

National Program 108
Food Safety
2006-2010 Action Plan

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Introduction

Food Safety falls under Goal 3 of the Agency Strategic Plan: **Enhance Protection and Safety of the Nation's Agriculture and Food Supply**. For the Nation to have affordable and safe food, the food system must be protected at each step from production to consumption. The production and distribution system for food in the United States has been a diverse, extensive, and easily accessible system. This open system is vulnerable to introduction of pathogens and toxins through natural processes and global commerce and by intentional means. In response to these threats, crop and livestock production systems must be protected from the ravages of diseases whether domestic or exotic in origin. The food supply must be protected during production, processing, and preparation from pathogens, toxins, and chemical contamination that cause disease in humans.

Food safety research seeks ways to assess and control potentially harmful food contaminants. Research to ensure a secure agricultural production system refers to work that reduces or eliminates factors that threaten the ability of U.S. agriculture to produce enough food and fiber, year to year, to meet the needs of American consumers. ARS will conduct research designed to generate knowledge regarding new and improved management practices, pest management strategies, sustainable production systems, and control of potential contaminants for farms of all sizes. These activities will ensure a secure production system able to provide a safe, plentiful, diverse, and affordable supply of food, fiber, and other agricultural products.

ARS will provide scientific information and technology to producers, manufacturers, regulatory agencies, and consumers to support their efforts to provide a secure, affordable, and safe supply of food, fiber, and industrial products.

Relationship of the NP 108 to the ARS Strategic Plan: Outputs of NP 108 research support the ‘Actionable Strategies’ associated with the performance measures shown below from the ARS Strategic Plan for 2003-2007, Objective 3.1: Provide Science-Based Knowledge on the Safe Production, Storage, Processing, and Handling of Plant and Animal Products and on the Detection and Control of Toxin-producing and/or Pathogenic Bacteria and Fungi, Parasites, Chemical Contaminants, Mycotoxins, and Plant Toxins so as to Assist Regulatory Agencies and the Food Industry in Reducing the Incidence of Foodborne Illnesses.

Performance Measures 3.1.1: Develop new on-farm preharvest systems, practices, and products to reduce pathogen and toxin contamination of animal- and plant-derived foods. **Target:** Develop practices and/or products that reduce preharvest contamination of major animal- and plant-derived food products.

Performance Measures 3.1.2: Develop and transfer to Federal agencies and the private sector systems that rapidly and accurately detect, identify, and differentiate the most critical and economically important foodborne microbial pathogens. **Target** Develop practices and/or products that reduce postharvest contamination of major animal- and plant-derived food products.

Mission Statement:

To provide through scientific research, the means to ensure that the food supply is safe and secure for consumers and that food and feed meet foreign and domestic regulatory requirements. Food safety research seeks ways to assess, control or eliminate potentially harmful food contaminants, including both introduced and naturally occurring pathogenic bacteria, viruses and parasites, toxins and non-biological-based chemical contaminants, mycotoxins and plant toxins. Since food safety and food security are global issues, our research program involves both national and international collaborations through formal and informal partnerships. Our accomplishments and outcomes are utilized in national and international strategies delivering research results to regulatory agencies, commodity organizations and consumers.

Vision Statement:

For the Nation to have a safe, plentiful, diverse, affordable, quality food supply, protected during production, processing, and preparation from pathogens, toxins, and chemical contamination that cause disease in humans.

The research components of this program include:

- [Microbial] Pathogens, Toxins and [non-biological-based] Chemical Contaminants: subdivided in Pre-harvest and Post-harvest
- Mycotoxins and Plant Toxins

Component 1.1. Pathogens, Toxins and Chemical Contaminants - Preharvest

The challenge to preharvest food safety research is to develop information and products that will assure the safety of food products from animal agriculture. Animal food products must be safe for the consumer and produced with sensitivity to animal welfare concerns, and they must be competitive in the national and international market place; to make this possible research must develop interventions, that is effective products and practices which will be used. Profit margins in animal and poultry production are variable and often small, and producers will not increase their cost of doing business, by using a new vaccine or handling manure in a different manner, for instance, unless they are convinced it will bring them a return in the health of their animals, the price they will receive in the marketplace, or the acceptability of their animals to meet regulatory agency standards.

The effectiveness of interventions during production must be considered at the slaughterhouse level as well as at the farm itself. We need to know both (1) if on farm interventions have really decreased the pathogens in the GI tract of the food producing animal or bird at the time of slaughter, and (2) has this made a difference in the pathogen load of the animal products leaving the slaughter/packing plants. Production of food products with the minimum of both pathogens and chemical residues is important even if current regulatory guidelines are being met. For example, the Food and Drug Administration has requested that on farm practices be optimised to decrease the number of pathogens in raw milk, even though pasteurization has been considered successful for many years in providing safe milk for the American consumer. There are certain cheese products, primarily soft cheeses that consumers prefer to be made from unpasteurized milk, and they are available either in imports or from ethnic markets. Some consumers also continue to obtain raw milk for home consumption. Thus we must have the knowledge to produce milk in the safest manner possible. Another reason to further assure the microbiological safety of raw milk, or any other food product of animal origin, is to provide a hedge against accidents or errors occurring during the pasteurization, handling and transportation processes which would provide an opportunity for pathogen growth.

Problem Statement 1.1.1: Methodology

Rapid, accurate, and sensitive methodology is needed for identification and quantitation of epizootic pathogens and chemicals that may be transmitted to humans, as well as for other bacteria that may affect their occurrence in the animal and their environment. Previous standards for the sensitivity of assays will not be acceptable in the future. For example, acceptable precision and accuracy for all dioxin congeners can now be achieved. This achievement has made a study with dioxins conducted less than 10 years ago now obsolete.

State of the art methodology is needed for research that is conducted to identify and quantify the pathogens and chemicals in both animal fluids and tissues, and in their environment, including animal manure; to establish the ecology and epidemiology of the pathogens; to delineate the host-pathogen relationship of these zoonotic pathogens in food producing animals; and to develop effective interventions that can be easily instituted by the livestock and poultry producer.

Research Needs

Methods for accurate, timely and cost effective specific identification and quantitative enumeration of pathogens and chemical contaminants with appropriate speciation, are necessary not only for epidemiology, ecological and intervention research, but also to support the risk assessments which are carried out by the regulatory agencies, that is, FDA and FSIS. New methods must provide a wide range of molecular genetic, genomic and proteomic information on epizootic pathogens that in turn enable the development, validation and standardization of new rapid test methods, new drugs and vaccines, insights into key immune responses, and improved intervention strategies.

Outputs

- Methods to recover, identify and quantitate pathogens and chemical residues in the tissues and matrices, including animal manure, where they are expected to be found, or are needed for regulatory purposes.
- Methods to recover and enumerate 'non-culturable' pathogens from all environmental niches and to selectively enhance the growth of extremely slow growing organisms.
- Methods that are sufficiently rapid and cost effective to be useful to both producers and regulatory agencies.
- Genetic and serologic bases for new methods that will provide differentiation of bacterial clones to the extent needed for attribution of these sources.
- Highly specific serological tests with the necessary sensitivity to detect animal carriers.
- Development of microarrays to assess gene expression in various environments.

Impact

ARS will lead in developing and validating methodologies that have regulatory, industry and research uses for solving preharvest pathogen and chemical contamination problems. Fortunately there is a commonality of interests among these groups which allow similar methods to be used by each. Having reliable, cost effective identifications of pathogens and chemicals in food animal tissues and environmental samples will assist in carrying out risk assessments, in developing and validating microbial models that predict the growth and virulence of pathogens, and in identifying areas where interventions are most critically needed. This information will assist FSIS, FDA and the food animal industry in the production of safe food products.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; National Animal Disease Center, Ames,

Iowa; Swine Odor and Manure Management Research Laboratory Ames, Iowa; Meat Animal Research Center, Clay Center, NE; Red River Valley Agricultural Research Center, Fargo, ND; Western Regional Research Center, Albany, CA.; Southern Plains Agricultural Research Center, College Station, TX; Livestock Issues Research, Lubbock, TX; and Richard Russell Research Center, Athens, GA.

Problem Statement 1.1.2: Epidemiology

Epidemiology determines the origin and routes of transmission of epizootic pathogens. The livestock industry continually incorporates additional mobility, moving animals one or more times during the production cycle, in order to benefit from large scale operations. Animal mobility complicates the challenge to trace the flow of both pathogens and specific identified genes, through the production cycle from application of manure to crop or grazing lands, to the raising and slaughter of meat animals, or to production of milk and eggs. For instance, less than half of the dairy calves now grow and mature at the premises where they are born, and thus whatever pathogens are associated with them at birth or at common rearing facilities may be introduced to the milking herds where they spend their productive lives. Also cross-contamination occurs in multiple sites as animals are transported to slaughter plants, at the plant through contact with workers and the plant environment, and during fabrication in plant and in retail supermarkets, particularly those that further grind or fabricate on site. We need to be able to evaluate the contribution of each of these sites.

