



E. coli O157 in Central Coast Wildlife

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Overview

•Intrusion by wild or feral animals into produce fields or surrounding watersheds is now considered a significant risk factor for produce contamination of leafy green vegetables California.

•Some on-farm food safety practices to reduce wildlife intrusion into production fields may be detrimental to water quality and environmental conservation best practices.

Mechanisms of foodborne pathogen transmission from domestic animals or wildlife to leafy greens

- Direct transmission: fecal defecation or runoff (for example from a cattle operation) onto the plant.
- Indirect transmission: fecal contamination of water (surface or well), soil, sediment, and/or bioaerosols that may later contaminate the plant.







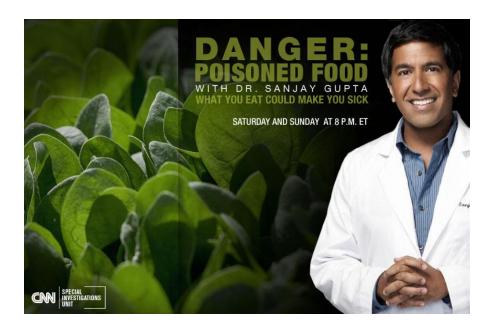


Animal Intrusions

Deer feces observed on Romaine lettuce

Canada geese traveling between adjacent strawberry and Romaine lettuce fields

Outbreak Investigations





Spinach Outbreak 2006

Food safety: No guarantees



USA TODAY

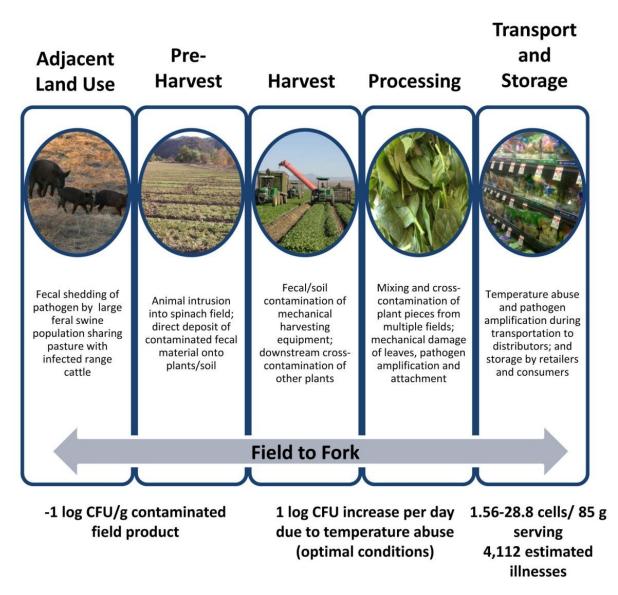
Photo by Richard Green, The Californian

2006 *E. coli* O157 Spinach Outbreak Farm Investigation, San Benito County

Sample	No. tested	<i>E. coli</i> O157 (%)	Outbreak strain
Cattle feces	77	26 (33.8)	15
Cattle water trough	10	0	
Compost (chicken pellets)	1	0	
Feral pig feces	47	11 (23.4)	6
Feral pig colon	40	2 (5)	2
Other animal samples*	26	0	
Surface water	79	3 (3.8)	2
Soil/sediment	37	3 (8.1)	3
Well/irrigation water**	18	0	

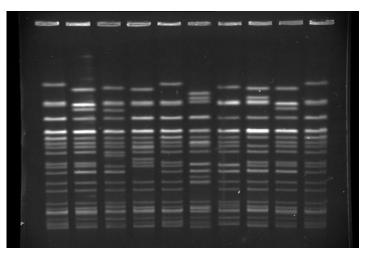
*Coyote (1), deer (4), dog (1), horse (2), sheep/goat (3) waterfowl (2), owl pellet (2), unknown (11); **100-1000 ml grab or 40,000 ml ultrafiltration

Hypothetical Model of Fecal Deposition In-Field Contamination



<u>2011 E. coli O157</u> Outbreak, Strawberries (Oregon, Roadside Stands): deer droppings in fields at Jaquith Strawberry Farm in Washington County contained the same strain of bacteria that sickened 15 (2 deaths)







<u>2008 Campylobacter Outbreak, Raw Peas (Alaska, Farmers Market)</u>: 63 confirmed cases, 1 with paralysis(GBS), *Campylobacter jeuni* found in 14/14 (<u>100%</u>) Sandhill crane fecal samples collected at farm implicated outside Anchorage; 2 samples contained the outbreak strain.







