Final Report for, "Source Identification, Optimized Monitoring, and Local Outreach for Reducing Animal Agricultural Inputs of Pathogens into the Sacramento-San Joaquin Delta Estuary" (04-122-555-0)

Watershed:

Sacramento / San Joaquin Delta

Project Type:

Water Quality Monitoring

Funding Source:

Funding for this project in the amount of \$899,776 was provided by the State Water Resources Control Board and came from Proposition 50, the Water Security, Clean Drinking Water, Coastal and Beach Protection Act of 2002.

Date: Oct. 2004 through June 2010

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Executive Summary

Achieving the water quality goals for the Sacramento-San Joaquin Delta Estuary (Delta) is challenging due to the complexity and large size of this watershed. Agricultural and anthropogenic processes that occur throughout the Delta exert a variety of stresses on the water quality and overall health of the ecosystem. On November 5th, 2002 the voters of California approved Proposition 50 to ensure water safety and security for future generations of Californians. In order to fulfill this promise, funding was provided through Proposition 50 for a thorough examination of the use of indicator bacteria to identify pathogens in Delta waterways as well as determining potential transport pathways for pathogens. This purpose of this project, administered through the California State Water Quality Resources Control Board (SWQRB), was to study the effects of pathogen transport and how anthropogenic processes, specifically agricultural operations, influence water quality of the Delta watershed.

Intensive sampling was conducted for two years from June 2006 through December 2008 on the Northeastern, Southeastern and Northwestern portions of the Delta. The chosen sampling locations provided a broad cross-sectional view of potential influences of agriculture on the water quality of smaller watersheds, which in turn affect larger systems. We monitored 88 sites each year located throughout the sloughs of the Delta. The sites were surveyed once a month for bacterial indicators and pathogens. We developed a watershed monitoring strategy that allows for detection of shifts in microbial water quality facilitating the detection of non-point sources of pollution such as animal agriculture.

This study addressed the bacterial indicators, *E. coli* and *Enterococcus*, and priority bacterial pathogens potentially shed by livestock populations located in the Delta: *Salmonella*, Shiga toxin-producing *E. coli* (STEC), and *Campylobacter*. Bacterial contaminants are priority pollutants of the CALFED Drinking Water Quality Program with agricultural impacts on water quality a priority issue for RWQCB5 and the CALFED Watershed Program. We also monitored 63 meteorological and physical parameters in conjunction with each water sample. A database was compiled to analyze which parameters correlated with bacterial indicators and pathogen loading.

Statistical analysis revealed that bacterial indicator exceedance was closely tied to the presence of livestock as well as marsh and wooded habitat. The Cache Slough region and areas east of the town of Locke appear to have higher levels of bacterial indicators and often exceed the single sample maximum (SSM). Cache Slough likely receives irrigated pasture runoff in summer and precipitation runoff in winter. Similarly, Locke and nearby sloughs such as Meadows appear to have multiple opportunities for human fecal inputs which may explain those higher levels, along with wooded/marsh areas that function as wildlife habitat. The elevated recreational use of the Locke Slough region during the summer months may increase the risk to human health.

We used this information along with our experience with beneficial management practices to conduct training with local land owners, Resource Conservation Districts operating in the Delta and its tributaries, (e.g., Solano, Dixon, San Joaquin, Lower Consumnes, Contra Costa), community stakeholders, county and state regulatory agencies (e.g., Regional Water Quality Control Boards, Natural Resources Conservation Service staff, non-profit watershed monitoring groups, and agricultural commodity groups with members in the Delta (e.g., California Cattlemen's Association (CCA), California Wool Growers Association). Training consisted of a series of day-long workshops ranging from Salinas to the south, Stockton to the east, Woodland to the west, and Petaluma and Redding to the north.



Figure 1. Map of sampling locations (red dots).

Table 1. GPS coordinates for all sampling sites.

Site	Site	Lat.	Long.	Site	Site Code	Lat.	Long.	Site	Site	Lat.	Long.	Site	Site	Lat.	Long.
	Code	North	West			North	West		Code	North	West		Code	North	West
1	01SYSD	38	121	16	16LOSD	38	121	31	33FMSD	38	121	46	46HNCD	38	121
		08.543	29.885			16.221	27.347			00.820	22.693			03.772	27.579
2	02SYSD	38	121	17	17MESD	38	121	32	34TUCD	38	121	47	47DISD	38	121
		08.384	27.832			15.961	30.315			00.580	23.525			03.038	27.399
3	03SYSD	38	121	18	18SNSD	38	121	33	18SNSD	37	121	48	48DISD	38	121
		08.744	26.169			16.929	30.069			59.917	24.486			02.400	28.795
4	04HOSD	38	121	19	19SNSD	38	121	34	19SNSD	37	121	49	49LCSD	38	121
		09.976	29.112			18.635	30.517			59.541	27.238			03.305	30.011
5	05HOSD	38	121	20	20LPSD	38	121	35	35TUCD	37	121	50	50LCSD	38	121
		10.155	27.870			06.150	29.492			58.585	28.506			04.159	30.059
6	06HOSD	38	121	21	21WHSD	38	121	36*	36WKSD	37	121	51	51LPSD	38	121
		10.459	26.443			05.490	29.075			57.679	27.853			04.653	30.086
7	07BESD	38	121	22	22WHSD	38	121	37*	37WKSD	37	121	52	52LPSD	38	121
		11.680	28.556			04.217	27.734			56.458	26.225			05.506	29.770
8	08BESD	38	121	23	23WHSD	38	121	38	38EMCD	37	121	53	53POSD	38	121
_		12.133	27.618			05.005	26.202			58.257	29.580			04.606	30.554
9	09BESD	38	121	24	24BICD	38	121	39	39LASD	37	121	54	54POSD	38	121
		12.254	26.054			04.825	24.915			58.583	30.769			05.308	31.202
10	10SNSD	38	121	25	25BICD	38	121	40	40LASD	37	121	55	55POSD	38	121
		14.046	30.026	~ ~		03.869	25.092			59.731	30.361	*		05.233	33.765
11	11LKSD	38	121	26	26BICD	38	121	41*	41HOCD	38	121	56*	56GESD	38	121
10	421465	15.076	30.135		2051465	02.810	25.168	40*	120000	00.431	34.790		FREED	07.787	35.041
12	12LKSD	38	121	27	29FMSD	38	121	42*	42COSD	38	121	57	5751SD	38	121
10	121.000	15.794	29.919	20	2051465	02.522	25.579	40	420000	00.548	33.180	50	FOCTOD	11.025	39.007
13	131020	38	121	28	30FIVISD	38	121	43	43CUSD	38	121	58	282120	38	121
14	141.000	16.115	29.615	20	2451465	02.775	24.173		441460	00.259	31.//1	50	FOCTOD	13.303	36.159
14	14LUSD	38 16 194	121	29	3TEINI2D	38 01 FF0	121	44	44LASD	38	121	23	292120	38 15 265	121
15		20.184	28.700	20	2751450	20	23.950	45	AFCUCD	00.988	31.097	60	GOSTOD	15.205	35.904
15	TOINID	3ð 15 900		30	32510130	30	121	45	450000		121	60	002120	38	
		12.899	27.584			00.905	23.110			01.554	30.251			18.206	34.054