Research Needs

Small scale, limited time frame and end point epidemiology studies can be carried out by one research scientist with limited assistance, but increasingly large scale research programs are necessary to meet the needs of today's agriculture which produces food for millions of consumers. Large studies are needed in order to have the statistical power to sample and analyze appropriate biological samples, recover the isolates and determine the identity and numbers of pathogens, and perform the necessary statistical analyses of the large volumes of data, in order to reliably correlate laboratory data with herd information and relevant production practices. A second vital consideration in epidemiology is the need to conduct studies over an extended time frame. Data from single time points do not reflect the large inherent variation that is a part of bacterial reservoirs and amplification cycles, and do not serve the needs of the animal industry. One program now in the initial stages that addresses this deficiency is the Collaboration for Animal Health and Food Safety Epidemiology (CAHFSE), a multiagency program utilizing the resources of ARS, FSIS and APHIS in a single coordinated framework to conduct epidemiology with repeat sampling over an extended time frame. (See additional information under 'Antibiotic Resistance'). Information-gathering projects that can detect early trends and warning signals are also vital to biosecurity systems for animal product food production.

Outputs

- Development of coordinated food animal surveillance and epidemiology programs for food animal species which recognizes early warning signs of pathogen infection.

- Estimates of the basic reproductive ratios (Ro's) for epizootic pathogens in various food animal species for epidemiologic modeling and more accurate prediction of the effectiveness of interventions.
- Epidemiological studies that elucidate chemical or pathogen presence and behavior in particularly important, suspected biological niches, including animal manure and other environmental niches.

Impact

Scientists will gain an additional understanding of the transmission and dissemination of food safety pathogens from the farm through slaughter environments from the epidemiology and surveillance data of food safety pathogens in animal production. The contribution from all aspects of the environment, including feed and water can also be evaluated. This information will provide data needed to carry out risk assessments, to develop and validate predictive microbial models, and assist in determining critical directions for intervention strategies to reduce the spread of pathogens both in food animals and poultry and to humans. CAHFSE will enable USDA to identify and track emerging diseases and identify and implement mitigation strategies in a timely manner thereby averting negative effects on the economy, animal health and public health. CAHFSE will also serve as a model for future surveillance efforts on a national level, thus assisting FSIS, FDA and the food animal industry in the production of safe food products.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; National Animal Disease Center, Ames, Iowa; Meat Animal Research Center, Clay Center, NE; and Richard Russell Research Center, Athens, GA.

Problem Statement 1.1.3: Ecology, Host Pathogen and Chemical Comtaminants Relationships

The challenge of the future is to determine the attributes of the ecological communities in which epizootic pathogens are found in animals and their environments. It is not sufficient to know that food animals may be carrying one or more specific epizootic pathogens; we need to know their relationship to and the attributes of the microbial communities in which pathogens live, particularly the gut of food producing animals. To develop interventions that can control these bacteria and protozoa we need to identify sites and mechanisms of colonization in animals, to identify and characterize virulence attributes and immunogenic responses, to learn how animals and microbes affect one another and how this relationship is affected by animal well being. We need to know where pathogens reside in the host, particularly during the carrier state when they may be particularly difficult to isolate. For instance, E. coli may be found in the gall bladder and Salmonella in the protozoa of the rumen. Similarly we need to know the metabolism, transport, sites of deposition and excretion of chemical residues.

Research Needs

New methods are needed for describing the exceedingly large number of very diverse organisms that inhabit the GI tract of animals in order to gain an understanding of bacterial and fungal community composition, for example, oligonucleotide fingerprinting of rRNA gene clones followed by sorting into taxonomic clusters. We need to more clearly understand what factors, including nutritional, allow animals to start shedding epizootic pathogens in their feces when under stress, and particularly when they are transported to slaughter. Similarly, information on diet or other factors affecting the duration and fate of chemical residues is needed. This basic information is necessary for development of interventions that reduce both chemical residues and colonization and shedding of pathogens in food animals.

Outputs

- Use of molecular methods and genomics to assess the dynamics of the microbial intestinal flora and to more thoroughly understand the dynamics of various gut environments of food producing animals in order to elucidate effective means for improved control of food pathogens in the pre-harvest stage.
- Use of such techniques as oligonucleotide fingerprinting of rDNA genes to assess species diversity and identify novel genes in order to identify opportunities for interventions to control pathogens.
- Locating sequestration and deposition sites, and metabolic pathways to determine factors affecting both holding and release of chemical residues and pathogens.
- Identification of SNP's (single nucleotide polymorphisms) that identify genes associated with host resistance and pathogen virulence and shedding.
- Identification of environmental sites, both physical and biological, which provide pathogen reservoirs and sites of amplification.
- Knowledge of the interrelationships among production practices, animal well-being and pathogen shedding.

Impact

Information gained from study of the ecology, genomic, host pathogen and chemical residue relationships of food safety pathogens in animal production will help scientists gain an additional understanding of the transmission and dissemination of these pathogens and toxins in and among food producing animals. An important underlying assumption for this type of research is the “pre-harvest hypothesis” that is “live animal” control efforts (implemented in or on the animal itself or in its agricultural environment) will reduce downstream post-harvest carcass and meat contamination and ultimately reduce human zoonotic and food-borne disease risk. This information will provide critical data to help reduce fecal shedding and transmission of pathogens to food, to carry out risk assessments, to develop and validate predictive microbial models, and assist in determining critical directions for intervention strategies to reduce the spread of pathogens in food animals and poultry and to humans. This research will also lead to both a core understanding and improved methods to control stress and enhance well-being in livestock resulting in improved and commercially viable handling, feeding, therapeutic, and transportation practices which together will help to reduce or eliminate food borne pathogen infection during production and transportation stages in food

producing animals. Thus it will assist FSIS, FDA and the food animal industry in the production of safe food products.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; National Animal Disease Center, Ames, Iowa; Swine Odor and Manure Management Research Laboratory Ames, Iowa; Livestock Behavior Research Unit, West Lafayette, IN; Meat Animal Research Center, Clay Center, NE; Red River Valley Agricultural Research Center, Fargo, ND; Western Regional Research Center, Albany, CA.; Southern Plains Agricultural Research Center, College Station, TX; Livestock Issues Research, Lubbock, TX; Poultry Production and Product Safety Research Fayetteville, AR; and Richard Russell Research Center, Athens, GA.

Problem Statement 1.1.4: Intervention Strategies

Intervention strategies, including both products and practices, are needed to reduce colonization and shedding of epizootic pathogens by food producing animals. Successful interventions must cost the producer almost nothing. Optimally an intervention could increase the productivity of the animals in addition to decreasing the number of pathogens, e.g., decreasing Salmonella while also increasing the rate of gain and/or feed efficiency of the animals. Interventions that can be used just prior to slaughter are particularly useful, as they can act almost as a preharvest HACCP step with the animals having a very limited opportunity to acquire additional infection prior to slaughter. Industry has been reluctant to change rations and apply practices late in the production cycle. The use of Optaflex, a new feed additive, by the beef feeding industry starting 28 days before slaughter has now opened new opportunities for other additives to be given to cattle during this period just before slaughter.

With swine the present industry practice of holding animals in lairage for at least 2 hours prior to slaughter provides an opportunity for considerable reinfection and rapid systemic transit through the body followed by excretion into the feces. This could limit the value of a preharvest killing step unless it were combined with alternative methods of transporting swine to slaughter. Chickens and cattle have the opportunity to introduce pathogens into the slaughterhouse from their hide, hair and feathers even when pathogens from their GI tract may have been killed by chemical or biological intervention. Residues of any drug used just prior to slaughter are also of great concern, thus drugs used at this time must not be absorbed from the GI tract, or must be otherwise eliminated extremely rapidly, and/or must be extremely non toxic to the animal.

Bacteriophage and bacteriocins both hold a great deal of theoretical promise but these treatments, at least for food producing animals and birds, have not yet been subjected to Food and Drug Administration review. Past history for the use of competitive exclusion cultures (CEC) and the bacteriocin nisin as methods of control, indicates there will be significant regulatory hurdles before approval for the use of either bacteriophage or other

new bacteriocins. Although bacteriophage also have a high level of specificity which may limit their broad application, chemical or microbial treatments with narrow pathogen targets, and specificity of action are sought after, as they are associated with less antibacterial resistance and less disruption of normal gut function.

Research Needs

The best interventions are simple, and changes in management practices to control a pathogen would be ideal. However preventing exposure to pathogens that are almost ubiquitous, as are most of the food safety pathogens, is extremely difficult, and cleaning of the environment and ration changes prior to slaughter have not yet been successful. Production operations need products that are both effective and can be approved by applicable regulatory agencies to successfully emphasize food safety.

Outputs

- Bacterial metabolic targets that allow interventions to be closely focused to specific metabolic pathways.
- Interventions targeting specific metabolic endpoints.
- Vaccines that decrease shedding of epizootic pathogens at the time of slaughter.
- Effective interventions that also increase animal or bird productivity.
- Effective modifications of production practices that particularly address animal welfare concerns and /or dietary components in reducing pathogens.
- Interventions that prevent colonization or exclude pathogens from the gut.

Impact

The development of intervention strategies to control food safety pathogens in animal production will provide technologies to help producers clear these pathogens from food animals during on-farm rearing. Again an important underlying assumption of intervention strategies is the “pre-harvest hypothesis” that is, “live animal” control efforts (implemented in or on the animal itself or in its agricultural environment) will reduce downstream post-harvest carcass and meat contamination and ultimately reduce human zoonotic and food-borne disease risk. These intervention strategies will provide environmentally compatible technologies to significantly decrease or eliminate pathogens food animals during critical periods of on-farm rearing to help reduce fecal shedding and transmission of pathogens to food. The strategies can add critical information to predictive microbial models, in order to reduce the spread of pathogens in food animals and poultry and to humans. These interventions will assist animal producers in meeting FSIS, FDA and the food animal industry goals for the production of safe food products.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville MD; National Animal Disease Center, Ames, Iowa; Swine Odor and Manure Management Research Laboratory Ames, Iowa; Livestock Behavior Research Unit, West Lafayette, IN; Meat Animal Research Center, Clay Center, NE; Red River Valley Agricultural Research Center, Fargo, ND; Western Regional Research Center, Albany, CA; Southern Plains Agricultural Research Center, College Station, TX; Livestock Issues Research, Lubbock, TX; Poultry Production and

Product Safety Research Fayetteville, AR; and Richard Russell Research Center, Athens, GA.

