

Salmonella in Eggs: Issues and Research

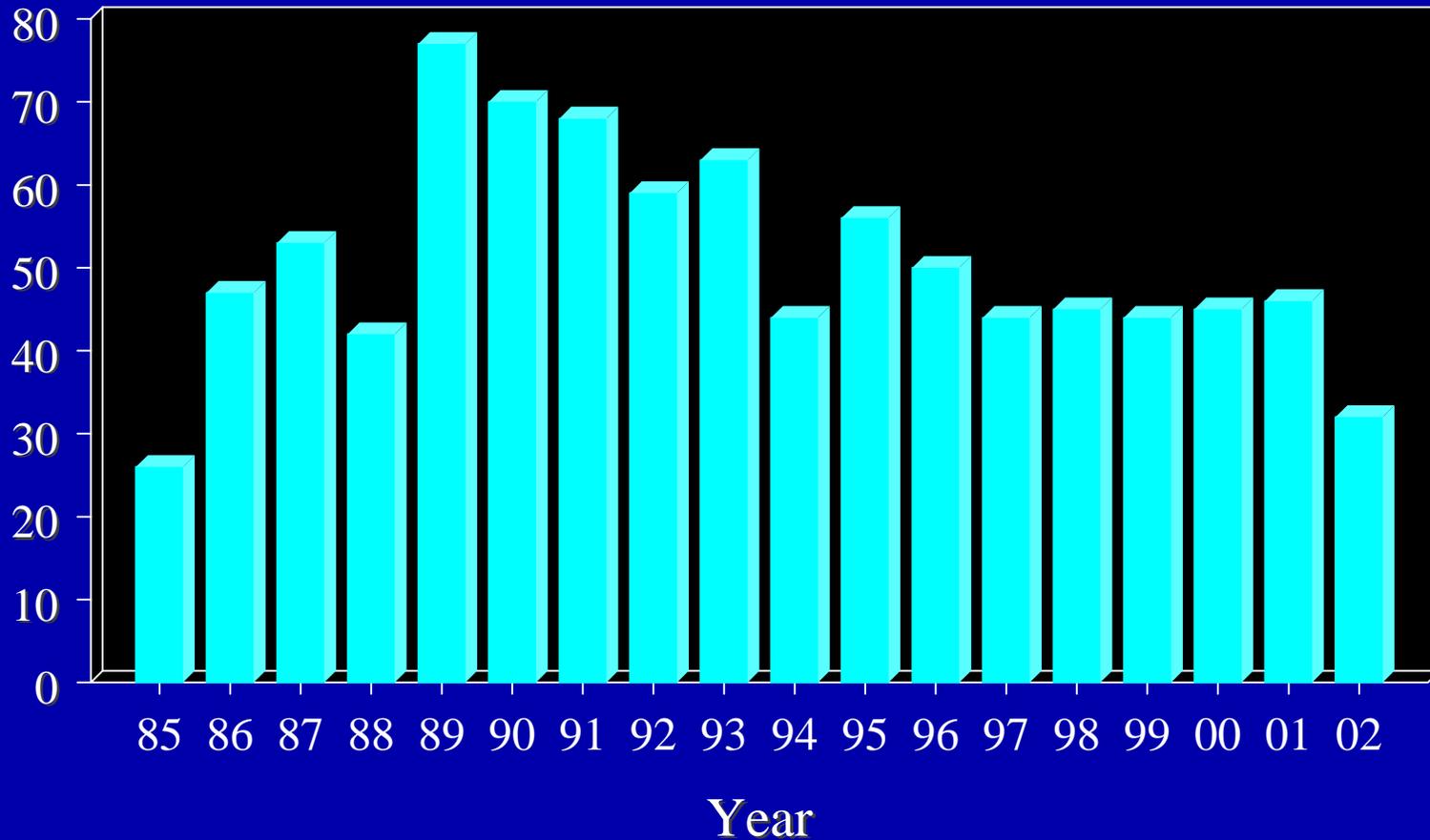
Richard K. Gast



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S. enteritidis Outbreaks in the USA

