

Appendix A

Vermillion River Watershed TMDL Workplan and Amended Workplan

Since the acceptance of these documents by the PCA, the Vermillion River Watershed TMDL was changed to the “Vermillion River Watershed Fecal Coliform Bacteria Study: A Technical Report Supporting the Regional TMDL Study.”

VERMILLION RIVER WATERSHED TMDL PROJECT WORK PLAN

April 2000

Applicant Information

Organization Name: Vermillion River Watershed Management Organization
Organization Type: Joint powers organization
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Project Information

Project Title: Vermillion River Watershed TMDL Project
Impaired Uses: Swimming due to fecal coliform bacteria
Requested Amount: \$46,842
Project Dates: May 2000 – December 2002

Project Summary:

In 1998, the Vermillion River main stem from Empire Township to Hastings was listed on the Federal Clean Water Act's 303(d) list of impaired waters for fecal coliform bacteria which impairs the river's swimming use. Also in 1998, the Vermillion River was placed on the Minnesota Pollution Control Agency's (PCA) list of waters in need of a total maximum daily load (TMDL) study for fecal coliform. Last year the PCA with the help of local agencies and citizens collected fecal coliform samples throughout the Vermillion River watershed to begin determining the extent of the bacterial problem. These data indicate that the river and its tributaries have bacteria levels in excess of the PCA's state standard of 200 organisms/100 ml of sample.

This project will include additional fecal coliform monitoring throughout the watershed to better identify highly impaired reaches and possible sources of the bacteria. Landuse and landcover GIS data will also help identify possible sources of non-point source pollution. Using GIS data and lab results, a TMDL model will be produced and various loading scenarios will be drafted. Reduction goals and an implementation strategy will be the final outcome of the project. Public participation will be solicited throughout the project in the form a citizen advisory committee, press releases, newsletter articles, web site updates and other avenues.

Background Information

Watershed History and Landuses:

The Vermillion River Watershed encompasses 372 square miles, mostly located through central Dakota County just south of the Twin Cities metropolitan area. The main stem originates in Scott County to the west and flows generally northeast to the City of Hastings. In Hastings, the river drops 90 feet before its confluence with the Mississippi River 20 miles downstream. Pre-settlement vegetation in the watershed

consisted primarily of a mixture of prairie, oak savanna and forests. In the mid 1800's settlers began to alter the landscape, tilling up prairie and draining low areas for agriculture. Current land use in the watershed is still dominated by agriculture with suburban areas and smaller urban growth centers interspersed throughout the watershed. Residential, commercial and industrial areas in upstream portions of the watershed continue to grow rapidly. Several major transportation routes bisect the watershed and may result in more land development.

There are 21 local units of government in the watershed. These entities created a joint powers organization, the Vermillion River Watershed Management Organization (WMO), as required by the Metropolitan Surface Water Management Act of 1982. The WMO's responsibilities include controlling excessive volumes and rate of runoff, improving water quality, preventing flooding and erosion, promoting groundwater recharge, and protecting and enhancing fish and wildlife habitat and water recreation.

There are six wastewater treatment plants (WWTP) in the watershed. Two of these plants discharge directly to the main stem of the Vermillion River and both are in the process of upgrading to increase capacity and volume of discharge. One WWTP discharges to the South Branch, and two more discharge directly to the Mississippi River.

Problem Statement

In 1998 the Vermillion River main stem from Empire Township to the dam in Hastings was listed on the Clean Water Act's section 303(d) list of impaired waters for fecal coliform. The river is not meeting its swimming use due to high bacteria levels.

Since the 303(d) listing, and after reviewing fecal coliform data from the "milestone monitoring site" on the Vermillion River in Empire Township, the PCA decided to take a closer look at the bacteria problem and placed the river on the 1998 Minnesota TMDL list. In 1999 the PCA, with the help of local agencies and citizens, sampled 10 sites in the watershed for fecal coliform five times a month. The results indicate that many reaches and tributaries exceed the PCA standard of a geometric monthly mean of 200 organisms/100 ml.

Additional monitoring is necessary to further define the extent of the problem, determine possible sources of bacteria, calculate an acceptable load of bacteria for the various reaches, and draft a series of reduction goals for future implementation.

Project Activities and Schedule

Quarterly reports on the project's progress will be provided to the PCA throughout the duration of the project. These reports will include a description of completed or ongoing tasks and preliminary findings as well as a budget summary detailing the amount of money spent on each task during the quarter.