SWRCB Agreement Number 04-122-555-0

Proposition 50/CALFED Drinking Water Project

Site	Site	Lat.	Long.	Site	Site Code	Lat.	Long.	Site	Site	Lat.	Long.	Site	Site	Lat.	Long.
	Code	North	West			North	West		Code	North	West		Code	North	West
61	61SUSD	38	121	71	71PRSD	38	121	81	81CACD	38	121	91 ^{**}	91CACD	38	121
		19.793	34.810			16.048	40.207			15.629	46.621			15.528	48.209
62	62SUSD	38	121	72	72BDCD	38	121	82	82BASD	38	121	92**	92BASD	38	121
		18.376	35.898			17.560	39.631			15.688	46.581			15.990	47.990
63	63SUSD	38	121	73	73BDCD	38	121	83	83BASD	38	121	93	93LKSD	38	121
		16.541	36.098			20.462	38.903			16.358	47.709			15.794	29.919
64	64ELSD	38	121	74	74LICD	38	121	84	84CASD	38	121				
		22.486	32.809			17.994	40.027			17.473	44.004				
65	65ELSD	38	121	75	75LCED	38	121	85	85CASD	38	121				
		21.253	33.934			20.145	39.792			18.006	45.290				
66	66ELSD	38	121	76	76LCWD	38	121	86	86CSCD	38	121				
		20.036	35.034			20.168	40.339			17.455	46.328				
67	67MISD	38	121	77	77SHSD	38	121	87	87HASD	38	121				
		17.565	37.906			19.201	41.569			17.672	43.560				
68	68MISD	38	121	78	78SHSD	38	121	88	88HASD	38	121				
		14.725	39.725			16.390	41.652			18.471	43.997				
69	69MISD	38	121	79	79LISD	38	121	89 ^{**}	89BDCD	38	121				
		14.102	39.950			14.798	42.112			21.305	38.522				
70	70PRSD	38	121	80	80LISD	38	121	90**	90SHSD	38	121				
		14.392	40.913			15.435	43.764			17.777	41.557				

*Sites relocated/renamed after sampling year one

**New location/name for sites discontinued after year one

Problem Statement & Relevant Issues

The Sacramento-San Joaquin Delta Estuary (Delta) is the largest estuarine system in the Western North American Continent. (Norgaard et al. 2009) The Delta is an inland delta at the culmination point of three major rivers in Northern California, the Mokelumne, Sacramento and San Joaquin. The water that flows into the Delta is responsible for serving two thirds of California's population with freshwater. Water quality goals have been exasperated by many challenges that still exist today due to the vastness of this watershed. Agricultural and anthropogenic processes have deleterious effects on water quality and overall health of the ecosystem throughout the Delta. Undesirable water quality conditions stress native populations of fish and wildlife, an ecosystem already strained by the demand for fresh water and the extensive levee system. Failure of the levee system could affect the health and wellbeing of hundreds of thousands of humans as well. There are publically funded steps for improving this most important resource in California, and ensuring that this ecosystem is safe for future generations.

Microbial contamination of California's Delta continues to impact the many beneficial uses of these waters. A particular concern addressed by this project is the risk of pathogen contamination of the Delta via irrigated and non-irrigated runoff from animal agriculture. It is well established that infected livestock can shed a wide variety of pathogenic bacteria and protozoa in their manure and many of these pathogens can be transmitted to humans, other domestic animals, and wildlife through the water. Excessive amounts of pathogenic microorganisms in municipal water supplies not only increases the risk of waterborne infectious disease in humans through consumption of contaminated drinking water, but also jeopardizes the public's health during activities such as swimming, bathing, and eating fresh fruits and vegetables irrigated with contaminated water.

Long-term reduction of pathogen pollution in the Delta from animal agriculture requires an integrated approach to be successful. Success is achieved by using adaptive management strategies to ensure that this multifaceted system is well balanced. In order to address the many facets of pathogen pollution in the Delta, an integrated approach would need to combine source identification, on-farm beneficial management practices (BMPs), agricultural community outreach and training, and monitoring protocols that can detect trends in recovery or degradation of microbial water quality.

Extensive numbers of dairy cattle, beef cattle, and sheep graze throughout the Delta (RWQCB 5), often in close proximity to the complex network of tidal sloughs and tributaries that make up this portion of the estuary. Primary routes of waterborne contamination from animal agriculture tend to occur by two main processes: (1) during the winter rainstorm season when pasture runoff is contaminated with livestock fecal material and (2) during summer from a combination of direct fecal deposition by grazing livestock and from contaminated runoff from either irrigated pastures or row crops fertilized with animal manure. Examples of the potential for high microbial loading of surface water from agricultural runoff are the high concentrations of fecal coliforms (>10,000 cfu/100 ml) we observe in winter runoff from California dairy operations (Lewis 2001), high concentrations of the protozoal parasite, *Cryptosporidium parvum*, in runoff from grazed rangeland (Tate 2000), and the frequent isolation of *Salmonella* in agricultural canals in the San Joaquin Valley (Barnett 2001).

This project, with the help of interested parties, was meant to develop an integrated approach for water quality management of the Delta. Specifically, we monitored 88 sites throughout the sloughs of the Delta which were surveyed once a month for bacterial indicators and pathogens. We conducted intensive sampling over a two year period to develop a watershed monitoring strategy that allows for detection of changes to microbial water quality. Additionally, we developed beneficial management practices for reducing environmental loading of *Cryptosporidium* in the Delta. Further, we examined whether valid correlations exist between bacterial indicators and the presence or absence of bacterial pathogens. We also conducted training workshops to extend all of this information and methodology to such entities as local land owners, county and state regulatory agencies, NRCS staff, and agricultural commodity groups active in the Delta.

