FSIS Draft charge to NACMCF on new technologies

Food Safety Inspection Services

9/22/06

Introduction

- "The overall goal is to obtain recommendations from NACMF for the most appropriate technologies for FSIS and the public health community to yield the best available microbial analysis.
- "FSIS expects that this charge will be a long-term project for NACMCF."

The Most Appropriate Technologies

Overview

- Microbial analysis at FSIS
  - Programs
  - Data applications
  - Laboratory methodology
- Important analysis parameters
  - Time and Expense
  - Sensitivity and specificity
  - Scope of analysis
- Considerations
  - Data transfer and acquisition
  - DNA vs protein
  - Genotype vs serotype
  - Different applications (in-plant vs. laboratory, baseline vs. regulatory)
- Charge questions

FSIS microbial analysis programs

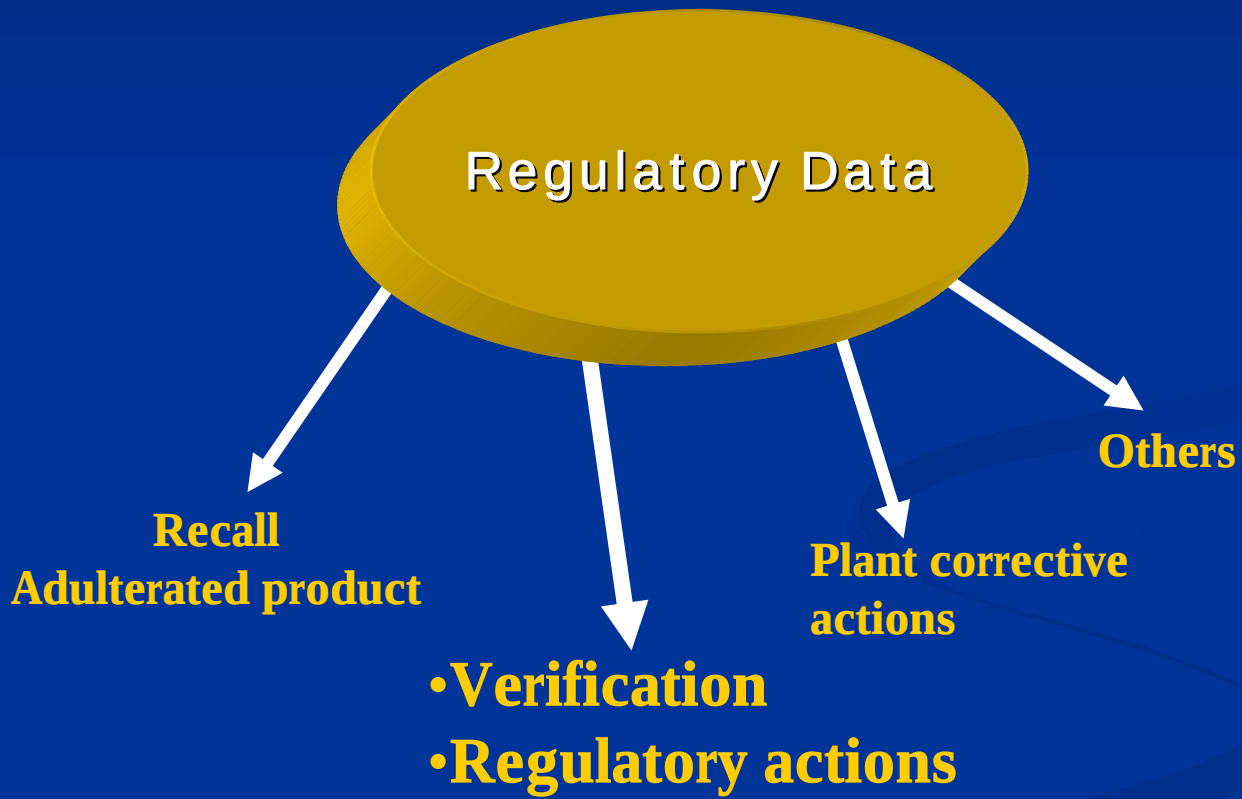
Regulatory Sampling Programs

Sampling from FSIS inspected establishments to verify product safety.