Number of outbreaks



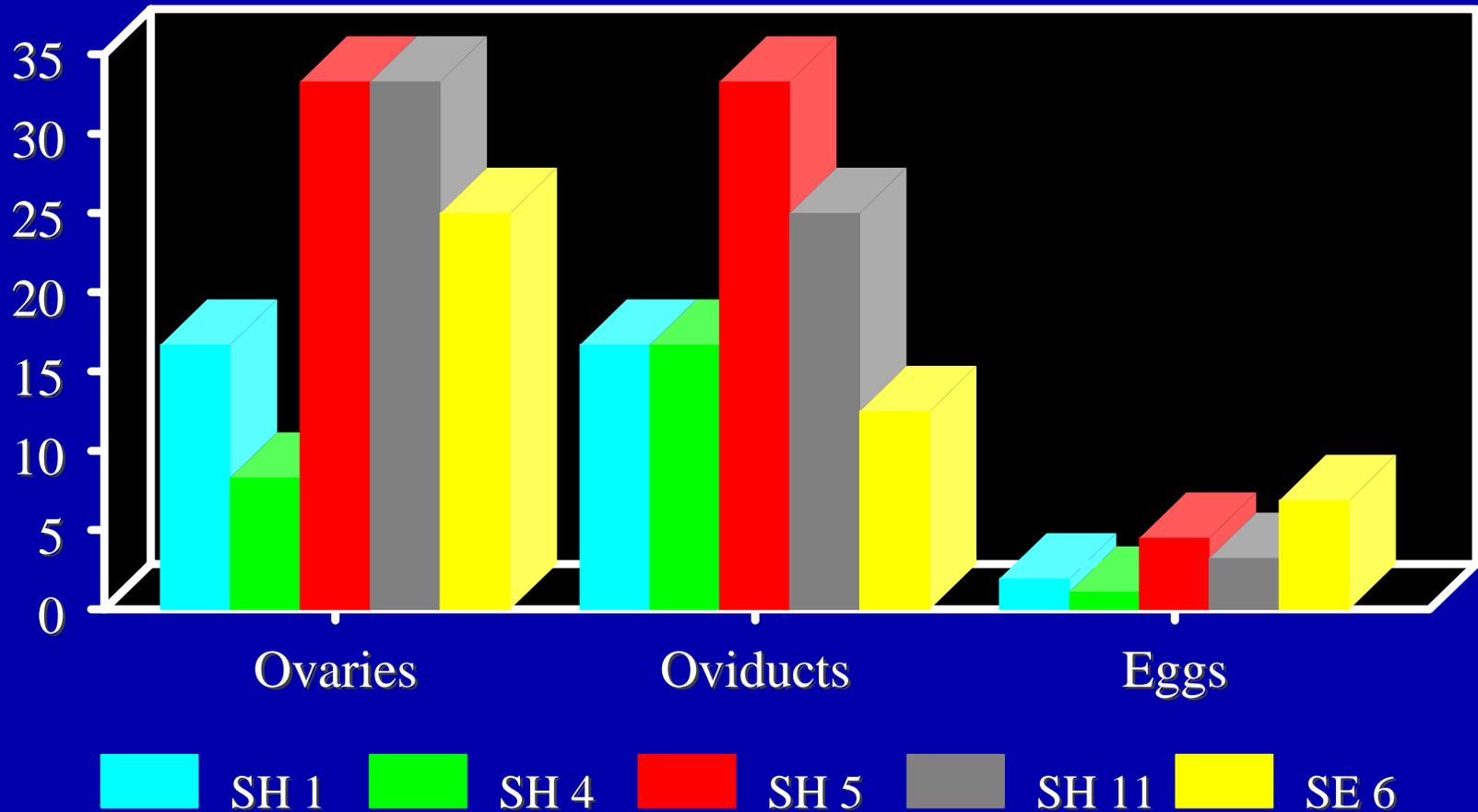
The association of eggs with human *Salmonella enteritidis* and *S. heidelberg* infections:

- 80% of human *S. enteritidis* outbreaks in the USA (1985-1999) for which a food source could be identified were attributed to eggs or egg-containing foods.
- 23% of *S. heidelberg* outbreaks in the USA (1973-2001) were attributed to eggs or egg-containing foods.

The deposition of *Salmonella* in eggs is a consequence of the colonization of reproductive organs, particularly the ovary and upper oviduct, in systemically infected hens.

Salmonella Recovery from Reproductive Tissues and Egg Contents

% positive



What is the incidence of
S. enteritidis infection in
laying flocks?

7% of commercial laying houses in the
USA were environmentally positive for
S. enteritidis in a 2000 survey
conducted by the USDA National
Animal Health Monitoring System.

What is the incidence of *S. enteritidis* contamination of eggs?

- In field studies of environmentally positive flocks in Pennsylvania and California, the prevalence of egg contamination with *S. enteritidis* was approximately 5 in 20,000 (0.025%).
- The 1998 USDA *S. enteritidis* Risk Assessment Report estimated the overall national egg contamination prevalence to be 1 in 20,000 (0.005%).

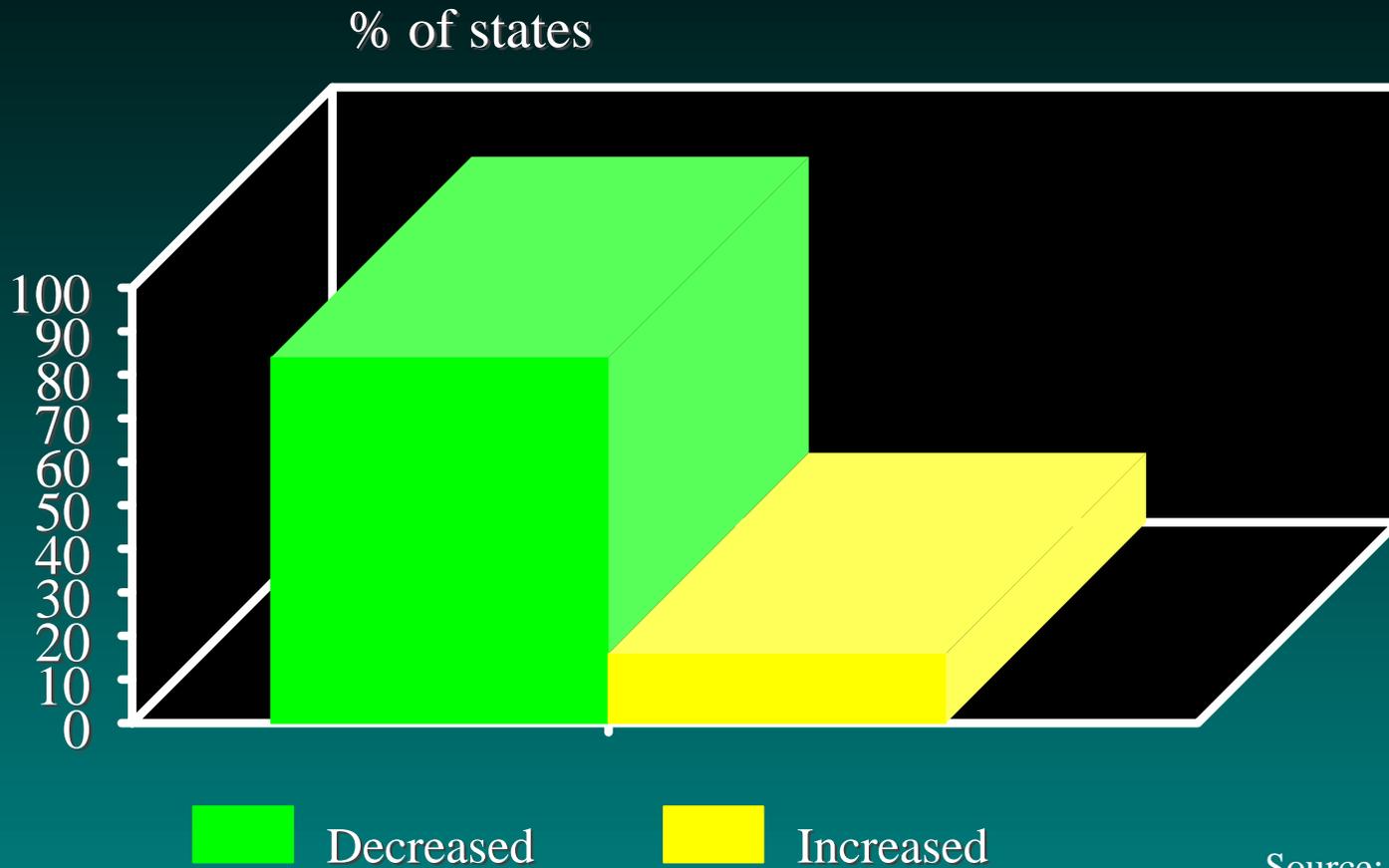
Recent Events in S. enteritidis Epidemiology and Control

