Action Plan National Program 104 – Veterinary, Medical, and Urban Entomology (August 2024 – August 2028)

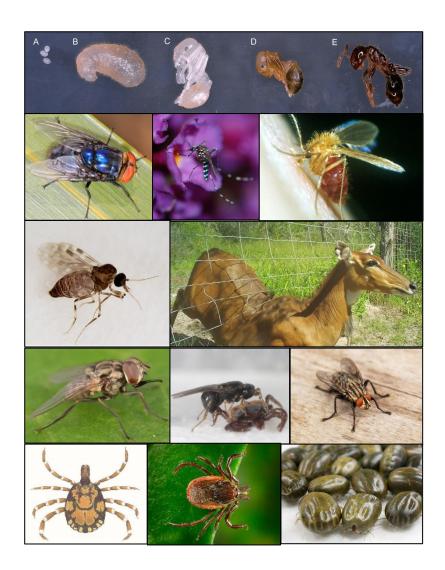


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Vision Statement

Keeping humans and other animals safe from arthropods.

Mission Statement

Manage arthropod vectors and the diseases that they transmit to livestock, humans, and other animals and reduce their economic impact.

Approach

Develop novel and/or improved predictive risk assessment, surveillance, control, and monitoring tools for ticks, mucoid flies, mosquitoes, midges, sand flies, and ants of veterinary, medical, and urban importance.

Relationship to the USDA Strategic Plan

This Action Plan outlines research that supports Objective 2.2 in the USDA Strategic Plan for FY 2022-2026. Strategic Goal 2 is to ensure America's agricultural system is equitable, resilient, and prosperous. Within this Strategic Goal, Objective 2.2 is to protect plant and animal health by minimizing major diseases, pests, and wildlife conflicts. Controlling the pests and diseases of humans, livestock, and structures is the major thrust of the NP 104 Action Plan. NP 104 also supports Strategic Goal 3: Fostering an Equitable qnd Competitive Marketplace for All Agricultural Producers.

Relationship to the USDA Science and Research Strategy, 2023-2026

This NP 104 Action Plan outlines research that supports the USDA Science and Research Strategy, 2023-2026 Priority 2 Driving Climate-Smart Solutions, Objective 2.1 Climate Change Impacts and Priority 4 Cultivating Resilient Ecosystems, Objective 4.1 Genomics and Genome Editing, Objective 4.2 Microbiome Research and Objective 4.4 Infectious Diseases and Pests.

Relationship to the USDA Agricultural Research Service (ARS) Strategic Plan 2023-2026

The NP 104 Action Plan addresses the high level goals and objectives of the <u>2023-2026 ARS Strategic Plan</u> as described in the section discussing the ARS Program Area for Animal Production and Protection and in Strategic Goal Area 3 Make Safe, Nutritious Food Available to All Americans.

Introduction

Damage and disease associated with biting and stinging arthropods affect humans and livestock in the United States and around the world. Economic losses from arthropod damage, including crop losses, exceed \$100 billion annually. Global and local movement of humans, international trade, and altered ecosystems facilitate the introduction of new disease vectors and pathogens into the United States, promote new parasitic arthropod-wildlife-livestock interactions, enable atypical arthropod vector-host-pathogen interactions, and expose humans to exotic vectors and pathogens. The research seeks to reduce arthropod damage to animals, humans, and structures. This work will target (1) arthropods transmitting pathogens that cause diseases in humans and animals, (2) pests that directly harm human health, and (3) ants that damage physical infrastructure, outdoor urban areas, and agriculture production. Due to the interconnection between the health of animals, humans, and the environment, the program employs a "<u>One Health</u>"¹ approach. Through this effort, USDA supports sustained health outcomes for animals,

¹ The "One Health" approach is the collaborative effort of the human health, veterinary health and environmental health communities. Through this collaboration, USDA achieves optimal health outcomes for both animals and people.

people, and the environment.

Big Data and Data Automation

NP 104 conducts research that requires big data and data automation capabilities.

This research includes:

- Models that use data on environmental or climatic events to forecast disease outbreaks;
- Databases and apps used to manage cattle fever tick outbreaks;
- Reverse vaccinology²;
- Genomic studies such as genome sequencing of arthropods of medical and veterinary importance to help identify and elucidate gene activities in target organisms. ARS scientists used this process to discover aquaporin and found it to be a good vaccine candidate for the control of the cattle fever tick after computer modeling predicted it to be highly antigenic. This work recently received a patent.
- Genomic techniques that provide information on population genetics and support the following:
 - o identifying invasive insect source populations and migration pathways;
 - $\circ \quad$ developing gene targets for editing, disruption, or RNAi gene suppression;
 - o identifying pest species that are difficult to identify morphologically; and
 - using next-generation sequencing and metagenomics to discover tiny natural enemies parasitizing their host.
- Research on fly, tick, and ant metagenomes (the entire genomic complement found in the environment) and microbiomes (the universe of microbes living in association with each pest) generates information about pathogens and commensal and symbiotic microorganisms that can be used to solve agricultural problems associated with flies, ticks, and ants. For example, the red imported fire ant (*Solenopsis invicta*) virus 3 (*Invictavirus solenopsae*) was discovered using metagenomics. This virus causes shifts in ant feeding behaviors, which result in the collapse of infected fire ant colonies by starvation. It is successfully being used as an augmentative biocontrol agent against red imported fire ants in Florida and California. Scientists have worked with U.S. university partners and international institutions to generate 'omic resources such as transcriptomes, microbiomes, and genomes for several livestock and human pests, including biting midges, mosquitoes, and house flies. Sequenced mosquito mitochondrial genomes support the development of databases for rapid identification of field samples. Sequencing of pooled field mosquito samples is performed for the real-time identification of hosts and pathogens. RNA sequencing is valuable for gene expression analysis of pathogens that cause disease in humans, livestock, and other animals.
- The <u>Partnerships for Data Innovations (PDI)</u> and <u>SCInet</u> are resources ARS scientists can use to collect and handle large data sets more efficiently.

Artificial Intelligence

NP 104 conducts research that involves artificial intelligence (AI). Robots and drones used for cattle fever tick surveillance and monitoring are an offshoot of new management and production strategies rooted in precision agriculture. Advanced neural networks (ANN) have led to the discovery of new long-lasting mosquito repellents and more potent and selective insecticides. The ANNs were trained using data from

² Reverse vaccinology is an approach that employs bioinformatics to screen the entire pathogenic genome. This approach led to development of a vaccine for Serogroup B meningococcus in 2000, avian influenza A (H7N9) virus in 2019, and AI H5N1 in 2022.

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tens of thousands of compounds and insect responses. Al facilitates translating digital animal monitor outputs into behaviors associated with stress and defense and is also used to interpret digital imagery for quantifying ectoparasite infestation levels. Researchers use systems networks and mathematical modeling to model the spread of pathogens in U.S. vector-borne disease introduction scenarios. They also use particle filters (an algorithm for solving non-linear estimation problems) and learning algorithms to combine disease data and environmental parameters to retrospectively determine basic reproductive rates and transmission parameters for mosquito vectors that infect humans with pathogens that cause disease. This information can be used to forecast mosquito density and disease risk in the future.

Globalization

Globalization, the increasing connectedness and interdependence of world cultures and economies, is leading to a worldwide increase in invasive species through the movement of people and products. Security challenges resulting from a globalized economy include preventing the introduction of exotic pest species and pathogens that damage humans, plants, animals, and infrastructure. In recent years, new mosquitoborne pathogens, i.e., Chikungunya and Zika viruses, were introduced into the United States through the movement of people traveling from locations where these diseases are endemic. In the mid-1980s the Asian tiger mosquito (Aedes albopictus) was introduced into the southeast United States in shipments of tires from northern Asia and into California in 2001 from shipments of ornamental bamboo. The New World screwworm, a pest completely eradicated from the United States by 1985, was introduced in the Florida Keys in 2016 through human activity in that area. By April 2017, ARS and USDA-APHIS had successfully eradicated this pest from the United States once again. In 2017, the invasive East Asian tick (Haemaphysalis longicornis) was discovered in New Jersey and has since been found in 18 states; studying archived samples, scientists determined the tick entered the United States before August 31, 2010, on an imported animal from northeast Asia. The invasive red imported fire ant was introduced into the United States in the 1930s in ships' ballast, but the rate at which its U.S. range was expanding declined over the past few decades. However, within the last 15 years it has been detected in Australia, Taiwan, and mainland China and several port introductions were recently discovered in Japan and South Korea. These are just a few examples of how globalization facilitates the spread of arthropods and the diseases they transmit. These introductions and re-introductions reinforce the need for new and improved strategies and tools to detect, survey, control, and monitor arthropod pests to protect against diseases and other damage.