Ecology and epidemiology of *Escherichia coli* O157:H7 in fresh produce production regions of Salinas, California/Central California Coast, 2008-2010

USDA ARS (R. Mandrell, PD; M. Cooley) UC Davis (R. Atwill, Co-PD; M. Jay-Russell, R. Larsen) USDA Wildlife Services (D. Orthmeyer, S. Chandler)

- (1) Determine risk factors for in-field contamination of lettuce with enteric pathogens
- Sampling water, soil, produce, livestock and wild animals (>12,000 samples) (2)
- (3) Disseminate recommendations for risk reduction to prevent pre-harvest contamination









FOOD & AGRICUITURE



Methods

- Confidential enrollment of growers and ranchers – Monterey, San Benito, San Luis Obispo
- Leafy greens sampling during growing season (young and mature plants) with paired soil sample
- Cattle sampled quarterly
- Wildlife hunted or trapped fecal, distal colon, and /or swabs under DFG Scientific Collection Permit with USDA Wildlife Services

Methods

- *E. coli* O157 (Cooley et al, 2007)
 - Culture: selective enrichment/plating, IMS
 - Confirmation: PCR
 - DNA fingerprinting: MLVA, PFGE (PulseNet)
- Salmonella (Gorski et al, 2011)
 - Culture: frozen enrichments, IMS
 - Confirmation: PCR
 - DNA fingerprinting PFGE

Methods - Sample Type

Таха	Cloacal Swab	Colon/Feces (necropsy)	Feces	Unknown	Grand Total
Avian	384	60	14	0	458
Carnivore	89	171	1	0	261
Large game animal	0	309	25	1	335
Marsupial	16	33	0	0	49
Rodent/lagamorph	105	168	2	0	275
Grand Total	594	741	42	1	1378

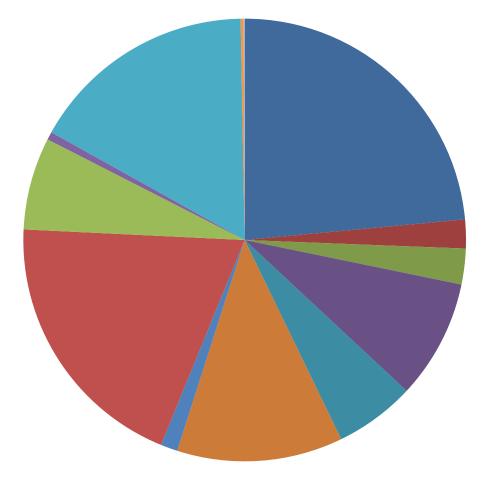
Collection Method

Таха	Feces from ground	Hunt	Road Kill	Trap	Grand Total
Avian	14	183	0	261	458
Carnivore	0	145	0	116	261
Large game animal	26	249	0	60	335
Marsupial	0	7	0	42	49
Rodent/lagamorph	2	269	2	2	275
Grand Total	42	853	2	481	1378

Sample Location

Таха	Monterey	San Benito	San Luis Obispo	Grand Total
Avian	444	14	0	458
Carnivore	122	139	0	261
Large game animal	287	46	1	334
Marsupial	14	35	0	49
Rodent/lagamorph	229	25	21	275
Grand Total	1096	259	22	1377

Habitat Type



- Feedlot
- Field crop
- Golf Course
- Grassland
- Landfill
- Oak Woodland
- Orchard
- Pasture
- Residential
- Riparian

