genetic isolation. For instance, although the central region of Virginia is heavily developed and features large interstate highways, potentially separating coastal covote populations from those in the interior, no genetic evidence of separation among the populations was observed. At least for coyotes, results suggest that habitat fragmentation and connectivity across the Mid-Atlantic region may not be influencing coyote genetics. Contact: Eric Gese

Contact. Enc dese

**Dietary Overlap Between Kit Foxes and Coyotes.** Range expansions by generalist species can alter animal communities and introduce competition. In the Great Basin Desert, coyotes have become more common, causing concern that they may outcompete the smaller kit fox for food and other resources. Increased coyote abundance has been linked to declining kit fox populations in some areas. Since both species feed on similar prey items, dietary or other resource partitioning may allow the species to coexist. NWRC, university, and National Park Service scientists analyzed coyote and kit fox diets in the Great Basin Desert and compared the results to an earlier study. The analysis showed a high level of dietary overlap during both historical and contemporary sampling periods. This suggests that both species target the same prey and that prey may be abundant enough to support both species populations. Researchers note that other factors, such as changes in habitats from desert vegetation to exotic grasses, may be influencing declines in kit fox populations instead of food resources. *Contact: Eric Gese* 

Impacts of Changing Climate on Bear Hibernation. Hibernation is an important activity that enables animals to conserve energy during seasonal food shortages or severe weather. When, where, and how long animals hibernate is influenced by changing patterns in land use and climate. For example, warmer winter weather has resulted in some animals emerging earlier from winter hibernation. To better understand the impacts of human development, natural food conditions, and weather on the start, duration, and end of black bear hibernation, NWRC and Colorado Parks and Wildlife researchers followed the hibernation activities of 131 radio-collared female black bears. Of the habitat conditions evaluated. researchers found that warmer temperatures tended to be most influential, delaying the onset of hibernation in the fall, expediting emergence from hibernation in the spring, and reducing the overall duration of hibernation. The increased availability of natural and human foods had similar effects, as good natural food conditions and high use of human foods both delayed the start of hibernation and reduced its duration. Given that warmer temperatures and human development both reduced hibernation, researchers note that predicted climate and land use change may increase the length of the active bear season, with the potential for more human-bear conflicts and bear mortalities.

Contact: Stewart Breck



To better understand the impacts of human development, natural food conditions, and weather on the start, duration, and end of black bear hibernation, NWRC and Colorado Parks and Wildlife researchers followed the hibernation activities of 131 radio-collared, female black bears. *Photo by U.S. Fish and Wildlife Service* 

• Calculating Bird Mortality at Wind Farms. Collisions with wind turbines are one source of mortality for some bird species. To estimate the number of birds killed at wind farms, trained observers often search for bird carcasses and adjust calculations to account for any carcasses that may have been removed by scavenging animals. Formulations used to make carcass adjustments often are based on carcass persistence data from surrogate domestic birds. Research by NWRC; Montana Fish, Wildlife and Parks; University of Georgia; and Environmental Solutions and Innovations, Inc., showed that carcass type influences the probability of scavenging and carcass persistence. In a 14-day study, 35 percent of American kestrel carcasses were partially scavenged compared to 64 percent of chicken carcasses. Additionally, 67 percent of northern bobwhite carcasses were completely

scavenged (removed) compared to only 14 percent of red-tailed hawk carcasses. Researchers note that the use of surrogate species to quantify carcass removal at wind turbines may lead to inaccurate mortality estimates. *Contact: Travis DeVault* 

Using Biomarkers To Age and Sex Cormorants. Animals age because cells and tissues are damaged by the accumulation of certain proteins called advanced glycation end products (AGEs). AGEs are irreversible, stable, and accumulate in animal tissues. Pentosidine and hydroxyproline are two AGEs whose concentrations vary with age. To aid in the development of new methods to estimate age distributions of certain wildlife populations, NWRC researchers investigated the relationship between the accumulation of pentosidine and hydroxyproline with the age, sex, and breeding status of double-crested cormorants. Using skin samples from free-ranging cormorants that were banded as fledglings, scientists found a correlation between pentosidine