Climate Change, Altered Ecosystems and the Impact on Vector-Borne Disease

Ecosystem components include physical habitats, potential hosts, and pests, and the characteristics of physical environments significantly affect the potential establishment and success of an invasive arthropod species or pathogen. The pathogenic landscape concept describes and explains ecosystem attributes affecting spatial variations in arthropod-borne disease risk or incidence; for instance, invasive weeds can facilitate the survival or invasion of exotic arthropod disease vectors. In some cases, small changes in climate dramatically affect the distribution and range of a pest species, which in turn affects a vector's ability to sustain disease cycles. Many distributions and ranges for established pests and vectors are based on climatic conditions, while urbanization and other human activities alter pest ecosystems, which affect the ability of pest populations to colonize and persist in specific locations and ecosystems.

Research in NP 104 is conducted with cooperators to develop models that predict disease risks based on vector populations and environmental conditions. Maintaining disease cycles requires certain conditions that may help pest vectors and hosts reach threshold populations supporting sufficient vector access to hosts. For instance, though Zika and Chikungunya viruses were recently introduced into the United States, several factors reduced the local transmission of the pathogens that cause these diseases. These factors included insufficient populations of *Aedes aegypti* mosquitoes, which spread diseases, and insufficient

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contact between these vectors and their human hosts. The latter results from a combination of circumstances, including screened windows, availability of air conditioning inside dwellings, and use of mosquito repellents to prevent mosquito bites while outdoors. Neither virus has been locally acquired in the United States since 2018. Zika still has some locally acquired transmission in U.S. Territories, but the numbers are low and decreasing each year.

The physical environment affects arthropod vectors of disease in many ways. Cattle fever ticks on both sides of the Rio Grande transmit babesiosis (cattle fever) to livestock, but efforts to eradicate the tick are complicated by interactions with its feral hosts, white-tailed deer and exotic nilgai. The challenge is the absence of standard treatment methods to control ticks on nilgai, which are highly mobile and contribute to the dispersal of cattle fever ticks beyond the Rio Grande in south Texas, especially along the Gulf Coast. Control efforts are further impeded by stands of invasive Spanish cane known as giant reed (*Arundo donax*) that provide shelter for cattle fever ticks. Biological control agents have been released and physical topping of the cane have helped to open the canopy and allow native vegetation to grow and reduce tick habitat.

Global sea rise due to glacier melt will not be even. The Texas gulf region is expected to have a disproportionate sea level rise. This in combination with stronger and more frequent hurricanes can damage infrastructure, limit access, and make eradication efforts more difficult. Another example of the environment affecting arthropod dispersal is through changes in average temperature. In recent years, a small increase in the average temperature in Costa Rica mountain ranges and changes in economic practices have resulted in an explosion of stable flies at pineapple and coffee plantations and on livestock at nearby ranches and dairies. U.S. producers have also reported an increase in stable fly populations in recent years.

Climate change has a powerful influence on the distribution of invasive pest ant species. For example, the Asian needle ant was first reported in the United States in 1930, but only in the past 8 years have their populations increased, causing them to be classified as an invasive pest ant. Suitable habitat for this ant is expected to increase by 75 percent around the world in the next 50 years due to climate change.

Cross-Cutting Research.

The Veterinary, Medical, and Urban Entomology National Program conducts cross-cutting research with other ARS national programs. Greater livestock protection is achieved by research that results in disease reduction (which is a focus of NP 103, the Animal Health National Program) in concert with NP 104 research on arthropod control for reducing direct damage and stress to animals. A more specific example of the association between NP 104 and NP 103 is in their aligned focus on livestock disease vectors, (biting midges, mosquitoes, ticks) and the diseases that they transmit, *e.g.*, blue tongue, Rift Valley fever, and babesiosis. Scientists in NP 104 cooperate with scientists in the European Biological Control Laboratory (EBCL) on Crop Protection and Quarantine National Program (NP 304) research to develop solutions for protecting deployed U.S. military personnel using spatial repellents. Other research projects in both NP 104 and 304 are searching for new biopesticides for the control of insects. These biopesticides could potentially be used to control pests of interest in both programs. All these efforts lead to solutions that increase and enhance animal production, which is a focus of the Food Animal Production National Program (NP 101).

Stakeholder Input

A series of stakeholder workshop webinars/conference calls in February 2023 gave stakeholders from a wide range of sectors (private conservation organizations, industry, academia, and government) an opportunity to provide input on priority research for NP 104. Their input was carefully considered, and a significant effort has been made to address as many of their concerns as possible in this plan.

NP 104 is one of 15 national programs within ARS, and this NP 104 Action Plan, which extends from 2024-

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2028, will set important research priorities as ARS enters its eighth decade. NP 104 research focuses on developing solutions for current agricultural problems and threats caused by arthropods of medical, veterinary, and urban importance in the United States, and on potential threats developing in other countries. Each problem statement will focus on addressing at least one of the four elements of the Integrated Pest Management (IPM) strategy discussed in more detail below.

Integrated Pest Management

IPM is an ecologically based approach to the management of pest populations. The approach was developed in response to overuse and overreliance on chemical pesticides and their declining effectiveness in reducing pest populations and associated diseases and damages. Effective IPM strategies can be classified into four elements: (1) risk assessment and biology; (2) surveillance; (3) control; and (4) monitoring and sustainability. These elements are listed in the order in which they would be applied operationally, *i.e.*, identifying the problem and threat, assessing the scope of the problem, mitigating the problem, and developing an effective long-term adaptable solution. Each element is discussed in more detail below.

Risk Assessment and Biology

Risk assessment and basic biological studies are critical first steps in developing an IPM program. Defining the problem and selecting appropriate control strategies requires obtaining background information on pest identification (systematics and taxonomy), distribution (spatial and temporal), behavior (particularly behaviors that cause or have the potential to cause damage), genetics, and ecology (bionomics). Fundamental research on pest biology generates information that can be used to identify vulnerabilities of the pest; findings can also be used to help develop models that assess entomological and/or epidemiological risks to host populations. Studies focused upon the relationship between vector and pathogens fall under this element.

Surveillance

Surveillance provides assessments of the composition and abundance of pest populations. This information is needed to determine pest risk levels, identify when control measures are required, select appropriate control measures, and estimate the efficacy of alternative control strategies. Surveillance can also be conducted to determine the extent of pest damage; track infection levels associated with disease vectors; and assess environmental conditions, such as soil and moisture content, which affect larval habitats. Accurate surveillance data is critical to overall IPM programs, and research on developing efficient traps for collecting target pest species is an important component of this work.

Control

Pest control is a key outcome of IPM; it is a very broad category of activities that essentially consist of management tools, including conventional chemical controls, which prevent pests from accessing structures or hosts. These efforts often prevent or mitigate the harm pests can inflict on living organisms (e.g., disease transmission or injury from parasitic activity) and reduce pest damage to structures and associated developments. Effective pest control with a single intervention is often risky and unattainable, so multiple measures are often combined to achieve optimal success and minimize negative environmental effects. In some cases, control is obtained by reaching targeted thresholds for suppressing pests and in other cases control is achieved by killing the pest. Common control classifications include the following:

- Cultural control, *e.g.*, practices to reduce pest establishment, reproduction, dispersal, and survival.
- Physical control (source reduction and mechanical control), *e.g.*, reduction of breeding areas, and/or installation of barriers such as screen windows.
- Biological control (*e.g.,* introducing specialist parasitic insects that feed on or otherwise inflict

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damage to invasive arthropods, plants, or pathogenic environments).

• Chemical control using synthetic insecticides or bio-rational compounds (*e.g.*, botanical compounds, double-stranded RNA, etc.).