- January, 2005: The Risk Assessment Division of USDA-FSIS estimates that 182,060 human illnesses were caused by consumption of *S. enteritidis*-contaminated shell eggs in 2000.
- September, 2004: FDA proposes regulations for shell egg production that include mandatory testing of environmental samples (and eggs, if necessary) from commercial laying flocks.

*Common Risk Reduction Practices for *S. enteritidis* in Poultry Flocks*

- Using uninfected chicks
- Effective pest control
- Cleaning and disinfection between flocks
- Heightened biosecurity
- Washing and refrigeration of eggs
- Vaccination

Changes in Human S. enteritidis Incidence in 31 States after Implementation of Egg Quality Assurance Programs

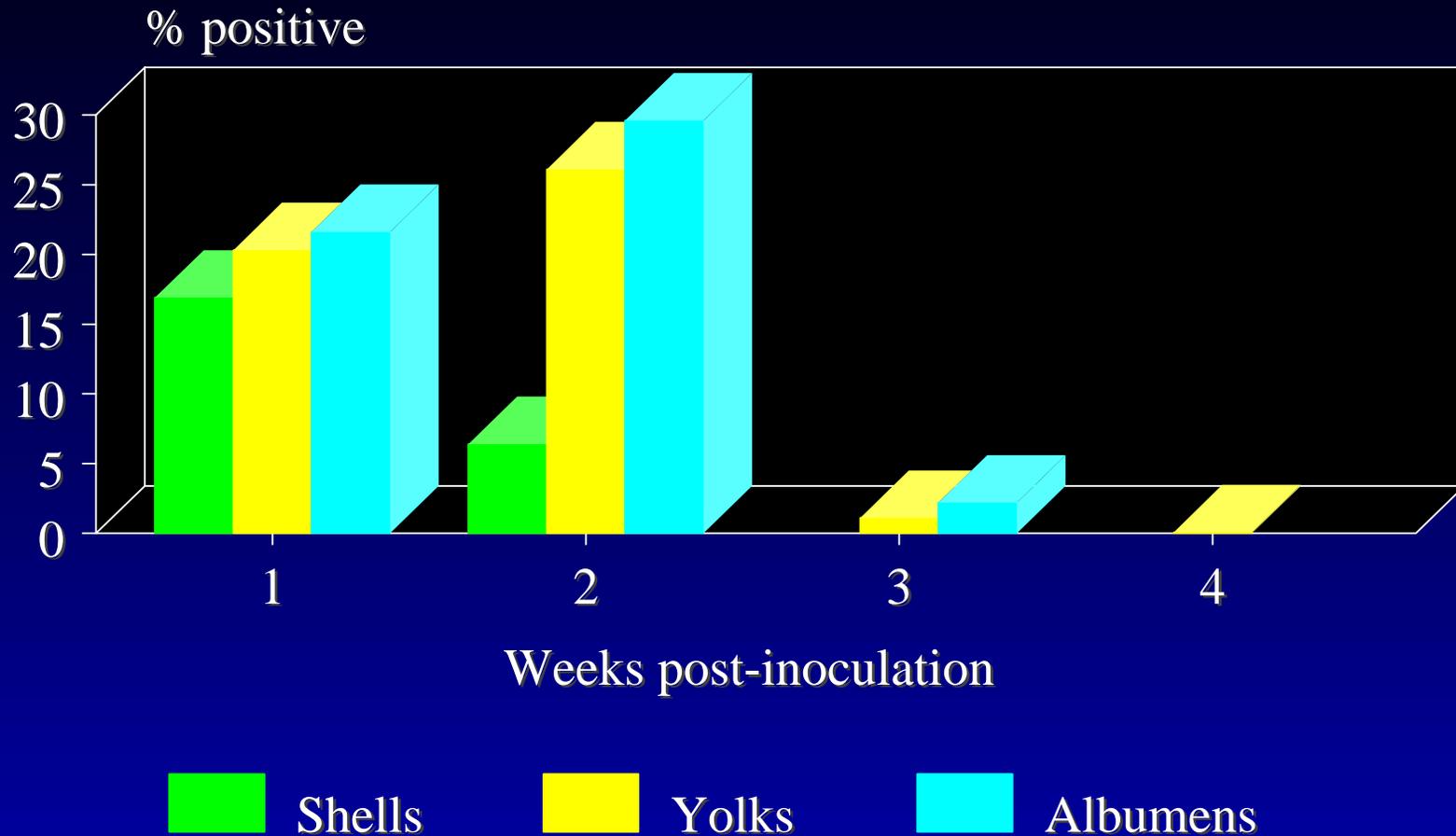


Source: CDC

Proposed FDA Regulations for Shell Egg Producers

- Purchase chicks from uninfected (NPIP-certified) breeder flocks.
- Maintain pest control and biosecurity programs.
- Test laying house environmental samples for *S. enteritidis* at 40-45 weeks of age and after induced molting.
- If environment positive for *S. enteritidis*, clean/disinfect thoroughly between flocks and test eggs. If eggs positive, divert for pasteurization.
- Store eggs under refrigeration at 45° F.