Problem Statement 1.1.5: Antibiotic Resistance

The emergence of antimicrobial resistance (AR) among food-borne and commensal bacteria associated with food animal production has become an important global issue. Despite growing concerns, information regarding the development, prevalence, spread and persistence of AR in food-borne and commensal bacteria is limited, and AR's impact on human health is poorly understood.

Some facets of antimicrobial resistance (AR) cut across all the components of preharvest food safety. AR is considered separately because of its extreme visibility and importance to food safety, human illness and the livestock industry. Both the CDC and the FDA believe that the use of antibiotics in animal production, in particular antibiotics used in feed, and for growth promotion, is a major cause of antibiotic resistance in humans with resulting longer duration of illness and/or untreatable illness with more severe sequelae. The result is that it is more and more difficult for antibiotics for animals to receive approval from the FDA, with the result that the livestock industry is being deprived of valuable tools they have previously relied upon to maintain healthy animals and flocks, and provide consumers with safe and affordable animal food products.

Based on the precautionary principle, the EU has implemented a complete ban on subtherapeutic use of most antimicrobials, although these drugs are still widely used by prescription. Conversely, the US continues to utilize subtherapeutic antimicrobials in animal production. Without continued surveillance and accurate resistance data, trade negotiations will be negatively impacted.

Research Needs

Throughout the farm-to-fork continuum, there are numerous processes that affect the degree of microbial contamination of foods (with any accompanying antimicrobial resistance), including the rearing, transport, slaughter and processing of food animals, and the packaging and preparation of foods prior to consumption. Research is necessary to provide the necessary data to analyze specific resistance to antibiotics in food animals as they are presented for slaughter, so that animals and drugs that are not contributing to human resistance are not unfairly penalized.

CAHFSE, first considered under 1.1.2 Epidemiology will enable ARS, collaborating with other USDA agencies, to identify and track emerging diseases and identify and implement mitigation strategies in a timely manner which can avert economic, animal health and public health consequences. Without continued surveillance, research and testing of interventions, it would be impossible to determine on-farm practices affecting food borne bacterial populations, including the antimicrobial resistance associated with these bacteria. Such a surveillance program will also be vital to the National Biosurveillance Integration System of the DHS.

Outputs

- Development of food animal surveillance and epidemiology programs, particularly the Collaboration for Animal Health and Food Safety Epidemiology (CAHFSE), together with other USDA agencies to assure early detection of epizootic pathogens and unique antibiotic resistance patterns.
- Information about rates and patterns of spread of AR gene flow, among pathogenic and commensal microorganisms and in different environments of various food producing animals, including animal manure. This information can then be used for population genetic studies to map the spread of specific clones in populations across the U.S., and in risk assessments to help assess the effects of antibiotics in animal medicine.
- Identifying linkages of bacterial AR genes to other genes affecting other aspects of bacterial physiology, particularly pathogenicity factors.
- Combining information on the AR of isolates with genetic or other attributes to obtain specific identification and exclusion characteristics for isolates. .

Impact

Identification of factors responsible for mediating AR in food-borne and commensal bacteria will provide vital information on AR mechanisms as well as the incidence, persistence, and rates of dissemination of AR. Scientists will gain an additional understanding of the transmission and dissemination of these pathogens and their accompanying resistance from the farm through slaughter environments. The contribution from all aspects of the environment, including feed and water can also be assessed. This information will provide data to carry out risk assessments, to develop and validate predictive models of microbial and antimicrobial resistance, and assist in determining critical directions for intervention strategies to reduce the spread of AR in food animals and poultry and to humans. Information gained from epidemiology and surveillance of antibiotic resistance (AR) of food safety pathogens in animal production using epidemiologic and molecular and population genetic approaches will help. This will also help prevent physicians from assuming that antibiotic resistance problems can be solved by banning of certain antibiotics from animal use, thus postponing their examination of the role of human antibiotic prescription and use practices in AR. Thus it will assist FSIS, FDA and the food animal industry in assuring production of safe food products.

Resources

ARS scientists assigned to projects under this Problem Statement are located at the National Animal Disease Center, Ames, Iowa; Southern Plains Agricultural Research Center, College Station, TX; and Richard Russell Research Center, Athens, GA.

Component 1.2 Pathogens, Toxins and Chemical Contaminants - Postharvest

In order to direct the research program towards improving public health the direction of the food chain must be reversed: plate-to-farm, not farm-to-plate. This considers the consumer first, not the producer or the processor, and is in-line with countries in the European Union and Australasia. Additionally, the artificial barrier between pre- and post-harvest is reconsidered, with food safety treated as a single entity. NP108 Food Safety is a single integrated National Program, however, for convenience and clarity, within the Action Plan the Program is subdivided into various components and subcomponents, including pre and post-harvest research issues.

Within the postharvest food safety program there are specific research problems that have direct relevance for individual parts of the food chain: these include, but should not be limited to the following: detection; [on-line] sensing systems; production and processing ecology; processing intervention strategies; omics; [safety] and health; risk [assessment]; pathogenicity; and [food] security. Some problems have a higher priority than others, while some, for example health issues and food security are new, and require additional funding to be implemented. The Action Plan is pragmatic, outlining the needs and subsequent outputs that can likely be achieved during the next 5 year cycle. The Action Plan is a working document, subject to realignment and modification when and where necessary.

Problem Statement 1.2.1: Detection and [Validation]

Detection and quantitation of pathogens, toxins and chemical contaminants are the central challenge to any food safety [and food security] system. Diagnostic tools must be developed for the entire food chain which allow the highest detection capability, and guarantee a maximum degree of prevention. Where practical however, detection as early as possible is best, thus avoiding or preventing the need for significant processing interventions or even the recall of food products from purchase endpoints. The term “diagnostic” implies analytical methods, and denotes the method/mechanism to establish epidemiological priorities.

Research Needs

Development of technologies and techniques is complicated, expensive and time consuming. Therefore collaborations with other research entities, both known, and those not previously considered, but who have the technology or capacity to supply or assist innovative ideas must be made. Most methods currently available were developed for a particular target, and are not intended to screen for multiple agents. The research program must be more forceful in having a systems approach, utilizing all its expertise. Pragmatically, not every method developed has, or will find utility. For regulatory use any developed methods must be validated in various food matrices, and where necessary refined. There is simply no point in developing any detection system or technique that will have no use in the real world. Validation of utility may require multidisciplinary, multi-location, and possibly multi-agency research efforts, in particular direct interaction with FSIS, FDA and EPA. The specific pathogens, toxins and chemical contaminants are

not articulated since food safety and food security priorities and needs shift with time. Decisions have to be made about what is really critical, especially in light of limited fiscal resources, time, rapidity etc.

Outputs: (developing in various sub-components for both food safety and security)

Sampling:

- Best practice sample collection protocols for various food systems and matrices.
- Innovative approaches to sample processing, for example universal separation and concentration steps.
- Mathematical algorithms to maximize the probability of detection.

Detection

- Rapid systems for target amplification (genotypic/phenotypic) to maximize detection potential.
- Multiple platform systems that will be combined with multiple target assays (multi-platform devices for universal contaminant detection).
- Systems that function in real or near real-time, or have the potential to be automated for high-throughput.

Validation

- Determined under both laboratory, pilot-processing, and commercial-processing conditions, and where appropriate in association with the specific stakeholder end-user, and/or development partner.
- The protocol for method validation for any specific pathogen, toxin or chemical contaminant detection will be different. However, the following generalities hold. The fundamental parameters for method validation will include evaluation of (1) sensitivity and selectivity; (2) accuracy, precision, recovery, and reproducibility; (3) stability and calibration curve (4) cost, robustness, and environmental impact.
- Measurements for each (pathogen/toxin/chemical contaminant) in the matrix will be validated. In addition, the stability of the (pathogen/toxin/chemical contaminants) in spiked samples will be determined.

Impact

ARS will lead in developing and validating methodologies that have both regulatory, industry and research use: a commonality of interests between government and stakeholders. Having reliable, cost-effective quantitative measurements of pathogen/toxicant in all food types can provide data to carry out risk assessment, to develop and validate predictive microbial models, and to identify areas where interventions are most critically needed. This information will assist the implementation of HACCP programs by FSIS, FDA, and their regulated industries.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; Eastern Regional Research Center, Wyndmoor, PA; Meat Animal Research Center, Clay Center, NE; and the Western Regional Research Center, Albany, CA.

Problem Statement 1.2.2: [On-line] Sensing Systems that Assist Processing, and Have Application in Food Security

With more focus on HACCP and HIMP (HACCP-based inspections models project) both the FSIS and FDA have placed more of the burden of inspection responsibility on the producers and processors. Plants are also responsible for meeting other consumer protection (OCP) issues as determined by regulatory agencies. In essence producers/processors assume the responsibility for inspection, and the regulatory agencies perform oversight and verification to ensure standards are met. Under HACCP, HIMP or GMP consumers demand safe, high quality food, however, consumer demand for more food increases the need for, and pressure on inspectors. Balancing consumer needs with the capabilities of the inspection agencies and the producers/processors will not be easy.