Project Goals Met

Our goals for this project were to develop monitoring protocols, conduct outreach, and identify agricultural inputs of bacterial indicators and enteric pathogens for the sloughs and tributaries of the Delta. The long-term success of reducing impairments to beneficial uses of the Bay-Delta rely in part on reducing local agricultural inputs of bacterial indicators and pathogens to the network of sloughs and cuts of the Delta. We developed management recommendations for land owners on how to minimize animal infection with waterborne pathogens such as *Cryptosporidium parvum* and developed management recommendations on how to reduce the discharge of pathogens from livestock production systems into California water supplies and extended this information on BMPs to land owners and regulatory agencies through workshops (Task 2.6). We determine that commonly used bacterial indicators such as *Enterococcus* and *E*.

coli would not function as reliable predictors of bacterial pathogens such as *Salmonella*, *Campylobacter*, and *E. coli* O157 for the waters of the Sacramento/San Joaquin Delta. We ascertained if there were correlations between bacterial indicators and pathogens such as *Salmonella*, *Campylobacter*, and *E. coli* O157 during different hydrological seasons of the Delta.

CALFED Drinking Water Quality Program

"The CALFED Program's Water Quality Program is the continuous improvement of Delta water quality for all uses and to advance efforts to provide safe, reliable and affordable drinking water to millions of Californians who rely on waters from the Delta watershed through cost-effective continuous improvement of source water, water management and treatment. To that end, the Water Quality Program invests in projects to improve water quality from source to tap to benefit more than 25 million Californians who obtain at least some of their water from the Delta." <u>http://calwater.ca.gov/calfed/objectives/Water_Quality.html</u>

Activities Completed

During the first year of this project we intensively monitored 88 sites in order to find areas in the Delta with higher levels of bacterial indicators, i.e. E. coli and Enterococcus, and occurrences of the priority bacterial pathogens Salmonella, Shiga toxin-producing E. coli (STEC) and Campylobacter. Based on year one data, 5 sites were relocated to areas of especially high bacterial counts and given new names. Thus the number of sites was consistent from year to year (88) but a total of 93 sites were sampled during the two-year period. Bacterial contaminants are priority pollutants of the CALFED Drinking Water Quality Program, with agricultural impacts on water quality a priority issue for RWQCB Region 5 and the CALFED Watershed Program. We assessed how land uses surrounding and within the Delta might impact microbial water quality and the potential correlation between agricultural processes and overall ecosystem health. The optimized monitoring system we employed at each site, including land use surveys during our project allowed us to address the primary goal of the CALFED Drinking Water Quality Program to reduce microbial loading into the sloughs and rivers of the Delta. Knowledge of how management practices affect drinking water constituents of concern (DWCC) from agricultural systems is paramount in understanding how to best optimize monitoring strategies. We enhanced, through a series of workshops, the ability, capacity, and coordination of local communities, conservation organizations, county and state regulatory agencies, and agricultural commodity organizations to more effectively monitor water quality and develop on-farm management practices that reduce agricultural impacts on microbial water quality of the Delta. The monitoring and workshops that took place during this project are consistent with the CALFED drinking water priority of providing an intergraded strategy for the protection and mitigation of deleterious constituents.

Project Assessment and Evaluation Plan (PAEP)

Goal 1

We characterized potential agricultural sources of bacterial indicators and bacterial pathogens that discharge into the sloughs and local tributaries of the Delta.

Success: We monitored 93 sites for these sources over a two year period and correlated bacterial levels with nearby land uses to achieve this goal.

Benefits: These results will help regulators or stakeholders' best target intervention and remediation efforts and help prioritize local sites for installation of beneficial management practices.

Shortcomings: Sites should have been monitored more frequently to better match regulatory methodology, i.e. 5 samples in 30 days. Using the geometric mean of a consecutive sampling pattern was too difficult to achieve so we resolved to sample our 93 sites with the single sample grab method which suited the projects scope better.

Goal 2

We developed an improved watershed monitoring strategy for these hydrologically dynamic systems of the Delta.

Success: We successfully monitored our 93 sites over a two year period using well established *in situ* techniques coupled with data at nearby California Irrigation Management Information System (CIMIS) sites provided by the California Department of Water Resources.

Benefits: An optimized sampling strategy will allow regulatory agencies and watershed monitoring groups to better detect the recovery or degradation of microbial water quality for sloughs draining into the Delta.

Shortcomings: Although our project was well orchestrated, the two years that covered our project were relatively dry. More seasonal variation would have given us better insight into how our sampling sites changed with lower temperatures, more precipitation, and lower ultraviolet light.

Goal 3

We developed beneficial management practices that reduce environmental loading of fecallyderived pathogens such as *Cryptosporidium parvum* from livestock herds in the Delta and elsewhere.

Success: We partnered with the Natural Resources Conservation Service, USDA, for this effort and created a document of considerable length to address waterborne pathogens of animal origin in agricultural watersheds.

Benefits: The detailed literature review and methods utilized in this technical document will enable stakeholders, including state, federal, and agricultural organizations better assess regulatory and operational needs for the betterment of the Delta estuary.

Shortcomings: The manual does not provide succinct recommendations for the agricultural and governmental communities. Better approaches are being developed to bridge this gap.

Goal 4

To develop guidance material for RWQCB and SWRCB regarding the efficacy of using bacterial indicators to denote the presence or absence of specific pathogenic bacteria.

Success: Using statistical analyses we have concluded that elevated counts of indicator *E. coli* and *Enterococcus* do not reliably predict the presence of *Salmonella, Campylobacter,* nor Shigatoxin producing *E. coli*. However we did find seasonal trends in the occurrence of Shiga-toxin producing *E. coli* that appear related to exceedance of the *Enterococcus* single sample maximum (61 CFU/100ml). These results were compiled in a guidance document (Deliverable 2.7.2, Attached DVD) along with suggestions for regulatory managers.

Benefits: The information contained in the guidance document should prove helpful to regulators when determining monitoring and/or management strategies for the California Delta.

Shortcomings: Statistical analyses fell short of determining causal relationships between the occurrence of indicators bacteria, pathogenic bacteria, and environmental conditions. At this time we cannot support the use of indicator bacteria as the sole measure of the presence of pathogenic bacteria, nor can we offer an alternative standard.