Tasks and Products:

1. *Task:* Involve local citizens and stakeholders in all aspects of the project including monitoring, data analysis, TMDL development, and development of reduction scenarios and implementation strategy. Establish a local team consisting of agency staff, local citizens, and WMO representatives.

Product: Involving local citizens and landowners is crucial to the overall TMDL project. They can help to collect samples, reducing the amount of funding needed for collections, they can help determine possible sources of bacteria, and they can help draft an implementation plan that is acceptable to local landowners. The TMDL team for the Vermillion River will offer technical and local support for the project, acting as a sounding board and analyzing methods and results.

2. *Task:* Monitor 15 sites throughout the watershed for fecal coliform bacteria. Sites will be monitored 5 times per month, either May – July, August – September, or both. (See Table 1.) SWCD staff, PCA staff, and local citizens will take the samples. Samples will also be collected after major runoff events. Samples will be analyzed by a state certified laboratory. Quality assurance and quality control procedures will be followed including collecting field blanks, rinsing collection buckets with distilled water, keeping samples on ice, and analyzing samples within 24 hours of collection.

Product: Collecting additional data will greatly aid in the determination of highly contributing reaches and will help to identify possible sources of bacteria. More actual data means less reliance on modeling of the problem and sources.

3. *Task:* Conduct a pilot project studying landuse and landcover in a small area of South Creek to determine the source of elevated bacteria levels between Cedar Ave. to the west and just east of the Flagstaff Ave. to the east.

Product: This task will better prepare us for full-scale data analysis using landuse and landcover data and field surveys. Results from 1999 sampling indicate a great disparity between south Creek at Cedar Ave. and approximately 1.5 miles downstream.

4. *Task:* Collect landuse and landcover data in GIS format for areas that are not already mapped, but that indicate a bacteria problem that needs identification. (Most of the watershed is being mapped through other grants.)

Product: Landuse and landcover data will help determine possible sources of bacteria throughout the watershed and aid in setting reduction goals and implementation strategies.

5. *Task:* Analyze and interpret fecal coliform data and landuse/landcover data to identify highly contributing sub-watersheds.

Product: This task will identify highly contributing sub-watersheds and distinct possible sources of bacteria. These may include locations such as feedlots, livestock access to waterways, fields with land-spread manure, unsewered communities, failing septic systems, etc.

6. *Task:* Calculate appropriate and acceptable loadings from each sub-watershed and develop load reduction scenarios.

Product: This task will list acceptable loadings for specific reaches and sub-watersheds and reduction scenarios for each possible source and sub-watershed.

7. *Task:* Develop implementation strategy for reaching reduction goals in each reach or sub-watershed. Identify specific actions at specific locations. Incorporate a plan to pay for reduction activities including cost-share programs, possible grants, WMO funds, and other programs.

Product: This task will produce an implementation strategy and overall project report identifying specific actions for specific locations and the estimated costs associated with each strategy.

8. *Task:* Send TMDL and implementation strategy to the EPA.

Product: Completed TMDL project.

Table 1. Fecal coliform monitoring locations and schedule for 2000

Site ID	Location	May - July	Aug. – Sept.	Runoff events
VNC-170	North Fork of North Cr., 170 th St., Lakeville	X		X
VNC-175	South Fork of North Cr., S. of 170 th on Pilot Knob Rd.		X	X
VNC	Mouth of North Cr., W. of Hwy. 3, near Farmington	X		X
VMC-190	Upper reach of Middle Cr., stormwtr structures, 190 th St.			X
VMC	Mouth of Middle Cr., W. of Hwy. 3, near Farmington	X		X
VMCpk	Middle Cr., at Pilot Knob Rd., N of Hwy. 50, Farmington		X	X
V23	Main stem at Hwy. 23 (Cedar Ave.), Eureka	X		X
Vwec23	“West Eureka Cr.” at Hwy 23 (Cedar Ave.), Eureka	X		X
Veec235	“East Eureka Cr.” at 235 th St. (Hwy 78)		X	X
Vbisc*	Main stem at Biscayne Ave., 2 mi. E of Farmington	X	X	X
VSB3	South Branch, at Hwy 3, ½ mi. S of Farmington		X	X
VSBtrib	Small tributary to South Br., at Hwy 79, W of Hampton	X		X
V47*	Main stem at Hwy. 47 near Hastings	X	X	X
V54	Main stem at outlet to Miss. R., at Hwy. 54		X	X
Vsec31	Main stem at outlet of dry run, Sec. 31, 3 mi. S of Hwy 54 bridge, Ravenna		X	X
Vblack	Small creek outlet near Blackbird Tr., W of Hwy 54			X
V18	Main stem at Hwy. 18, Prairie Island, westerly main stem	X		X

*Sampled weekly by Met Council. We will collect once per month to make 5 samples/month total.