Collisions with wind turbines are an important source of mortality for some bird species. To estimate the number of birds killed at wind farms, trained observers often search for bird carcasses. NWRC research shows that the use of surrogate carcasses to quantify carcass removal by scavengers may lead to inaccurate mortality estimates. *Photo by Wikimedia Commons, Walter Siegmund* 

and hydroxyproline concentrations and age, and pentosidine concentrations and sex. In about 84 percent of cases, researchers were able to accurately determine whether a cormorant was a breeder or a nonbreeder. Given the prevalence of these biomarkers across animal groups, their use in estimating wildlife population characteristics could offer a powerful tool in animal ecology, conservation, and management. *Contact: Brian Dorr* 

 Habitat Use by American Beaver. NWRC and university researchers used a combination of modeling techniques along with global positioning system (GPS) locations, presence-absence data, and vegetation characteristics to determine if beaver habitat selection is consistent between landscapeand fine-spatial scales. Contrary to the researchers' predictions, results showed that woody wetland edge density, shrub edge density, and open water edge density were important in beaver habitat selection at both spatial scales. Researchers also note that scarce food resources including woody plants may limit beaver fitness and subsequently determine beaver habitat selection during winter and spring. *Contact: Jimmy Taylor*



NWRC researchers are investigating the relationship between the accumulation of pentosidine and hydroxyproline in tissue cells with the age, sex, and breeding status of double-crested cormorants. Photo by USDA, Wildlife Services

Rodenticide Use in Marijuana Fields Threaten Wildlife. California is the largest producer of marijuana in the United States, with a mix of illegal and legal fields grown for medical and recreational purposes. One environmental side effect of marijuana production is the extensive use of anticoagulant rodenticides (AR) to prevent rodent damage to marijuana plants. Because marijuana is classified by the Federal Government as an illegal substance, no rodenticides are currently registered for use on marijuana crops. For these reasons, regulatory compliance of rodenticide use at marijuana growing operations (MGO) is uncertain and assumed to be low. Rodenticide use at illegal MGOs is even more concerning.

NWRC, Colorado State University, U.S. Geological Survey, and the Integral Ecology Research Center compiled information about the use of ARs on MGOs in California. In some instances, up to 25 kilograms of ARs have been found at illegal sites on public lands. Non-target wildlife killed from AR poisoning were found at 22 percent of 41 MGOs investigated in Humboldt, Trinity, and Siskiyou counties. Species affected included bears, foxes, fishers, squirrels, deer, and passerine birds. In addition, liver residues in wild rodents at MGOs also tested positive for ARs. Recently, NWRC researchers and partners conducting owl surveys in California reported brodifacoum AR exposure in a northern spotted owl found dead in an area near seven active MGOs. The researchers note the exposure was likely caused by the owl feeding on AR-contaminated prey. The proliferation of MGOs and their use of ARs in forested California landscapes may serve as an added stressor to this threatened species and other wildlife. Contact: Alan Franklin

 Invasive Rodents in Puerto Rico's Caribbean
 National Forest. Black rats are now documented as the most widespread invasive rodent species in Puerto Rico. In the Caribbean National Forest



Rodenticide use and secondary hazards to nontarget wildlife at illegal marijuana growing operations are a growing concern in the United States.

Photo by Wikimedia Commons, Colorado National Guard

(CNF) in northeastern Puerto Rico, black rats threaten many native species, including the endangered Puerto Rican Parrot. Norway rats are generally restricted to urban areas and possibly agricultural settings in Puerto Rico, whereas house mice are found in and around suburban areas. To determine the presence and distribution of invasive black rats, Norway rats, and house mice across elevations and habitats within the CNF, NWRC and University of Puerto Rico researchers established a series of 104 tracking tunnels (i.e., baited ink cards placed in tunnels to capture footprints of animal visitors) along an elevation gradient from sea level to 1,070-meter El Yunque Peak. Previous CNF studies determined that invasive black rats are common within closed-canopy forests at about 300 to 600 meters in elevation, but no other rodent species were found in these habitats.