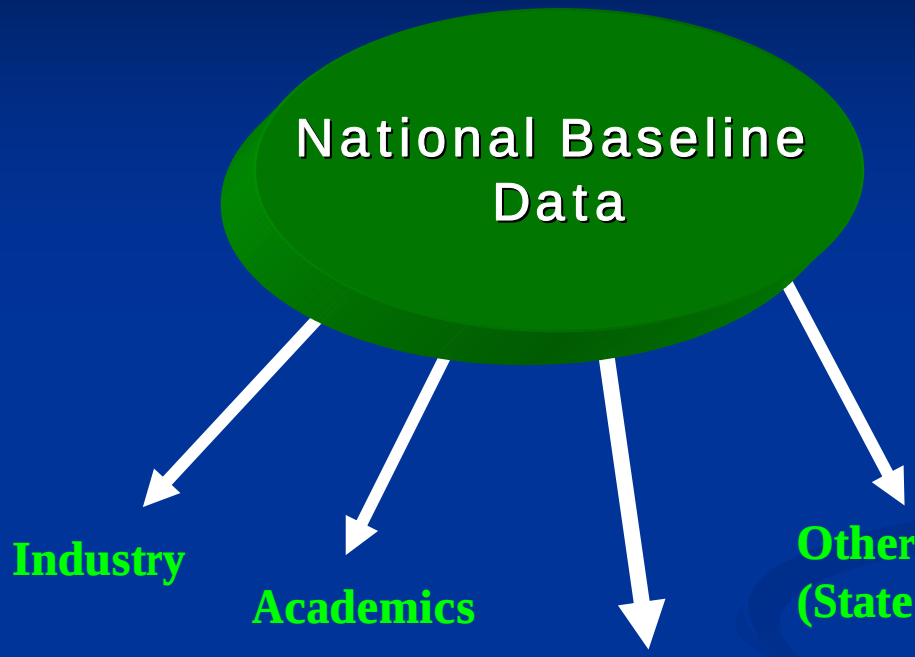
National Baseline Studies

Determine the nationwide prevalence of pathogens and other microorganisms in meat and poultry products

Applications of FSIS regulatory data



Applications of FSIS baseline data



- Performance standards
- Science based programs and policies
- Risk analysis

Microbial analysis is central to the FSIS mission

FSIS Laboratory methodology

Enrichment
(specific growth requirements)

PCR screen
(specific primer pairs)

Confirmatory testing
(species specific)

Further Characterization
(Serotype,
antimicrobial resistance,
PFGE)

Day 1 Enrichment
(specific growth requirements)

Day 2 PCR screen
(specific primer pairs)

Days 3-5 Confirmatory testing
(species specific)

Further Characterization
(Serotype,
antimicrobial resistance,
PFGE)

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Important analysis parameters
(and current FSIS standards)

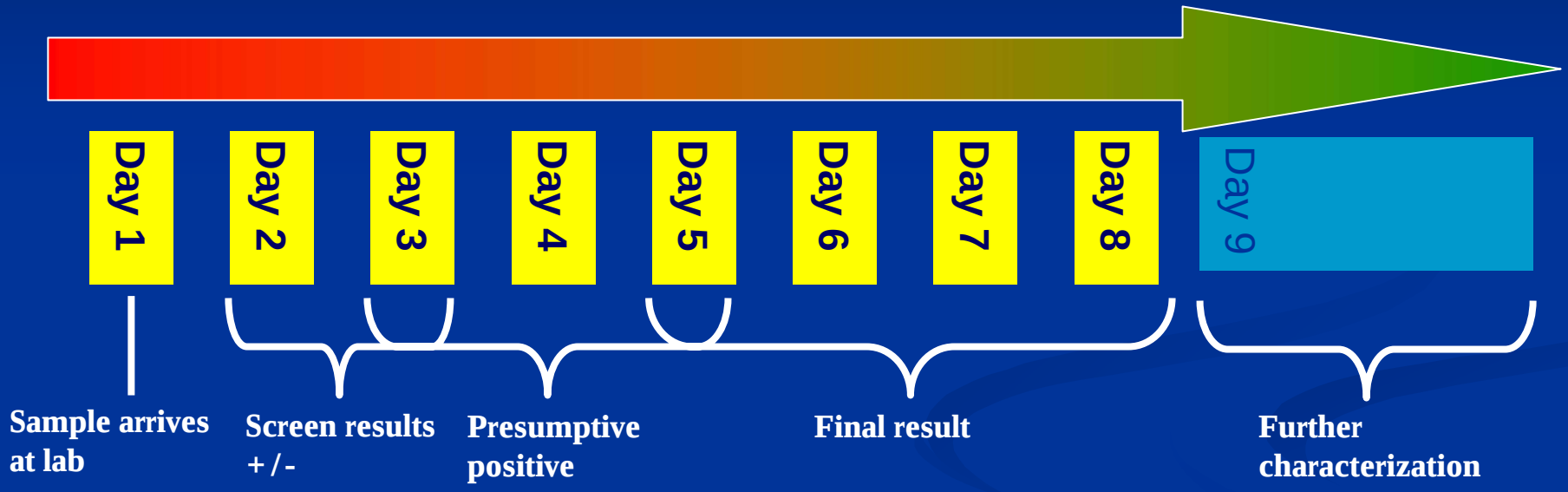
- **Time and expense**
- **Sensitivity, specificity and selectivity**
- **Scope of analysis** (type and amount of data collected)

