V(A). Planned Program (Summary)

Program # 12

1. Name of the Planned Program

Food Safety - Agricultural Biosecurity

☑ Reporting on this Program

V(B). Program Knowledge Area(s)

1. Program Knowledge Areas and Percentage

KA Code	Knowledge Area	%1862 Extension	%1890 Extension	%1862 Research	%1890 Research
211	Insects, Mites, and Other Arthropods Affecting Plants	13%		5%	
212	Pathogens and Nematodes Affecting Plants	12%		50%	
213	Weeds Affecting Plants	10%		0%	
712	Protect Food from Contamination by Pathogenic Microorganisms, Parasites, and Naturally Occurring Toxins	10%		35%	
903	Communication, Education, and Information Delivery	55%		10%	
	Total	100%		100%	

V(C). Planned Program (Inputs)

1. Actual amount of FTE/SYs expended this Program

Year: 2012	Extension		Research	
	1862	1890	1862	1890
Plan	1.0	0.0	3.0	0.0
Actual Paid Professional	0.3	0.0	3.0	0.0
Actual Volunteer	0.0	0.0	0.0	0.0

2. Actual dollars expended in this Program (includes Carryover Funds from previous years)

Extension		Research	
Smith-Lever 3b & 3c	**1890 Extension**	**Hatch**	**Evans-Allen**
15000	0	123307	0
1862 Matching	**1890 Matching**	**1862 Matching**	**1890 Matching**
15000	0	123307	0
1862 All Other	**1890 All Other**	**1862 All Other**	**1890 All Other**
50000	0	682079	0

V(D). Planned Program (Activity)

1. Brief description of the Activity

*Establish the **Oklahoma National Institute for Microbial Forensics and Agricultural Biosecurity**, a multi-disciplinary unit to support and address issues of crop and food biosecurity, and their impacts.

*Conduct **scientific research** targeted specifically towards plant pathogen forensics, sociological impacts of terrorism, and other areas of agricultural biosecurity

***Develop an academic "track"** for students seeking M.S. or Ph.D. degrees in established programs such as Plant Pathology, Biochemistry, Plant Sciences or Forensic Sciences, who seek plant pathogen forensics

Offer a short course on microbial forensics to prepare State educators, diagnosticians, researchers, extension agents, students and postdocs, producers and first detectors/responders

Develop an **undergraduate course in Agricultural Biosecurity**

2. Brief description of the target audience

Key members of National and Oklahoma homeland security community (DHS, FBI, CIA, etc)
Key members of National and Oklahoma agricultural leaders and representatives
Oklahoma extension personnel
Master gardeners
Oklahoma producers and crop consultants
OSU students and faculty
Professional/scientific societies
Key industries
The public

3. How was eXtension used?

eXtension was not used in this program

V(E). Planned Program (Outputs)

1. Standard output measures

2012	Direct Contacts Adults	Indirect Contacts Adults	Direct Contacts Youth	Indirect Contacts Youth
Actual	500	100	45	0

2. Number of Patent Applications Submitted (Standard Research Output)

Patent Applications Submitted

Year: 2012

Actual: 0

Patents listed

3. Publications (Standard General Output Measure)

Number of Peer Reviewed Publications

2012	Extension	Research	Total
Actual	0	17	17

V(F). State Defined Outputs

Output Target

Output #1

Output Measure

- Number of OSU faculty and staff affiliated with the new Oklahoma Center for Agricultural Microbial Forensics Biosecurity

Year	Actual
2012	40

Output #2

Output Measure

- Number of grant/contract proposals submitted in agricultural microbial forensics and biosecurity

Year	Actual
2012	40

Output #3

Output Measure

- Number of journal articles submitted with emphasis on agricultural microbial forensics and biosecurity

Year	Actual
2012	17

Output #4

Output Measure

- Workshops to develop the discipline of plant pathogen forensics, train "first responders", and state and national stakeholders

Year	Actual
2012	3

V(G). State Defined Outcomes

V. State Defined Outcomes Table of Content

O. No.	OUTCOME NAME
1	Number of invitations to agricultural biosecurity team members for participation in initiatives, programs, presentations, and consultations related to agricultural biosecurity and microbial forensics
2	Number of forensics-relevant journal articles published
3	Percentage of agricultural producers, handlers and processors employing at least one new (to them)practice to enhance biosecurity