Recovery of S. enteritidis from eggs laid by experimentally infected hens



What are the research needs of regulatory agencies and the poultry industry regarding *Salmonella* in eggs?

Sources of Information:

- 2004 USDA-FSIS Risk Assessment for *Salmonella enteritidis* in Eggs
- Egg Nutrition Center Food Safety Advisory Panel
- American Egg Board Open House

Post-Harvest (Egg Processing) Research Needs

- Prevalence of pathogens on egg shells before and after processing
- Effectiveness of egg sanitizers
- Validation of current and prospective regulatory provisions

Egg Processing Safety, Quality, and Security

Deana Jones – Food Technologist
(Lead Scientist)

Michael Musgrove – Food Technologist

Effects of commercial cool water washing on pathogen detection in shell eggs

- Egg surface temperature was significantly reduced when eggs were washed with a combination of warm and cool water.
- *Enterobacteriaceae* frequency was not different between wash water temperature treatments for offline eggs.
- *Listeria* was detected in cool wash water at the offline facility but not on or in any eggs.
- *Salmonella* and *Campylobacter* were detected within the shells of 3 and 2 (of 384) samples, respectively. All positive samples were from cool water treated eggs.
- More work needs to be conducted to determine the feasibility of cool water washing of shell eggs, including examining detergents designed for cooler temperatures.

Correlation of shell strength and *Salmonella* Enteritidis infection

- Eggshell quality has always been difficult to quantify due to variable nature of eggs
- Advent of more objective detection devices has eliminated much of the testing variability
- A highly invasive inoculation method was utilized, yet it was very difficult to detect SE in the egg. Positive egg contents were only found after pre-enrichment.
- Correlation analysis did not find any strong positive or negative correlations between shell strength and SE contamination within the range of shell strengths detected in the current study

Recovery of *Salmonella* from commercial shell eggs by two methods

- Utilized shell rinse and shell crush and rub
- Monitored for naturally occurring *Salmonella*
- Shell rinse recovered 4.8% positives
- Shell crush and rub recovered 5.3% positives
- Occasionally only one method was positive for *Salmonella* per sample
- Using both methods increased sensitivity, although in most cases, crushing provided more sensitive detection

Impact of commercial processing on the incidence of *Salmonella* on shell eggs

<i>Salmonella</i>	No. of isolates	Sample type
Typhimurium	21	Eggshell
	1	Tap water
Typhimurium (Copenhagen)	4	Eggshell
4-12:i:-monophasic	2	Eggshell
Heidelberg	9	Eggshell
Kentucky	1	Eggshell
	1	Wash water
Total	39	

Pre-Harvest (Egg Production)

Research Needs: Stress and Immunity

Issues

- Effects of induced molting (by feed withdrawal and alternative methods) on immune responsiveness and susceptibility to *Salmonella* infection
- Stress associated with other management practices and effects on immune responsiveness
- Protection by vaccination and stress effects on vaccine efficacy

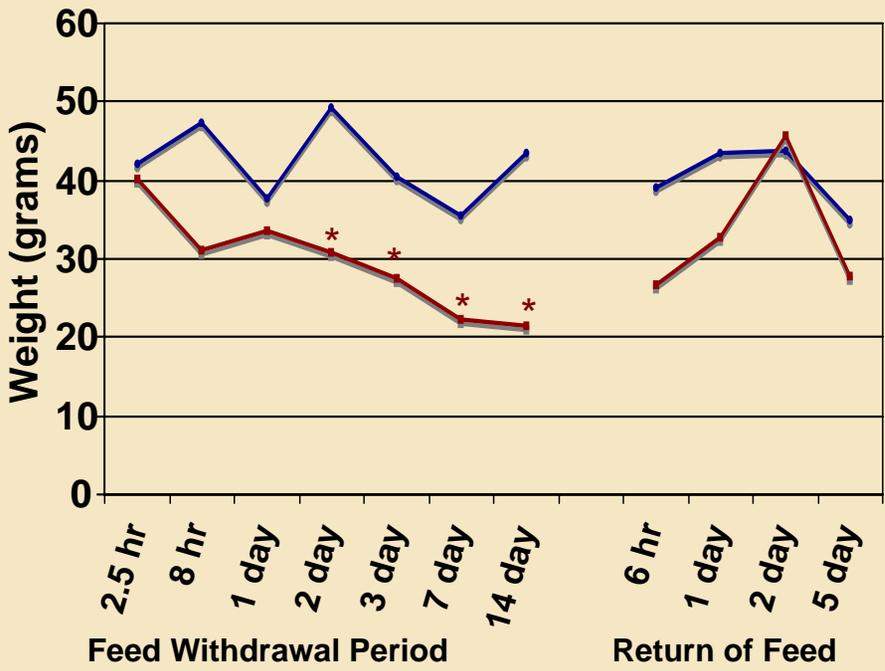
Stress Effects on Immunity and Physiology of Poultry

Peter Holt – Immunologist
(Lead Scientist)