Time of analysis

Time analysis

- **Response time in outbreak situations**
- **Product recalls**
- **“Test and hold” situations**

Current FSIS time for analysis
(where we are now)



Expense

- FSIS has finite resources and an obligation to spend tax dollars effectively
  - Given the sample numbers (10^4 – 10^5 samples analyzed/year) and importance of FSIS microbial analysis this is a key area for cost benefit analysis
  - Increased Public health benefit/dollar
- Where we are now
  - $88.00 – $98.00/sample

Sensitivity and specificity

Sensitivity = % of true positives a test identifies

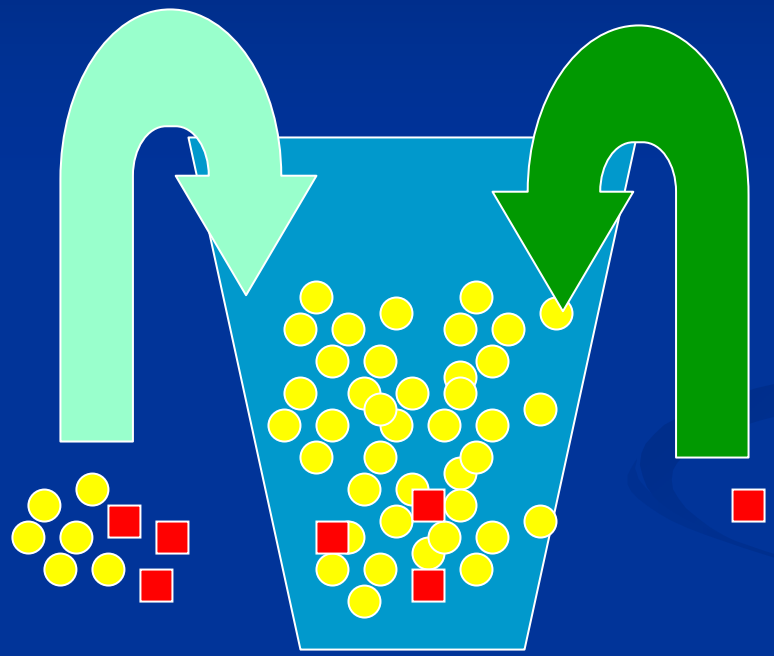
Related to the limit of detection CFU/g

Specificity = % of true negatives a test finds

Positive Predictive Value (rate of false positives)