Goal 5

Through as series of workshops we enhanced the ability, capacity, and coordination of local communities, conservation organizations, county and state regulatory agencies, and agricultural commodity organizations to more effectively monitor water quality and develop on-farm management practices that reduce agricultural impacts on microbial water quality of the Delta.

Success: We held six "Rangeland Water Quality" workshops across the State of California including Browns Valley, Yuba Co. (1-14-09), Red Bluff (1-16-09), Woodland (1-24-09), Stockton (1-29-09), Paso Robles (2-18-09), Salinas (3-18-09), and Petaluma (4-29-2010). The workshops were organized in cooperation with other CALFED funded projects. A total of approximately 560 people attended the workshops. Attendees included ranchers, growers, policy makers, regulators, and scientists.

Benefits: Sixty-eight percent of attendees strongly agreed that information was delivered well; with the rest of attendees (32%) agreeing that the information met their expectations. 100% agreed or strongly agreed that they would use the information and 100% of participants agreed that they would apply the information presented in the next 12 months.

Shortcomings: It would have been useful to have a panel discussion between project members and various stakeholders as part of the outreach workshops.

Project Tasks

Planning, Research, Monitoring and Assessment

Task 1.0 Developed a Quality Assurance Project Plan and Monitoring Plan

Task 2.1 Developed a PAEP

Task 2.2 Develop an Advisory Committee of Affected Parties (ACAP)

Task 2.3 Identified 93 monitoring sites and installed monitoring protocols

Task 2.4 Conducted monitoring

Task 2.6 Developed Beneficial Herd Management Practices for Protozoal Pollution and fecal contamination

Task 2.9 Draft and Final Project Reports

Education, Outreach, and Capacity-Building

Task 2.5 Developed a Pathogen Monitoring Protocol for the Sacramento-San Joaquin Delta Estuary

Task 2.7 Developed Regulatory Guidance for Bacterial Indicators

Task 2.8 Conducted Local Outreach, Training and Workshops

Deliverables Table

		% of Work	Date
Work Item	Items for Review	Complete	Submitted
EXHIBIT A	1.1 QAPP	100%	5/23/2005
	1.2 MP	100%	6/2/2006
	2.1 PAEP	100%	5/17/2006
	2.2.1 List of ACAP members	100%	5/16/2008
	2.2.3. ACAP meeting documentation	100%	5/16/2008
	2.3.2 Install monitoring equipment and/or	100%	6/2/2006
	access at eighty (80) monitoring sites		
	2.3.3 Prepare summary document and	100%	6/2/2006,
	database of Global Positioning System		revised
	coordinates for each sampling point		3/5/2008 and
			5/22/2008
	2.3.4 Submit photo documentation of	100%	5/22/2008
	each sampling site		
	2.4.3 Prepare summary report on ambient	100%	6/2/2010
	monitoring		
	2.5.3 Develop standardized monitoring	100%	6/2/2010
	protocol for quantifying bacterial loads		
	2.6.1 Obtain approved landowner	100%	5/22/2008
	agreements		
	2.6.4 Prepare manual on on-farm BMPs for	100%	9/23/2009
	minimizing protozoal infection & pasture		
	runoff		
	2.7.2 Develop guidance for use of bacterial	100%	6/2/2010
	indicators as proxies for pathogens		
EXHIBIT A (cont)	2.8.1 Develop fliers and news releases to	100%	1/14/2009
	announce workshops		
	2.8.2 Develop mailing list	100%	1/14/2009
	2.8.3 Develop three (3) training modules	100%	1/14/2009
	for microbial water quality monitoring		
	2.9.1 Draft project report	100%	06/2/2010
	2.9.4 Final project report	100%	06/16/2010

		% of Work	Date
Work Item	Items for Review	Complete	Submitted
EXHIBIT B	1.1 Invoices	100%	1-07/07/06
			1 - 07/07/06
			1 – 07/07/06
			1 – 07/07/06
			1 - 07/07/06
			2 – 07/07/06
			3 - 01/11/07
			4 - 04/11/07
			5 – 02/08/07
			6 - 06/19/07
			7 – 09/12/07
			8-01/10/08
			9 - 04/15/08
			10 - 08/15/08
			11 - 11/15/08
			12 - 02/15/09
			13 - 05/07/09
			13a-05/14/10
			14 - 05/11/2010

		% of Work	Date
Work Item	Items for Review	Complete	Submitted
	6.1 Progress Reports	83%	1-2/24/06
	(20th of the month following the end of the		2 – 2/24/06
	calendar quarter)		3 – 2/24/06
			4 – 2/24/06
			5 – 2/24/06
			6-4/18/06
			7 – 7/18/06
			8 - 12/8/06
			9 – 2/20/07
			10 - 7/12/07
			11 – 7/16/07
			12 – 12/30/07
			13 – 12/31/07,
			rev 6/4/08
			14 - 6/9/2008
			rev 6/18/08
			15 –7/7/08
			16 - 11/5/08
			17 – 6/12/09
			18-6/2/10
	6.2 Expenditure/invoice projections	100%	NA
	6.3 Grant summary form	100%	2/17/2006
	6.4 Natural resource projects inventory	100%	6/18/2010
	project survey form		

Project Description

Our project was 4-years in duration starting in 2006 and continuing through Spring of 2010, with watershed monitoring, source identification, and BMP development occurring in years 1 through 3, with community outreach, workshops, and stakeholder training occurring in years 2 and 3. During year 1, we first installed an Advisory Committee of Affected Parties (ACAP) with a variety of local stakeholders (Task 2.2). Following input from our ACAP, during year 1 we identified eighty-eight monitoring sites throughout the many sloughs draining sections of the eastern Delta that were mostly dominated by agriculture (Task 2.3). This region was primarily in Sacramento and San Joaquin County, with secondary locations in Yolo and Solano Counties. Monitoring ranged from Lindsey and Cache Slough and associated tributaries that drain irrigated agriculture and beef cattle and sheep grazing operations in the northern section the Delta, locations such as Snodgrass, Hog, Beaver, and Sycamore Slough and associated tributaries which drain irrigated agriculture, beef cattle and dairy farming operations in the NE section of the Delta, to waterways such as Paradise Cut, Honker Cut and Whiskey Slough in the SE Delta which drain a variety of agriculturally-dominated lands, including dairy farming, seasonal sheep grazing, and beef cattle operations. In addition, with input from our ACAP, we enrolled beef cattle operations in order to develop BMPs that reduce environmental loading of protozoal pathogens such as Cryptosporidium from beef cattle herds in the Delta (Task 2.6). The goal for these BMPs are to reduce the high rates of environmental contamination attributed to fecal shedding of pathogens in young stock (Atwill 1998), complement our existing BMPs for beef cattle (Atwill 1999, Hoar 2000), and help form a multi-barrier approach to pathogen pollution (Atwill 2002) by combining all available information regarding livestock BMPs with information generated in our report to NRCS regarding riparian or vegetated buffer strips for filtering microbial pathogens in runoff from rangeland and irrigated pasture (Atwill 2010).