Genetic studies of pests are rapidly generating a wealth of information that can be used to develop new and adaptive pest control measures using transgenic, symbiont-mediated (negatively affecting a closely associated organism important to the pest organism's survival), or gene-drive strategies. Driving female lethal genes into mosquito populations is one example. In practice, each genetically based control method is species and population specific and it is challenging to project if or how laboratory results will be replicated in integrated field studies. Developing new genetic and chemical pest control measures that result in commercial IPM products often requires coordinating stakeholder efforts, funding, and other resources.

Monitoring and Sustainability

Continuous monitoring of IPM performance is essential for assessing the efficacy of control interventions. Sustaining interventions can be challenging because they are often expensive and are usually discontinued once success is achieved. However, pest populations that survive an initial IPM effort may develop resistance to chemical control methods, and pests can also be reintroduced after control efforts are no longer active. Therefore, sustained pest monitoring is necessary for early detection and rapid mitigation. Monitoring tools for pest, disease, and/or damage surveillance need to be and developed and deployed using a comprehensive strategy.

All research conducted within NP 104 supports one or more of the four elements of IPM as discussed, and the NP 104 research program covers the spectrum from fundamental to applied studies. Fundamental research provides information that identifies and explains previously unknown or misunderstood elements of pest biology, pest behavior, pest responses to control methodologies, and the impacts of control strategies on surrounding ecosystems. Translational research is also needed to develop or enhance management strategies or technologies that target known pest vulnerabilities. Finally, applied research is conducted to demonstrate the efficacy of strategies or technologies for pest control, or to demonstrate the level to which an integrated approach is achieving the overall goal of reducing pest populations and/or disease risk.

Research Components

The NP 104 Action Plan contains general strategies and specific actions within the following organizational hierarchy:

- 1) <u>Components</u> are general categories of research areas developed with input from stakeholders.
- 2) <u>Problem Statements</u> indicate the specific nature and scope of problems to be solved by ARS.
- 3) <u>Research Focus</u> identifies the needs and the research that ARS will perform to solve the problem.

The components of the program have been modified to clarify the organization of the program relative to its name, National Program 104: Veterinary, Medical, and Urban Entomology. Additionally, the sole focus of the urban entomology program is invasive ants; therefore, the title of this component is named to better describe that focus.

The components of the program are:

Component 1: Veterinary Entomology Component 2: Medical Entomology Component 3: Fire Ants and other Invasive Ants

Component 1: Veterinary Entomology

Veterinary entomology research within NP 104 is comprised of research with a primary focus on arthropods that harm livestock and negatively impact animal agricultural production. The goal of this research is to protect animals from damage and diseases with an ultimate impact of improved animal health and wellbeing, increased productivity, and greater economic yields. There is a broad range of livestock and other animals that need protection from a vast range of arthropods and arthropod-borne diseases. Control of arthropods and reduction of disease risk to animals is a more complex problem than control of arthropods involved in purely anthroponotic cycles. Zoonotic cycles are more difficult to interrupt because animals tend to remain in the outdoor environment, providing arthropods with perpetual access to host animals. Animals are more susceptible to arthropod-borne infections and suffer direct damage from bites and infestations. In addition, there are arthropods that are not vectors of livestock pathogens or parasites, but still negatively impact productivity and economic yield due to stress from arthropod bites. When the disease risk is not present, it is common to accept a higher bite threshold for animals than the threshold set or tolerated by humans. Humans can convey their objection to nuisance biting in a direct manner compared to animals, where humans must interpret the nuisance from the animal's behavior. With respect to control methods and the use of conventional chemical control, animals are subjected to greater exposures of pesticides in the environment, and in some cases, there are pesticides used directly on animals to prevent arthropod attack. Animals are eaten as food; therefore, chemical residues within animal food products are of concern because they can accumulate in humans through consumption of treated animals. There have been successes in the field of veterinary entomology, and some of these are the control of cattle lice, sheep mange mites, cattle bot flies, and the successful elimination of cattle fever ticks and screwworm flies from the United States. The latter two are still foci of ARS programs where continual effort is necessary to keep the United States free from these pests. There are many livestock pests that are not well controlled. Customer/stakeholder input indicates that cattle fever ticks, screwworm flies, stable and horn flies, house flies, biting midges, and mosquitoes are the arthropods of veterinary importance that are of greatest concern, so this is where ARS efforts are concentrated. Additional research on invasive ticks is added to address the expansion of Lyme disease and discovery that the East Asian tick is present in the United States.

Problem Statement 1A: Integrated Pest Management of Ticks of Veterinary Importance

Ticks bite livestock and transmit pathogens that cause disease. *Babesia* pathogens transmitted by cattle fever ticks are of great concern because they cause bovine babesiosis, an expensive and potentially fatal disease in cattle. Strategies to better protect cattle and other livestock from the bites of ticks will lead to improved livestock health and production. Keeping cattle fever ticks outside of the United States is a national priority for the livestock industry.

Cattle fever ticks (*Rhipicephalus* (*Boophilus*) annulatus and *Rhipicephalus* (*Boophilus*) microplus) can be infected with *Babesia* parasites and transmit them to cattle when they feed. Bovine babesiosis leads to production losses and death in cattle. The Cattle Fever Tick Eradication Program (CFTEP) was established in 1906 as the first U.S. livestock pest eradication program. Successful eradication of cattle fever ticks was accomplished in Texas by 1943, and in the 1960s for the rest of the United States. Since then, APHIS and the Texas Animal Health Commission have maintained a quarantine zone in southeastern Texas where Mexican cattle and wildlife frequently cross the international border with attached ticks. The mitigation strategies used within this zone continue to keep cattle fever ticks out of the United States, but sustainability is an issue due to the expense of dipping infested cattle in acaricides, quarantining pastures, and by tick populations moving on much-expanded populations of native white-tailed deer and exotic ungulates (e.g., nilgai). Cattle fever ticks are also present in U.S. territories such as Puerto Rico and the U.S. Virgin Islands. NP 104 has made progress toward overcoming these challenges and needs to continue those efforts.

Additional threats to livestock by high-consequence foreign pests are always present and one example of

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this is the recent discovery of the East Asian tick, *Haemaphysalis longicornis* which has now spread to 19 states: New Jersey, Virginia, West Virginia, Arkansas, North Carolina, New York, Pennsylvania, Maryland, Connecticut, Rhode Island, Indiana, Kentucky, Tennessee, Ohio, Georgia, Missouri, Delaware, Massachusetts, and South Carolina. There are other potentially invasive tick vectors (*e.g., Amblyomma variegatum, Hyalomma* spp., *Rhipicephalus appendiculatus*) and species that might expand their ranges within the United States (e.g., *A. americanum, A. maculatum, A. mixtum*). Likewise, there are high-consequence foreign tick-borne pathogens, like African swine fever, that could devastate U.S. animal agriculture if their emergence involved transmission by native tick species (Note: ASF can also spread by direct animal-to-animal transmission).

Research Focus

Risk Assessment and Biology

- Assessment of ecological drivers for range expansion, fluctuation of cattle fever outbreaks in the United States, and potential introduction of invasive ticks to the United States.
- Identification of pathways for invasion by foreign ticks transported into the United States.
- Determination of pathogenic landscapes (interactions between land, people, disease vectors, and their animal hosts) contributing to suitable tick habitats.

Surveillance

• Evaluation of strategies to improve detection of cattle fever tick infestations and other tick infestations to eliminate or reduce the need for animal inspections.

Control

- Development of methods to mitigate impact of invasive weeds that facilitate cattle fever tick survival and reinvasion of the United States.
- Development of novel technologies for eradication and control of cattle fever ticks and other ticks, including formulations used against ticks that have developed resistance to current commercial acaricides and biocontrol agents.

Monitoring and Sustainability

• Development of improved methods to prevent introduction of exotic ticks.

Anticipated Products

- Decision-making tools to lessen the burden of ticks and the risk for tick-borne disease transmission.
- Sustainable technologies for control of cattle fever ticks and other invasive ticks.
- Countermeasures to mitigate the threat of invasive tick species and the diseases they can transmit.

Potential Benefits

- Improved surveillance and detection of invasive ticks.
- New control technologies for cattle fever ticks and other ticks infesting livestock and wildlife.
- Decreased economic losses due to bovine babesiosis.
- More effective prevention of cattle fever tick entry into the United States.