Outcome #1

1. Outcome Measures

Number of invitations to agricultural biosecurity team members for participation in initiatives, programs, presentations, and consultations related to agricultural biosecurity and microbial forensics

2. Associated Institution Types

- 1862 Research

3a. Outcome Type:

Change in Condition Outcome Measure

3b. Quantitative Outcome

Year	Actual
2012	16

3c. Qualitative Outcome or Impact Statement

Issue (Who cares and Why)
A biological attack on United States crops, rangelands or forests could have severe impacts. Biocrimes, perpetrated for economic gain, are even more likely. Preparedness requires a strong national security plan that encompasses microbial forensics and criminal attribution. However, U.S. crop producers, consultants and agricultural scientists, unaccustomed to the possibility of intentional pathogen introduction, traditionally focus disease management strategies on prevention, rapid eradication or long-term management. New information, technologies and resources in microbial forensics (human, livestock and plant) are needed to enhance the nation?s preparedness and responsiveness to plant health emergencies.

What has been done
Scientists make presentations to technical groups to advise of research advances and as educational programs.

Results
(1) OSU researchers developed and automated a coding method that captures the results of the commonly used microbial DNA fingerprinting method, Amplified Fragment Length Polymorphism (AFLP), in a portable format that is both reliable and highly discriminatory. The method will be of value for the attribution of pathogenic organisms that might be used in a biocrime.
(2) Bioinformatics research focuses on dealing with large-scale sequencing data, developing novel algorithms and converting them into useful knowledge and products. NIMFFAB bioinformaticists developed two prediction systems for the community. AP-iNET (http://bioinfo.okstate.edu/AP-iNET/) is useful for predicting genome-wide host-pathogen interactions in the model system Arabidopsis-Pseudomonas syringae. This system will be extended to agricultural host-pathogen interaction systems to support the improvement of agricultural productivity, e.g. by reducing yield losses due to pathogens. Another tool, LigPred

is useful in the discovery of novel biomass degrading enzymes. This system could identify and classify "unknown" lignin related enzymes into any of the 50 functional classes of lignin based on EC number. LigPred, which works on full length as well as metagenomic samples, is already actively used by researchers and bioenergy community worldwide.
(3) A new polymerase chain reaction (PCR) protocol for specific detection of a fungal plant pathogen, Fusarium proliferatum, was developed and validated to forensic standards for delivery to the National Bioforensic Analysis Center, Department of Homeland Security.
(4) Bioinformatic pipelines were created for use on mock sample genomic databases to simulate nucleic acid sequencing (454 format) of plant pathogens. Electronic probes (e-probes) were designed and used for BLAST database searches. They were validated with sequencing data sets containing genome sequences of plant pathogenic viruses, bacteria and fungi. Representatives of all pathogen groups were detected in metagenomic samples containing mixed microbial and plant sequences when the pathogen reads were 1.34% or greater.

4. Associated Knowledge Areas

KA Code	Knowledge Area
212	Pathogens and Nematodes Affecting Plants
712	Protect Food from Contamination by Pathogenic Microorganisms, Parasites, and Naturally Occurring Toxins
903	Communication, Education, and Information Delivery

Outcome #2

1. Outcome Measures

Number of forensics-relevant journal articles published

2. Associated Institution Types

- 1862 Research

3a. Outcome Type:

Change in Knowledge Outcome Measure

3b. Quantitative Outcome

Year	Actual
2012	5

3c. Qualitative Outcome or Impact Statement

Issue (Who cares and Why)
Important national research and policy initiatives in homeland security have engendered a need for additional personnel, trained and experienced in fields such as microbial forensics and food safety. A new national priority for enhanced food safety requires novel diagnostics, advanced

traceback capabilities in the event of foodborne disease outbreaks, and mitigation strategies has been set. Federal agencies, such as FDA, CDC, USDA, and private fresh produce industries are all impacted.