During year 2 we began the 24-month water quality monitoring effort across our network of sloughs. Water samples were collected about every month from each of our sites, with sampling occurring at different tidal stages and at base-flow and storm-flow conditions. This resulted in approximately 900 samples collected during year 1. For each sample we enumerated for the concentration of *E. coli, Enterococcus, Salmonella,* and *Campylobacter* and detected SLTEC, all of which can be shed by livestock (Atwill 1997). These bacteria were enumerated or detected using membrane filtration or modified multiple-tube methods, in combination with enrichment broths and selective culture media, with biochemical confirmation. For each sampling event, we measured flow in the slough channel using a flow meter, in addition, we characterized the water chemistry of each sample (nutrients, temperature, pH, conductivity, TSS fractionated into organic and inorganic, DO, salinity, turbidity), tidal stage, adjacent agricultural composition, antecedent 24-hour, 5-day, and annual

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precipitation, antecedent 24-hour, 5-day solar radiation, slough channel morphology, air temperature, barometric pressure, antecedent 24-hour, 5-day wind direction and speed, depth of water column and land uses at each side of the levee.

During year 3, we focused our monitoring efforts on those sloughs discharging higher amounts of bacterial indicators and pathogens, targeting seasonal runoff and irrigation return flows from specific agricultural operations. This was achieved by placing additional sampling sites (5) in the sloughs with the highest average bacterial counts; additionally, we eliminated several sites that had the lowest bacterial counts. The microbiological methods, pathogen load calculations, and water chemistry measurements were conducted as in year 2. This targeted sampling helped identify regions, land uses or agricultural sources associated with the largest loads of bacterial indicators and pathogens in the sloughs and local tributaries of the eastern portion of the Delta (Task 2.7). This data set also allowed us to identify climatic and environmental factors that may predispose animal agriculture to discharge higher bacterial loads into the Delta.

Using linear and logistic mixed-effects regression models, the 24-month longitudinal dataset of microbial water quality enables us to develop predictive models for bacterial indicators and bacterial pathogens discharging through the large network of sloughs in the eastern Delta. We have found during previous projects on waterborne pathogens that these statistical methods can be effective for developing predictive models for waterborne bacterial contamination. Serial correlation, clustering of data and heteroscedasticity errors are typical problems inherent to monitoring data, all of which can be handled with these mixed-effects models (Pinheiro and Bates 2000). These analyses will form the basis of a new pathogen monitoring protocol tailored to the eastern Delta (Task 2.7) and allow for the development of new regulatory guidance for bacterial indicators (Task 2.7). Completion of these tasks will help develop a standardized watershed monitoring strategy for these dynamic systems and will enhance the ability, capacity, and coordination of local communities, conservation organizations, county and state regulatory agencies, and agricultural commodity organizations to more effectively monitor water quality and assess the effectiveness of on-farm BMPs that reduce agricultural impacts on microbial water quality of the Delta.

In order to develop multiple barriers to pathogen pollution from animal agriculture, we developed BMPs that reduce the rate of environmental loading and therefore the potential for waterborne contamination from beef cattle in the Delta. During years 2 and 3 we conducted a longitudinal epidemiologic study on how calves might become infected and spread *Cryptosporidium throughout the environment* (Task 2.6). We have calculated previously that environmental contamination from infected calves constitutes over 99% of a herd's total load of *Cryptosporidium parvum* (Atwill 2003). Minimizing infection among young-stock will reduce this pathogen load up to 99% and thereby reduce the risk of waterborne transmission to

humans. Combining these new BMPs with those developed in other projects (BMPs for minimizing pathogens in rangeland and irrigated pasture runoff) will form an integrated, coordinated approach to minimizing microbial water quality impacts from animal agriculture. Results are to be peer-reviewed.

During years 3 and 4, with input from members of our ACAP, we used a combination of technical workshops, newsletters, and training modules to enhance the ability, capacity, and coordination of local communities, watershed groups, county and state regulatory agencies, and agricultural commodity organizations to more effectively monitor water quality and to develop effective site-specific intervention strategies that reduce agricultural impacts on microbial water quality in the Delta. For example, our workshops pooled the results and conclusions from several SWQCB-funded projects with existing knowledge regarding BMPs for water quality. Information presented included a general review of California surface water quality concerns and the major water quality contaminants of concern; reviewing priority pathogens of concern from livestock; ambient water quality conditions of generic *E. coli*, *Enterococcus, Salmonella*, STLEC, and *Campylobacter* for the eastern Delta; statistical associations between bacterial indicators and bacterial pathogens both in the Delta and in the foothills of the Sierra Nevada; using constructed wetlands to improve water quality for irrigated agriculture; and reviewing a variety of on-farm BMPs for improving water quality and minimizing microbial contamination from animal agriculture and irrigated pastures.

Outreach efforts enhanced the ability of stakeholders and affected parties to identify and prioritize which local agricultural sources are discharging the largest loads of bacterial indicators and bacterial pathogens into the eastern portion of the Delta, how to develop a watershed monitoring strategy for these hydrologically dynamic systems, a detailed manual regarding priority pathogens and intervention strategies for reducing these waterborne hazards in agricultural watersheds, and regulatory guidance regarding the efficacy of using bacterial indicators as proxies for the presence or absence of pathogenic bacteria of animal agricultural origin.

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Project Type

This project was an assessment of water quality parameters throughout the Northern and Southeastern portions of the Delta. This area encompassed approximately 940 square miles. This project supports the goal of long-term reduction of pathogen pollution in the Delta from animal agriculture by using an integrated approach that combines source identification of indicator bacteria and pathogens, on-farm beneficial management practices, agricultural community outreach and training, and developing monitoring protocols that detect recovery or degradation of microbial water quality.