Problem Statement 1B: Integrated Pest Management of Stable Flies that Feed on Livestock

Stable flies feeding on livestock cause significant animal stress and loss of vigor leading to production losses, increased susceptibility to disease, and in extreme cases, death. Strategies to better protect

livestock from stable flies and strategies to reduce stable fly populations will result in improved livestock health, production, and welfare.

Although stable flies are not normally considered a vector of disease for cattle and other livestock in the United States, their painful bites have direct impacts on cattle, such as reduced weight gain and in some cases death due to stress from the bites. Stable flies, native to southern Asia and Africa, were introduced to the United States with the first European colonists. The genus Stomoxys includes more than twenty species in Africa and southern Asia. At least four species are pests of cattle. Stable flies have been implicated as mechanical vectors of several livestock pathogens including trypanosomes, equine infectious anemia and lumpy skin disease viruses, and anthrax outside of the United States. In recent years, stable flies developing in crop residues have produced serious "outbreaks" outside of the United States. The risks of introduction of exotic species of Stomoxys and Stomoxys-borne diseases or outbreaks resulting from changing agronomic practices to the United States are unknown. Controlling this fly has been challenging because the larvae develop in a broad range of habitats consisting of decaying vegetation, often contaminated with animal urine and feces. Adult stable flies can fly long distances (up to 70 miles), infesting livestock many miles from their developmental sites. Because stable flies feed on their hosts for only a short period of time, and usually on the lower legs, control techniques such as ear tags, topical applications of insecticides, residual insecticides, and traps have had limited success. Larval control options are limited to sanitation (removal of developmental substrates) and the treatment of developmental substrates with insect growth regulators such as Cyromazine (a strategy discovered by ARS).

Research Focus

Risk Assessment and Biology

- Estimation of the risk of introduction of exotic species of stable flies and their potential ranges in the United States.
- Determination of environmental factors that lead to stable fly population increases and outbreaks.

Surveillance

• Development of new methods for adult surveillance control based on attractants and traps.

Control

- Development of new methods of bite prevention based on repellents.
- Development of new strategies for larval control based on stable fly biology, development, environmental substrates used for breeding, and microbial interventions.

Monitoring and Sustainability

• Evaluation of genetic approaches for use in sterile insect technique and other control approaches.

Anticipated Products

- Improved traps for surveillance and control of adult stable flies.
- New attractants and attractive surfaces or fabrics that increase efficacy of attract-and-kill devices.
- Long term, low maintenance attract-and-kill devices.
- Novel management tools for control of larval and adult stable flies.
- Novel control technologies discovered by mining the stable fly genome.

Potential Benefits

- Decreased stable fly populations.
- Increased animal health and well-being.
- Increased animal production.

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Problem Statement 1C: Integrated Pest Management of House Flies that Harm Livestock

House flies are ubiquitous pests of veterinary importance that can harbor and transmit pathogens to livestock. Because many of these pathogens are zoonotic to humans, flies are equally important to both preharvest and postharvest food (food safety). Studies to better characterize the role of houseflies in disease transmission and strategies to better control houseflies and reduce disease incidence will result in improved livestock health and production.

Flies can cause significant animal stress due to their activities, such as congregating on animals to feed on secretions from eyes, nares, teats, and open wounds. Flies also associate with refuse and manure and aggregate on feed boxes, providing a direct bridge between these environments and transmitting microbes in the process. These pests are known to be mechanical and/or amplifying vectors of pathogens and these flies are found often near livestock where massive populations can emerge due to abundant access to food and larval habitat. Bacteria that cause mastitis, pink eye, respiratory diseases and other illnesses are routinely acquired and carried by house flies, implicating them as both reservoirs and disseminators in livestock operations. Since flies sometimes travel great distances from larval sources, agricultural operations that produce them are blamed for flies affecting humans living nearby. Sanitation is usually a part of fly control, but farming operations often cannot remove the many larval sources associated with animal waste, spilled feed, etc. Traps, residual insecticides, attractive baits, and biological control are strategies employed to counter the nuisance and vectoring impacts of house flies. However, widespread resistance to pyrethroids and other pesticide classes has been reported on a global scale. Larval control remains largely experimental because the maggots reside below the surface of media. Basic research on flymicrobe interactions across life history and surveillance of microbes harbored and transmitted by flies is needed to improve risk assessment and inform novel control strategies that collectively result in reduced disease transmission.

Research Focus

Risk Assessment and Biology

- Evaluation of the economic impact of flies on agriculture.
- Assessment of the risk of bacterial transmission by studying the interactions of flies and bacteria, and the interaction of flies and microbes throughout life history.

Control

- Determination of factors that favor or limit the effectiveness of parasitoids for fly management, including temperature, substrates, and effects of other fly management tools.
- Development of novel methods to control adult flies with emphasis on limited use of pesticides and resistance management.

Monitoring and Sustainability

- Evaluation of novel microbial methods for managing adult flies and their compatibility with other management components.
- Development of new methods for larval surveillance and control based on volatiles.

Anticipated Products

- Improved systems for house fly management based on trapping, source reduction, and beneficial microbes.
- Better larval detection and control strategies based on larval-produced volatiles.
- Improved management strategies for confined livestock operations that are based on research findings on the risks house flies pose in harboring and transmitting pathogenic bacteria.

• New targets for controlling larvae based upon improved understanding of use of microbes in the developmental substrate (e.g. manure).

Potential Benefits

- Decreased nuisance effects from houseflies.
- Decreased incidence of disease.

Problem Statement 1D: Risk Assessment, Biology, and Control of Horn Flies

Horn flies take multiple bloodmeals from cattle and other animals per day. The bites cause discomfort to the animals, reduce productivity, and can result in secondary infections that damage leather quality. Strategies to better protect cattle and other animals from horn flies will result in improved animal health and decreased production losses.

Horn flies were first reported in the United States in 1887. A 12 percent decrease in average daily growth rate of nursing calves has been reported for cows that were not treated. Despite direct treatment of livestock and use of feed supplements that regulate growth, horn flies remain a major problem for producers in the United States and southern South America. Horn fly adults remain on cattle and oviposit in freshly dropped feces, then return to cattle for feeding.

Research Focus

Risk Assessment and Biology

- Determination of the economic impact of horn flies in the United States.
- Determination of the genetic basis for variation in infestation levels among individual cattle.

Control

- Mining of the horn fly genome for development of biotechnology-enhanced sterile-insect technique approaches.
- Examination of horn fly biology to develop new control strategies, including interruption of pathogen transmission.
- Discovery of antigens for novel anti-fly vaccine formulations.

Anticipated Products

- Novel attractants and repellents for use in horn fly surveillance and control.
- Control technologies with new modes of action.
- Integrated approaches to manage insecticide-resistant horn fly populations.

Potential Benefits

- Decreased damage to cattle from horn fly bites and associated diseases.
- Improved cattle health.

Problem Statement 1E: Integrated Pest Management of the New World Screwworm.

Screwworm flies infest animals by ovipositing their eggs in an animal wound. The larvae that develop from the eggs consume the host animal tissue as they develop, resulting in hide damage and death to the animals. Sterile insect technique (SIT) is relied upon to control screwworms and keep animals in the United States free from their attack. Improvements to the SIT program are needed to enhance sustainability of this program.

ARS developed the screwworm SIT technique in the 1930s and implemented it to control the New World screwworm fly (*Cochliomyia hominivorax*). By the 1960s, this pest was eradicated from the southern United

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States. Eradication in Mexico was achieved in 1993. By the early 2000s screwworms were eradicated throughout all Central America. Currently, the U.S. government (USDA APHIS) funds 90 percent and the Panamanian government funds 10 percent of a binational commission responsible for the production and distribution of millions of radiation-sterilized screwworm flies in eastern Panama. This activity forms a barrier at the neck of the land bridge between Central and South America that prevents reentry of the damaging species into Central and North America. One of the most successful entomological programs of all time, this operation continues to require technical support from ARS to address new problems and to reduce costs. In 2016, there was an infestation of screwworms found in Key Deer in the Lower Florida Keys. ARS cooperated with APHIS to successfully eradicate screwworms during this outbreak.