Timeline:

2000

January	February	March	April	May	June	July	August	September	October	November	December
Begin stakeholder involvement, put together TMDL team, meet to discuss workplan											

Conduct South Creek Pilot Project

Analyze fecal data and landcover to identify highly contributing sub-

2001

January	February	March	April	May	June	July	August	September	October	November	December
Continue to involve stakeholders and get feedback											
Keep team informed on progress											

Calculate acceptable loadings from each sub-watershed and develop load reduction scenarios

Develop implementation strategy for reduction goals

2002

January	February	March	April	May	June	July	August	September	October	November	December
Continue to involve stakeholders and get feedback											
Keep team informed on progress											

Continue and complete implementation plan

Submit project results and implementation plan to team and others for review / revise

Submit final project report to EDA

Project Budget

Total amount requested: \$46,842

Estimated Breakdown of Costs:

Monitoring

Weekly Sample Collection	½ hr/sample * 5 samples/month * 5 mo. * 4 sites/run * \$45/hr	\$2,250
Storm Event Collections	½ hr/sample * 17 samples * 5 storm events * \$45/hr	\$1,912
Citizen Collection Coordination	½ hr/week * 22 weeks * \$45/hr	\$495
Sample Analysis	305 samples * \$14/sample	\$4,270
Miscellaneous Supplies	Distilled water, ice, permanent markers, labels for sample bottles	\$150
	SUBTOTAL	\$9,077

Stakeholder Involvement

Meeting Facilitation	4 meetings/year * 2.75 years * 5 hr/mtg (prep. included) * \$45/hr	\$2,475
Mailings and Supplies	4 meetings/year * 3 years * \$30/meeting	\$360
	SUBTOTAL	\$2,825

Data Analysis/Modeling

South Creek Pilot Project	25 hours * \$45/hr	\$1,125
Analyze data	100 hours * \$45/hr	\$4,500
Digitize landcover for necessary areas	5 mi ² * 13 hours/mi ² * \$45/hr	\$2,925
Calculate TMDL & reduction goals	100 hours * \$45/hr (SWCD staff) 220 hours * \$79/hr (professional engineer)	\$4,500 \$17,380
Develop implementation strategy	100 hours * \$45/hr	\$4,500
	SUBTOTAL	\$34,930

TOTAL \$46,842

Responsible Parties

Although the WMO is the primary responsible party in this project, Dakota County Soil and Water Conservation District (SWCD) staff will complete the majority of the tasks. The WMO will be represented on the Project Team by one or more WMO Board members, and the WMO Board will be given timely updates on the progress of the Project and may advise accordingly.

<u>Name</u>	<u>Title</u>	<u>Responsibilities</u>
Laura Jester	Watershed Conservationist, SWCD; administrative assistant and technical advisor to WMO	Project Management Sample collection Data analysis Public outreach TMDL development Reduction goals Implementation Plan
SWCD employee – to be hired	Conservation Technician, SWCD	Sample collection Data analysis
MVTL	Minnesota Valley Testing Laboratories, Inc.	Sample analysis
Dave Holmen	GIS Technician, SWCD	Landuse/landcover mapping
Sabrina Cook	Principal Engineer, Montgomery Watson	TMDL development
Ed Matthiesen	Supervising Engineer, Montgomery Watson; WMO engineer	TMDL development Reduction goals

VERMILLION RIVER WATERSHED TMDL PROJECT WORK PLAN *AMENDMENT*

March 2002

Applicant Information

Organization Name: Dakota County Soil and Water Conservation District
Organization Type: Local Unit of Government
Project Manager: Laura Jester, Dakota County Soil and Water Conservation District
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Project Information

Project Title: Vermillion River Watershed TMDL Project
Impaired Uses: Swimming due to fecal coliform bacteria
Additional
Requested Amount: \$10,000
Requested Extension: December 2003

Updated Project Summary:

The Vermillion River Watershed TMDL Project has stayed within budget and on schedule since it began almost 2 years ago. Monitoring was completed in 2000 and, through a consulting firm, watershed flows were modeled, a literature review was conducted, the fecal coliform spreadsheet was calibrated, and load estimates were calculated. In addition, watershed citizens, local governments and the general public have been continuously updated on the progress of the TMDL through a variety of media avenues and meetings. The PCA has been updated on the TMDL's progress through a series of quarterly reports.