Negative Predictive Value (rate of false negatives

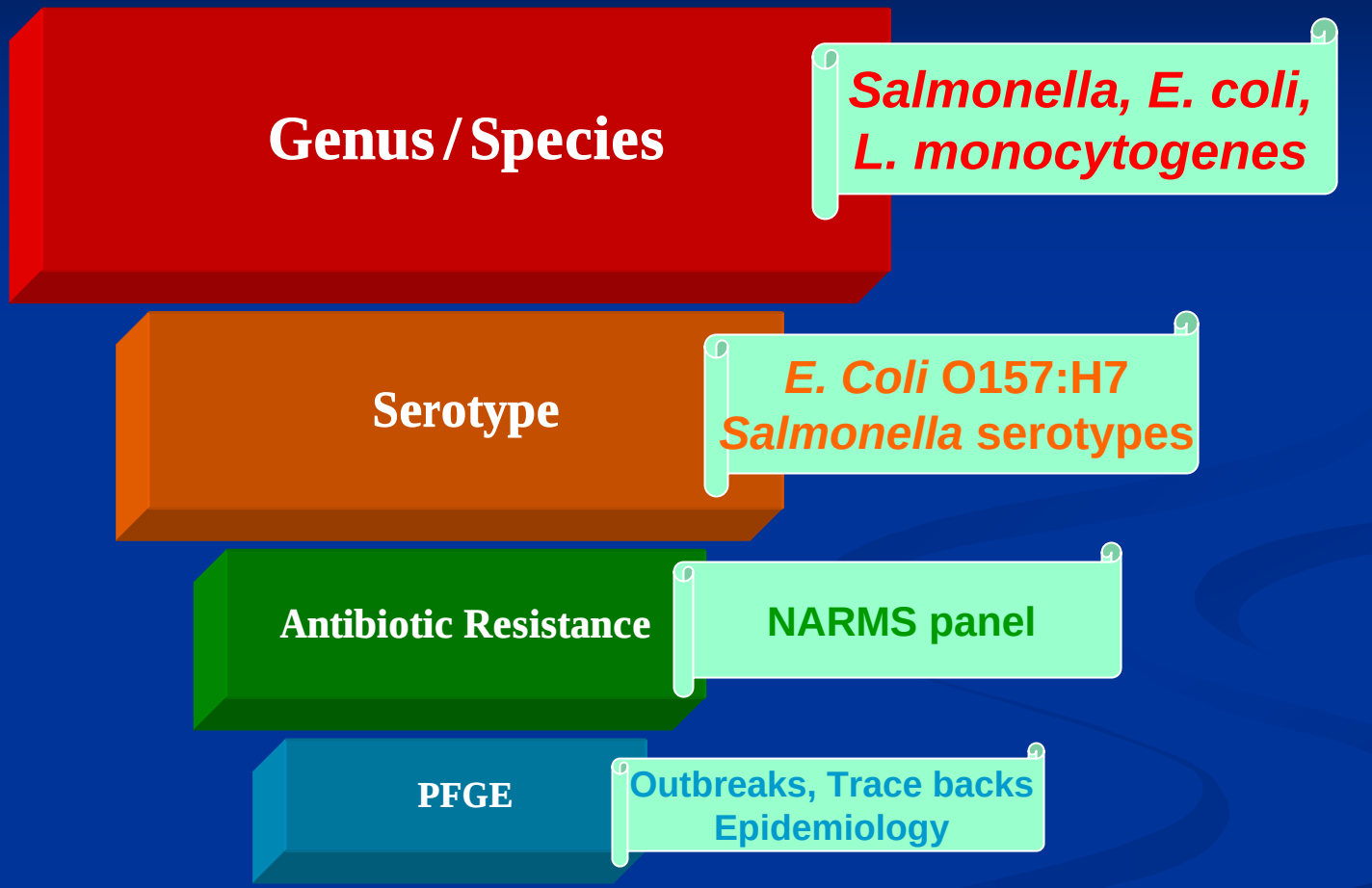
Sensitivity and specificity



Sensitivity and specificity
(where we are now)

- FSIS methods have very high sensitivity and specificity
  - For most tests over 99% and a limit of detection of app. 1CFU/25g