Project Partners

Government	Agricultural Commodity Group	Academic
Regional Water Quality Control	California Cattlemen's	Dr. Ken Tate- University of
Board (Region 5)	Association	California, Davis
Natural Resources Conservation	California Wool Growers	Morgan Doran- UC Cooperative
Service	Association	Extension-Solano County
Department of Water Resources,	Western United Dairymen	Theresa Becchetti UC
California Irrigation		Cooperative Extension-Stanislaus
Management Information		County
System (CIMIS)		

Project Costs

With operating costs (travel and supplies) of \$350,835.00 and personnel costs of \$548,941.00, the total cost of the project was \$899,776.00. There were no matching funds used for duration of the project. Funding for this project was provided by the State Water Resources Control Board and came from Proposition 50, the Water Security, Clean Drinking Water, Coastal and Beach Protection Act of 2002.

Monitoring Practices *

*see Deliverable 2.5.3 on the Attachments DVD, for in-depth monitoring methods

Field Methodology

Sample sites were approached from a downstream direction so as not to disturb bottom sediments. An anchor was carefully lowered fore and aft in order to keep the boat in position over the sample site. Meteorological and water-quality data (dissolved oxygen, water/air temperature, wind speed, etc.) were taken prior to the collection of water samples. After removing sample bottles from the sampler housing, the rubber stoppers were sterilized with 70% ethanol and dried before next use. With multiple depth samples, care was taken not to disturb the bottom sediment and thus bias the sample.

Collection Steps

Samples were taken at the channel's deepest point, or thalweg, determined by a GPS finder. Position the boat over the sample area. Prior to anchoring turn the DO meter on to ensure proper functionality of electrode. Lower DO probe into the water column until value on the display is stable and record the values of all water-quality parameters. Place the flow meter into the water column at approximately 1 to 2 meters below the surface of the water column above the sample site. The average flow velocity should be recorded after a minute in the water column. The direction of flow should be recorded by placing a 360 degree pivoting object (string or vane) on the flow meter at the surface to determine the correct direction of flow.

Unscrew the lids and load a sterile 1L sterile Nalgene bottle into place, making sure to test plunger for headspace to allow water to flow into bottle. Lids should be in a dry sterile place facing up. Lower the Bond sampler vertically through the water column to the correct depth using the markings on the lowering rod. Once at the correct depth, pull up on the handle attached to the trigger lines to disengage plungers and hold for 10 seconds, then release trigger lines to reengage plungers. Pull the sampler out of the water vertically to avoid contamination and place on deck so that the bottles are vertical. Take the bottles out of the Bond sampler and make sure there is headspace for proper homogenization in when shaken, replace the tops. Place sample bottle upright in a cooler of ice and drain excess water from bottom to ensure there is no sinking of sample over the cap line.

Collection and Chain of Custody

Water samples need to be analyzed within 6 to 30 hours after collection. Longer times for analysis may be necessary if too numerous to count (TNTC) colonies need to be re-plated. Each sample set needs to be accompanied by a written record of time, date and environmental data reproduced in triplicate for distribution.

Table 2. Summary of Monitoring Parameters

Parameters	Method of Analysis	Type of Monitoring ^a	Frequency of Monitoring ^b
Water velocity	Global Flow meter	F	М
Antecedent 24-hour, 5- day and annual cumulative ppt	CIMIS Website Station 122	L	Μ
Temperature	YSI Temp Probe	F	М
Dissolved Oxygen (DO)	YSI DO Probe	F	М
Salinity	YSI Conductivity Probe	F	Μ
Turbidity	Lamott Nephlometer	L	Μ
Total Suspended Solids (TSS)	Standard Methods Method 2540 D	L	Μ
рН	YSI pH Probe	F	М
Electrical conductivity	YSI Conductivity Probe	F	Μ
Nitrate (N)	QuikChem Method 10-107-04-1-A	L	Μ
Ortho-phosphate (P)	Method 4500-P G	L	М
E. coli	Membrane Filter, EPA Method 1603	L	Μ
Enterococcus	Membrane Filter, EPA Method 1600	L	Μ
Salmonella	Membrane Filter, Modified MPN Procedure	L	Μ
Campylobacter	Membrane Filter, Campy Line Agar	L	Μ
Stx- <i>E. coli</i>	PCR presence/absence	L	Μ

^aType: F: Field Analysis, L: In-house Lab Analysis; ^bFrequency: M: monthly.

Laboratory Methodology

Membrane Filtration for E. coli, Enterococcus, Campylobacter

Depending on the expect number of bacteria, a set of three different volumes (1, 10, 100 ml) were filtered through 47 mm filters with a 0.45µm pore size (APHA, 2005). Membranes were then incubated for the recommended time and temperature on selective agar or broth as recommended by the manufacturer, followed by colony counts and two suspect colonies subjected to biochemical confirmation. Manifolds and magnetic filter funnels were sterilized by either autoclaving or placing them into an ultraviolet sterilizer for two minutes.

Shiga-toxin *E. coli* and *Campylobacter* Real-time PCR Procedure:

Real-time polymerase chain reaction (qrtPCR) was used to detect genes for Shiga toxin 1 and 2. The real-time multiplex polymerase chain reaction (qrtPCR) method was based on a previously described procedure by Sharma et al., 1999, with modifications. For *Campylobacter* we used the method described by Fermer and Engvall 1999, for the identification of thermophilic *Campylobacter* with modifications.

Salmonella Enumeration using an MPN Procedure

Membrane Filtration as described above for *E. coli* was used for each sample using the three volumes listed below:

a. 10 mL x 4 b. 50 mL x 4 c. 100 mL x 4

Membrane filters are placed into a 12-well plate containing 3 mL of Buffered Peptone Water. Incubate at 37° C for 16-20 hrs. Pipette 0.01 ml of each sample into 1 mL of Rappaport-Vassiliadis and incubate at 42° C for 24-48 hrs. Take 3-5 μ l of culture and streak onto Xylose Lysine Deoxycholate agar plate. Incubate at 37° C for 24 hrs. After incubation store the plates in room temperature for 2-3 days. Restreak positive colonies onto XLD plates for colony isolation and biochemical confirmation.