Findings from this study not only support the establishment of invasive rats in the CNF, but also indicate they are active at all elevations (1 to 1,070 meters) and in all habitats (disturbed and undisturbed). In addition to confirming the establishment of rats across the CNF, this is the first time that house mice have been documented in the CNF. These mice appear to be restricted to the forest edges near the main road (Highway 191). This knowledge of invasive rodent distributions aids in developing management strategies for rodent control and for protecting native species. *Contact: Aaron Shiels* 

Predicting the Spread of Invasive Lizards. Tegu lizards are widely distributed in their native range in South America, but have become established in Florida due to the release of unwanted pets. Tegus are opportunistic feeders, eating a variety of foods including plant matter, fruit, insects, mollusks, every class of vertebrates, and carrion. This and other characteristics make it a successful invader, likely impacting native plants and animals. NWRC, U.S. Geological Survey, and university scientists developed species distribution models to predict the potential spread of tegu lizards in the United States. Results suggest that much of the Southern United States and Northern Mexico contains suitable habitat for one or more of the three tegu species currently found in Florida. Maps highlighting suitable habitats will help inform policies or management actions aimed at keeping tegus from establishing in the American Southwest. Contact: Page Klug

# **Registration Updates**

• Field Study of New Toxicant for Feral Swine Control. Feral swine are an invasive species in the United States capable of causing significant damage to agriculture, natural resources, and private property, as well as spreading disease to livestock and people. NWRC is developing and evaluating a new toxicant for use in feral swine control efforts. To this end, APHIS received a U.S. Environmental Protection Agency (EPA) Experimental Use Permit for a field study of the toxicant on free-roaming feral swine. The NWRC Registration Unit supported NWRC scientists and their private-sector partner



Tracking tunnels with inked tracking cards and peanut butter bait (ready to be inserted into the tunnel) were used by NWRC researchers to determine the presence and distribution of invasive rodents on Puerto Rico. *Photo by USDA, Wildlife Services* 

in developing study protocols and final reports and consulted with EPA to ensure the studies met that agency's specifications. This field study was the first landscape-scale trial and a pivotal step in developing the tool for control of invasive feral swine. *Contact: Jeanette O'Hare* 

- New DRC-1339 Bird Control Pesticide Label. Before 2018, WS Operations used five DRC-1339 pesticide labels and multiple Special Local Need (SLN: Section 24c) labels for the control of problem birds. The NWRC Registration Unit and WS Operations drafted a new DRC-1339 label named "Compound DRC-1339 Concentrate -Bird Control" (EPA Reg. No. 56228-63), which combines uses from many of the old DRC-1339 Section 3 labels (Feedlots, Staging Areas, Pigeons, and Gulls) and SLN labels. The new label was approved by EPA in December 2017 and provides greater clarity for applicators, lowers costs to WS Operations by eliminating the need for multiple State pesticide registrations and associated fees, and reduces time and costs related to the regulatory maintenance of multiple labels. Contact: Jeanette O'Hare
- New Sources of DRC-1339. The NWRC Registration Unit identified three chemical companies as potential new sources of DRC-1339 used in APHIS'

DRC-1339 pesticide products. The Unit also worked with NWRC chemistry experts to develop the required data package and reports to support the registration of these companies as DRC-1339 manufacturers. The final reports were submitted to EPA in April 2018 for review and approval. One of these suppliers met all of EPA's requirements and has been approved as a new manufacturer for DRC-1339 in APHIS' DRC-1339 products. *Contact: Jeanette O'Hare* 