Ranch Types

Таха	Produce Farm	Cow-Calf	Feedlot	Wildlife Services	Grand Total
Avian	46	106	276	30	458
Carnivore	20	32	13	196	261
Large game animal	28	72	15	220	335
Marsupial	9	4	0	36	49
Rodent/lagamorph	96	101	20	58	275
Grand Total	199	315	324	540	1378

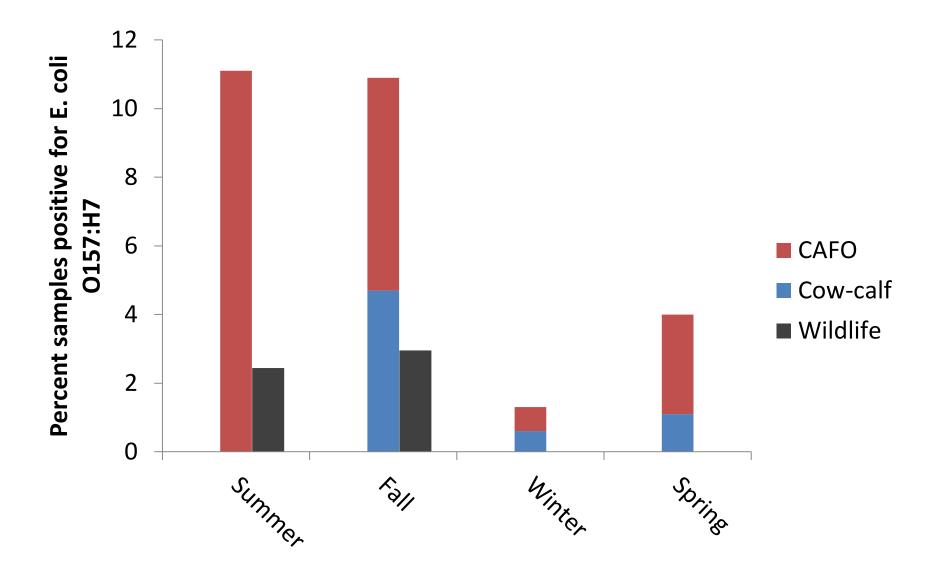
Results

- A total of 1,378 samples collected from 18 avian and mammalian species at 49 different locations in 3 counties, April 2008-November 2010
- *E. coli* O157 detected in American crow, brown-headed cow bird, coyote, and feral pig
- Seasonal and geographical variations

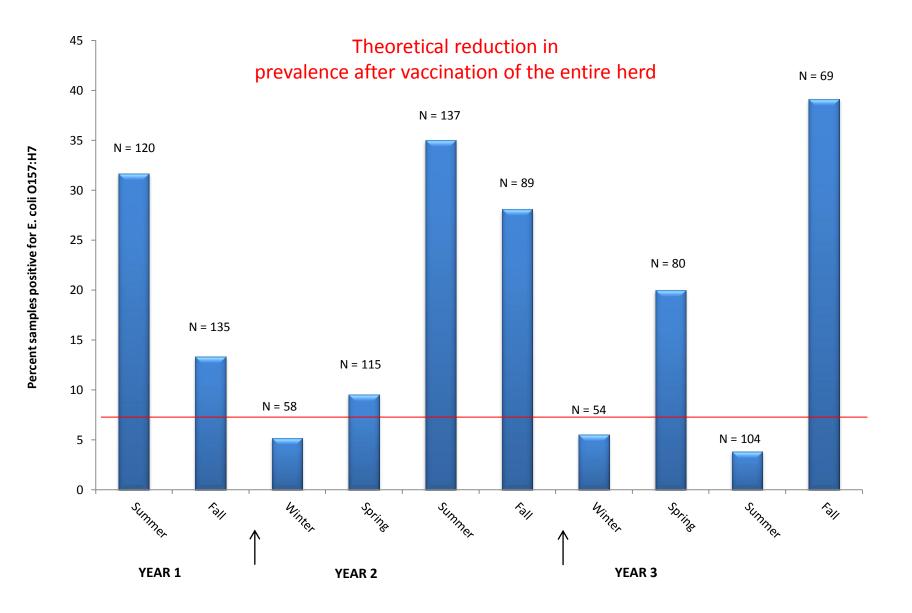
Species	<i>E. coli</i> O157 positive	Percent	Grand Total
American crow	5	3.6	137
Blackbird	0	0	129
Brown-headed cow bird	2	3.3	60
Canada goose	0	0	32
Cottontail rabbit	0	0	92
Coyote	2	1.4	145
Black-tailed deer	0	0	27
Feral pig	11	3.6	306
Ground squirrel	0	0	128
Gull	0	0	19
Jack rabbit	0	0	45
Mallard duck	0	0	33
Opossum	0	0	49
Rabbit	0	0	10
Raccoon	0	0	31
Striped skunk	0	0	85
European starling	0	0	48
Tule elk	0	0	1
Unknown feces	0	0	1
Grand Total	20 (1.5)	1.5	1378