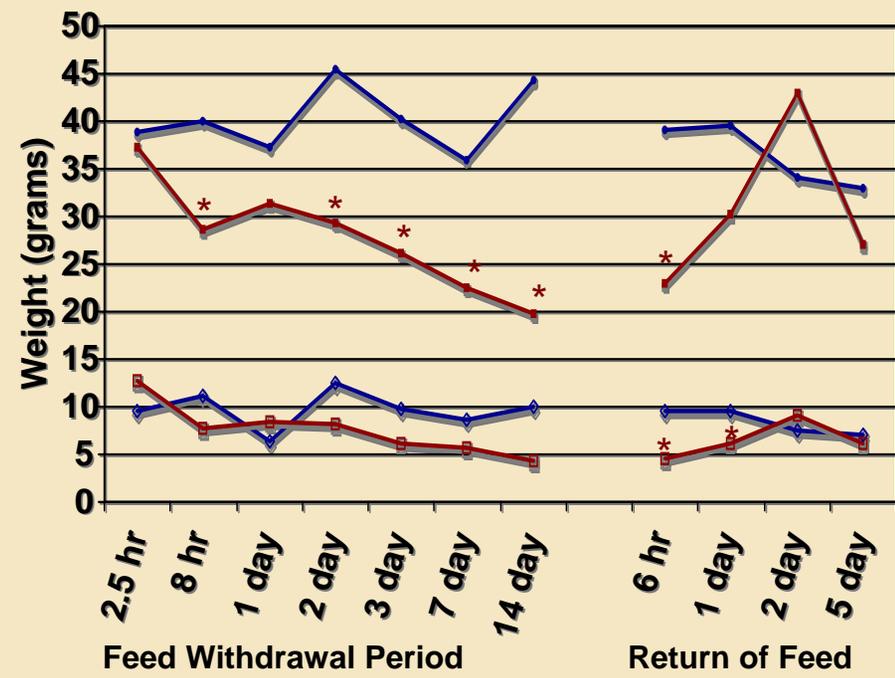
Randle Moore – Veterinarian/Physiologist

EFFECT OF FEED DEPRIVATION AND RECOVERY FROM FEED DEPRIVATION OVER TIME ON:

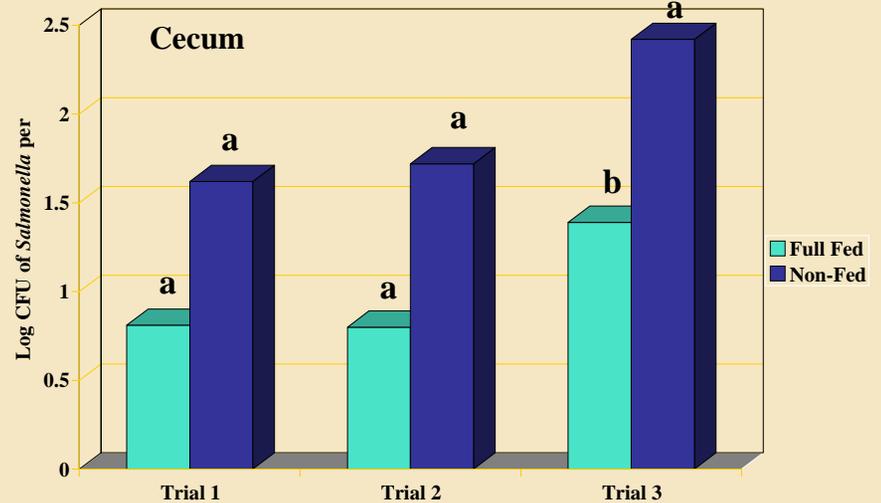
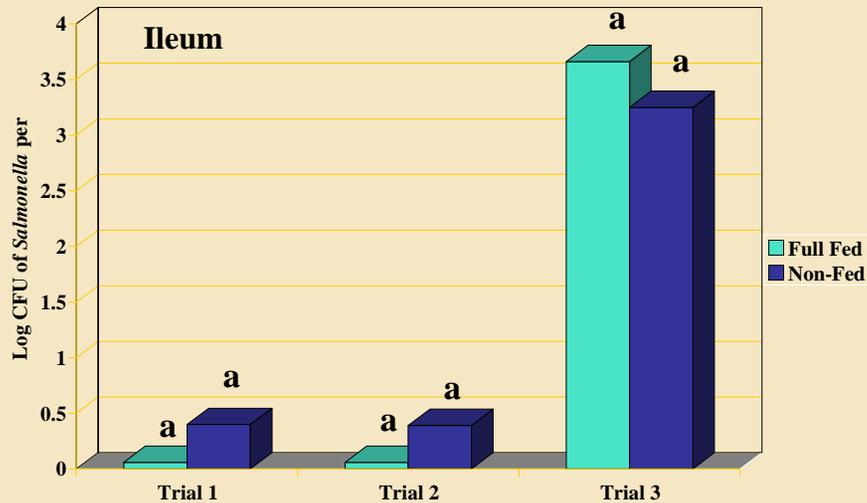
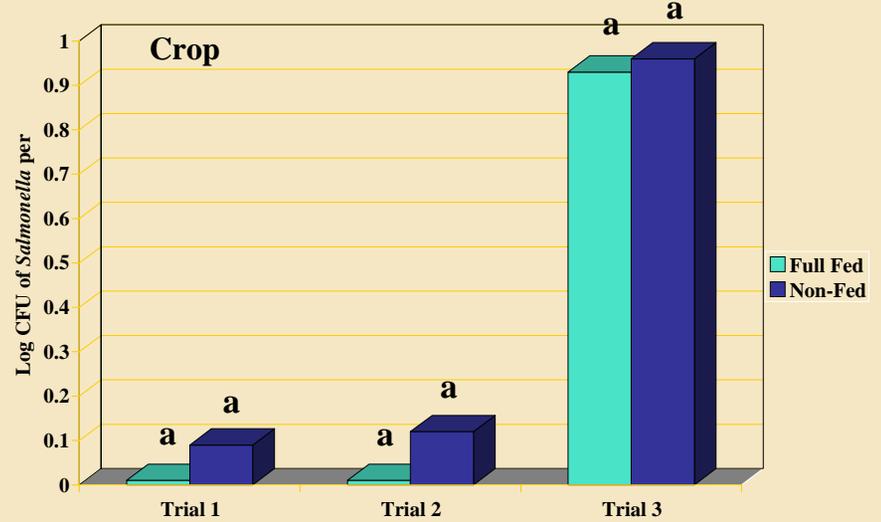
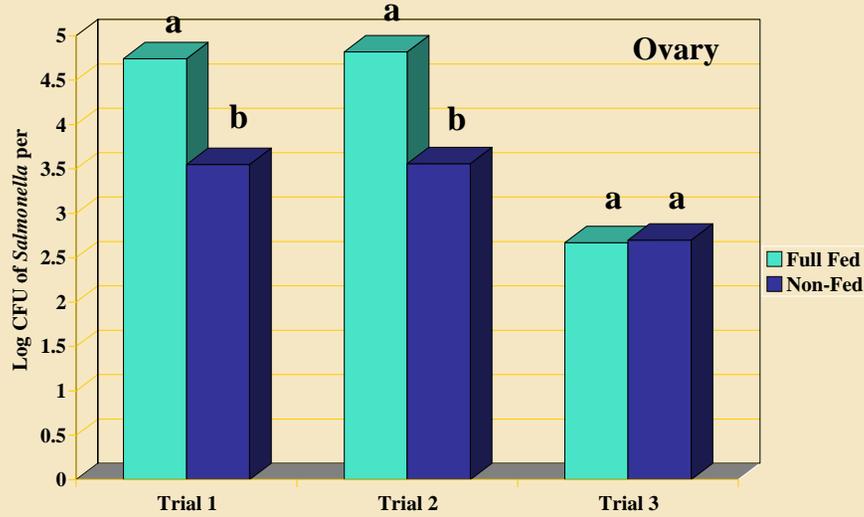
Intestinal Length