# **Technology Transfer**

- **Operationalizing Brown Treesnake Aerial Baiting.** The Technology Transfer Program helped procure manufacturing equipment for WS efforts to conduct invasive brown treesnake aerial baiting on Guam. This involved four site visits at Applied Design Corporation to conduct milestone inspections and final signoff of the completed aerial bait cartridge winding station, as well as obtaining U.S. Department of Defense (DoD) funds to purchase two stations. These activities have advanced WS' ability to manufacture hundreds of thousands of bait cartridges for aerial baiting programs on the island. *Contact: John Eisemann*
- Patents, Licenses, and New Inventions. In 2018, the U.S. Patent and Trademark Office allowed one NWRC patent (USPTO Patent #9,999,220) for development of a rodent repellent. NWRC also prepared and presented four new invention disclosures to the USDA Forest Service's Technology Transfer Office for consideration to patent. These include a feral swine feeder, avian perch deterrent, wildlife deterrent, and improvements to pyrotechnic pistols. *Contact: John Eisemann*
- Technology Transfer Agreements. WS forms partnerships through a variety of legal agreements. In 2018, NWRC entered into 3 Memorandums of Understanding, one Cooperative Research and Development Agreement, 14 Material Transfer

and Research Agreements, 7 Material Transfer Agreements, 3 Data Sharing Agreements, and 8 Confidentiality Agreements. *Contact: John Eisemann* 

### Awards

- 2018 Regional Partnership Award. In July 2018, NWRC and its private partner Arkion Life Sciences, LLC, received the Federal Laboratory Consortium (FLC) Mid-Continent Region's Regional Partnership Award for their role in developing, testing, registering, manufacturing, and distributing a suite of anthraguinone-based repellents for reducing bird and mammal damage to crops. The partnership has resulted in five co-owned patented technologies and associated repellent products that are cost effective, practical, environmentally safe, and socially responsible and are currently marketed and sold nationally and internationally. Recent advances have also led to the development of a new repellent application strategy that takes advantage of both visual cues and post-ingestive consequences (e.g., an unpleasant taste or sickness in the birds that eat it). The results of the NWRC-Arkion partnership not only impact wildlife conservation and crop and disease protection in the United States, but also food production in lesser developed countries.
- 2018 NWRC Publication Award. Each year, the NWRC Publication Awards Committee, composed of NWRC scientists, reviews over 100 publications generated by its NWRC colleagues. The resulting peer-recognized award honors outstanding contributions to science and wildlife damage management. In 2018, the award was presented to Drs. Nathan Snow and Kurt VerCauteren and their external partner for their 2017 article "Interpreting and predicting the spread of invasive wild pigs" (*Journal of Applied Ecology* doi: 10.1111/1365-2664.12866).

- 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 toward the NWRC mission. The winners this year are:
  - Travis DeVault, Research Grade Scientist; Understanding, Preventing, and Mitigating the Negative Effects of Wildlife Collisions With Aircraft, Other Vehicles, and Structures Project; Sandusky, OH
  - Are Berentsen, Support Scientist; Methods and Strategies for Controlling Rabies Project; Fort Collins, CO
  - Stacey Brummer, Technician; Developing Control Methods, Evaluating Impacts, and Applying Ecology To Manage Carnivores Project; Logan, UT
  - Jennifer Edwards, Information Services Unit; Fort Collins, CO



An anthraquinone-based bird repellent is being used by rice farmers in the Republic of Ghana. The repellent is the result of a partnership between NWRC and Arkion Life Sciences, LLC. It received the Federal Laboratory Consortium Mid-Continent Region's 2018 Regional Partnership Award. Photo by Wikimedia Commons, African Rice Center

# **2018** Publications

The transfer of scientific information is an important part of the research process. NWRC scientists 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: 2017 publications that were not included in the 2017 NWRC accomplishments report are listed here.)