Last year the University of Minnesota chose the Vermillion River watershed for a pilot project associating DNA fingerprinting with sources of fecal coliform pollution. Last summer (2001) University staff and Dakota County SWCD staff began collecting fecal coliform samples once a month at ten sites within the watershed. Collections will resume again during the field season in 2002. Final results of this work will be available in December 2002.

We feel that the DNA data is very important to the fecal coliform TMDL we are completing for this watershed and would like to include the data in the final loadings model and reduction scenarios. As the Vermillion River TMDL is scheduled to be complete by December of 2002, we are seeking a one year extension to the TMDL project and additional funds to incorporate the DNA data.

Additional Project Activities and Schedule

Quarterly reports on the project’s progress will be provided to the PCA throughout the duration and extension of the project. These reports will include a description of completed or ongoing tasks and preliminary findings as well as a budget summary detailing the amount of money spent on each task during the quarter.

Additional Tasks and Products:

- 9. *Task:* Involve local citizens and stakeholders in new aspects of the project related to the DNA fingerprinting study.

Product: Involving local citizens and landowners is crucial to the overall TMDL project.

- 10. *Task:* Monitor 10 sites throughout the watershed for fecal coliform bacteria concentrations and DNA strains associated with that bacteria. Sites will be monitored 1 time per month, April – October in 2001 and 2002. Samples will be analyzed by a state certified laboratory.

Product: Collecting DNA data will greatly aid in the determination of possible (probable?) sources. More actual data means less reliance on modeling of the problem and sources.

- 11. *Task:* Re-calibrate spreadsheet model as necessary using new DNA data. Calculate appropriate and acceptable loadings from each sub-watershed and develop load reduction scenarios.

Product: This task will list acceptable loadings for specific reaches and sub-watersheds and reduction scenarios for each possible source and sub-watershed.

Revised Project Budget

Additional amount requested: \$10,000

Estimated Breakdown of Costs:

Monitoring

Sample Collection	Sample collection by SWCD staff can be accomplished through the previous budget	\$0
Sample Analysis	Samples will be analyzed through the U of M Study at no cost to the TMDL project.	\$0
	SUBTOTAL	\$0

Stakeholder Involvement

Meeting Facilitation	2 additional meetings * 5 hr/mtg (prep. included) * \$45/hr	\$450
Miscellaneous Supplies		\$25
	SUBTOTAL	\$475

Data Analysis/Modeling

Calculate TMDL & reduction goals	80 hours * \$45/hr (SWCD staff)	\$3,600
	75 hours * \$79/hr (professional engineer)	\$5,925
	SUBTOTAL	\$9,525

TOTAL \$10,000

Responsible Parties

<u>Name</u>	<u>Title</u>	<u>Responsibilities</u>
Laura Jester	Watershed Conservationist, SWCD; administrative assistant and technical advisor to WMO	Project Management Sample collection Data analysis Public outreach TMDL development Reduction goals Implementation Plan
SWCD employee – to be hired	Conservation Technician, SWCD	Sample collection Data analysis
LeeAnn Johnson	Senior Scientist, Microbial ecology, University of Minnesota	DNA study
MCES	Metropolitan Council Environmental Services – Analytical Laboratory	Sample analysis
Joe Bischoff	Aquatic Modeler, Montgomery Watson	TMDL development
Ed Matthiesen	Supervising Engineer, Montgomery Watson; WMO engineer	TMDL development Reduction goals

Appendix B

Raw Bacteria Data by Subwatershed and Site

Lab Codes:

- MVTL = Minnesota Valley Testing Laboratory
- MDH = Minnesota Department of Health
- MCES = Metropolitan Council Environmental Services
- U of M/MCES = Sample collected through University of Minnesota DNA Fingerprinting Study; sample analyzed at MCES
- WOMP/MCES = Sample collected through Met Council's Watershed Outlet Monitoring Program; sample analyzed at MCES
- P.I. = Prairie Island Indian Community Wastewater Treatment Plant Laboratory

Notes:

Shaded boxes indicate samples that were taken on the same day, often by different agencies at different times. Samples taken on 7/26/99 and 9/21/99 were intentional duplicates for QA/QC purposes. Please see Appendix E for full QA/QC results.