Intestinal Weight

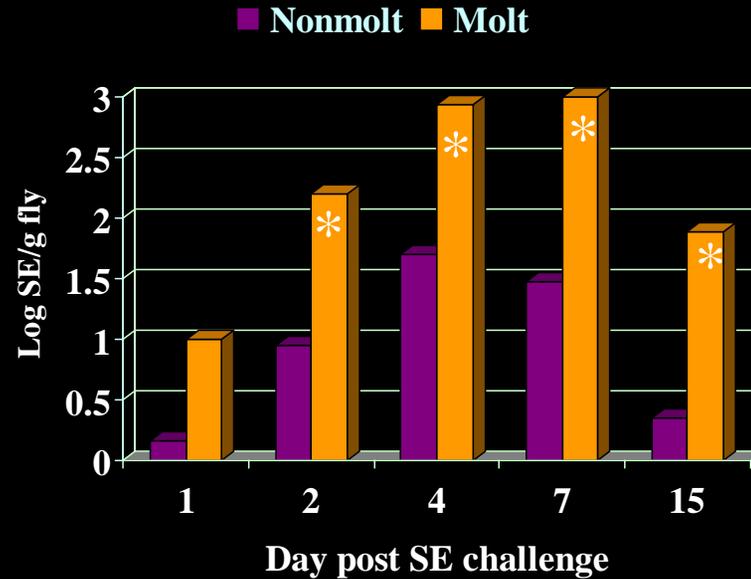


Effect of Feed Deprivation on *in Vitro* Tissue Invasion of *Salmonella enteritidis*

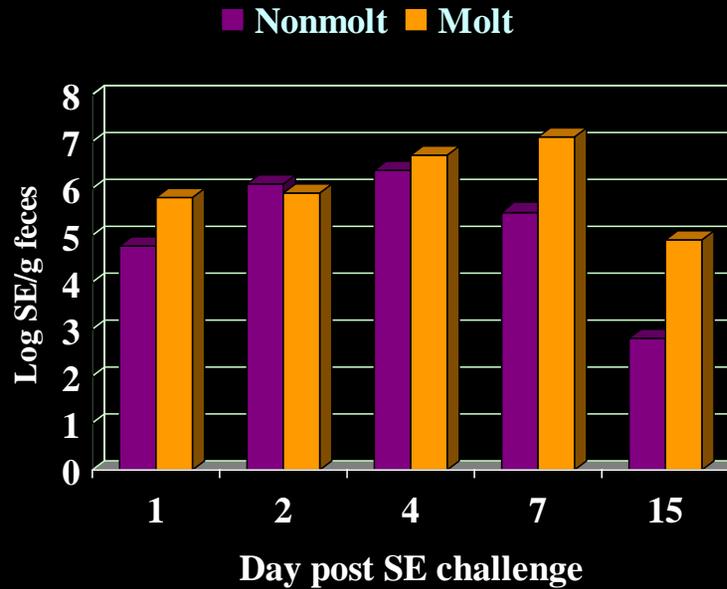


Effect of Induced Molting on Levels of SE in Flies vs on Floor

SE levels in flies



SE levels on floor



Isolation of *S. Enteritidis* from Fly Interior vs Exterior

<u>Treatment</u>	<u>Exterior</u>	<u>Interior</u>
Nonmolt	8/10	9/10
Molt	9/10	9/10

Isolation of *S. Enteritidis* from Internal Organs

<u>Treatment</u>	<u>Sal. Gland</u>	<u>Crop</u>	<u>Gut</u>
Nonmolt	0/10	1/10	10/10
Molt	0/10	3/10	8/10

Pre-Harvest (Egg Production)

Research Needs: Pathogenesis Issues

- Prevalence, level, and site of egg contamination; growth kinetics of *Salmonella* in eggs during storage; effects of refrigeration on bacterial growth in eggs
- The evolution (and identification) of *Salmonella* strains able to contaminate eggs
- Genetic differences between *Salmonella* strains that can be used to establish epidemiological relationships (including sources and routes of transmission)

*Controlling Egg Contamination
with Salmonella enterica by
Understanding its Evolution and
Pathobiology*