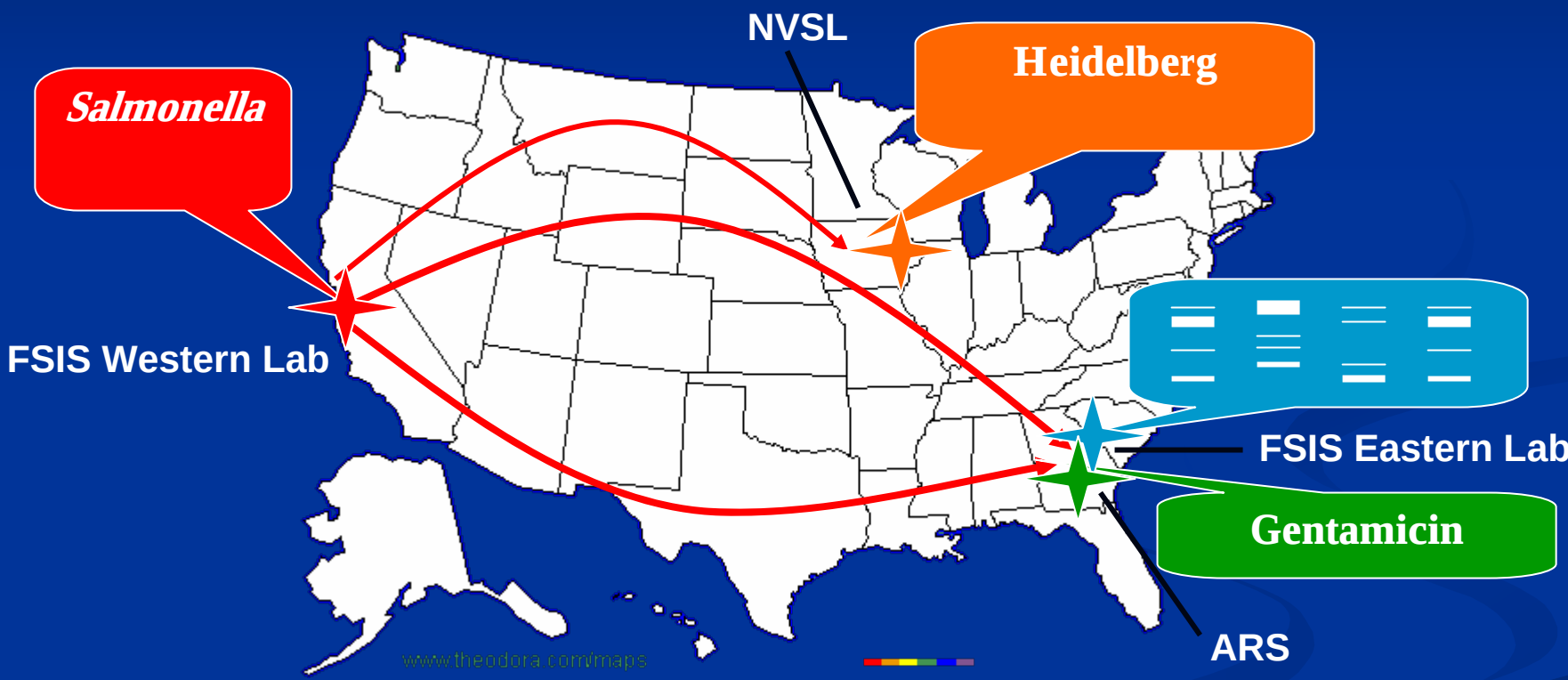
Scope of FSIS Microbial Data



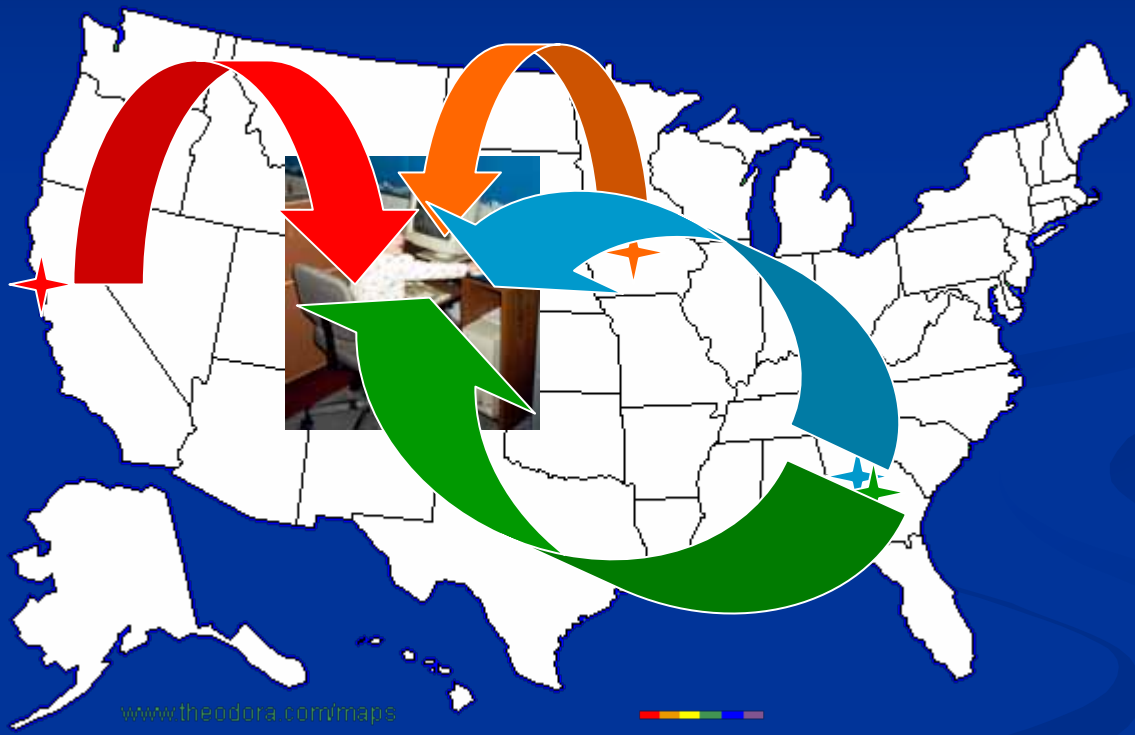
Considerations

- Data acquisition and transfer
- DNA vs. protein
- Genotype vs. serotype
- Different applications
  - In-plant vs. laboratory
  - Baseline vs. regulatory

Microbial data acquisition



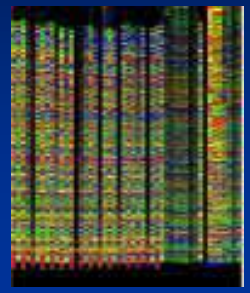
Data transfer and aggregation

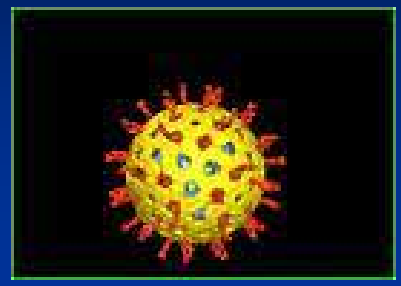


Detection of DNA vs. Protein

- **Excels at detecting large scope of microbial traits**
  - Probably more feasible with current technologies to perform comprehensive microbial analysis
- **Excels at speed**
  - very rapid detection possible (binding kinetics faster than amplification or DNA Hybridization)
  - Detection demonstrates expression

Genotype vs. serotype





- Genotype has the potential to deliver detailed information about pathogenicity and virulence
- Less cost
- Less time

Specific applications

- In-Plant vs. laboratory
- National Baseline studies vs. Regulatory programs

Balancing public health with burden

Charge question 1

- What are the most appropriate technologies FSIS should consider for improved laboratory and in-plant microbiological analyses?

Validation?

Implementation Models?

Charge question 2

- Consider specifically the accuracy, applicability, and validation of an assay capable of detecting thousands of single nucleotide polymorphisms (SNPs) in a single reaction. Would such an assay be timely, cost effective, and capable of screening specimens to monitor process control?

Large scale genotype assay

- **Large Scale Genotype assays identify 1000's of different DNA sequences in a single sample**