Research Needs

During the past 5 year research cycle the Program has been pre-emptive in devoting resources to developing automated, low cost, accurate, on-line and hand-held, computerized inspection [sensing] systems for poultry, beef and produce. These automated systems operate with minimum human intervention and are able to function despite changes in physical plant structure, and environmental conditions. The specific need is to continue development and validation where appropriate with the goal of commercial implementation asap. Future research with regulatory agencies, security agencies, and industry collaboration and support will focus on the following:

Outputs

- Commercial implementation for detecting pathophysiological abnormalities, and feces/ingesta in poultry combined and integrated with on-line washing/intervention systems developed by ARS.
- Development, evaluation, validation, refinement, and commercial or near-commercial implementation of computerized, on-line detection/sensing systems for whole beef and pork carcasses; feces and defects on apples and other fresh fruits and vegetables; physical hazards (bones, glass, plastic, stones and metal) in foods; and physical defects in shell eggs.
- Development, evaluation, validation, refinement and commercial implementation of head-gear systems; for use by small producer/processing operations. Expand their use to include other areas, for example: small and medium size processing facilities, cleaning and sanitation issues both in production and retail facilities.
- Development of innovative sensing capabilities for use in military and food security applications. Any work should specifically be coordinated with needs in the Food [Security] problem 1.2.9 of the Action Plan.

Impact

On-line, computerized sensing-systems placed or used strategically in food processing plants will assist and improve the regulatory and in-house inspection system; minimizing the problems of human error and variability; and increasing commercial processing productivity and profitability.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; and the Richard Russell Research Center, Athens, GA.

Problem Statement 1.2.3: Production and Processing Ecology

Elucidating the ecology of pathogens was one of the major thrusts in the previous Action Plan, and there is no doubt that understanding microbial ecosystems is extremely useful when developing intervention strategies to minimize contamination, to prevent the growth of pathogens, and kill or remove pathogens at various stages of production processing, transportation, marketing, and preparation for consumption. Such studies are also a means utilize and validate any new, improved and innovative methods to detect, differentiate, type, and quantify pathogens on some foods, and within some production and processing environments. Research also facilitates the identification of critical control points during food production and processing; and allows development of alternative HACCP/GMP systems.

Microbial ecology is perhaps however, the most contentious of the problems in the post-harvest food safety program. Not unexpectedly, such studies are often seen as collecting base-line data, which is not part of ARS' mandate. However, these studies are appropriate if for example, the data have a research purpose, and contributes to improved assessment of the public health risk posed by the pathogen, or to form and/or implement regulation(s).

Research Needs

The Program must build on previous ARS studies directed towards understanding for example, biofilms especially in processing plants; physiological status and survival in niches; attachment; quorum sensing; influence of environmental stresses; or processing or packaging technology. It is critical that such studies be integrated within other problem areas of the research program, specifically, detection, omics, pathogen modeling and food security. Collecting baseline data should be undertaken with extreme caution; and only after specific regulatory agency request. Studies would likely require direct collaboration with FSIS/FDA laboratories. New priority areas for baseline studies could include the following: plant environment sampling especially in blade or pin tenderiser equipment (specific FSIS need); expanding the hide and carcass mapping previously done in cattle to include swine and turkey carcasses (specific FSIS need); foods produced with minimal processing (RTE foods) or when prepared are subsequently associated with increased public health risk (deli-meats) (specific FSIS and FDA need); foods produced by new or alternate processing and/or packaging technologies; exotic species (alligator, beefalo ratites: specific FSIS need) ethnic food products (meats and produce); products produced

and/or processed by alternate methods (kosher, halal); and organic foods (USDA priority).

Outputs

- Determine why certain, and often very specific pathogens (types/subtypes) persist not only on food but in the food processing environment.
- Determine why certain types/subtypes are better adapted to production environments rather than the food processing environment, and vice versa.
- Determine correlations between specific genetic types or clonal groups and virulence characteristics; and any relationships between the virulence characteristics and human infections.
- Understand biofilm formation structure and composition both on foods and within processing, and transportation environments. Elucidate any differences between biofilms found on product, those found on equipment producing the product, and containers. Specifically addressing the formation and behavior of pathogens in biofilms associated with raw fruits and vegetables, both conventionally and organically produced; and elucidating the differences in biofilms on containers and equipment used in the produce industry.
- Determine the genes associated with altered physiological states, adaptation to extrinsic/intrinsic stresses, and quorum sensing.
- Provide baseline data to regulatory agencies to write Performance Standards.

Impact

Exposure of animals, seafood and produce to pathogens during production, slaughter and processing operations, and transportation can be a significant source of contamination. Research into the microbial ecology of pathogens on various surfaces is critically important in order to understanding the phylogenetic relationships between and within species, and how it may explain the variability of types/strains encountered in food, the environment and in humans, and the range of virulence characteristics expressed by the different strains under varying conditions and stresses. Studies will assist in understanding how biofilms are formed by elucidating the genes and physical/chemical structures on the surface of pathogens responsible for attachment and colonization. This will ultimately allow development of improved food production, processing, handling, distribution, and storage techniques, including identification of the factors that contribute to the spread of pathogens and techniques for the elimination of cross contamination

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; Eastern Regional Research Center, Wyndmoor, PA; Meat Animal Research Center, Clay Center, NE; Richard Russell Research Center, Athens, GA; and the Western Regional Research Center, Albany, CA.

Problem Statement 1.2.4: Processing Intervention Strategies

New/alternative food processing technologies all have the ability to inactivate microorganisms to varying degrees. However, the high treatment intensities required for inactivation usually result in adverse functional and/or sensory properties, significantly reducing food quality. Quality and safety are intimately associated, especially considering the change in consumer demands for more fresh (minimally processed) and natural food products. Apart from irradiation, all of the new technologies (used alone) are too costly, too energy expensive, and cannot guarantee safety to be of practical use. Irradiation has high potential, however, development and commercialization of irradiators (including electron beam) has been hampered by unfavorable public perception, despite endorsements by national and international organizations, and increased industry interest.

Research Needs

A research strategy for the future needs to be based on future trends (reverse food chain): consumer issues and concerns; traceability, regulatory needs; and current/future fiscal and personnel resources. Previously research has focused on commodities: poultry, produce, ready-to-eat foods etc., future research must address very specific needs with high impact. Therefore, the intervention program needs to be re-defined, to focus on utilizing and integrating all aspects of ARS food technology capabilities. Process engineering may play a larger role than before. However, the caveat is that some lines of research have limited applicability due to cost and practicality. Research involving processing technologies that are unlikely to be implemented by industry will have limited or no value and impact.

The need for more fresh (minimally processed) and natural food products could for example be achieved in part by revisiting the (under-utilized) hurdle effect concept which in and of itself is a minimal processing technology that exploits interactions between preservation treatments. Intelligent application of hurdle technology could readily be combined with current (and alternate) processing technologies. There also needs to be an increased effort to coordinate intervention(s) research with other ARS Units/Centers viz-a-via: CEMMI to address modelling and risk [assessment], and ERRC/RRRC food processing suites; and external to ARS with the FDA-National Center for Food Safety and Technology, Chicago, or University collaborators.

Specific needs articulated by both FSIS, FDA and commodity stakeholders include developing, and validating intervention strategies for specific foods and food types, and those useful for implementation in small and very small processing plants. Other critical areas of focus include the following: validating specific food cooking temperatures for food handlers at retail, water and water reuse, air-decontamination in processing facilities, shell egg processing, liquid egg processing, juices (in particular orange-juice), carbonated and fermented beverages, organic foods, and carcass dehairing. Specific processing technologies to be utilized would include, but not be limited to: thermal and non-thermal technologies such as radio-frequency (RF), electric field (EF), microwaves, vacuum-steam-vacuum (where appropriate), irradiation, high pressure, pulsed light, microfiltration, GRAS status chemicals, and competitive exclusion using both bacteria and bacteriophage.

Outputs

- Evaluate, develop, and validate through both laboratory, pilot-plant processing and commercial processing facilities the effect of single and combinations of intervention technologies (multi-target approach) on pathogen reduction. Ensure that lethality/intervention treatments do not negatively impact product quality.
- Determine whether combinations of non-thermal technologies can be incorporated in the hurdle concept; and determine whether single or combinations of non-thermal technologies are more effective if used in combination with traditional interventions.
- Increase fundamental understanding of the mechanisms, modes and sites of action at the cellular level of various intervention (inactivation) processes, and combination(s) thereof.
- Develop methods to prevent the growth of pathogenic and spoilage microorganisms in minimally preserved, brined, and fresh-cut foods. Develop predictive models for growth survival and inactivation of pathogens critical to Problem Statement 1.2.7.
- Utilize the inactivation data to model pathogen and non-pathogen behavior in complex food systems. These types of studies are fundamental to developing HACCP systems and regulations, and are critical to Problem Statement 1.2.7.
- Develop of post-harvest interventions options for small and very-small FSIS regulated plants.