Public Outreach

A workshop for members of the California Cattleman's Association (CCA) was held on June 26th 2008 at their Mid-Year Annual Meeting, Joint Water and Environmental Quality Committee meeting. We reviewed the leading zoonotic pathogens that can be shed by livestock and transmitted through drinking or recreational water to humans. This is a leading threat for the beneficial uses of the water passing through the Sacramento-San Joaquin Delta Estuary. We reviewed the methods that are being used to monitor bacterial water quality and the potential impacts from animal agriculture including the numerous pumping stations that discharge surface water into the Delta from the surrounding islands and livestock being allowed direct access to Delta water. Cattle access to the banks of the Delta was highlighted as a key concern for water quality regulators and public health officials. The use of bacterial indicators for signaling the presence of microbial pathogens from animal agriculture was also reviewed, along with new summary statistics regarding E. coli exceedances from Year 1 data and the apparent lack of correlation between these exceedances and either the concentration of Salmonella or the presence of Shiga toxin-producing E. coli. Results of Cryptosporidium monitoring from three cow-calf herds were discussed and the surprising finding that none of the animals were found to be shedding *C. parvum*, which is the strain of this parasite known to be infectious for humans. Beneficial management practices were discussed regarding this waterborne parasite in the context of the conclusions from the Coarsegold RCD project (contract 04-118-555-0). It was emphasized that the water quality benefits from implementing the recommendations from this southern Sierra research site concur with previous research done at other locations in California, such as recent work at the Sierra Foothill Research and Extension Center, indicating that short grassland or rangeland buffers can dramatically reduce the likelihood that cattle contaminate surface water with pathogens infectious for humans. It was emphasized to the CCA members that these buffers appear to be effective for livestock-derived pathogens such as Cryptosporidium parvum, Giardia duodenalis, Salmonella enterica, and enterohemorrhagic E. coli like E. coli O157:H7. Throughout 2009 and into 2010 a series of workshops were held to ensure that public and private stakeholders had access to project outcomes and water quality methods being developed during this project for improving drinking water safety from irrigated agriculture and animal agriculture around rangelands and pastures. Presented below are the techniques that were shared.

Public Outreach Using a Series of Workshops: Drinking Water Safety, Range, Pasture, and Livestock Management (Training Modules Developed)

Purpose

A series of workshops were conducted throughout California in order to disseminate results of this project and related work funded by the SWRCB. These workshops focused on: 1) ambient water quality conditions for a variety of surface water sources ranging from Sierra Foothill Research & Extension Center (SFREC) study plots, rivers and creeks in Northern and Central California downstream, to the eastern region of the Sacramento/San Joaquin Delta, 2) potential risks which range and pasture management can pose to drinking water safety; 3) opportunities to modify range and pasture management to mitigate these risks; and 4) use of vegetative filter strips and wetlands to clean-up runoff from range and pastures. To provide practical management options which can be implemented to protect drinking water safety, and are key to compliance with water quality regulatory programs.

Audience

Anyone interested in the impact on surface water quality and drinking water from irrigated agriculture, livestock production, and range and pasture management, including local land owners with livestock, range, or irrigated pasture, as well as members and staff of agricultural water quality coalitions, agricultural and environmental advocacy organizations, irrigation districts, resource conservation districts, municipal water districts, water quality regulatory agencies, natural resources management and conservation organizations, and environmental consulting firms among others.

Dates and Locations of Workshops

Jan. 14, 2009 UC SFREC, Browns Valley – ½ day presentations, lunch, plus tours of field sites
Jan 16, 2009 Red Bluff Area – ½ day presentations, lunch, no tours
Jan. 24, 2009 Woodland – ½ day presentations, lunch, no tours

Jan. 29, 2009 Stockton – ½ day presentations, lunch, no tours Feb 19, 2009 Paso Robles-½ day presentations, lunch, no tours Mar 18, 2009 Salinas-½ day presentations, lunch, no tours April 29, 2010 Petaluma-¹/₂ day presentations, lunch, no tours

Presenters

- **Dr. Rob Atwill**, Professor of Environmental Animal Health and Medical Ecology, School of Veterinary Medicine, UC Davis
- **Dr. Randy Dahlgren**, Professor of Biogeochemistry, Department of Land Air and Water Resources, UC Davis
- **Dr. Toby O'Geen**, Soil Resource Specialist, Department of Land Air and Water Resources, UC Davis
- Dr. Ken Tate, Rangeland Watershed Specialist, Department of Plant Sciences, UC Davis

Agenda

Topic 1 General review of California surface water quality concerns and the major water quality contaminants of concern. Material covered, included animal-derived microbial pollutants of concern for drinking water; which pathogens cause waterborne outbreaks in the U.S; nutrient pollutants of concern for drinking water safety; background and constituents of concern (e.g., carcinogenic by-product formation from DOC/DON, nitrate and ammonia toxicity); data to indicate this is or is not a drinking water safety problem.

Topic 2 Review priority pathogens of concern from livestock and key aspects about their biology. Contrast protozoal parasites to bacterial pathogens and why which pathogens utilize a waterborne route of infection. Discuss the risks that livestock production, wildlife, range and irrigated pasture management could pose to drinking water safety and surface water quality. Provide overview of how infectious key pathogens are for humans.

Topic 3 Discuss ambient water quality conditions of generic *E. coli, Enterococcus, Salmonella,* STLEC, and *Campylobacter* for the eastern Delta and foothills of the Sierra Nevada; statistical associations between bacterial indicators and bacterial pathogens both in the Delta and various locations in the foothills of the Sierra Nevada. Explore the validity of using bacterial indicators to signal the presence of pathogens and why indicators often do not work. **Topic 4** Review and discuss the variety of on-farm BMPs for improving water quality and minimizing microbial contamination from animal agriculture and irrigated pastures. Topics reviewed include: the use of vegetative filter strips and grassed waterways to filter pollutants in runoff from rangeland and irrigated pasture; management of residual dry matter and use of rotational grazing to reduce pathogens in pasture runoff; the use of constructed wetlands to improve water quality for irrigated agriculture and tail-water discharges. Discuss various case studies as examples of how one can implement various combinations of management measures based upon site specific opportunities and constraints to create multiple barriers to water contamination (e.g., improve irrigation management to reduce runoff, rotate cattle out of pasture before irrigation, construct a wetland to capture pasture runoff, using electric fencing to create grassland buffers, linking rotational grazing to aging of fecal pats to reduce bacterial indicators in runoff from grazed pasture.

Follow-up Activities

Follow-up activities will focus on correlations between land uses and pathogen prevalence. Using data collected over the two year period we will further analyze environmental conditions to determine if they influence changes to microbial water quality around agricultural operations.