What has been done

Results

(1) Greater national attention is now directed towards understanding routes of contamination of fresh produce, including wildlife and insects. In efforts to understand the factors involved in vector competence of human pathogens, OSU researchers compared transmissibility of two human foodborne pathogens, E. coli O157:H7 and Salmonella enterica, using two filth fly species and determined that E. coli was transmitted 10-15 x the level of Salmonella. They also observed bacteria like organisms on 100 percent of the tarsi and labellae of blow flies exposed to both Salmonella and E. coli O157:H7.
(2) NIMFFAB, in conjunction with American Phytopathological Society, hosted the Human Pathogens on Plants Workshop in February 2012. Over 100 university, government, and industry representatives attended this 3 day conference. Attendee survey tools indicated changes in both knowledge and attitudes, attributable to the workshop (% agreement after workshop/% change from pre-workshop survey): awareness of cross-disciplinary research (94/10); interest in collaboration with other discipline (94/3); aware of key research areas (55/28); familiar with FSMA (77/24); familiar with FDA efforts (88/10); know two other professionals within discipline (91/11); know two other professionals outside of discipline (95/25); opportunities for new collaborations (88/3); annual conference is best way to network (87/8); annual conference is best way to exchange knowledge (87/5); poster sessions increased my knowledge (95/4); themed sessions were effective (90/2).
(3) The recent Listeria outbreak in cantaloupe, and previous associations of melons with Salmonella food poisoning motivated OSU researchers to study the interaction of Salmonella with a common melon plant pathogen. They found that the mesocarp of a low percentage of cantaloupe fruits that developed from flowers inoculated with Salmonella Poona (SP) alone, or from those receiving SP-Erwinia tracheiphila (Et) mixtures, were PCR positive.
(4) OSU researchers also found that inoculation of fruit rind cracks with SP-Et, but not SP alone, caused water-soaked lesions on fruits and SP was recovered from the mesocarp of 17% of co-inoculated fruits.
(5) Researchers concluded that the presence of the cucurbit bacterial wilt pathogen, Erwinia tracheiphila (Et), influenced SP survival on or in the blossoms or fruit. Salmonella population increased by 39% and 45% from the initial inoculum level on SP only and SP+Et inoculated blossoms even though the flowers had dehisced. Et inoculated into the flower was later detected in fruits and stems. Inoculated flowers fell prematurely, some fruit rinds had watersoaked lesions and plants wilted and died.
(6) Within the Food Safety research group in NIMFFAB, the research of Ph.D. student Dhiraj Gautam was completed and one Masters student (Chris Timmons) graduated and thesis was published.
(7) Our research on E. coli 0157:H7 and Salmonella reveals the nature of human pathogen - plant interaction. Whether the bacteria can be disseminated by insects, colonize plant surfaces, invade interior plant tissues, move systemically, increase in titer, and persist are critical to assuring safe produce.
(8) Five food safety research papers were submitted and accepted or published.

4. Associated Knowledge Areas

KA Code	Knowledge Area

212	Pathogens and Nematodes Affecting Plants
712	Protect Food from Contamination by Pathogenic Microorganisms, Parasites, and Naturally Occurring Toxins
903	Communication, Education, and Information Delivery

Outcome #3

1. Outcome Measures

Percentage of agricultural producers, handlers and processors employing at least one new (to them)practice to enhance biosecurity

2. Associated Institution Types

- 1862 Extension
- 1862 Research

3a. Outcome Type:

Change in Condition Outcome Measure

3b. Quantitative Outcome

Year	Actual
2012	1

3c. Qualitative Outcome or Impact Statement

Issue (Who cares and Why)
Important national research and policy initiatives in homeland security have engendered a need for additional personnel, trained and experienced in fields such as microbial forensics and food safety. The Bureau of Justice Statistics (BJS) recently identified an increasing backlog of forensic casework due to a shortage of forensic analysts. The BJS's call for 20% more traditional analysts is compounded by the additional need for analysts in the newly emerging sub-disciplines of microbial and agricultural forensics. In addition, there is a need for those in federal agencies, Cooperative Extension, and law enforcement, to receive training specifically related to response in a plant health emergency, and to practice their respective roles and responsibilities.
Note: Most of the OSU faculty and staff on the Agricultural Biosecurity Team have appointments with high percentages in research. The metrics required for the NIFA report involve documentation of changes in methods, practices, suppliers, etc. through surveys, personal contacts, etc. In the past, research personnel have not used surveys to measure and document such changes. Although many of the Ag Biosecurity team members have plans to use such surveys in conjunction with extension/outreach activities, we do not have such results for 2012.