Impact

Postharvest operations of all sizes (large to very small) provide an opportunity to remove or inactivate pathogens and their toxins acquired during the production and processing phases. Pathogens may develop resistance to antimicrobials from traditional measures used for pathogen control. Successful technologies and strategies to eliminate, reduce, or suppress human pathogens are needed for foods and food types associated with foodborne illnesses, or at risk of becoming vehicles for human pathogens. Development of individual or combinations for new or innovative intervention technologies for (minimal) processing will be developed based on an understanding their modes of action and effects on the microbial ecology of a food product; inadequate suppression of spoilage could create an opportunity for human pathogen growth and toxin production.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; Eastern Regional Research Center, Wyndmoor, PA; Meat Animal Research Center, Clay Center, NE; North Carolina State University, Raleigh, NC, Richard Russell Research Center, Athens, GA; and the Western Regional Research Center, Albany, CA.

Problem Statement 1.2.5: Omics

In 2001 the Program initiated an “omics” (genomics/proteomics) research effort to sequence and annotate the genomes, construct microarrays, conduct gene expression analysis, and construct data bases of several critically important food-borne bacterial pathogens. Although this research could be viewed in-part as a sub-component of Detection and Validation 1.2.1, the rewards of carrying out this work are far greater as are indicated in the Outputs. It is critically important that this work be done collaboratively where possible and appropriate by combining resources and expertise between different ARS Research Units, Federal agencies, ARS and University partners and internationally. While ARS may not have the capabilities to undertake complete genome sequencing, outsourcing of this task has proven to be very cost effective. The sequence data are directly provided to ARS for annotation, since we understand the biology of the organism. Lack of understanding of the organism’s biology by the sequencers was a critical and severely limiting problem with previous microbial genome analysis.

Research Needs

Initially the “omics’ research focus was directed towards food safety: the development of detection methods, phylogenetic analysis, and elucidation of the biology of the organism at the molecular level under advantageous and disadvantageous environments. However, future work will also include microbial forensics as part of both food safety and food security. This is a direct request from various Federal agencies. This specifically implies the directed use of information for the development of new assays and physical analysis methods, and for the identification of biocrime organisms.

Future initiatives progress from easier to more complex challenges and should be directed towards undertaking the following: Complete genome sequence analysis and annotation of 10-20 additional bacterial strains from specific genera/species, and at least 3-5 close relatives. This should also include the small viral genomes. Finished sequences should be the goal rather than draft. Developing single (genera or species) and multiple (Pan-genera and species) arrays for all critical food-borne pathogens. Developing protein microarrays: where possible, allowing for loss of sensitivity and discrimination due to conformation changes that occur during attachment. To meet this need multiple toxin microarrays may initially be of the most utility and value. Utilizing innovative (new technology) approaches to detect genera and species, and differentiate both gross and subtle changes in serotypes/strains. Establish a food-borne pathogen database based on nucleic acid, DNA/RNA profiles, proteomics and other phenotypic properties.

Outputs

- Improved science-based risk assessment decisions to control foodborne pathogens. In particular, allowing comparisons (molecular systematics) of strains, providing information on genes that contribute to pathogenicity in human and/or animal hosts
- Allow the study of expression of genes of interest, including, but not limited to genes involved in virulence and/or viability (growth and survival) in foods.

- Increase understanding of population genetics and epidemiology, by assisting development of improved ways of molecular tracking of pathogens through the food chain.
- Allow incorporation of sequence information into the design and optimization of nucleic acid arrays and chips.
- Facilitate gene expression studies for identification of sequences expressed or repressed in response to external cues such as those likely to be encountered by the specific bacterium in the environment, foods, and/or humans.
- Make possible more rapid and less costly diagnostic methods and vaccine development, and allowing analyses of resistance to antimicrobials, toxins, and disinfectants.
- Reveal any novel genes associated with specific ecological niches, and allow for comparative genomic analyses to determine critical areas for targeting strategies for interventions and controls.
- Identify regions of the genome that may have variations in the rate of nucleotide substitution or in the rate of intergenic recombination.

Impact

Genome and proteome analysis are critical to the development of rapid diagnostic tests/methods used by regulatory agencies, industry and research agencies. Methods development subsequently allows for the detection and enumeration of pathogens during processing and storage (microbial ecology and epidemiology) which leads to the development of predictive models and risk assessments, critical in assisting both regulatory agencies in making food safety decisions that impact public health, and industry for the development of HACCP plans. Microbial models and risk assessment, combined with genomics and proteomics will lead to the development of innovative intervention strategies, and therapeutic agents. These in turn will allow development of better HACCP plans which directly improve the safety of the food supply. Genomics and proteomics will also allow a greater understanding of beneficial genes and their products, which can be utilized for the development of new agriculturally based products. These products could be used for the development of alternate/innovative intervention strategies, particularly in ready-to-eat foods.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; Eastern Regional Research Center, Wyndmoor, PA; Meat Animal Research Center, Clay Center, NE; National Center Agricultural Utilization Research, Peoria, IL; Richard Russell Research Center, Athens, GA; and the Western Regional Research Center, Albany, CA.

Problem Statement 1.2.6: [Safety] and Health

Many health problems result from food safety events, and food borne pathogens may have a role in 2-3% of chronic diseases, if not more. Increasing consumer awareness of the link between diet and health has promoted the introduction of components such as non-pathogenic microorganisms used to produce fermented foods (probiotics) and certain

food compounds (prebiotics) to produce “functional foods”. These foods appear to benefit health through mitigating diseases. Countries within the European Union, Asia and Australia have significant research programs directed towards addressing health benefits of pre- and pro-biotics. Two years ago NP107/108 (post-harvest) began an inter-program collaboration on the role of probiotics in health using pigs as a model system. Research has proceeded rapidly, and the collaboration has developed into an excellent model of Cross-Location/Cross-National-Program projects. While this research has had a positive outcome and will continue with strong support from NPS, it is unlikely that in the foreseeable future there will be any significant new funds. Irrespective, of the funding issue, the problem will remain a component of the Action Plan, and the NPL will endeavour to obtain funds through external (international) collaborations.

Research Needs

Probiotics are a class of microorganisms that can establish and grow in a compartment in the host after consumption and provide some positive health benefit. Benefits claimed include protection against pathogens, stimulation of the gut immune system, correcting some bowel diseases, reduced allergic disease and protection against carcinogens. However, the basis of such claims is often confounded by a lack of demonstrable growth and function of the probiotic in the gut. Scientific validation of many of the claims of probiotics activity is missing. Consumer concern about probiotic reliability, efficacy and safety can be addressed by hypothesis-based and statistically validated testing of probiotics under controlled experimental conditions. Therefore, the need of this research is to test if dietary probiotics can enhance immune function against infectious agents, prevent the onset of allergenic disease, and improve the function of the GI tract.

Outputs

- Develop animal model(s) to test the unique nature of various probiotic strains and their efficacy against particular disease conditions will benefit the functional food industry.
- Provide information on the prophylactic use of dietary probiotics to enhance the mucosal immune system and to provide a more robust response to infection especially for neonates and the elderly.
- Provide a description of biomarkers that associate probiotic consumption with improved immune function would benefit human health.
- Provide practical recommendations for dietary changes to improve health status and reduce the cost of health care.
- Determining the efficacy of probiotic treatment of swine on resistance to infection will have inherent value to the swine industry for control of these infections.

Impact

Studies will provide information on the relationship between probiotics, diet, and immune function and identify biomarkers of nutritional status. Sound scientific evidence for probiotic efficacy will provide the food industry and regulatory agencies with relevant information for concerned consumers. Those at risk of chronic nutrition-related diseases will directly benefit. Research will also impact development of value-added food products, such as those containing probiotics or plant sterols. Other areas impacted will

be by increasing opportunities for initiating joint (international) programs in omics to develop useful bacterial strains for food use; and future development of probiotic strains to produce innovative foods with increased bioactive capabilities useful for improved food safety and [food] security.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; and the Eastern Regional Research Center, Wyndmoor, PA.

Problem Statement 1.2.7: Risk [Assessment]

ARS does not conduct risk assessment (RA) per se. The Program provides data, models, and expertise in order for the regulatory/action agencies to conduct their RA. ARS is recognized as a world leader in the development of food-related predictive models, and these models are an integral part of microbial risk assessment used to support food safety measures adopted by member countries of the WTO. Also, these models assist in identifying specific food processing steps, and associated Critical Limits that can serve as Critical Control Points in HACCP systems, as well as to facilitate food safety decisions when process deviations occur.

Research Needs

There are a number of diverse critical issues in RA that must be addressed in future ARS research. Broadly, they include research that expands exposure assessment, dose response, risk characterization, and overall Quantitative Microbial Risk Assessment (QMRA), including the development of more robust predictive and process risk models that specifically address hazards in complex food matrices. Specific outputs include, but are not limited to those listed below:

Outputs

- Determine the influence of the food matrix on the infective dose of a specific pathogen, and the subsequent biological response. Examine the uncertainties that surround dose-response models derived from animal studies, and determine which models more accurately function as surrogates for humans. Determine if dose-response data from epidemiological investigations are ultimately better.
- Develop and validate models that consider the inherent complexities of intrinsic and extrinsic parameters; or the macro- and/or micro-environment in food matrices. Determine the strategies needed to extrapolate model predictions to more complex foods.
- Determine and elucidate objective measures to assess model performance versus observation. Develop alternate responses or options, for example iterative approaches for specific food applications.
- Determine what strategies should be used to predict the distribution of lag times, and the worst-case scenario, since lag time has the highest uncertainty in predictive modelling.

- Develop data, and use these data for the development of predictive models for growth survival and inactivation of pathogens critical in minimally processed, brined, and fresh-cut produce.
- Determine the strategies for converting and integrating ARS models into fully functional process risk models.
- Compile quantitative food microbiology data into a database so that they can be exploited for model development and validation, especially when resources are limited.