	All Locatio	ns	E. coli O157 positive locati	ons
Species	No. sites	No pos./No. tested (%)	Code (ranch, county)	No. pos./No. tested (%)
	10		A (feedlet D(ee))	F (47 (10 C)
American crow	12	5/137 (3.6)	A (feedlot, Mon)	5/47 (10.6)
Brown-headed cow bird	1	2/60 (3.3)	Α	2/60 (3.3)
	24	2/145 (1.4)	B (WS, SB)	1/2 (50)
Coyote			C (WS, Mon)	1/34 (2.9)
	22	11/306 (3.6)	Α	1/15 (6.7)
			В	3/3 (100)
			C	1/6 (1.7)
			D (WS, Mon)	3/118 (2.5)
			E (WS, Mon)	1/24 (4.2)
			F (produce, Mon)	1/11 (9.1)
Feral pig			G (cow-calf, Mon)	1/34 (2.9)

Seasonality of *E. coli* O157, 2008-2010



Could a Cattle *E. coli* O157:H7 Vaccine Reduce Risk of Environmental Dissemination to Wildlife Water, Dust?



E. coli O157 Survey of Rodents





Species	Total trapped	<i>E. coli</i> O157: H7 (%)
Brush mouse (Peromyscus boylii)	6	0
California Pocket mouse (Chaetodipus californicus)	10	0
Deer mouse (Peromyscus maniculatus)	777	1 (0.1)
California Ground squirrel (Otospermophilus beecheyi)	22	0
Harvest mouse (Reithrodontomys megalotis)	12	0
House mouse (Mus musculus)	36	0
Kangaroo rat (Dipodomys californicus)	47	1 (2.1)
California Vole (Microtus californicus)	11	0
Norwegian rat (Rattus norvegicus)	2	0
California Parasitic mouse (Peromyscus californicus)	87	0
wood rat (Neotoma fuscipes)	34	0
Unknown	13	0
Total	1057	2 (0.2)

Survey of Amphibians and Reptiles





- There is epidemiological and experimental evidence that captive amphibians and reptiles can shed *Escherichia coli* O157:H7 and *Salmonella* and cause human illnesses and outbreaks
- The relative significance of coldblooded vertebrate species in the contamination of fresh produce or waterways in their natural habitat is unclear

Methods

- Confidential enrollment of growers, ranchers, conservation properties in CA and GA
- Wildlife agency permits and IACUC approval to catch and release common species in CA and GA
- Multiple samples per animal as appropriate (cloacal swab, ventral swab, "PBS" water bath)
- Standard culture methods for *Salmonella* and STEC
- Extensive characterization of associated waterbodies including temperature, pH, turbidity, DO, and chemical analysis

Passive trapping







Trapping – Nets, Seines, Nooses





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Sampling – Cloacal and Ventral Swabs





Sampling – PBS "Bath"









Water Sampling

Recovery of foodborne pathogens from amphibian and reptile samples in California and Georgia produce production regions, March – October 2011.