Jean Guard-Bouldin –

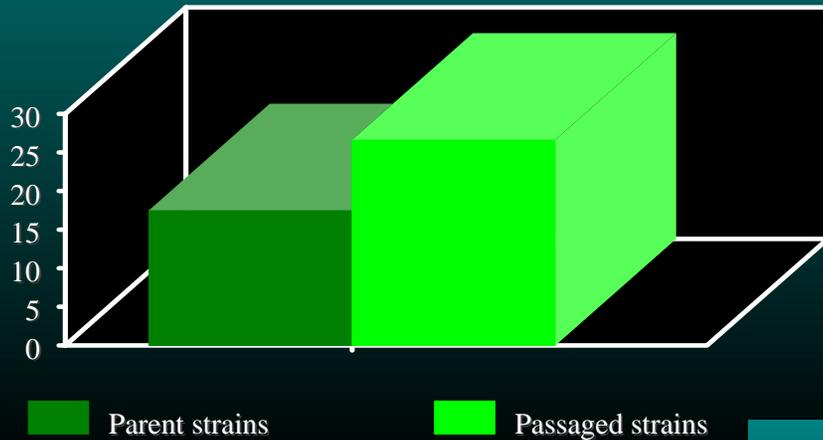
Veterinarian/Molecular Biologist (Lead Scientist)

Richard Gast - Microbiologist

(Research Leader)

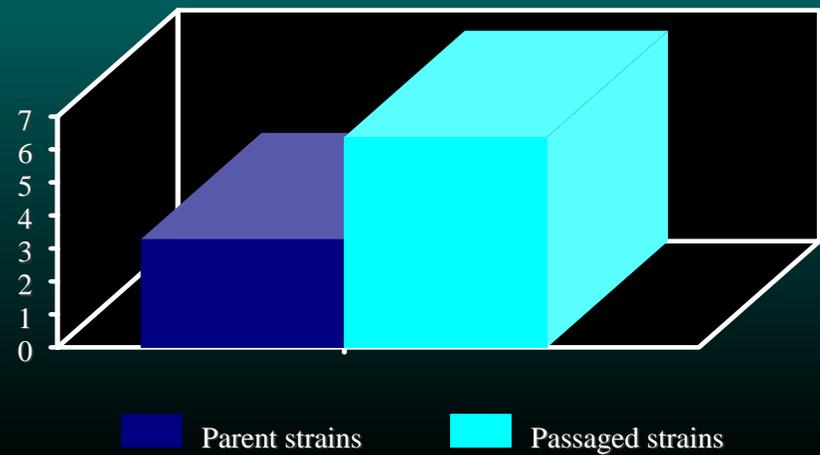
Mean Duration of Salmonella Recovery from Fecal Samples