Appendix C

Subwatershed Maps

Appendix D

Summarized Results of the DNA Fingerprinting Study

(See Figure 3.1 for sampling site locations and subwatershed delineations)

Due to the limitations of the DNA study methods, the following data is only a cursory evaluation of the DNA associated with some types of animals in the Vermillion River Watershed. These data are preliminary in nature and may only hint at some possible sources of bacteria contamination. The large stakeholder and technical advisory panel minimally considered these data when they assigned relative loads to the various sources in each subwatershed.

The results shown are those of a Jackknife analysis on groupings of possible animal sources using a Pearson product-moment curve correlation coefficient to compare fingerprints of isolates in the control library to the isolates from the Vermillion samples. Of the 1,776 isolates from the Vermillion, 80% met the raw similarity and quality factor cutoff for analyses. Results are shown by sampling site in each subwatershed.

Middle Creek Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
VMC	34%	6%	18%	10%	32%	6%	94%
VMCwest	27%	18%	23%	13%	18%	18%	82%
VMCeast	36%	6%	19%	16%	23%	6%	94%

South Branch Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
VSBtrib	31%	12%	19%	12%	26%	12%	88%
VSB	44%	6%	14%	12%	24%	6%	94%

South Creek Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
No sites							

North Creek Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
VNC	30%	5%	17%	12%	24%	5%	95%
VNC175	16%	10%	25%	19%	20%	10%	90%

Eureka Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
V31	26%	10%	22%	16%	26%	10%	90%

Hastings Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
V47	31%	9%	20%	16%	23%	9%	91%

Farmington Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
No Sites							

Empire Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
No Sites							

Goodwin Subwatershed

DNA Fingerprinting Data (2000 – 2001)

Site	Livestock	Human	Pets	Waterfowl	Wildlife	Human	Non-Human
Vverm	36%	6%	18%	14%	26%	6%	94%

Appendix E

QA/QC Results and Data Quality

RESULTS FROM 1999 QA/QC PROCEDURES

July 26, 1999 Storm Event Sampling

	MVTL	MVTL-24	MDH	Blank MVTL	Blank MDH	P.I.	Sample Time	MVTL - 26th	MVTL - 27th	MDH - 27th	RPD
V9head	2,000		1,800 ^a				12:00	17:10		14:00	11%
Veureka	2,000	3,600	7,900 ^a	<10	<4		11:40	17:10	16:30	14:00	119%
Vscnb	2,600		3,100 ^a				12:15	17:10		14:00	18%
VNC+VMC	2,400		12,000 ^a				12:35	17:10		14:00	133%
VNC	3,600		8,000 ^a				12:45	17:10		14:00	76%
VMC	3,000		7,200 ^a				12:40	17:10		14:00	82%
Vbisc	3,900		7,100 ^a				13:00	17:10		14:00	58%
V79	3,000	2,400	5,000				13:30	17:10	16:30	14:00	50%
VSB	3,400		8,700				12:48	17:10		14:00	88%
VSBhead	5,600		5,200				13:05	17:10		14:00	7%
VSBtrib			67,000				13:15	17:10		14:00	
Vverm	4,000	2,700	13,000				12:27	17:10	16:30	14:00	106%
V47	6,000		13,000				12:00	17:10		14:00	74%
Vmouth			900 ^b			180	10:35			14:00	133%

Notes:

MVTL = Minnesota Valley Testing Laboratory

MDH = Minnesota Department of Health

P.I. = Prairie Island Wastewater Treatment Plant

MVTL-24 = Triplicate samples with deliberate delay in analyses beyond 24 hours

^a Samples delivered to lab with high temperature (above 15⁰C); data suggests slightly higher RPD

^b Sampler accidentally touched the inside lip of the sample bag

MVTL-26th = Samples analyzed at MVTL on July 26th

MVTL-27th = Samples analyzed at MVTL on July 27th

MDH 14:00 = Samples were analyzed between 13:50 and 15:05 on July 27th

RPD = Relative Percent Difference = Difference/Average between MVTL and MDH analyses

Typical Variation $[1/(MVTL/MDH)] * 100$ indicates that MDH was running about 277% higher in average values than MVTL. MVTL holding time is 6 hours; MDH holding time is 24 hours.