Research Focus

Risk Assessment and Biology

• Determination of the population genetic structure of screwworm populations in the Americas and in the Caribbean.

Control

- Mining of the screwworm genome for biotechnology-enhanced sterile insect technique and other approaches for optimal eradication.
- Monitoring and sustainability
- Improvement of rearing procedures and diet to increase screwworm production efficiency.

Anticipated Products

- Genetic database of screwworm populations for regions where it remains endemic in the Americas and in the Caribbean.
- Next generation genetic systems for screwworm eradication.
- Decreased cost of rearing male screwworms for the SIT program.

Potential Benefits

- Determination of infestation source.
- Improved production of male screwworms.
- Reduced program cost.
- Continued reduction of screwworms from the Caribbean and other regions of the Americas.

Problem Statement 1F: Integrated Pest Management of Mosquitoes of Veterinary Importance

Mosquitoes bite animals and transmit pathogens that can result in disease and death in these animals. Climate change may influence the impact nuisance mosquitoes have on farm production in unforeseen ways . Also of great concern are invasive mosquito-borne pathogens that have detrimental impacts once they become established in the United States. A greater understanding of the threats that the United States faces is required to assess risk and develop strategies to mitigate these threats.

West Nile virus entered the United States in 1999 and spread rapidly from the U.S. east coast to the west in about 3 years. The United States faces additional disease threats that have the potential to be a greater societal detriment than West Nile virus. Livestock and poultry are heavily exposed to mosquitoes but, with few exceptions, there is little understanding of their current impact. Research emphasis is placed typically on disease rather than nuisance vectors. However, with the introduction of an invasive pathogen, a nuisance mosquito can instantly become a disease vector. The potential for the introduction of damaging invasive species is particularly broad due to globalization. Rift Valley fever and Japanese encephalitis are viruses that can be transmitted by many species, including *Aedes taeniorhychus, Aedes vexans* and *Culex*

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tarsalis. Further studies are needed to define the risk to the United States based upon increased understanding of basic biology and to develop mitigation strategies for the potential vectors and for nuisance biting mosquitoes.

Research Focus

Risk Assessment and Biology

- Use of population-environment modeling to identify ecological determinants of the spatial and temporal ranges of subpopulations of pestiferous or disease-vector mosquito species.
- Determination of biological characteristics of pestiferous or disease vector mosquito species that are likely to harm animal production through bites or pathogen transmission.
- Investigate whether invasive species, such as *Culex coronator*, pose more of a health risk than the native species.

Surveillance

• Evaluation of surveillance methods for use against pestiferous and disease vector mosquitoes.

Control

• Evaluation of management strategies for use against pestiferous and disease vector mosquitoes.

Anticipated Products

- Quantitative assessment of mosquito damage to U.S. agriculture.
- Predictive models that provide early warning of unusually large populations of mosquitoes and the environmental conditions that lead to increased vectorial capacity.
- Population distribution projections (up to three decades) into the future based upon climate change models for mosquito populations.
- Novel mosquito surveillance and management tools.
- Improved control strategies specific to habitat, ecological region, and established mosquito species.

Potential Benefits

- Identification of disease threat potential for the United States.
- Improved control strategies for potential vectors of invasive pathogens.
- Improved protection from mosquito bites to improve animal production.

Problem Statement 1G: Risk Assessment, Biology, and Control of Biting Midges of Veterinary Importance.

Hematophagous *Culicoides* spp. (biting midges) are a threat to livestock and wildlife, and vector orbiviruses, orthobunyaviruses, and rhabdoviruses. Additional research on vector innate immunity, gut microbiomearbovirus interactions, and arbovirus transmission zone surveillance is needed to reduce disease risk to livestock and wildlife.

Although arboviruses transmitted by *Culicoides* can cause devastating disease in mammals, their infections in midges are almost always nonpathogenic and persist for the life of the midge. Relatively little is known about how the *Culicoides*' environmental microbiome affects arbovirus survival in the midge, and how specific environmental conditions can influence transmission-competent midge populations.

Research Focus

Risk Assessment and Biology

• Correlation of midge surveillance to environmental conditions in arbovirus transmission zones.

- Characterization of biting midge developmental sites.
- Determination of the effects of larval substrate on gut microbiome of midges.

Control

• Development of larval habitat treatments for biting midge population management.

Anticipated Products

- Knowledge of biotic and abiotic characteristics of developmental sites can help in predicting location of midges and will inform larval control strategies.
- Novel control based on behavior that can be targeted to either increase lethality of arboviruses to *Culicoides* midges or decrease arbovirus transmissibility by these vectors.
- New control products for larval habitat treatments to reduce biting midge populations.

Potential Benefits

- Reduced ability of *Culicoides* midges to maintain persistent, productive arbovirus infections, and target specific habitats in arbovirus transmission zones.
- Reduced attack rate of and virus transmission to livestock and wildlife.

Component 1 Resources

- Agroecosystem Management Research Unit, Lincoln, NE
- Arthropod-borne Animal Diseases Research Unit, Manhattan, KS
- European Biological Control Laboratory, Thessaloniki, Greece
- Invasive Insect Biocontrol and Behavior Laboratory, Beltsville, MD
- Livestock Arthropod Pest Research Unit, Kerrville, TX
- Cattle Fever Tick Research Unit, Edinburg, TX
- Veterinary Pest Genetics Research Unit, Kerrville, TX
- Mosquito and Fly Research Unit, Gainesville, FL
- Crop Bioprotection Research Unit, Peoria, IL

Component 2: Medical Entomology

Medical entomology research in NP 104 is comprised of research with a primary focus on arthropods that harm humans with the potential to cause disease because of their bites. The USDA has been involved in medical entomology and public health since its inception in 1862. Many public health approaches of arthropod control are adapted from the agricultural sector. The USDA emphasis on medical entomology was established when the Department of War requested the assistance in developing repellents and chemicals to protect service personnel while on missions. NP 104 continues this medical entomology effort with a mission to protect both U.S. service personnel and U.S. civilians from arthropod attack at home and abroad. Scientists within the program work closely with other government agencies, such as the U.S. Environmental Protection Agency, to develop products to protect humans from arthropod attack.

Based upon customer/stakeholder input, the medically important arthropods of greatest concern are mosquitoes, house flies, ticks, and sand flies. Based upon the increase in tick-borne Lyme disease, additional resources will be directed at tick research. As a result, sand fly research has been decreased and bed bug research eliminated from the NP 104 Action Plan.

Problem Statement 2A: Integrated Pest Management of Mosquitoes of Medical Importance

Mosquitos are vectors of pathogens that cause disease and death in humans. The development of insecticide resistance makes control approaches less effective. Improved surveillance, and sustainable

control for mosquitoes is necessary to reduce risk to U.S. military and civilians at home and while traveling abroad.

Worldwide, mosquitoes are the insects that threaten human health most severely. The most severe diseases caused by mosquito-borne pathogens are malaria, dengue, Japanese encephalitis, and yellow fever. There are about 3,400 species of mosquitoes described, but only 200-300 affect human health. Most mosquito studies have been concentrated on the important vectors and species that are easiest to manipulate in the laboratory, even though other species transmit pathogens, serve as enzootic vectors, or cause critical annoyance problems.

The recent introductions of West Nile virus and Zika virus into the United States reinforces the need to continue research on the arthropod vectors that transmit disease pathogens. Introduction of Rift Valley fever into the United States remains a significant threat. This disease has devastating impacts on animals and humans. Additionally, the corresponding expansion of *Aedes aegypti* into areas in the United States where it was previously eradicated exposes even more people to disease.

Anopheles mosquitoes transmit the parasites that cause malaria. The United States had severe problems with malaria through the 1920s and it is still a principal cause of morbidity and mortality throughout the tropics and in parts of temperate Asia. The importance of malaria prevention is increasing as the U.S. military shifts its emphasis to Africa and eastern Asia. In addition, there is a threat of invasive Anopheles species that could increase the likelihood of malaria epidemics, such as the outbreak that occurred when An. arabiensis invaded Brazil in the 1930s and Egypt in the 1940s.