days postinoculation



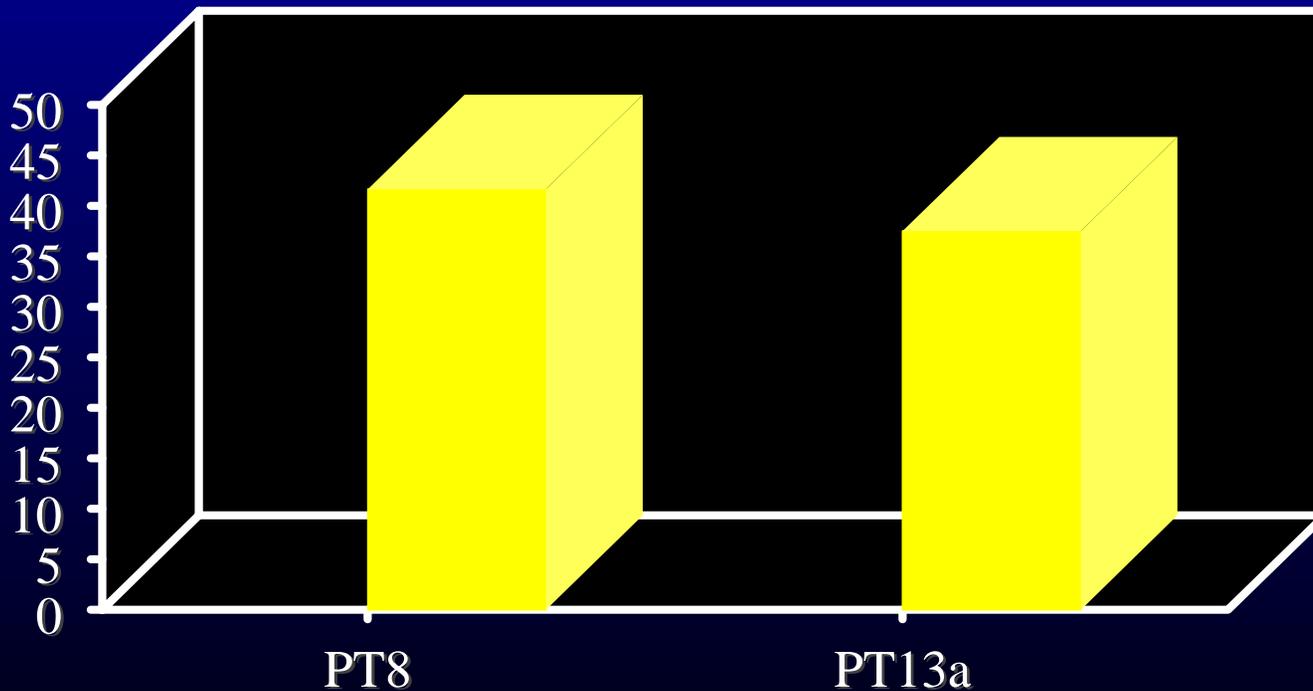
Salmonella Recovery from Eggs

% positive



S. enteritidis penetration into egg yolks

% positive



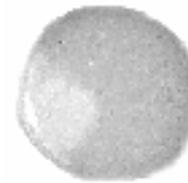
7° / 24 hours

30° / 24 hours

Phenotype can vary dramatically within serotype and phage type

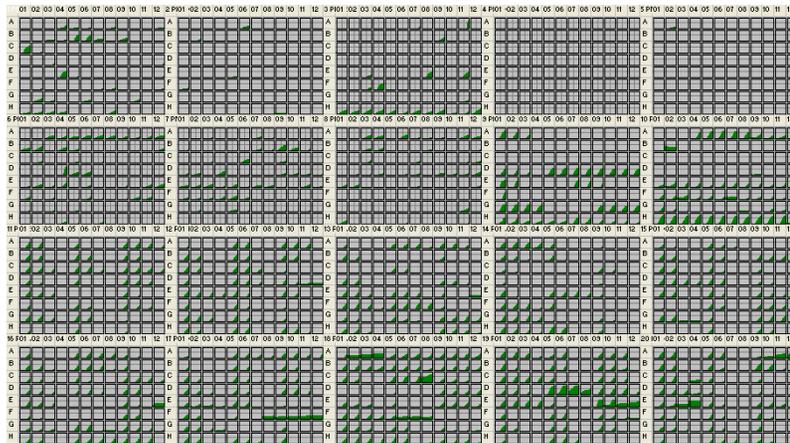


PT13A

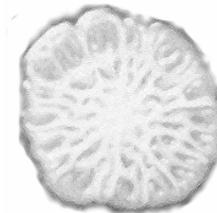


≠ Egg
Contamination

biofilm
negative



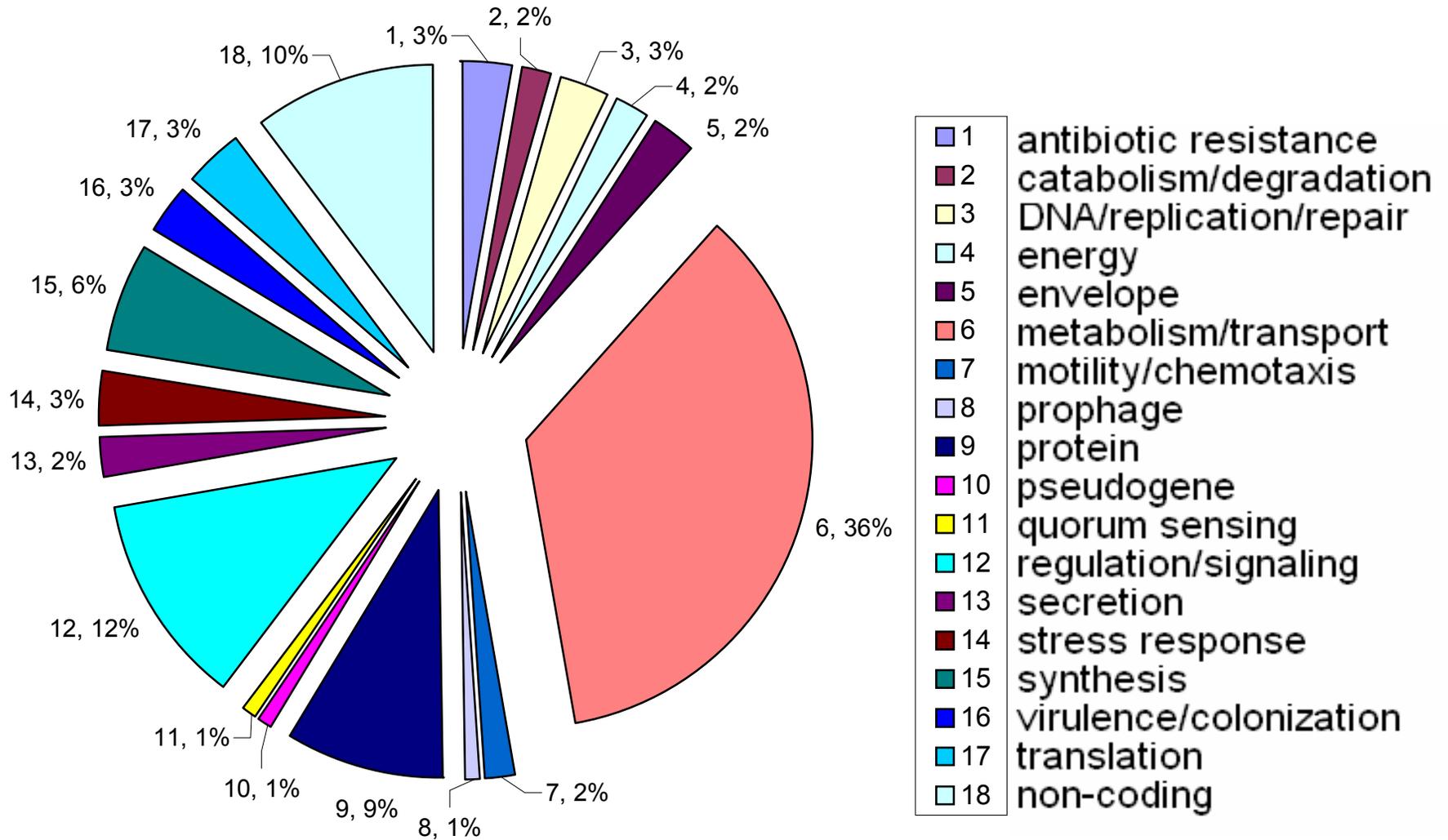
PT13A



≠ Egg
Contamination

biofilm
positive

SNP portfolio of *Salmonella* Enteritidis



Work in progress from Jean Guard Bouldin, DVM, Ph.D., USDA-ARS-ESQRU, Athens GA

The Future of Salmonella Control in Egg-Laying Chickens?



Detecting and protecting against specific Salmonella serotypes can be essential for responding to urgent or severe threats to public health, but the most cost-effective and sustainable approaches to controlling food-borne disease are based on risk reduction practices that address a broad spectrum of current and emerging pathogens.