September 21, 1999 Duplicate Sampling

	VSBtrib	Vmouth
MVTL	260	40
MDH	280	14
P.I.	30	

Citizen Sampler Blanks

Veureka	VSCnb	VNC+VMC	Vbisc	VSB	VSBtrib	Vverm
7/19/99 <10	5/18/99 <10	6/9/99 <10	8/19/99 <10	6/14/99 <10	9/21 <4	7/12/99 <10
7/26/99 <10	9/21/99 <10			7/20/99 <10		8/3/99 <10
8/17/99 <10				9/14/99 <10		
9/28/99 <10						

FECAL COLIFORM BACTERIA AND DATA QUALITY (A citizens guide to understanding sampling protocol and monitoring results)

Sources of Error:

Bacteria reproduce and die off at non-linear rates. Unlike chemical pollutants, a split sample for bacteria from the same container will often yield quite different results. Since the late 1800's, research efforts have been conducted to establish a set of sampling protocols and laboratory methodologies to minimize this sample result variability. By the late 1930's, drinking water studies had suggested a protocol that bacterial water samples be delivered to the lab in "preferably less than 6 hr."¹ because variability in the lab results with longer holding times increased considerably. Subsequent laboratory research, conducted in the 1950s, helped to establish the need for preserving fecal coliform samples on ice and provided good comparisons on 6 to 24 hour and greater holding times. One study found that within 6 hours, the percentage of samples that either doubled or halved (most probable number) in counts ranged from about 10% to 19% depending upon if it was refrigerated or not. When analyzed at 24 hours, the percentage of samples showing a doubling or halving changed by about 25% for refrigerated samples to 35 % for unrefrigerated samples. It was also found that within the first 6 hours, the refrigerated fecal coliform counts had about six times as many significant decreases (halvings) as significant increases, but unrefrigerated, there were only about twice as many decreases as increases. Another study found that after 20 to 24 hours of storage, about 6 % of the samples had "significant increases" and about 14 % had "significant decreases".

Studies on chlorinated drinking water (de-chlorinated by adding sodium thiosulfate at the time of sampling) in the 1980' s reiterated the need to have samples iced and analyzed as soon as possible. Bacterial losses of up to 23% occurred within 24 hours, and declined rapidly after that. However, most of these samples were tested at very low counts relative to what is common in streams. Research using higher counts more typical of streams (several hundred to a few thousand organisms/100 ml) suggested that longer holding times could produce valid results. It has also been found that as concentrations of fecal bacteria in waters get higher, so does the number of non-coliform organisms. Samples that are over about 230 organisms/100ml may decline more rapidly in the first 24 hours than samples with relatively low counts (tens). Taking many of these research results into consideration, the following is an abbreviated list of reasons why there is so much variability in fecal coliform bacteria monitoring in streams.

Holding Times and Refrigeration:

With a 6 hour holding time, it is reasonable to expect that 10% of the samples will have at changed significantly in their counts. Although some counts will increase, statistically more will have decreased. Within 24 hours, 20% to 25 % of the samples will have changed significantly (more if left unrefrigerated).

Laboratories and Methodologies:

Another source of error can come from the variability between laboratories and methodologies. A study and literature review in 1977⁴ noted that an 80 % agreement between fecal coliform analysis

methodologies was allowable because of the lack of precision in the methods used in estimating the organisms in the laboratory. The two most common methodologies are the Membrane Filter (MF) method and the Most Probable Number (MPN) method. Translated, this means that there is an inherent 20% variability in fecal coliform counts for any individual sample tested.

Uneven Distribution of Bacteria in Water:

Another source of error comes from the inherent non-uniformity (or colonial “clumpiness”) of organisms within the sample of water itself. Bacteria grow in colonies and they do not evenly distribute themselves in a water sample even with shaking. For fecal coliform testing, a Relative Percent Difference (RPD) of 50 % can be acceptable for field splits using the same lab and the exact same lab methodology.

Field Methods:

Fecal Bacteria can survive in soils and river sediments and are generally present throughout the summer. Care must be taken not to allow sampling equipment to become contaminated. Care must also be taken in the field not to disturb the sediments so a representative water quality sample is obtained for comparison to all other water quality samples which are generally obtained by this standard non-disturbance protocol. It should be noted, however, that if samples are to be taken for swimming impairment, some disturbance of sediment in a quieter settings (backwater or lake) could be more representative of actual exposure conditions since swimmers will be disturbing sediments also.