Other genera of mosquitoes also include important species. *Ae. aegypti* remains the principal vector of dengue virus worldwide. This species is also the vector of Chikungunya and Zika viruses; it also vectors the Mayaro virus, a related pathogen of emerging significance. Many of the lessons learned on *Ae. albopictus* control apply to *Ae. aegypti. Culex tarsalis* is a major vector of viral encephalitides in the western United States and *Cx. pipiens/quinquefasciatus* is a significant vector throughout the United States and worldwide. Many floodwater *Aedes* and *Psorophora* species, including *Ae. vexans* and *Psorophora columbiae*, are severe pests and occasionally vector viruses. The threat of new invasive species is particularly severe for mosquitoes that develop in containers, like *Ae. notoscriptus* from Australia, or that transmit a virus, like *Cx. Tritaeniorhynchus*, a vector of Japanese encephalitis virus.

Research Focus

Risk Assessment and Biology

- Evaluation of the risk and economic impact of mosquito-borne pathogens during the next 3 decades, both domestically and in potential foreign military theaters, considering both human and animal diseases, as well as native, invasive, and potentially invasive vector species.
- Identification of species-specific microbiomes in mosquitoes and determining their effects on the vector competence (or vectorial capacity) of mosquitoes.

Surveillance

• Development of a standardized vector detection and surveillance system that is effective for all important groups of mosquitoes and provides rapid detection of vector species and pathogens, including methods that employ genomics and proteomics.

Control

- Development of biological control using a sterile insect technique against medically important biting insects.
- Development of integrated personal protection systems, including new topical repellents that use a

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lower percentage of active ingredients; spatial repellents that protect a room, a group of people, or an individual from point sources; and textile treatments that are alternatives to current pyrethroid impregnation.

- Development of new and improved tools for adult mosquito control, including adulticides, textile treatments (bed nets, curtains, clothing, military field materials), and larvicides that overcome current limitations on use.
- Development and evaluation of devices, techniques, insecticides, or spatial repellents that prevent mosquitoes from being transported on vehicles, aircraft, or ships.
- Coordination with military partners to evaluate effectiveness and use of ARS-invented technology under deployment conditions.

Monitoring and Sustainability

- Determining how pathogens of mosquitoes affect their susceptibility to control and personal protection techniques with emphasis on animal and human diseases such as Rift Valley fever, and other arboviruses recently introduced into the United States such as dengue, chikungunya, and Zika viruses.
- Determining how the main control chemicals (currently pyrethroids) affect important vector and pest species, including how the chemicals affect the development of resistance to these controls.

Anticipated Products

- Models developed to identify areas of the United States at high risk of introduction of invasive vector mosquitoes or invasive emerging mosquito-borne pathogens.
- Mosquito host range and resistance status maps of major vector and pest species in the United States and in foreign military theaters to enhance mitigation programs by public health agencies to control vector driven disease outbreaks.
- Knowledge regarding efficacy of novel and conventional control strategies specific to habitat, ecological region, and target insect populations relevant to U.S. military deployments.
- Improved surveillance technologies.
- Improved personal protection strategies.
- Bioinsecticides and biorepellents containing new natural active ingredient(s) for use in sustainable management and control of mosquitoes.
- New insecticides containing natural product synthetic analog active ingredients for use in the management and control of mosquito populations.
- Microbial bioinsecticides from bacterial and fungal isolates for use in the sustainable management and control of mosquito populations.
- Identification of U.S. military field materials capable of being treated at the factory with residual adulticides effective at reducing medically important mosquitoes across a range of environments relevant to U.S. military deployments.
- Spatial repellents to protect defined areas from key species of mosquitoes across a range of environments relevant to U.S. military deployments.
- Knowledge of efficacy of sterilization techniques, rearing capacity, survival behavior, and competitiveness in laboratory and field studies for potential use of SIT against natural populations of medically important biting insects in the United States.
- Database (catalog) of species-specific molecular characterization for the rapid identification of mosquitoes and associated pathogens as part of surveillance programs and as an early warning system for detection of invasive species and pathogens of humans and animals.
- Knowledge of distinct and shared microbiomes of mosquito vectors by comparing life stages, different physiological states, and pathogenic challenges to identify potential targets for

management strategies.

Potential Benefits

- Earlier detection of invasive species and more effective IPM of vectors and pest mosquitoes.
- Improved integrated personal protection from arthropod attack.

Problem Statement 2B: Integrated Pest Management of House Flies of Medical Importance

Due to their indiscriminate feeding habits, predilection for microbe-rich environments, and synanthropic behavior, house flies are cosmopolitan pests of medical importance. House flies have been shown to harbor and transmit hundreds of species of microbes, including those causing foodborne illnesses in humans, such as the pathogenic bacteria *Escherichia coli* O157:H7 and *Salmonella enteriditis*. Studies to better characterize the relationship between pathogens and adult flies are needed to develop effective strategies to reduce foodborne illnesses in humans.

"Filth flies" in the families Muscidae, Calliphoridae, and Sarcophagidae transmit foodborne pathogens like *Escherichia coli, Salmonella enteriditis,* and noroviruses. Recent work has shown that house flies (*Musca domestica*) amplify some pathogens in their guts. As a result, flies can be a major cause of diarrheal disease in humans. Little information exists on how pathogenic bacteria multiply within and on house flies, and this knowledge could improve our ability to limit their role as vectors of those pathogens. The military sees diarrheal disease as its third most significant infectious disease threat and routinely controls flies to protect deployed personnel. Unfortunately, state-of-the-art fly control is much less effective than mosquito control. The best strategy to separate flies from humans is uncertain, though dozens of products are used in specific situations. Among the gaps in these techniques are effective larval and adult control, interruption of fly movement from larval sources to humans, effective and maintenance-free interventions such as attract and kill devices, and insecticide resistance management.

Research Focus

Risk Assessment and Biology

- Quantification of the distribution, health threat, and associated costs of flies with respect to various populations (e.g., urban, agricultural, military), projected 3 decades into the future and incorporating factors associated with climate change.
- Evaluation of gaps in techniques available for integrated fly control, especially in a military setting and including establishment of realistic action thresholds.
- Assessment of house fly interactions with human pathogens including bacterial survival and transmission.
- Bioinformatic studies of the house fly genome and pathogen-induced transcriptomes.

Surveillance

• Development of improved traps and baits.

Control

• Development of biological control agents.

Monitoring and Sustainability

• Conduct physiological and other research that supports development of effective resistance management.

Anticipated Products

• Risk assessment of house flies as reservoirs and transmitters of pathogens affecting human health.

- Novel control methods based on disrupting pathogen transmission by targeting of key defense genes used during fly-microbe interactions.
- Improved traps and baits for use in sensitive areas, such as in food service and medical facilities.

Potential Benefits

- Reduced prevalence of fly-transmitted diseases.
- Improved human health especially in areas where flies have access to animal or human waste.
- Increased food safety.
- Improved surveillance and management systems using knowledge gained in fly behavior and trap development studies.

Problem Statement 2C: Integrated Pest Management of Sand Flies of Medical Importance

Sand flies bite humans and if infected can transmit several pathogens, such as Phlebovirus, Rift Valley fever virus, and *Leishmania* parasites, the latter leading to a disease called leishmaniasis. There are two forms of this disease, a cutaneous form that produces skin lesions and a visceral form that affects internal organs. Improved understanding of sand fly behavior in the ecosystem and novel control strategies are needed to reduce risk to U.S. military and civilians while traveling abroad.

Phlebotomine sand flies are a subfamily of Psychodidae that include the familiar and non-biting drain flies. Sand flies transmit pathogens causing leishmaniasis, bartonellosis, and sand fly fever. They can be infected with one of three serotypes of *Phlebovirus* (Naples, Sicilian, and Toscana viruses) that can be transmitted and cause Pappataci fever in human populations in the subtropical regions of the Eastern Hemisphere. Sand flies can also transmit another very dangerous *Phlebovirus*, Rift Valley fever virus, which threatens agriculture globally, putting national economies and domestic animal and human populations at elevated risk. Typical control techniques such as fogging and residual treatments are effective at controlling these pests, but not ideal. For the U.S. military, sand fly fever has been a major problem in particular regions, but most recently leishmaniasis was a major problem in Iraq and a threat in Afghanistan. Leishmaniasis also occurs in Africa and the Western Hemisphere, though it is a very rare disease in the United States. From the standpoint of NP 104, research on sand flies is conducted to protect U.S. military personnel when deployed overseas.