What has been done

Results

(1) The course, ENT/PLP 2143, "Global Issues in Agricultural Biosecurity and Forensics" was taught for the third time during the spring of 2012. The aim is to introduce undergraduate students to the fundamental components of an effective agricultural biosecurity system by providing insights and practical understanding about how all components of a biosecurity system operate and integrate, facilitating the understanding of the relevance of the sciences involved in agricultural biosecurity and microbial forensics. This course provides broad information to a variety of majors, but also serves as a springboard for students considering careers in agricultural biosecurity, microbial forensics, related research, or roles in agricultural biosecurity agencies. Areas covered are:

a. Scientific and political definitions of biosecurity in its broad sense, to include microbial forensics, bioterrorism, biowarfare, biosafety, invasion biology, emerging pathogens, and invasive species.

b. Quarantine, response and surveillance.

c. Detection, diagnostics and forensics technologies.

(2) The course, ENT/PLP 5560 "Human Pathogens on Produce" was taught for the first time by Dr. Li Maria Ma. The aim of this course was to introduce graduate students to concepts of food safety microbiology as it applies to the unique situations encountered by human pathogens intersecting with plants. A combination of field site visits, lecture, and discussion, graduate students get the latest food safety information and exposure to practices that prepare them for careers in food safety and food biosecurity.

(3) One workshop on primer design was presented at Oklahoma State University to research scientists affiliated with Pioneer International. The goal of the three day workshop was to teach researchers how to design more specific primers for more sensitive detection of pathogens. A similar workshop was held in Venezuela.

(4) A successful Bioinformatics workshop was held in August 2012 that combined efforts of R. Kaundal (ICREST) with those of the Bioinformatics Certificate Program and Entangled Genomes. Research scientists, post-docs, and graduate students benefitted from expertise from iPlant researchers to learn the latest techniques in next generation sequencing and bioinformatics analysis.

(5) The 2nd 4-H Summer Camp was held at Oklahoma State University in June 2012; attendance included 16 teenaged 4-H youth and six Extension Educators. Evaluation forms from both adults and teens were extremely positive.

4. Associated Knowledge Areas

KA Code	Knowledge Area
212	Pathogens and Nematodes Affecting Plants
712	Protect Food from Contamination by Pathogenic Microorganisms, Parasites, and Naturally Occurring Toxins

V(H). Planned Program (External Factors)

External factors which affected outcomes

- Natural Disasters (drought, weather extremes, etc.)
- Economy
- Appropriations changes
- Public Policy changes
- Government Regulations
- Competing Public priorities
- Other (exotic pathogens, terrorism)

Brief Explanation

(1) Increased restrictions on the ability of foreign nationals to work on certain biosecurity-related (and dual use) projects funded by federal security agencies has affected the ability of some NIMFFAB faculty to work on these projects and also has led to our stated preference for U.S. citizens when bringing faculty members, graduate students and postdocs into our programs.

(2) A number of high-profile food-borne pathogen outbreaks occurred in 2012, continuing a multi-year trend in the increase of foodborne illnesses, including Listeria monocytogenes, E. coli 0145, and Salmonella in various fresh fruits and nuts. As a result, food safety was again named as one of the major S&T focus areas for the Obama administration (and a priority for the White House Office of Science & Technology Policy). Further, food safety also was named as one of the five major Emphasis Areas for 2012 NIFA proposals. These factors place NIMFFAB food safety researchers in a position to take advantage of potential new funding streams.

(3) Significant cuts in Federal spending on biosecurity resulted in a change in our Memorandum of Understanding with the National Bioforensic Analysis Center. AlthoughNIMFFAB remains an official "Spoke Laboratory" of the NBFAC we are not currently receiving funding from them for specific research or deliverables. It is hoped that ourprojects can be picked up again when funds become available.

(4) The new Food Safety Modernization Act mandates that the FDA develop new regulatory policy on food contamination by human pathogens, intensifying the agency's desire for targeted research to provide the scientific foundation for new regulations and focusing more USDA and NIH funding attention on food safety research.

V(I). Planned Program (Evaluation Studies)

Evaluation Results

NIMFFAB was awarded 3rd place in the 2012 **OSU President's Cup Award for Creative Interdisciplinary Research.**

Key Items of Evaluation

The School of Forensic Sciences received a renewal of our accreditation from the American Academy of Forensic Sciences (2012-2017). Included among the items evaluated

was the overall quality of graduate student research, for which the School of Forensic Sciences was congratulated.