Impact

Dose-response models underpin the establishment of Food Safety Objectives, and will assist public health by determining host responses to specific challenges. Research will address realistic processing and handling conditions that provide greater value to food industries, risk assessors, and risk managers. The validation of current models; and development and validation (including accuracy) of new models is critical since there are no universally accepted criteria for determining safe prediction zones. Models with the greatest value must couple quantitative microbial ecology with a better understanding of the physiological responses of microorganisms to stressors (interventions) used in food processing. We will address through creative approaches the behaviour of Select Agents in foods since they may not follow conventional pathogen growth and survival patterns. A quantitative database can be achieved through further expansion of Combase through CEMMI.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Eastern Regional Research Center, Wyndmoor, PA; and the North Carolina State University, Raleigh, NC.

Problem Statement 1.2.8: Pathogenicity

Microbial genomic efforts, for example, the availability of genome (and gene) sequences and microarrays have allowed us to obtain an increased appreciation and understanding of pathogenicity and virulence. We have already begun to compare different strains and species to determine phylogenetic relationships, especially in relation to ecological niche; understanding the role of intrinsic and extrinsic stress responses; and understanding the role of quorum sensing. To achieve the “Outputs” for these research needs will require all of the omics and ecology capability and expertise that both currently exists, is made available in the future. Being able to identify the “bad bugs” will require also the ability to do functional studies in models that are relevant to human disease. This will be a particularly difficult but exciting area for research and will require researchers and collaborators both nationally and internationally with special expertise in many aspects of microbiology related to pathogenesis and virulence (attachment, invasion, cell culture, biochemistry, molecular biology), not just expertise in food microbiology.

Research Needs

Pathogens have the capacity to readily and rapidly adapt and evolve. We must increase our understanding of the genetic mechanisms that make one strain of a microorganism pathogenic, while others within the same species are not; in essence, how do microorganisms become pathogenic? Virulence is the degree of pathogenicity, so why are some pathogens highly virulent while others are less virulent? How or is virulence directly related to the infective dose? What other factors impact virulence? Understanding pathogenicity and virulence are basic and critical issues, however, data are implicitly needed for pathogen control and risk assessment. Control strategies that were once effective may not remain so, hence they force us to change or develop new production processes and products to maintain and improve both food safety and food security. Good risk assessment(s) are predicated on understanding the pathogen, dose response and the behavior in foods

Outputs

- Isolation and characterization of virulence factors such as toxins, adhesions and invasions, and determination of how they interact, for example, do various factors behave synergistically or antagonistically?
- Utilize genomics and bioinformatics to classify pathogens based on specific virulence factors, rather than by name or by non-virulent associated phenotypic traits. This will improve the evaluation of safety currently focussed on organisms that have differential pathogenicity.
- Determine the ecological basis for predominance, persistence and succession along the food chain to prioritize points for intervention application. (See Problem Statement 1.2.3)
- Utilize genomics and bioinformatics information to understand why evolutionary shifts occur, and identify the selection pressures responsible for the shifts.
- Utilizing bioinformatics to make predictions, for example on protein structure and function.
- Determine whether quorum sensing occur in foods, or on surfaces (biofilms), and if so, whether it is factor in determining or regulating virulence in foodborne pathogens
- Determine through analysis of gene/protein expression profiles the responses to intrinsic and extrinsic stressors imposed during production, processing and storage.
- Determine if stress-regulatory-response-systems may also be targets for the design of innovative antimicrobial agents.
- Though understanding virulence, develop improved detection and identification methods that enable differentiation of virulent from non-virulent.
- Initiate studies through collaborations with other Federal agencies, affiliated research establishments and Universities to determine the infectious dose of various pathogens and their toxins, in various food matrices, both pre and post stress.

Impact

Sequencing and annotating pathogen genomes will provide the ability to understand the nature and differences between pathogens and non-pathogens, and identify factors that encode for variations in virulence. Physiological characteristics of pathogens are known to be affected by stress. This work will determine the effect on virulence mechanisms which directly impacts pathogenicity and the emergence of new pathogen types. If virulence is correlated with quorum sensing this could potentially be a mechanism to disrupt or negate pathogenicity by targeting communication pathways with novel antimicrobials. Research will allow better risk assessment and allow more efficient allocation of risk management resources focussing on only the critical pathogens in the food supply.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; Eastern Regional Research Center, Wyndmoor, PA; Meat Animal Research Center, Clay Center, NE; National Center Agricultural Utilization Research, Peoria, IL; Richard Russell Research Center, Athens, GA; and the Western Regional Research Center, Albany, CA.

Problem Statement 1.2.9: [Food] Security

To enhance the effectiveness of food security a layered approach must be taken. Three areas need to be addressed: detection (and validation); prevention, and response and recovery, not all of which the NP108 Program can assist with. At present the Program can and will only focus on detection and prevention, with detection technologies and their validation in food systems being the principal directive. Detection of intentionally added biological and/or chemical agents is central to [food] security. ARS's contribution with its partners/collaborators (both government and industry) will be very high. Currently we have technologies that can detect many pathogens, toxins and other food (residue) contaminants. However, the breadth of these technologies must be expanded both in scope and limits of detection. Prevention is a proactive response, relying upon a high degree of intuition and awareness within food safety practices and procedures to prevent an event. The current evaluation of high risk foods (Food Shields) is a first-start, however, all individual HACCP programs, and critical points within the various food production, processing and transportation pathways must be re-evaluated, and points of concern identified and made invulnerable.

Research Needs

Currently, there are no validated methods for the detection of Select Agents in food; and there are no deployable and rapid biological detection capabilities for food and water. These needs were directed to ARS from several Federal Departments/Agencies and Food Security Committees, and include but are not limited to the following:

- New approaches to sampling protocols to maximize the probability of detecting intentional contamination.

- Innovative approaches to sample processing such as universal separation and concentration steps; automating systems for high-throughput.
- Rapid systems for target amplification to maximize detection potential allied to multi-platform devices for universal contaminant detection, and where possible develop robust systems that function in real or near real-time.
- Validating technologies to detect Select Agents in various food matrices in both in pilot plant and real world situations; conduct simulation and validation tests. Although assays for many Select Agents exist for clinical or environmental samples, few exist that are validated for use in the food matrices of concern.
- Elucidating if and how Select Agents are affected by intervention strategies in various food matrices, including their viability after-processing.
- Technologies to deactivate/neutralize of Select Agent hazards in food, and studies to determine the effect of these processes on food quality.
- Decontamination technologies to safely clean food processing, distribution, and storage operations contaminated with select agents.

Other potential research areas (based on needs articulated by Stakeholders) that could be incorporated into the Action Plan at a later date.

- With other government agencies and industry produce a best practices handbook which establishes standards and requirements for a secure food supply chain to reduce the probability, severity and extent of an event.
- Biosensors (tagging methods) for real-time integrated tracking of product that can be monitored through geospatial information systems (GIS).
- Infective dose studies of biological toxins in various food matrices. What is effect of pathogen and toxin combinations; examining synergy, and changes in lethal dose level? Examine the effects and markers of toxicological responses in model systems.

Outputs

- Develop best practice sample collection and analysis system for foods of concern.
- Validate methods and diagnostics (nucleic acid based, serological, analytical assays) specifically developed for the detection of select agents (biological agents, toxins, chemical residues and pesticides) in various food matrices.
- Develop systems and models that validate the behavior of pathogen in foods, and the effect of various intervention strategies on select agents.
- Identify, develop and validate decontamination systems for various food commodities, with particular reference to quantity, level and type of contamination. Develop simulation models for strategies of containment, decontamination and disposal of both of the affected food; the production and processing facilities, within the distribution system, and environmental fate, persistence and impact of the select agent.

Impact

Agencies within the Federal government have begun to implement strategic action plans to further ensure the safety of the nations food supply. It is apparent that even if a small number of contaminants were intentionally introduced into some part of the food chain, such an action would have the potential to seriously damage public confidence. Apart from the impact on health and safety, bioterrorism against the food supply would also directly harm the nation's economy. U.S. agriculture employs nearly one quarter of the workforce, and annually contributes over one trillion dollars to the gross domestic product. The food production industry annually exceeds two hundred billion, with exports over fifty-five billion.

There are several critical needs in order to implement a comprehensive food security strategy. The ARS Food Safety program's role is to establish methods to protect food that has been identified as at-risk; strengthen and expand laboratory preparedness; and to develop rapid and confirmatory laboratory methods to analyze suspect foods for Select Agents, toxins and chemical contaminants.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; Eastern Regional Research Center, Wyndmoor, PA; Meat Animal Research Center, Clay Center, NE; National Center Agricultural Utilization Research, Peoria, IL; North Carolina State University, Raleigh, NC; Richard Russell Research Center, Athens, GA; and the Western Regional Research Center, Albany, CA.

Component 2. Mycotoxins and Plant Toxins

Food products of plant origin may contain mycotoxins as a result of fungal growth/toxin production in/on the plant prior to harvest, as well as during transportation and storage. Intrinsic plant metabolites that are toxic to mammals and heavy metals from soil uptake can also be transmitted to food products. Contamination may make the plant commodity itself unacceptable for human and animal food, or it may result in residues of the toxins in food products of animal origin, primarily milk. The potential for the presence of mycotoxins in commodities is a threat to competitiveness of US agriculture in the world export market, where limits of acceptability are generally more stringent. Furthermore, the potential presence of mycotoxins in the human food supply is detrimental to the public perception of a safe and healthy food supply.