Research Focus

Risk Assessment and Biology

• Identification of the potential ranges of subpopulations of known vectors of leishmaniasis to humans (such as *Phlebotomus orientalis*) based on population-environment modeling.

Surveillance

• Development and evaluation of attractant-based surveillance devices for sand flies that provide a practical alternative to traps baited with carbon dioxide.

Control

- Discovery of personal protection products (e.g., topical repellents and treated bed nets) against sand flies.
- Improvement of sand fly IPM programs by evaluation of alternative toxicants and other control strategies.

Anticipated Products

- Predictive models that provide early warning of unusually large populations of sand flies.
- Control strategies with efficacy specific to habitat, ecological region, and target sand fly

populations.

Potential Benefits

• Fewer cases of leishmaniasis and sand fly fever in U.S. military personnel.

Problem Statement 2D: Surveillance and Control of Ticks of Medical Importance

Ticks bite humans and transmit pathogens that cause diseases such as Lyme disease and Rocky Mountain spotted fever. Lyme disease has been increasing over the past few decades and the *Ixodid* tick species responsible for infecting humans are widespread along the U.S. west coast and almost completely throughout the eastern half of the United States. Other tick genera, such as *Amblyomma*, transmit different human pathogens, including Mammalian Meat Allergy (MMA), also known as "alpha-gal", and are the presumed vector of a poorly defined illness named Southern Tick-Associated Rash Illness (STARI). Improved surveillance, control, and risk assessment strategies are needed to protect Americans from tick-borne disease.

Ticks transmit a wide variety of pathogens, many of which are associated with potentially fatal disease. Fortunately, most of the diseases are uncommon in the United States, including human babesiosis, Rocky Mountain spotted fever, ehrlichiosis, anaplasmosis, and Colorado tick fever. Lyme disease affects at least 20,000 people per year and is concentrated in the Northeast and the northern Midwest, though the Centers for Disease Control considers the disease vastly underreported and estimates that up to 300,000 people are infected annually. The public health and medical research communities are actively engaged with the clinical aspects of Lyme disease, but vector control and bite prevention are areas in which NP 104 has expertise. In addition, the Lone Star tick (*Amblyomma Americanum*) is assuming importance because of its geographic range, pathogen transmission, and association with other tick maladies. NP 104 will focus on the challenge of preventing tick bites by personal protection, particularly in and around areas with high disease prevalence and where ticks are rapidly expanding their geographic range. Therefore, it is important to document the geographical range of a potential vector and conduct a risk assessment of the importation threat for exotic ticks originating from Europe, Asia, and Oceania.

Research Focus

Surveillance

- Development and evaluation of new techniques for surveillance and control of *Ixodes scapularis*, *I. pacificus*, and *Amblyomma americanum*, *A. maculatum* and other human biting ticks; and assembly of available techniques into designs for integrated community control programs.
- Tick bite risk assessment for field workers in agriculture, forestry and other outdoor workers and protective measures that may help minimize the risk.

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Control

- Evaluate existing veterinary acaricide products and develop new systemic and topical acaricides for mice and deer-targeted tick control.
- Develop and evaluate new tick control strategies and products for personal protection.
- Develop and assess anti-tick antigens for vaccine development for use in wildlife (mice and deer) against *Ixodes, Rhipicephalus, Amblyomma* and other human biting Ixodid tick species.
- Assessment of the suitability of anti-tick vaccines for potential use in deer against *lxodes* ticks.

Anticipated Products

- Increased understanding of the role of individual tick species in disease transmission.
- Tools, including chemicals, to prevent tick bites.

- Novel vaccine formulations to control Ixodes ticks.
- IPM of deer (black-legged) ticks to prevent Lyme disease in suburban settings.

Potential Benefits

- Increased ability to control ticks.
- Fewer cases of Lyme disease and other tick-associated maladies in the United States.

Component 2 Resources

- Agroecosystem Management Research Unit, Lincoln, NE
- Arthropod-borne Animal Diseases Research Unit, Manhattan, KS
- European Biological Control Laboratory, Thessaloniki, Greece
- Invasive Insect Biocontrol and Behavior Laboratory, Beltsville, MD
- Livestock Arthropod Pest Research Unit, Kerrville, TX
- Veterinary Pest Genetics Research Unit, Kerrville, TX
- Mosquito and Fly Research Unit, Gainesville, FL
- Natural Products Utilization Research Unit, Oxford, MS
- Crop Bioprotection Research Unit, Peoria, IL

Component 3: Fire Ants and other Invasive Ants

Component 3 is comprised of research with an exclusive focus on ants that harm humans, livestock, crops, and structures. Pest ants cause severe damage in agricultural and urban sectors and concurrently negatively affect natural environments. Most pest ants are invasive species. In the agricultural sector, ants have been shown to reduce crop yields and harm livestock by attacking small or newborn farm animals, like chicks or calves. Such attacks lead to physical damage, increased stress, and the potential death of these animals. High-value crop plants such as soybeans, corn, okra, potatoes, almonds, and citrus have been directly affected by ant feeding. Even farm equipment and irrigation systems have been damaged by pest ant species. The predatory nature of ants may also harm plants or crop production by reducing the density of pollinators and other beneficial insects, such as natural enemies. Certain species increase the densities of sap-sucking pest insects (aphids and coccids) that transmit diseases directly or encourage growth of molds on important crops such as grapes, citrus, and cucurbits. Pest ant-contaminated shipments into domestic or foreign controlled areas impede commerce when they are subject to rejection and returned to their point of origin. This is of high concern in the plant nursery, sod, and forage industries. Ants also are the most common arthropod pests in urban environments and account for more complaints to pest control companies than any other insect group. Some ant species sting and are known to cause anaphylaxis and even death. In natural environments, invasive ants often displace native ant fauna, reduce arthropod biodiversity, and potentially disrupt entire ecological communities as invading "ecosystem engineers" due to their dominance and the varied functional roles they have in ecosystems.

Based upon customer/stakeholder input, the ant species of greatest concern are imported fire ants, Argentine ants, crazy ants, electric ants (little fire ants), carpenter ants, Asian needle ants, white-footed ants, odorous house ants, dark rover ants, Pharaoh ants, and big-headed ants.

Problem Statement 3A: Risk Assessment, Biology, and Control of Invasive Fire Ants

Invasive fire ants harm humans and animals and cause significant economic losses from damage to crops and structures. Sustainable, affordable approaches to long-term suppression of populations do not exist. Improved and novel control interventions and approaches are needed to control this pest.

Fire ants are considered major agricultural, medical, urban, and environmental pests in the United States. Imported fire ants currently infest more than 365 million acres from Virginia to Florida to California. Beyond

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the continental United States, these invasive ants have expanded into Puerto Rico, most of the other Caribbean islands, Australia, Taiwan, China, and India. Agricultural, economic, and medical costs due to fire ants exceed \$6 billion dollars annually in the United States and commercial chemical treatments for control are estimated to cost as much as \$40 per acre. The affected urban and agricultural sectors are broad in range and include households, schools and recreation areas, electric and communication equipment, and animal and plant agriculture. Fire ant control depends heavily on the use of synthetic insecticides. There are two major chemical methods available to control the red imported fire ant: baits using slow-acting insecticides and spray/drench/granule mound treatment using fast-acting contact insecticide. Both methods have problems. Current fire ant bait products lack specificity and can harm native ants as well as fire ants. For mound treatment, the major problem is the potential impact of synthetic insecticides on the environment. Both methods provide only temporary fire ant suppression. There is a need to develop new inexpensive, environmentally friendly, and fire ant-specific products and their delivery techniques. More than 80 years after its introduction into the United States, the red imported fire ant is still spreading. Novel detection methods to facilitate interception of fire ants at quarantine boundaries are needed to reduce continued spread of fire ants.