Lessons Learned

We were extremely impressed with how much data was able to be collected using a small amount of resources. Each water sample that we took carried with it 63 physical, chemical, and biological parameters. The strategy we employed was an efficient and effective method to qualitatively monitor an area of this extreme size. Monitoring should be conducted for more pathogens, such as human enteric viruses, because of the density of marinas in portions of the Delta and the occurrences of discharges in these areas. We could have moved additional sites to areas exhibiting high levels of indicator bacteria to better capture the potential relationship to pathogen prevalence. This study should be duplicated in smaller watersheds and/or at different estuaries.

Conclusion

Our goal for this project was to reduce agricultural inputs of bacterial indicators and enteric pathogens into the sloughs and local tributaries of the Delta. The long-term reduction of impairments to beneficial uses of the Bay-Delta relies on reducing local agricultural inputs of bacterial indicators and pathogens to this portion of the Delta. The following five outcomes allowed us to accomplish our goal for this project:

We characterized agricultural sources of bacterial indicators and bacterial pathogens that discharge into the sloughs and local tributaries of the Delta. We monitored 93 sites (Deliverable 2.3.2, Attached DVD) over a two year period to characterize existing water quality and determine regions of the Delta where single sample maximums (SSM) of bacterial indicators were regularly exceeded. As described in Deliverable 2.4.3 on the attached DVD, the occurrence of indicator bacteria in water samples at levels that exceed the SSM was greatest in a region heavily utilized by livestock. The presence of extensive marshland and riparian areas also appears to be associated with bacterial indicator exceedances. Given the extensive acreage dedicated to agricultural production in the Delta, it is no surprise that the majority of water samples were taken within one mile of land cultivated for food production. However, the presence of row crops and/or orchards did not appear to significantly influence the occurrence of indicator bacteria.

These results will help regulators and stakeholders target their intervention and remediation efforts and prioritize local sites for installation of beneficial management practices. For example, given the apparent relationship between exceedances and rangeland, woodland and marshes, an effective strategy might emphasize the construction of management practices in high risk areas where livestock and wildlife are likely to congregate. These measures may prove especially helpful in the Cache Slough region and areas east of the town of Locke, where water samples regularly exceeded the SSM for bacterial indicators. Cache Slough and the creeks that drain into it likely receive irrigated pasture runoff in summer and precipitation runoff in winter which may elevate bacterial indicators, while the Locke Slough area receives heavy recreational use increasing the risk to human health.

Using statistical analyses we have concluded that elevated counts of indicator *E. coli* and *Enterococcus* do not reliably predict the presence of *Salmonella, Campylobacter*, nor Shigatoxin producing *E. coli*. However we did find seasonal trends in the occurrence of Shigatoxin producing *E. coli* that appear related to exceedance of the *Enterococcus* single sample maximum (61 CFU/100ml). At this time we cannot support the use of indicator bacteria as the sole measure of the presence of pathogenic bacteria, nor can we offer an alternative standard.

We developed and improved a watershed monitoring strategy for these hydrologically dynamic systems that will allow regulatory agencies and watershed monitoring groups to better detect the recovery or degradation of microbial water quality for sloughs draining into the Delta. We developed a beneficial management practices manual (Deliverable 2.6.4, Attached DVD) for the reduction of environmental loading of pathogens derived from livestock herds. We also developed a guidance manual (Deliverable 2.7.2, Attached DVD) for RWQCB and SWRCB regarding the validity of using bacterial indicators to establish valid correlations between bacterial indicators and the concentration or presence/absence of specific pathogenic bacteria. Additionally, we enhanced the ability, capacity, and coordination of local communities, conservation organizations, county and state regulatory agencies, and agricultural commodity organizations to more effectively monitor water quality and to develop on-farm management practices (Deliverable 2.6.4, Attached DVD) that reduce agricultural impacts on microbial water quality of the Delta.

Appendices

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GRANT SUMMARY

Date filled out: 2/17/2006

Gr	ant Information: Please use complete phrases/sentences. Fields will expand as you type.				
1.	Grant Agreement Number: 04-122-555-0				
2.	 Project Title: Source identification, optimized monitoring, and local outreach for reducing animal agricultural inputs of pathogens into the Sacramento-San Joaquin Delta Estuary 				
3.	Project Purpose – Problem Being Addressed: Pathogen pollution in the Sacramento-San Joaquin Delta Estuary				
4.	Project Goals				
	a. Short-term Goals: Source identification of pathogen pollution, development of on-farm beneficial management practices, agricultural community outreach and training, and monitoring protocols that can detect trends in recovery or degradation of microbial water quality.				
	b. Long-term Goals: Reduction of pathogen pollution in the Delta from animal agriculture				
5.	Project Location: (lat/longs, watershed, etc.) Sacramento San Joaquin Delta Estuary				
	a. Physical Size of Project: (miles, acres, sq. ft., etc.) 1100 Square Miles				
	b. Counties Included in the Project: Sacramento, San Joaquin, Solano				
	c. Legislative Districts: (Assembly and Senate) Assembly: 8, 10, 15, 17 Senate: 2, 5, 14				
6.	Which SWRCB program is funding this grant? Please "X" box that applies.				
	□ Prop 13 □ Prop 40 □ Prop 50 □ EPA 319(h) □ Other				

Grant Contact: Refers to Grant Project Director.				
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Grant Time Frame: Refers to the implementation period of the grant.				
From: Oct 2004	To: June 2010			
Project Partner Information: Name all agencies/groups involved with project. University of				
California Cooperative Extension, Resource Conservation Districts, local agricultural owners				
Nutrient and Sediment Load Reduction Projection: (If applicable) N/A				

Attachments Listing For Review

Photo Documentation: (Deliverable 2.3.4) Available on DVD

Summary Report of Ambient Monitoring: (Deliverable 2.4.3) Available on DVD

Monitoring Methodology for Quantifying Bacterial Loads: (Deliverable 2.5.3) Available on DVD

Manual for on-farm BMPs to Minimize Protozoal Infection & Pasture Runoff: (Deliverable 2.6.4) Available on DVD

Guidance for Use of Bacterial Indicators as Proxies for Pathogens: (Deliverable 2.7.2) Available on DVD

Training Module for Microbial Water Quality Monitoring: (Deliverable 2.8.3) **Available on DVD**

To receive a copy of the Attachments DVD contact the author at:

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