Identify multiple pathogen species/strains simultaneously in a single sample

Rapid, Cost Effective, High Throughput

Identify virulence factors/antibiotic resistance genes

A large scale genotype assay

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Large scale genotype assay

3’ Extension Assay

- **Efficient detection of SNPs**
  - **Utilizing SNPs allows a very fine level of discrimination required for many of the features we want to detect**
- **High Throughput**
  - **Detect 1000’s of SNPs simultaneously, 1000’s of samples in parallel**
- **Quantitative PCR amplification of target sequence**
  - **Amplification with optimized universal primers may allow detection in crude matrix (i.e. meat) without culturing for single isolates**
  - **Amplification with uniform primers allows some quantification of SNP abundance (could be translated into relative pathogen load in sample…)**

Charge question 3

- Which of the recommended technologies are applicable for immediate use and which for future implementation?

Charge question 4

- What technologies will improve enumeration of pathogens and indicator organisms?

Charge question 5

- What is the type and format of analytical data that should be captured from laboratory analyses and from in-plant testing to be most valuable to improving food safety?

Charge question 6

- What technologies, especially from those suitable for FSIS testing, would provide the type of data useful in risk assessment attribution models for human illness? What tests could assist in yielding data that would translate into a risk profile for a given product/operation?

Thank You!!!!!!

All of RAD

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