A solution for alleviating the heavy dependence on synthetic insecticide is to implement biological control and develop biopesticides in fire ant management. Intercontinental comparisons between native (South America) and introduced (United States) fire ant populations have shown significant differences in the biology, genetics, and reproductive behavior of ants from the two areas. These comparative studies have also shown that numerous natural enemies (parasitoids, parasites, and pathogens) are associated with fire ants in South America, but only a few of these have been found from invasive fire ants in the United States. Extensive integrated studies are needed to understand biological differences between native and introduced populations and to identify the most appropriate natural enemies to release for sustainable biological control of fire ants.

Many natural materials have been reported to be toxic and/or repellant to fire ants, including citrus oil, mint oil, essential oil from the leaf of *Cinnamomum osmophloeum* Kaneh, sweet wormwood oil, Nootka oil, and sweet orange essential oil. Recently several naturally occurring benzoate analogs have been found by ARS scientists to be potent toxins against fire ants and one of these analogs has not been identified by OSHA as hazardous or toxic in water, indicating a great potential for its application in fire ant management. Natural products might be a promising source of new active ingredients for developing environmentally friendly fire ant control products. Interestingly, ARS research has also shown that some environmentally benign synthetic compounds are also potent fire ant toxins. These compounds may be also a source of safe active ingredients for fire ant control.

High selectivity is a desirable property for an ant control product. However, most active ingredients used in current ant control products are broad-spectrum insecticides that affect non-target ants and other insects. Genetic control strategies hold powerful potential for very specific intervention against invasive ants. Because gene sequences are extraordinarily unique, a gene disruption product hypothetically can target not only a single species, but a specific population within a species. Previous studies show a given ant species harbors hundreds to thousands of unique genes not found in other ants. A control product based on invasive ant genes will benefit the entire ecosystem disturbed by invasive species.

Research Focus

Risk Assessment and Biology

- Investigation of the ecological, behavioral, and competitive interactions of invasive pest ants that are sympatric with fire ants, e.g., the tawny crazy ant and the big-headed ant.
- Development of methods capable of identifying numerous fire ant species.

• Identification of essential molecular and/or receptor antagonist/agonists targets through molecular and metabolomic research

Control

- Discovery, collection, and evaluation of new natural enemies (parasitoids, parasites, and pathogens) of native South American fire ants for potential use as biological control agents of introduced fire ants in the United States.
- Improvement in virulence of fire ant pathogens by selection of strains in ants and in vitro.
- Design and evaluation of bait and mound treatment formulations with pathogens, natural products, and environmentally friendly synthetic chemical compounds as the active ingredients.
- Investigation of novel approaches to fire ant-specific control, including new active ingredients that are oil and water soluble, and attractants and phagostimulants for bait enhancement.
- Investigation of repellents and anti-phagostimulants, including those derived from plants, that exclude the ants from specific areas, and/or protect domestic and livestock food materials.
- Determination of gene function and use of existing genomic resources to develop fire ant-specific control methods and products.
- Development of new and improved insecticide delivery technologies to reduce the use of synthetic insecticide in fire ant management.

Anticipated Products

- Knowledge of the dynamics of pest ants that are sympatric with invasive fire ants.
- Field-ready rapid method to detect and discriminate red and black imported fire ants from other ant species.
- Classical biological control agents capable of providing sustained control of fire ants.
- Formulations capable of excluding the fire ant from areas designated as free zones.
- Novel fire ant control based on new biochemical and molecular targets.
- Novel and improved methods to deter fire ant invasion, spread, and methods and products to mitigate infestations and damage.

Potential Benefits

- Improved understanding of ants and their impact.
- Improved sustainable fire ant control throughout the United States.

Problem Statement 3B: Integrated Pest Management of Invasive Crazy Ants

Invasive crazy ants impact agriculture and human activities. This species was introduced into the United States about 15 years ago and therefore little is known about the biology of this ant species. Basic biological studies along with new and improved control approaches are needed to achieve adequate control.

The tawny crazy ant (*Nylanderia fulva*) is an invasive ant from South America that in the United States develops extremely dense populations that inundate and dominate natural, agricultural, and urban landscapes. Tawny crazy ants currently infest at least 28 counties in Florida, 29 counties in Texas, the island of St. Croix (U.S. Virgin Islands), and is spreading into Georgia, Alabama, Louisiana, and Mississippi. Tawny crazy ants impact agriculture by feeding on plant carbohydrate sources. They tend and protect pathogen-transmitting aphids and asphyxiate small livestock, e.g., chickens, and stress large livestock. They also affect natural resources, recreational areas, and residential environments by endangering wildlife, reducing biodiversity, infiltrating buildings, and decreasing the value and enjoyment of infested lands due to high ant density. In addition, high densities of these ants have resulted in excessive pesticide use, which poses a serious hazard to humans and the environment. Control options for tawny crazy ants are severely limited. There is over-dependence on residual insecticide sprays that are problematic in areas such as nature

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preserves where non-target damage to insects results. IPM strategies based on biology, baiting, and biological control are needed to obtain near and long-term control of invasive tawny crazy ants.

Research Focus

Risk Assessment and Biology

• Evaluation of the relationships between tawny crazy ants and other ant species through basic ecological and biological research.

Surveillance

• Investigation of the basic behavioral and reproduction ecology (biology) and chemical ecology of tawny crazy ants to improve surveillance.

Control

- Investigation of the basic behavioral and reproduction ecology (biology) and chemical ecology of tawny crazy ants to improve bait specificity and control.
- Discovery of natural enemies of tawny crazy ants suitable as biological control agents.
- Identification of effective bait active ingredients and formulations for tawny crazy ant control.
- Development of effective baiting strategies for tawny crazy ants such as timing bait applications relative to the seasonal phenology of brood production.
- Establishment of genetic resources as a foundation for developing next-generation genetic control for crazy ants.

Anticipated Products

- An understanding of how tawny crazy ant colonies grow, mature, and decline or migrate.
- An understanding of the dynamic interactions of sympatric pest ant species.
- Surveillance tools and improved bait specificity.
- Natural enemies for classical biological control (natural) of tawny crazy ants.
- Effective bait(s) for tawny crazy ant control.

Potential Benefits

- Improved understanding of tawny crazy ants and their impact.
- Improved and sustainable control of Nylanderia crazy ants.

Problem Statement 3C: Integrated Pest Management of Other Invasive Pest Ants

Other invasive pest ant species exist that harm humans and animals and damage crops and structures. The United States remains at risk from the introduction of invasive species. Basic biological studies, including improved surveillance, control, and understanding the ecosystem that the invasive ants thrive in, are needed to better protect the United States from invasive and native ants.

Numerous additional ant species are also serious pests of agriculture, residences, and the natural environment, including electric ants (little fire ants), Argentine ants, big-headed ants, white-footed ants, Pharaoh ants, and Asian needle ants. Many of the technologies developed for imported fire ants and tawny crazy ants may be adapted for use with these and other pest ants. A better understanding of the climatic and habitat requirements (applied ecology) of these pest ants will lead to improved predictions of their potential range and where they will cause the most problems.

Research Focus

Risk Assessment and Biology

- Development of models to predict the invasive ants likely to become future pests in the United States and to predict future range expansions of invasive ants already present in the United States.
- Expansion of the genetic understanding of invasive ants, individually and comparatively, through sequencing and recombinant technologies.
- Investigation of potential impacts of abiotic factors on invasive and native pest ants.
- Determination of comparative invasive pest ant characteristics, e.g., competitiveness, and reproductive biology through research on the basic biology and ecology of selected pest ants.

Surveillance

• Examination of chemical ecology and semiochemical approaches to enhance bait effectiveness and improve surveillance and detection methods for select pest ants.

Control

• Investigation of the chemical ecology and semiochemical approaches of invasive ants to improve bait specificity and control for select pest ants.

Anticipated Products

- An understanding of intraspecific and interspecific ant colony dynamics.
- Improved surveillance methods.
- Ant bait formulation(s) effective against selected invasive ant species.
- Natural enemies for classical biological control of an invasive ant species.
- New pest management and IPM strategies for selected invasive ant species.

Potential Benefits

- Improved understanding of invasive ants and their impact.
- Assessment of threat from invasive ants and development of management tools to control ants.

Component 3 Resources

- Biological Control of Pests Research Unit, Stoneville, MS
- Imported Fire Ant and Household Insect Research Unit, Gainesville, FL
- Natural Products Utilization Research Unit, Oxford, MS