Source	No. Tested	Salmonella (%)	<i>E. coli</i> O157 (%)
CALIFORNIA			
Frog	331	11 (3.3)	0
Toad	20	1 (5.0)	0
Newt	5	1 (20.0)	0
Salamander	6	0	0
Snake	39	23 (60.0)	0
Lizard	59	7 (12.0)	0
GEORGIA			
Frog	42	0	0
Toad	40	15 (37.5)	0
Salamander/siren	10	0	0
Turtle	125	23 (18.4)	0

Recovery of foodborne pathogens from water samples in California and Georgia produce production regions, March – October 2011.

Source	No. Tested	Salmonella (%)	<i>E. coli</i> O157 (%)	Non-O157 STEC (%)
CALIFORNIA				
Pre-irrigation reservoir	19	0	0	1 (5.3)
Non-irrigation*	99	16 (16.1)	1 (1.0)	6 (6.0)
Subtotal	118	16 (13.6)	1 (0.8)	7 (5.9)
GEORGIA				
Pre-irrigation pond	26	10 (38.5)	0	n/d

*Tailwater (sediment) ponds; natural ponds, greenhouse, grassed ditch, Salinas river, wetland/slough

Conclusions

•*E. coli* O157:H7 incidence in central coast (Monterey, San Benito, San Luis Obispo) cattle and avian/mammal wildlife species was low, but may vary by location and season. *Salmonella*, but not *E. coli* O157 were detected in wild-caught amphibians and reptiles.

•Some common wildlife species not on the Leafy Green Marketing Agreement "species of significant concern" list were positive (cowbird, crow, coyote, deer mouse)

•The findings emphasize the need to continue to follow food safety practices, but also balancing environmental stewardship goals.

Mitigation? Potential Conflicts with Environmental and Water Quality Goals

- Removal of sediment basins, waterbodies, near produce fields
- "Frog" fencing
- Bare ground buffers; between wetland/slough areas and produce fields; removal of vegetation strips



CA/AZ Leafy Green Marketing Agreement (LGMA) Wild and Domestic Animals

Environmental Assessments

- Pre-season and pre-harvest assessments are required to make sure conditions that can affect food safety, such as animal intrusions, flooding, proximity to animal feeding operations, etc. are not present, or have been properly mitigated.
- It is recommended that producers check for local, state, and federal laws and regulations that protect riparian habitat, restrict removal of vegetation or habitat, or restrict construction of wildlife deterrent fences in riparian areas or wildlife corridors.

Animals of significant risk: cattle, sheep, goats, pigs (domestic and wild), deer

Produce Safety Rule Domesticated and Wild Animals

- (Proposed §§ 112.83 and 112.112) If under the circumstances there is a reasonable probability that animal intrusion will contaminate covered produce, you would be required to monitor for evidence of animal intrusion immediately prior to harvest and, as needed, during the growing season.
- If you see evidence of animal intrusion, such as significant quantities of animals, animal excreta, or crop destruction via grazing, you must evaluate whether the covered produce can be safely harvested. For example, if you see evidence of bird excreta on a head of lettuce, you would not be allowed to harvest it.

Wildlife Management

<u>Lethal</u>:

- Legal SportHunting
- Depredation
 Permit
- Baiting

Non-Lethal:

- Fencing
- Buffers
- Scare tactics (noise makers)





Co-Management

"an approach to minimize microbiological hazards associated with food production while simultaneously conserving soil, water, air, wildlife, and other natural resources."

Acknowledgments

- University of California, Davis
- Rob Atwill
- Barbara Byrne
- Alexis Fisher
- Stephanie Huang
- Xunde Li
- Yingjia Liu
- John Madigan
- Jordi Montfort
- Eduardo Vivas
- Jessica Wheeler
- USDA ARS WRRC
- Robert Mandrell
- Michael Cooley
- Diana Carychao
- Lisa Gorski

University of Georgia, Tifton George Vellidis Paige Adams Debbie Coker

Joseph W. Jones Ecological Research Center

Lora Smith

USDA Wildlife Services

Dennis Orthmeyer Shannon Chandler Jerry Wiscomb

Ecological Studies Dawn Reis

<u>Funding</u> Center for Produce Safety FDA CFSAN Georgia Fruit and Vegetable Association JV Farms USDA NIFA