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# **Appendix 1**

More information about these projects is available on the NWRC web page at: www.aphis.usda.gov/wildlifedamage/nwrc

#### List of 2018 NWRC Research Projects

Methods Development and Population Management of Vultures and Invasive Wildlife *Project Leader: Bryan Kluever* 

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

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

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

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

Understanding, Preventing, and Mitigating the Negative Effects of Wildlife Collisions With Aircraft, Other Vehicles, and Structures *Project Leader: Travis DeVault* 

Improving Rodenticides and Investigating Alternative Rodent Damage Control Methods *Project Leader: Gary Witmer* 

Developing Methods To Evaluate and Mitigate Impacts of Wildlife-Associated Pathogens Affecting Agricultural Health, Food Security, and Food Safety *Project Leader: Alan Franklin*  Economics, Operations Research, and Social Dimensions of Wildlife Management *Project Leader: Stephanie Shwiff* 

Defining Economic Impacts and Developing Control Strategies for Reducing Impacts of Feral Swine and Other Ungulates *Project Leader: Kurt VerCauteren* 

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: Shane Siers* 

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

Chemosensory Tools for Wildlife Damage Management *Project Leader: Bruce Kimball* 

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

Development of Repellent Applications for the Protection of Plant and Animal Agriculture *Project Leader: Scott Werner* 

# Appendix 2

NAME	CONTACT INFORMATION	AREAS OF EXPERTISE	
Abbo, Benjamin	(970) 266-6122 benjamin.g.abbo@usda.gov	Chemistry	
Anderson, Aaron	(970) 266-6264 aaron.m.anderson@usda.gov	Economics	
Baroch, John	(970) 266-6308 john.a.baroch@usda.gov	NWDP: wildlife disease	
Berentsen, Are	(970) 266-6221 are.r.berentsen@usda.gov	Rabies	
Bevins, Sarah	(970) 266-6211 sarah.n.bevins@usda.gov	NWDP: wildlife disease	
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# Appendix 3

# **Acronyms and Abbreviations**

ADC	Applied Design Corporation	LPD	livestock protection dog
AGE	advanced glycation end products	LPE	Lincoln-Peterson estimator
AMR	anti-microbial resistant	LPS	lipopolysaccharide
APHIS	Animal and Plant Health Inspection Service	MGO	marijuana growing operation
AQ	anthraquinone	ml	milliliter
AR	anticoagulant rodenticide	NDV	Newcastle disease virus
ASF	African swine fever	nm	nanometer
BLM	Bureau of Land Management	NWDP	National Wildlife Disease Program
BMP-15	Bone Morphogenetic Protein-15	NWRC	National Wildlife Research Center
BSA	bovine serum albumin	ONRAB	Ontario Rabies Vaccine Bait
BTS	brown treesnake	ORV	oral rabies vaccine
CBC	conservation biological control	PAPP	para-aminopropiophenone
CDC	Centers for Disease Control and Prevention	PEDV	porcine epidemic diarrhea virus
cm	centimeter	PRRS	porcine reproductive and
CNF	Caribbean National Forest		respiratory syndrome virus
CVB	Center for Veterinary Biologics	PT0	U.S. Patent and Trademark Office
DNA	deoxyribonucleic acid	PVC	polyvinyl chloride
DoD	U.S. Department of Defense	RHS	relative hazard score
EPA	U.S. Environmental Protection Agency	RNA	ribonucleic acid
FLC	Federal Laboratory Consortium	SECR	spatially explicit capture-recapture
FRNA	F+ RNA	SLN	Special Local Need
GDF-9	Growth Differentiation Factor 9	SN	sodium nitrite
GPS	global positioning system	STEC	Shiga toxin-producing Escherichia coli
HP	highly pathogenic	TGEV	transmissible gastroenteritis virus
IAV	influenza A viruses	TNF	tumor necrosis factor
IDV	influenza D virus	USDA	U.S. Department of Agriculture
KLH	keyhole limpet hemocyanin	UV	ultraviolet
km	kilometer	VHF	very high frequency
LED	light emitting diode	WS	Wildlife Services
LH	luteinizing hormone		

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