Precipitation and Runoff:

Fecal bacteria sampling results can also change by orders of magnitude during precipitation events. Sample results will also vary depending on where in the hydrograph the samples were taken. The rising hydrograph (high flows) results may be larger (due to runoff) or smaller (due to dilution) and will likely be different from results of the falling hydrograph (still high flows). The data from this study showed a roughly 8.5% increase in the geomeans for the summer as the result of one storm event. Individual sites often showed a 20-fold increase in bacteria levels from this storm as well.

Cumulative Potential Error:

Current analytical protocol requires that samples be run within 8 hours (6 hrs. transit with two hours for lab analyses) and be held below 10°C. If you accept the premise that this is neither practical nor feasible for a citizen monitoring effort and aim at having a 24 hour analytical goal, then you theoretically (statistically) add about a 15% variability to your data if you wait the full 24 hours before the analyses is conducted. This is on top of an accepted laboratory methodology agreement variance in count estimates of 20 percent. The sampling protocol for this study was five times per month over the months of May through September (25 samples per site). By non-linear analyses of the data, even if we doubled 15 percent of the samples, then the calculated geometric mean would only vary by approximately 10 %. If we took a worst-case example and tripled the values of 15 percent of the samples, the geometric mean would only vary by about 18%. At a more realistic RPD of 50% (which is common although not necessarily desired), the geometric mean would only

vary by about 7%. Therefore, using non-linear analyses of the data (geometric means and not averages) significantly buffers the results from the effects of large spikes caused by storm event samplings.

Another factor to consider is that typical Standard Geometric Deviations (SGDs) for stream bacterial data are on the order of about 3.0 (with larger values when including data sets from major storm events). Statistically speaking, to actually say that any two samples are significantly different would require that they do not have overlapping SGDs. With a 95% confidence interval, a 25-sample data set for each site, and a SDG of 3.0, an interval of about 36 to 59 percent of the mean value can be expected. If you look at the bacterial values and deviations in terms of a less stringent 80% confidence interval, then the variability of the mean ranges would be more likely on the order of 25 to 34 percent of the geometric mean.

So what does all this mean? Individual sample results are highly variable and have a huge margin of error. Laboratory methods can add an intrinsic 20 percent variability in counts for any given sample. Holding times of up to 6 hours can add another 10 percent in overall variability. If the samples are analyzed at 24 hours, then the geometric means will have roughly 7 to 18 percent more variability than if they were analyzed within 6 hours.

So how good is our data?

Using a conservative 95% confidence interval, the data from this study indicates a typical range from -36 to +166 percent of the mean value. This suggests that a site with a geometric mean as low as 128 (below the 200 standard) could be considered as not significantly different from a site with a mean of 332 (well above the standard).

There was also a large variability in our sample splits between laboratories (which had an average relative percent difference of 76% in reported results). This is likely the result of both differences in laboratories and the inherent variability and non-uniformity of organism growth within a single volume (sample) of water. On the other hand, unlike many studies that use only one lab, this study obtained its results from three labs (MVTL, Met Council, & Prairie Island WWTF) and checked them against a fourth lab (MDH). While argued either way (creating more variability using more labs, or minimizing data skewing from any one particular lab) it can be argued that this was the best approach for shared data within this watershed. With only a limited number of sample replicates (3), it also appears that we averaged about a 33% difference in results from 6 hrs. holding time to 24 hours holding time (about double what we would expect from published literature). On the other hand, there were also two noted decreases and one noted increase which does typify the literature. In addition, the majority of the samples in this study were actually analyzed the same day, so consequently the variability is actually a much smaller percentage than the theoretical calculated errors noted above from assuming a full 24 hours before analyses. In fact, the bulk of the sampling for this study came close to the 8-hour timeframe.

It was also decided that for safety reasons, volunteers could gather their samples with a rope and bucket and then transfer the stream sample to the lab bottle. Our goal was to get a 10% number of sampler blanks (rinsate blanks) during the summer. While we only achieved about half of that, (16

samples), all of the blanks came back below detection limits, so there is no indication that there was any additional error added through cross contamination in the field.

To summarize, the citizen stream bacterial data is as good as it gets. A review of the data shows consistent relative trends between sites, relative contributions from tributaries, storm event dynamics, appropriate ranges in deviation, anticipated changes between sites, and seasonal variability. While agreement between the laboratories could have been better, the variability of this data set appears to be within the range of other fecal bacteria data sets collected in Minnesota.

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