Mycotoxins which are diverse chemically, are considered to be secondary fungal metabolites: that is having a role other than for primary energy and cell structure purposes. Mycotoxins have been thought to perhaps act as defence mechanisms, but their precise function(s) have never been elucidated. Evidence is now beginning to accumulate that at least one mycotoxin may affect the cellular redox homeostasis through action on

the mitochondrial antioxidative stress system as a metabolic target. Other physiological targets will undoubtedly be pinpointed for other toxins (they are not toxins as far as the producing fungi are concerned). Each fungus is adapted to grow well in certain crops and in certain ecological niches thus they have particular climatic optima, resulting in explosive growth and fungal production with favorable weather conditions.

The mycotoxins of most concern are aflatoxin, fumonisins, vomitoxin and ochratoxin, one or more of which may contaminate peanuts, corn, cottonseed, tree nuts and/or wheat. Fungi, particularly endophytic fungi with a very close relationship to the host plant, may produce metabolites that are essential to plant survival under stressed conditions, and at the same time are toxic to insect or mammalian herbivores. The mutualistic symbiosis of endophyte and grasses is beneficial to the host plant through insect and nematode resistance, drought tolerance, and improved competition with other plant species. Examples are the lolines and ergopeptine alkaloids in tall fescue.

Direct translocation of unsafe levels of chemicals from the soil on which they are grown, e.g., heavy metals and nitrates, or deposition by air currents, are other mechanisms that can result in toxins in edible plants. Toxic compounds may also be formed by the plant itself for fulfilling some physiological need, or as a defense mechanism, for example, against herbivores, both insect and mammalian. When plants cannot make their own necessary protective chemicals the need may be met by endophytic fungi as explained above. In either case the compounds can be toxic to insects, nematodes and/or mammals consuming the plant. In developing genetic solutions we must be careful to not inadvertently enhance factors which may be responsible for pest resistance in plants, and which simultaneously may have unanticipated consequences in animals and humans. (Residues of pesticides are not considered in the Food Safety National Program.)

Problem Statement 2.1.1: Toxin Methodology and Identification

Rapid and affordable chemical toxin identification and quantitation are the basis of both industry and regulatory food safety assurance activities, and they are now of vital importance to homeland security. In addition because mycotoxins are unevenly distributed in contaminated plant commodities, both regulatory agencies and the producing industries need methodologies to accurately determine the contamination of the entire tested lot, which may be thousands of tons, as in a loaded river barge.

Research Needs

Non-destructive, rapid, accurate, sensitive and affordable detection methods are needed. The most useful and effective methods are non invasive or require only a minimum of sample preparation, and preferably provide a quantitative assessment of the amounts of mycotoxins or plant toxins present on the food commodity. In addition, they must be compatible with today's high speed, high volume commerce. Possible methods include near infra red and multi spectral analyses, as well as the more traditional screening methods, that are rapid, sensitive, easy to perform and quickly learned, e.g., fluorescence polarization immunoassays, ELISAs, etc. To develop the necessary reagents for these

new assays we may need to use genetic components, computational chemistry approaches and novel synthetic chemistries.

To be truly effective in a high volume industrial operation sorting of a commodity with a spectral analysis for mycotoxin contamination must be combined with recognition and removal, such as a high volume optical sorter. Such a machine would in a single pass, simultaneously sort and reject mycotoxin-containing kernels from contaminated grain. Use of this technology to examine all product could overcome the very serious problem of sampling commodities with unequal distribution of toxins. Although more work will be necessary to improve the accuracy and specificity of NIR to detect very low amounts of mycotoxin contamination, it is an important first step in developing a useful online technology.

Outputs

- Rapid, accurate, sensitive and potentially affordable detection methods that are compatible with high speed, high volume commerce, that can be linked to optical sorters, and that are preferably non-destructive.
- Spectral analysis of contaminants to identify key optical signals that can be used for identification.
- Expand the use of genetic based determinants in PCR based tests including microarrays to rapidly identify mycotoxigenic fungi in contaminated food products.
- Novel mycotoxin-binding materials such as molecularly imprinted polymers, novel carbohydrates, and improved antibodies that bind mycotoxins.

Impact

Protection of both the human food supply and animal feeds requires the rapid detection of toxins, and in particular mycotoxins, in commodities, foods, and feeds. This includes both the commodities with which they are usually associated as well as those in which they have not traditionally been found, the latter as a tool in support of enhanced food security. ARS will lead in developing and validating methodologies and rapid screening assays including on-line methods that have regulatory, industry and research uses for solving mycotoxin contamination problems. Detection of toxins is the essential first step in the proper identification of contaminated materials, and ARS methods will aid, where necessary, the proper diversion of such materials to alternative uses. Rapid ARS methods will aid both today's high speed high volume commerce and the regulatory agencies. Fortunately, there is a commonality of interests among these groups which allow similar methods to be used by each. Reliable, cost effective identification methods for toxins in food animal tissues and environmental samples will provide data to help resolve food security concerns, to carry out risk assessments, to develop and validate predictive microbial models, and to identify areas where interventions are most critically needed. This information will assist FSIS, FDA and the food crops industries in continually assuring production and delivery of safe food products

Resources

ARS scientists assigned to projects under this Problem Statement are located at National Center for Agricultural Utilization Research, Peoria, IL; Western Regional Research Center, Albany, CA.; Southern Regional Research Center, New Orleans, LA; and the Richard Russell Research Center, Athens, GA.

Problem Statement 2.1.2: Crop/Fungal/Insect/Toxin Relationships

A thorough understanding of the very complex fungal/insect/crop/environment interactions during both fungal and plant growth and maturation is necessary to develop effective strategies to prevent toxin accumulation, whether of fungal or plant origin, in crop plants. We need to elucidate factors affecting, either positively or negatively, the interaction between soil and crops to identify interventions, such as, avoidance of certain soil series, use of soil amendments, crop production practices and/or development of crop varieties that do not absorb the toxic trace elements

Research Needs

In many cases mycotoxin producing fungi are introduced to the food crop by insects. Control of these insects can offer a practical avenue for controlling fungi on the crop - unless of course the weather pattern is so favorable for fungal growth and toxin formation that no method can be successful. As an example, Bt-resistant corn has significantly reduced levels of mycotoxins compared to non-Bt corn; but the degree of benefit is highly dependent on the timing and makeup of the insect pest complex in the particular crop year. Response to specific pheromones and kairomones by insect pests of tree nuts can also provide additional means for controlling specific fungus carrying insects.

Outputs

- Determining the effects of weather on toxin accumulation.
- Identify and develop commercially practical synthesis for specific pheromones and kairomones that can help control insect pests of tree nuts
- Developing mechanisms and products for preventing fungal introduction by insects.
- Determination of the sources and mechanisms of plant accumulation of toxic amounts of heavy metals or other toxins in food crop plants.

Impact

The thorough understanding gained through this research of the very complex fungal/insect/crop/environment interactions present during both fungal and plant growth and maturation will result in effective new strategies to prevent toxin accumulation, whether of fungal or plant origin, in crop plants. Knowledge of the biology of the production systems will provide data to help construct predictive systems, and to determine under what conditions is it economical to implement specific management strategies, particularly for insect control. Crops will be able to be produced in areas where it is not otherwise economical or even possible to do so. Having this thorough

understanding of crop/insect/ toxin/environmental relationships will help prevent food security concerns, carry out risk assessments, develop and validate predictive microbial models, and identify areas where interventions are most critically needed. This information will assist FSIS, FDA and the food crops industries in the continued production and delivery of safe food products

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; National Center for Agricultural Utilization Research, Peoria, IL; Western Regional Research Center, Albany, CA.; Southern Regional Research Center, New Orleans, LA; National Peanut Research Laboratory, Dawson, GA; and Richard Russell Research Center, Athens, GA

Problem Statement 2.1.3: Production Practices and Expert Systems

Optimal cultural and crop handling practices facilitated by expert systems, where they are available, can help decrease mycotoxin accumulation. Optimized practices are generally the least expensive methods for controlling aflatoxins. For instance, ARS research has demonstrated that the rehydration procedure used to facilitate cracking of closed-shell pistachios results in exceptionally high aflatoxin levels. In another crop ARS showed that the second phase of contamination of cottonseed, that is, between boll opening and harvest, is the most important factor predisposing the crop to aflatoxin contamination, rather than the earlier phase as was traditionally emphasized. This knowledge helps cotton breeders target events in plant growth and development cycle for reducing aflatoxin susceptibility of their crop. For corn ARS developed data and formulated a computer program to give useful predictions for mycotoxin occurrence in most corn hybrids in most years in the Midwest.

Research Needs

We need various good agricultural and handling practices to provide valuable comprehensive management information and tools for producers in order to help assure the lowest possible toxin levels in food crops. We also need to gain an understanding of the role of crop management practices, including use of rotation crops, on the ecology of mycotoxigenic fungi contamination in order to optimize the application of competitive exclusion products.

Outputs

- Identification of cultural crop production and handling practices that can assist in the reduction of mycotoxins in crops. This includes effects of various management strategies, e.g., rotations, tillage, and cover crops, and herbicide-resistant crops under different weather conditions, and insect control
- Formulation of user-friendly computer programs to provide useful predictions for mycotoxin occurrence.

Impact

We will be able to predict when mycotoxin problems may occur, and to determine under what conditions it is economical to implement specific management strategies, particularly for insect control. We will have effective strategies to prevent toxin accumulation, whether of fungal or plant origin, in crop plants. Corn will be less likely to exceed FDA tolerances and guidance levels for mycotoxin levels each year, and we will know what corn lines and treatments have insect resistance and thus tend to have reduced mycotoxin levels. This will result in corn more often being a viable economic crop for the south, especially in rotation with cotton and soybeans. This information regarding optimum production practices and the ability to predict when contamination may occur in the field will help develop and validate predictive microbial models and risk assessments, and will help prevent food security concerns. The information will assist FSIS, FDA and the food animal industry in the continued secure production and delivery of safe food products

Resources

ARS scientists assigned to projects under this Problem Statement are located at National Center for Agricultural Utilization Research, Peoria, IL; Western Regional Research Center, Albany, CA.; and Crop Genetics and Production Research, Stoneville, MS.

Problem Statement 2.1.4: Breeding Resistant Crops

Mycotoxin contamination of food crops cannot be completely prevented by improved cultural practices, insect control, and even competitive exclusion products. To economically produce crops that meet regulatory guidelines we need intrinsic crop resistance to colonization by the fungus and subsequent production of toxins. A thorough understanding and utilization of fungal genomics, including functional genomics offers an unprecedented opportunity to affect mycotoxin control through resistant crops. This genomic information can facilitate identification/ characterization of the complex set of genes involved in fungal virulence, and the signalling pathways between the fungus and the environment, and fungal reproduction/survival, all of which must be understood if fungal infection and mycotoxin production in crops is to be prevented.

A significant early development in this area is the determination that the effective aflatoxin resistance in the Tulare variety of walnut is due to high levels of hydrolyzable tannins in the seed coat that persist throughout the growing season, in contrast to other varieties. Fungal tannase hydrolyzes tannins to form gallic acid, which affects fungal response to oxidative stress and is a potent inhibitor of aflatoxin synthesis. This information explains the basis of a naturally occurring antifungal/antimycotoxin mechanism and points to the possibility of prevention of aflatoxin biosynthesis by disruption of oxidative stress response pathways. In addition proteomics has been used to identify several fungal resistance-related characteristics and stress-responsive proteins/genes in resistant lines of corn.

Research Needs

Both genomic and proteomic information are needed to guide traditional breeding, marker assisted selection and/or genetic engineering to develop aflatoxin-resistant varieties of crops. We particularly need molecular biological approaches to discover, introduce and evaluate plant-derived selectable markers for transgenic plant production.

Outputs

- Identification of unique fungal genes for specific biological and physiological functions for use in highly sensitive PCR based tests (including microarrays).
- Completion of the identity of the gene clusters and biochemical pathways required for the production of common mycotoxins.
- An understanding of how environmental factors affect the fungus, which genes are turned on during the plant-fungus interaction and mycotoxin production, and which are essential to fungal survival in the field environment.
- An understanding of the effect of the plant on the growth and sporulation of the fungi and on mycotoxin production under various crop production and stress conditions.
- Use of identified genes for marker assisted selection of corn resistant to mycotoxins.

Impact

Knowledge of fungal genomics will provide the basis to bring about mycotoxin control through resistant crops. Fungal genomics can help elucidate the dynamics of the fungal-crop relationship, including the role and mechanism of action of secondary metabolites in pathogenesis to the host plant, and the mechanism of transmission from one plant to the next. Fungal genomic information can help identify both the genes in the biochemical pathways that lead to mycotoxin formation, and the regulatory elements of mycotoxin production. Genomic information will also provide identification/ characterization of the complex set of genes involved in fungal virulence, and the signalling pathways between the fungus and the environment, and fungal reproduction/survival which will help prevent fungal infection and mycotoxin production in crops. This genetic information in turn will identify effective control strategies for mycotoxins during crop production, and, set the stage for providing farmers with crop varieties that will not support fungal growth and toxin production. A lower amount of crop will need to be discarded because it does not meet regulatory guidance levels. Thus, the results of this research will assist the food commodity industries in the secure production and delivery of safe food products with lower mycotoxin levels.

Resources

ARS scientists assigned to projects under this Problem Statement are located at National Center for Agricultural Utilization Research, Peoria, IL; Western Regional Research Center, Albany, CA.; Southern Regional Research Center, New Orleans, LA; Crop Genetics and Production Research, Stoneville, MS; and Richard Russell Research Center, Athens, GA

Problem Statement 2.1.5: Biocontrol Technologies

Biocontrol is the use of living organisms to control pests. The correct selection of organisms can provide a highly selective tool for controlling the pest of concern with minimal adverse effects on the environment. The simplest systems are where the organisms are amplified in the laboratory and then applied to the field; just as effective, and certainly more elegant and less expensive, are biocontrol organisms that amplify in the field to the necessary numbers to provide effective crop protection. Research to date has been highly successful in developing biocontrol strain technologies that use competitive exclusion to prevent aflatoxin in cotton and peanuts, and some promising trials have been carried out with corn and tree nuts. Biocontrol strains of organisms may be either bacteria or fungi, and they may be endophytic or confined to the outside of the plant.

Research Needs

New and improved biocontrol agents must be identified and developed for continually recognized new crop/fungal control opportunities. We need to understand the ecological relationship of the biocontrol fungi to other plant microorganisms, soil microorganisms, and the treated crops and the environment to help assure optimal delivery methods and timing. It is particularly important that ARS partner with commodity-producing industries, state boards or other development authorities, and/or crop protection industries to complete the development of promising biocontrol products. ARS must provide substantial assistance in interpreting the data and making it available to EPA in an understandable form in order to obtain the necessary, but often tedious, regulatory approvals. It may also be necessary to develop commercial scale production systems, including simple strain identification, starter culture procedures, scale-up procedures, quality control procedures and methods to assess efficacy for fungal biological control agents. Only following a prodigious amount of often frustrating work can products be made available to commodity producers.

Outputs

- New or modified effective biocontrol organisms and delivery systems that do not introduce toxic factors.
- Establishment of baseline levels of the atoxigenic biological control organisms to provide a basis for determining the influence of applied biocontrol products on natural microbial communities, particularly when applied over large crop acreages.
- Further delineation of the role of endophytic fungi in regulating plant metabolism and in providing effective defense against predators and stresses.
- Determination of the effect of control strain distribution on the ecology of the producing area.
- Safety, efficacy, and stability data necessary for maintenance and expansion of biopesticide registrations with EPA.

Impact

Effective and economical biocontrol strains that are approved by regulatory agencies and readily available to the producing industries, i.e., peanuts, corn, cottonseed and tree nuts

will provide control of aflatoxin in most years under most environmental conditions. Although there is occasionally a year with environmental conditions favourable to toxin production such that no single intervention can reduce losses from mycotoxin contamination, long-term use of competitive exclusion products will assist producers sufficiently that there is financial incentive to continue crop production and stay in business. The results of this research will thus assist the food commodity industries in the continued secure production and delivery of safe food products with lower mycotoxin levels.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Western Regional Research Center, Albany, CA.; Southern Regional Research Center, New Orleans, LA; Crop Genetics and Production Research, Stoneville, MS; National Peanut Research Laboratory, Dawson, GA; and Richard Russell Research Center, Athens, GA

Problem Statement 2.1.6: Toxicity Evaluations and Mechanism of Action

FDA has established guidelines for several mycotoxins in the US, but their toxicity continues to be assessed by the world community. Moreover new research including human epidemiology studies continually raises new questions of cause and effect links to human disease. There are also questions about the effectiveness of certain industrial crop handling and food product preparation procedures for irreversibly reducing mycotoxins. In addition the presence of heavy metals and plant toxins may be strictly regulated by the FDA, and also by the EPA, when there is evidence that they may occur in drinking water.

Research Needs

Critical data for science-based risk assessments world-wide are needed as the basis for regulatory decisions to protect consumers; and thus continued assessment of certain aspects of mammalian toxicity is necessary. Production of crops in developing countries is faced with constraints, e.g., severe drought without water for irrigation, that are of lesser importance in the U.S. Information is needed for the FDA and WHO to make solid recommendations regarding safe levels of mycotoxins in food. Toxicity of chemicals may be strongly affected by dietary constituents, and specific relationships need to be elucidated and recognized. We need to correlate the bases of identification tests for certain regulated chemicals with the active moiety of these chemicals as confirmed by tests of mammalian toxicity.

Outputs

- Chemical speciation of toxins following ingestion and elucidation of their specific biological effects with relevant dietary components.
- Mechanism of action based bioassays for 'mycotoxin - like' activity in foods and other matrices.

- Chemical analyses that accurately predict the biological activity of toxins as they are found in prepared food products, and assistance in the development of production procedures to achieve safe levels.
- Specific molecular endpoints of toxins in mammalian systems and downstream mechanisms of action using *in vivo* systems, plant models, and whole animal test systems; and validation of toxicological endpoints in whole animal feeding studies.

Impact

This research will provide information for the FDA and WHO to make solid recommendations with a scientific basis regarding safe levels of mycotoxins in food. The producing industries and the public will more likely accept regulatory decisions when they know and understand the basis of the regulatory guidance. Having a solid basis for identification tests for the active moiety of mycotoxins and plant toxins will assist in the identification and confirmation by the regulatory agencies of suspect food contamination, either naturally occurring or by bioterrorists. This will help assure food security, help consumers accept regulator's statement and believe in the safety of their food supply, and thus maintain healthy eating habits.

Resources

ARS scientists assigned to projects under this Problem Statement are located at Beltsville Agricultural Research Center, Beltsville, MD; and Richard Russell Research Center, Athens, GA.